Clinical Study Report

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Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Project code:	AIO-SUP-108
EudraCT:	2009-014725-16
Short title:	NA
Investigational substance:	Bevacizumab
Reference substance:	Placebo
Indication:	Symptomatic malignant ascites due to advanced-stage gastrointestinal cancer
Study phase:	Phase II
Inclusion of first patient:	01.06.2010
End of treatment of last patient:	25.09.2013
Date of final report:	09.04.2015

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GCP statement: This study was conducted in compliance with Good Clinical Practices (GCP) and the Declaration of Helsinki, and in accordance with applicable legal and regulatory requirements, including archiving of essential documents.

Signatures

Title of the trial: Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Trial substance: Bevacizumab

Trial code: AIO-SUP-108

The undersigned have read this clinical study report and hereby confirm that, to the best of their knowledge, it accurately describes the conduct and the results of the study.

Medical expert / LKP in accordance with §40 of the AMG (German Drug Law)

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Dr. Aysun Karatas

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Dr. Anne L. Kranich

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1 SYNOPSIS

Name of the sponsor: AIO-Studien-gGmbH	*Individual study table* *Referring to part of the dossier:*	*(For National Authority use only)*
Name of the finished product Avastin®	*Volume: N/A*	
Name of the active substances: Bevacizumab	*Page: N/A*	

Trial title: Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Study centres:
A total of 29 sites participated in this trial. Patients were included at 14 study sites. For a list of study sites, please refer to Appendix 16.1.4.

Trial duration:	**Phase of development:**
Inclusion of first patient: 01.06.2010 End of treatment of last patient: 25.09.2013	Phase II

Methodology: Double-blind, placebo-controlled, randomized, multi-centre phase II

Trial objectives:

Primary trial objective:

- To evaluate the paracentesis-free survival (ParFS) following intraperitoneal application of Bevacizumab/Placebo

Secondary objectives:

- To measure the frequency of paracenteses required for symptom control following intraperitoneal application of Bevacizumab/Placebo by assessing the longest paracentesis-free period within the 12-week main observation period (“best response”)
- To measure the volume of ascites following intraperitoneal application of Bevacizumab/Placebo
- To measure the effect of study treatment on the quality of life
- To assess feasibility and safety of intraperitoneal application of Bevacizumab including pharmacokinetic of Bevacizumab
- To evaluate the effect of an intraperitoneal application of Bevacizumab/Placebo on serum and ascites VEGF concentrations

Number of patients: 53

Included in the final evaluation:

Number of patients	**Total**
Recruited	53
Evaluable regarding toxicity	49
Evaluable regarding efficacy	49

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Diagnosis and key inclusion and exclusion criteria:

Inclusion criteria:

- Age ≥ 18 years
- Written informed consent has been obtained prior to inclusion into the study
- Patient is capable and willing to comply with the study
- Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma
- Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT, MRT or ultrasound
- Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
- Ascites clinically judged as not responsive to diuretics
- At the time of inclusion paracentesis required at least once within past 4 weeks. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase**.**
- Before inclusion of the patient into the study, a 4-week screening period allowed for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
- ECOG performance score 0-3
- Life expectancy > 12 weeks
- Laboratory parameters:
  Hematology
  - Neutrophils > 1,500/µl
  - Platelets > 100,000/µl
  - Hemoglobin ≥ 9 g/dl or 5.59 mmol/l

  Hemastaseology
  - INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

  Clinical chemistry
  - Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN
  - Serum bilirubin < 3.0 x ULN
  - Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN)

  Urine analysis:
  - Patients with < 2+ proteinuria on dipstick urinalysis.
  - Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.

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Exclusion criteria:

- Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
- Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion
- Hemorrhagic ascites (ascites hematocrit > 2%)
- ~~Transudative ascites (total protein in ascites < 30 g/l)~~
- Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion).
  Parallel treatment with Bevacizumab i.v. is not allowed.
- Therapy naïve patients
- Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
- Patients with extensive metastases of the liver making up >70% of the total liver mass
- Child C cirrhosis of the liver
- Occlusion or thrombosis of the portal vein.
- Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
- Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptysis, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage $\geq$ II.
- History of fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
- Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
- Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
- Serious non-healing wound, ulcer or bone fracture.
- Radiotherapy for purposes other than local control of symptoms.
- Evidence of bleeding diathesis or coagulopathy.
- Hematopoietic diseases.

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- Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
- History of chronic intestinal diseases associated with severe diarrhea.
- Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
- Known hypersensitivity to the test drug Bevacizumab
- Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicated the use of an investigational drug or put the patient at high risk for treatment-related complications.
- With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants was allowed as long as the INR or a PTT was within therapeutic limits (according to the medical standard in the institution) and the patient had been on a stable dose for at least two weeks at the time of randomisation.
- Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non-approved investigational drug (e.g. follow up within the trial is not exclusionary).
- Patients who have participated in this study before.
- Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[1]. [Women of childbearing potential had to have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
- Patients who were committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
- Patients who were underage or patients who were incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
- Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
- Patients who possibly were dependent on the sponsor or investigator.

[1] Methods allowed for birth control (with a failure rate of ≤ 1% per year) were implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

Treatment duration: Patients received paracentesis as needed for symptom control. In addition, patients received up to 4 intraperitoneal administrations of Bevacizumab/Placebo after paracentesis had been performed. During the 8-week treatment period, a minimum interval of 14 days was kept between applications of the study medication. End of treatment

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(EOT) was set at eight weeks after application of the first paracentesis within the treatment period for both arms of the study.

Trial medication, dose and method of administration:

Product: Bevacizumab, supplied in ready to use vials, containing 25 mg/mL

Dosing schedule: Patients received a maximum number of four cycles of intraperitoneal administrations of Bevacizumab/Placebo depending on clinical necessity of paracentesis for symptom relief. Study drugs were administered after placement of an intraperitoneal catheter and draining the largest possible volume of malignant ascites. Bevacizumab or placebo were applied through the same catheter at a total volume of 100 ml. Bevacizumab was administered at an absolute standardized dosage of 400 mg.

Evaluation criteria:
Primary parameter:
Efficacy:

- Paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurred first)

Secondary parameter:
Efficacy

- Best Response (BR) representing the longest period of time (in days) from
  - one paracentesis until next paracentesis within the treatment period
  - or, if longer, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up)
  - or, if longer, from the last paracentesis performed within the treatment period until death (before end of the standard 4 week follow-up)
  - or, if longer, from the last paracentesis performed within the treatment period until 4 week follow-up
- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites as indicated by body weight
- Quality of life as assessed by standardized questionnaires
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5.

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- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events).
- All adverse events.
- Changes in laboratory values and vital signs.
- Changes in ECOG performance status.

Pharmacokinetics of Bevacizumab and VEGF concentrations:

- Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.
- Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis was not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements were performed at bi-weekly intervals from last paracentesis until EOT and a final sample was collected at the standard follow-up 4 weeks after EOT.

Statistical methods:

Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms.
Best response was analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison.

Secondary analyses: Time to first subsequent paracentesis as well as best response was compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Additional response criteria were defined and analyzed as follows: Complete response (CR) was reached if no additional paracentesis needed to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) was reached if less than 3 additional paracenteses were performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions were compared using an exact version of the Cochran-Armitage test for trend.

Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT were calculated and were compared to volumes of the two

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most recent paracenteses before inclusion into the study, applying the same statistical test.

In addition, both groups were compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test).

Quality of life as assessed by the standardized questionnaires (FACIT-AI) was compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Body weight assessed throughout the study was analyzed in a comparable manner.

Other analyses were performed descriptively:

- Overall survival was analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status were displayed by frequency tables
- Essential laboratory values and/or vital signs were compared to baseline and displayed by shift tables.

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

Interim analysis: No interim analysis was performed.

Summary: In this unfavorable group of terminally ill patients intraperitoneal Bevacizumab was well tolerated but did not result in a significantly better symptom control of malignant ascites compared to the control arm.

Demographic data and baseline data:

53 Patients were randomized, 49 patients were evaluable (33 Bev/16 Pl arm). The majority of the patients were more than 60 years of age, with an overall median of 63 years. (mean: 61.7). 61% of the patients were male. More than half of the eligible patients included in the study suffered from pancreatic cancer (55%) followed by gastric carcinoma (20%) which was less common in the placebo group. Most of the patients, namely in the Bevacizumab arm, suffered from disseminated disease from the beginning. If UICC staging was reported, it was stage IV in almost each case. Nine out of ten patients had received at least one previous antineoplastic treatment regimen, being more frequent in the Bevacizumab group. In the remaining cases, the patient's therapy was still ongoing. There were no therapy naïve patients

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in the trial. While most of the patients in the placebo arm had an ECOG score of 2, the verum arm showed a broader distribution.

Efficacy results:

Primary endpoint: 49 patients received at least one application of the study drug and qualified for the intention to treat analysis. The median ParFS was 14d (CI: 11-17d) in the Bev arm and 10.5d (CI: 7-21d) in the Pl arm (hazard ratio 0.74, CI: 0.40-1.37; p= 0.16).

Secondary endpoints: The BR was 19d for the Bev arm (range 6-66d) and 17.5d for the Pl arm (range 4-42d) with a p value of 0.85. Median OS was 64d (CI: 45-103d) for the Bev arm and only 31.5d (CI: 20-117d) for the Pl arm (p=0.31).
An efficient downregulation of VEGF in the patients' malignant ascites was observed after intraperitoneal treatment with Bev. Furthermore, Bev treatment stabilized the expression of pro-inflammatory, and possibly tumor-promoting, cytokines / chemokines in the peripheral blood. Finally, patients with strong VEGF neutralization evidenced an improved ParFS.

Toxicity: The proportion of patients with at least one CTC grade 3-5 event occurred was similar with 20/33 (61%) in the Bev arm and 11/16 (69%) in the Pl arm.

Quality of life: While the baseline FACIT-Al form is available for the majority of patients, the relative numbers drop quickly less than half during the course of the study treatment period (in parallel to the death events). Thus, the remaining data definitely reflect a positive selection. Secondly, this dropout mechanism leads to a loss of power hypothesis testing. Between baseline and week 2, with corresponding pairs of data available in a total of 28 patients, there is no significant difference regarding the FACIT Ascites Summary Index. When comparing baseline data and week 4 data, with corresponding pairs of data available in a total of 16 patients, there is no significant difference between the two groups.

Analysing the DGHO palliative questionnaire, the number of available forms was distinctly reduced during the course of treatment, comparable to the assessment of FACIT-AI. All symptoms (except for weakness) were only rarely reported and of mild severity or not even present at all. While antiemetics were applied quite frequently (41% at week 0), only few patients received antidepressants, corticosteroids, or laxatives. Analgesic treatment including opioids were applied in 27% at study therapy onset. Several patients were recorded to receive additional palliative procedures.

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4 ABBREVIATIONS

β-HCG	Beta human chorionic gonadotropin
AE	Adverse event
AIO	Arbeitsgemeinschaft Internistische Onkologie
ALT (SGPT)	Alanine aminotransferase
AMG	Arzneimittelgesetz (German Drug Law)
ANC	Absolute neutrophil count
aPPT	Activated partial thromboplastin time
AST (SGOT)	Aspartate aminotransferase
BR	Best Response
CHF	Congestive heart failure
CHO	Chinese hamster ovary
CNS	Central nervous system
CR	Complete response
CRF	Case report form
CT	Computed tomography
CTCAE	Common terminology criteria for adverse events
DGHO	Deutsche Gesellschaft für Hämatologie/Onkologie
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDTA	Ethylenediaminetetraacetic acid
ELISA	Enzyme-linked Immuno-absorbant Assay
EOT	End of treatment
ESF	Eligibility screening form
FACIT-AI	Functional Assessment of Chronic Illness Therapy - Ascites Index
GCP-V	GCP-Verordnung
h	Hour
IC	Informed consent
ICH	International Conference on Harmonization
IMP	Investigational medicinal product
INR	International normalized ratio
ITT	Intent to treat
iv	intravenous
LD	Longest diameter
LDH	Lactate dehydrogenase
LKP	Leiter der klinischen Prüfung (Co-ordinating Investigator)
m2	Square meter (body surface area)
mg	Milligram
min	Minute
mL	Milliliter
MRI	Magnetic resonance imaging
NB	Nota bene (please note)
NCI	National Cancer Institute
NCT	National Center for Tumor Diseases
NSAIDS	Non-steroidal anti-inflammatory drugs
NYHA	New York Heart Association
PVS	Peritovenous shunting
PD	Progressive disease

PR	Partial response
RECIST	Response Evaluation Criteria in Solid Tumors
rHuMAb	Recombinant humanized monoclonal antibody
rpm	Rounds per minute
SAE	Serious adverse event
SD	Stable disease
SmPC	Summary of product characteristics
UICC	International Union Against Cancer
ULN	Upper limit of normal
VEGF	Vascular Endothelial Growth Factor
VPF	Vascular Permeability Factor
w/wo	With or without

5 ETHICS AND AUTHORITIES

5.1 Independent Ethics Committee

Before recruitment of patients, the Clinical Study Protocol and other appropriate documents were submitted to the Independent Ethics Committees (IECs) responsible for the participating investigators. The ICEs/IRBs did not raise principal objections against the study, but requested some changes to the protocol version 1.0 and to the Informed consent form. The study documents were revised, and all changes requested were taken into consideration. The revised documents were re-submitted to the IECs/IRBs and final approval was obtained. The protocol was later on amended and submitted to the IEC. Changes were requested to the amended protocol by the IEC, which were taken into consideration and the amended protocol was also approved by the IEC. A detailed description of amendments is given in Section 9.8.

Final approval of the IECs/IRBs was obtained for the following documents:

Document	Date of approval by IEC
Protocol version 2.0 (07.12.2009)	14.01.2010
Protocol version 3.0 (07.06.2010)	26.07.2010
Protocol version 4.0 (18.04.2012)	13.06.2012
Administrative Change to Protocol version 4.0 (15.10.2012)	08.11.2012
Patient informed Consent Version 1.4 (08.12.2009)	14.01.2010
Patient informed Consent Version 1.5 (07.06.2010)	26.07.2010
Patient informed Consent Short Version 1.0 (18.04.2012)	13.06.2012
Patient informed Consent Version 1.6 (15.10.2012)	08.11.2012
Patient informed Consent Version PK samples (02.12.2009)	14.01.2010

A list of the IECs/IRBs involved is presented in Appendix 16.1.3.

5.2 Ethical conduct of the study

The study was conducted in conformity with the locally legally valid requirements, the German Drug Law (AMG 1976 and amendments), the principles for the proper conduct of clinical trials for medical products (Federal Gazette no. 243, dated 30/12/1987), the ICH Harmonised Tripartite Guideline for Good Clinical Practice (1996), and the “Ethical principles for medical research involving human subjects” of the 18th World Medical Association General Assembly in Helsinki ((1964), and amended by the 29th, 35th, 41st, 48th, and 52nd World Medical Association General Assemblies (Tokyo 1975, Venice 1983, Hong Kong 1989, Somerset West 1996, and Edinburgh 2000)), the Note of Clarification on Paragraph 29 added by the World Medical Association General Assembly, Washington 2002, and the Note of Clarification on Paragraph 30 added by the World Medical Association General Assembly, Tokyo 2004 – as applicable in the respective countries.

The study was submitted to the Paul-Ehrlich-Institut on 11 December 2009 and approved on 22 December 2009. The applicable local regulatory authorities were informed about the participation of the sites in the conduct of the study according to 67 Abs. 1 AMG.

In conformity with ICH guidelines and in accordance with §40 AMG (section 1, clause 8) and §3 AMG, patients participating in the study were covered by an insurance policy that was taken out by the sponsor or the contract research organisations (CROs) on behalf of the sponsor. The insurance was taken out with HDI Gerling Industrie Versicherung AG, Niederlassung Dortmund, Märkische Straße 23-33, 44141 Dortmund, Germany, Tel.: +49 231 5481-492, Fax.: +49 231 54481 302, policy number: 48158388 03055 390

5.3 Patient information and informed consent

Before being enrolled in the clinical trial, each patient was informed that participation in the trial was voluntary and that he/she could withdraw from the study at any time without giving any reasons and without having to fear any detrimental effects on his/her medical care.

The patient was informed about the study medication and the possible side effects. At the same time, the purpose, significance, and scope of the study were explained to him/her. The explanation also included informing the patient about the insurance protection and the obligations of the insured.

The patient had sufficient time and opportunity to clarify any unresolved questions. Furthermore, the patient was given a copy of the Patient Information Form, containing all the important information in written form (in the local language) and a copy of the signed informed consent. A sample patient information/informed consent form is included in Appendix 16.1.3.

The patient’s consent had to be obtained in writing before the start of the study. By signing the informed consent form, the patient declared that he/she was participating voluntarily and intended to follow the study protocol instructions and the instructions of the investigator, and to answer the questions asked during the course of the trial. The investigator kept the signed patient informed consent form in the designated place in the investigator’s file.

By giving consent, the patient also agreed to the storage of his/her medical data in the context of the trial and its forwarding to third parties in pseudonymised form for checking by the sponsor. He/she also consented to the forwarding of his/her personal data for review by the supervisory authorities or to persons authorised by the sponsor to check the proper conduct of the clinical trial.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

The trial received Ethics approval for 30 sites, 29 sites were initiated and 11 sites were already deregistered during the study. 53 patients were recruited in 14 active sites.

The number of patients who were enrolled at each study site is shown in Table 1. The list of study sites is attached in Appendix 16.1.4.

Table 1: Study sites and recruitment

Centre	Location	No. of patients recruited	Start of treatment	Last contact
01	Halle/Saale	6	15.06.2011	08.07.2014
02	Hamburg	7	29.06.2010	20.05.2014
03	Berlin	2	14.01.2011	10.06.2014
05	Hildesheim	3	31.05.2011	24.06.2014
07	Frankfurt	7	18.11.2010	15.05.2014
10	Hamburg	6	31.08.2010	21.07.2014
12	Wendlingen	1	10.02.2011	21.07.2014
13	Wetzlar	7	20.07.2010	14.05.2014
19	Leverkusen	2	17.03.2011	06.08.2014
23	Berlin	3	01.06.2010	21.05.2014
24	Essen	4	09.08.2010	25.08.2014
25	Essen	1	19.06.2012	11.06.2014
26	Deggendorf	3	19.07.2010	11.06.2014
27	Berlin	1	18.07.2013	05.09.2014
	total	**53**		

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7 INTRODUCTION

Growth of tumors in serous cavities such as the peritoneum is often accompanied by the accumulation of a protein-rich exudates and formation of malignant effusions is a common problem for patients with advanced-stage cancer. Malignant ascites is defined as an abnormal accumulation of fluid in the peritoneal cavity as a consequence of cancer occurring in association with a wide variety of neoplasms such as colorectal, stomach, pancreatic, ovarian, breast, and lung cancer [1].

Accumulation of massive amounts of malignant ascites is a significant cause of morbidity and mortality in patients with intra-abdominal tumors [2]. Accordingly, mean survival is only 20 weeks after ascites has been discovered [3]. By increasing abdominal pressure malignant ascites causes severe symptoms [2]. Palliation of the symptomatic patient is the foremost goal and elimination of fluid accumulation in a patient with these symptoms will certainly improve the patient's quality of life and may even prolong survival [4]. Unfortunately, no single method has been developed that works satisfactorily for the majority of patients and, accordingly, effective management of malignant ascites has been a frustrating problem for many physicians and their patients [4]. Currently, treatment modalities commonly applied for patients with malignant ascites include diuresis, salt restriction, paracentesis, and peritoneo-venous shunts, however, evidence for each of these treatment options is weak, and there are no randomized controlled trials evaluating their safety and efficacy. In addition, results achieved by the application of these conventional methods are variable, only occasionally provide prolonged relief from symptoms, and do not improve survival [5]. Accordingly, there is no generally accepted and evidence-based guideline for the management of malignant ascites [6].

Paracentesis has been the most common treatment option offering the advantage of a quick, simple, and relatively low-risk procedure with immediate symptom relief. However, as the patient's disease progresses, the frequency of hospital visits for the procedure increase as well. In addition, repeated paracenteses subject the patient to risks such as bleeding, infection, visceral perforation, and hypotension associated with invasive fluid depletion, and renal impairment [1]. Finally, the procedure itself is painful, inconvenient, and, most importantly, only temporarily effective [4].

Recently, the tri-specific antibody catumaxomab has been introduced to the treatment of malignant ascites [7, 8]. It is thought to stimulate the T cellular immune system as well as to induce MHC-unrestricted cytotoxicity and phagocytosis of tumor cells [9, 10]. Cohorts of patients suffering of ovarian cancer and gastric cancer have experienced symptom relieve after catumaxomab application and, more recently, a phase II/III trial has assessed catumaxomab in the treatment of malignant ascites, with significant results regarding to puncture-free survival and quality of life [11].

In conclusion, catumaxomab might represent a new approach for the therapy of malignant effusions. However, the needs for placement of an intraperitoneal catheter for several days limit its potential use in patients with end-stage cancer who require palliative treatment [8]. In addition, significant grade III-IV hepatic toxicity has been when catumaxomab was applied intravenously or intraperitoneal, sometimes even associated with an impaired hepatic function [8, 12]. Therefore, catumaxomab seems to be better suited for patients with a relatively good performance state [12].
In conclusion, effective palliation of malignant ascites remains a difficult management issue. As patients are expected to survive only for a very limited period of time, a desirable treatment should (1) effectively alleviate associated symptoms, (2) be minimally invasive, (3) allow for rapid discharge from the hospital, (4) be relatively simple with low associated risk of complications, and (5) be of tolerable cost to the patient and his/her family [13].

VEGF and malignant ascites

Tumor-induced angiogenesis has been suggested to contribute to the development of ascites [14, 15]. In 1983, Senger et al. isolated vascular permeability factor (VPF) from ascites of tumor-bearing animals and hypothesized that this factor secreted by tumor cells in a paracrine fashion was responsible for the cancer-related fluid accumulations [16, 17]. A few years later, vascular endothelial growth factor (VEGF) was discovered as a potent stimulator of angiogenesis and was subsequently recognized to be identical to VPF [18, 19]. In addition, peritoneal mesothelial cells [20], monocytes/macrophages infiltrating malignant effusions [21], and even tumor-infiltrating T cells [22] are capable of producing VEGF and VEGF acts on endothelium both normal and newly induced by tumor angiogenesis [23]. Angiogenesis, the development of new blood vessels from pre-existing vasculature, is an essential component of solid tumor growth and metastasis [24, 25, 26, 27]. However, in addition to its ability to promote angiogenesis, VEGF is also capable of markedly augmenting the permeability of pre-existing microvasculature [16, 23, 28].
VEGF is over-expressed in a variety of tumors causing malignant ascites [29, 30, 31] and intra-tumoral VEGF expression correlates with an increased metastatic potential [31, 32] and poorer survival rates, among others, in gastrointestinal tumors, ovarian, breast, and lung cancer [30, 31, 33, 34, 35, 36, 37, 38].
Initial studies had already indicated that the accumulation of malignant ascites results in large parts from an increased permeability of peritoneal lining vessels [39, 40]. It was shown that VEGF protein accumulated in the leaky blood vessels that line the peritoneal cavities of mice bearing ascites tumors [23, 41, 42] and that in low nanomolar or picomolar concentrations VEGF increased the permeability of venules and small veins for plasma proteins with a potency 10.000 times higher than histamine [16]. Furthermore, the expression level of VEGF by cancer cells has been shown

to directly correlate with the tumor cell-induced production of ascites in the animal model [43, 44, 45]. Finally, direct transfection of mouse peritoneum with VEGF was sufficient to cause an accumulation of ascites [46]. In contrast, transfection of tumor cell lines with VEGF antisense oligonucleotides resulted in a reduced formation of malignant effusions in the mouse [42, 47, 48]. In numerous human studies, markedly increased concentrations of VEGF have been found in malignant pleural effusions and ascites derived from patients with a large variety of solid tumors, such as ovarian cancer, gastric cancer, colorectal cancer, pancreatic cancer, breast cancer, and lung cancer. In a very recent study by Atanackovic et al. concentrations of 21 different cytokines/chemokines were simultaneously analyzed in malignant and non-malignant ascites and it was clearly shown that VEGF is the one cytokine most strongly over-expressed in ascites related to cancer [49]. Concentrations of VEGF within human malignant effusions correlate with their capability to induce vascular leakage in an experimental model, an effect that can be blocked by treatment with an antibody directed against VEGF receptor Flk-1 [50, 51]. Most importantly, concentrations of VEGF in malignant ascites have recently been shown to correlate with chemosensitivity and represent an independent predictor of progression-free and overall survival of cancer patients [51, 53, 54].

Inhibition of VEGF activity as a potential therapy for malignant effusion

It has repeatedly been shown in *in vitro* experiments that the capacity of VEGF present in the supernatant of tumor cell lines or in malignant ascites to induce vascular hyperpermeability can indeed completely be neutralized using an antibody directed against VEGF [16, 17, 41, 55, 56]. Furthermore, it was already shown in an initial animal study by Senger et al. that an anti-VEGF antibody is able to block the increased peritoneal influx associated with the intra-abdominal presence of VEGF-secreting tumor cells *in vivo*. Since then, a number of studies have clearly demonstrated that the intraperitoneal application of anti-VEGF antibodies is safe and leads to impressive and often complete remissions of the local fluid accumulations in mice following inoculation with different carcinoma or sarcoma cell lines [57, 58, 45, 59, 60]. A comparable preclinical efficacy was seen with tyrosine kinase inhibitors targeting VEGF receptors [61, 62] or with a soluble VEGF decoy receptor inhibiting VEGF [63, 64], and after intraperitoneal infusion of a VEGF antisense oligonucleotide [65], but not with conventional chemotherapy applied intraperitoneally alone [43].

Despite the very strong preclinical evidence for an obligatory role of VEGF in the formation of malignant ascites and for a possible therapeutic efficacy of anti-VEGF therapies in the setting of malignant effusions, there are currently no reports from clinical studies addressing this point in cancer patients. However, recently a number of articles reporting on small series of patients with

malignant effusions treated off-label with Bevacizumab have presented impressing results. It was first reported by Pichelmayer et al. that Bevacizumab might be active in malignant ascites [66]. Following their observation of a marked response to treatment with Bevacizumab in a patient with benign pleural effusion [67], they decided to apply a single dose of Bevacizumab intravenously at 15 mg/kg to two patients with malignant ascites due to colorectal cancer and adenocarcinoma of unknown origin, respectively. They found that both patients, in whom paracentesis was previously required at least every second week, treatment with Bevacizumab was safe and highly successful. They observed significant reductions in ascites volume resulting in a discontinuation of repeat paracentesis. Moreover, both patients had a marked decrease in their VEGF plasma levels after treatment. [54]. In agreement with these early observations, Numnum et al. reported the intravenous application of Bevacizumab (15 mg/kg every 3 weeks) to 4 heavily pretreated patients with end-stage ovarian cancer with the intent to palliate symptomatic ascites. In all 4 patients repeatedly applied paracenteses could be discontinued because of dramatically reduced levels of ascites after initiation of therapy with Bevacizumab [68].

In a very recent publication, Hamilton et al. reported on the treatment of an 88-year-old patient receiving home hospice care with refractory ovarian cancer, a very poor functional status, and severe symptomatic ascites. They performed paracentesis and treated the patient with two subsequent doses (5 mg/kg) of intraperitoneal Bevacizumab with dramatic improvement in her ascites and the quality of her final weeks of life [69]. The largest series of patients treated with intraperitoneal Bevacizumab has recently been presented by El-Shami et al. who evaluated the safety and efficacy if intraperitoneal administration of Bevacizumab (5 mg/kg every 4 weeks) to a total of 9 patients with refractory ascites due to colorectal, breast, uterine, or ovarian cancer. Impressively, malignant ascites resolved after a single intraperitoneal dose in every single patient without reaccumulation or repeat paracentesis over a median observation period of more than two months. Moreover, no grade 2-5 adverse events were observed [70].

Study Drug Bevacizumab

Bevacizumab (Avastin®) is a recombinant humanized monoclonal antibody (rHuMAb) to VEGF composed of human IgG1 framework regions and antigen-binding complementarity-determining regions from a murine antibody (A.4.6.1) that blocks the binding of human VEGF to its receptors [71] (further details can be found in the Investigator's Brochure). Bevacizumab has the capability to reduce the vascularization of tumors, thereby inhibiting the growth of the malignancy. In numerous mouse models, Bevacizumab has clearly demonstrated an inhibition of human tumor growth. The administration of Bevacizumab in hetero-transplant models of colon carcinoma has shown to result in a reduction in microvessel formation and number of metastases.

A number of properties make some agents more favorable than others for intraperitoneal therapy. One important characteristic might be that the drug has activity in the malignancy to be treated. The efficacy of Bevacizumab when used in combination with chemotherapy has been demonstrated in several prospective, randomized phase III studies [72, 73, 74, 75]. In Germany, Bevacizumab has been approved for first-line treatment of metastatic colorectal carcinoma (in combination with 5-FU/folic acid or 5-FU/folic acid/irinotecan [FOLFIRI]) in January 2005. Since January 2008, Bevacizumab has been approved for the treatment of mCRC in combination with any fluoropyrimidin-based chemotherapy.

Bevacizumab is generally well tolerated and has an acceptable toxicity profile consisting primarily of hypertension and proteinuria. Other rare but important adverse effects, however, include delayed wound healing, arterial thrombosis, and bleeding [76]. Another potentially serious adverse effect of Bevacizumab is gastrointestinal (GI) perforation and, although comparably infrequent, this potentially life-threatening complication has generated significant clinical interest. Overall, GI perforation was found to be an uncommon but well-documented side-effect of treatment in the phase III trials of Bevacizumab, as well as in subsequent surveillance trials, with a reported incidence of 1% to 2% [73, 74, 77].

Rationale

Malignant ascites represents a severe clinical problem for physicians and patients being confronted with this common symptom of advanced-stage gastrointestinal cancer. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites and therapies which are currently being used are, if anything, only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still

an urgent need for more effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers.

Preclinical data strongly suggested that Bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites treated with Bevacizumab added further support to this idea. Bevacizumab had been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. While there are, so far, no reports on GI perforation resulting from intraperitoneal application of Bevacizumab, there might still be a significant risk of such adverse reactions. However, we believe that palliative intraperitoneal treatment with Bevacizumab is still indicated in these patients with advanced-stage gastrointestinal cancer patients who are capable of providing informed consent and who often severely suffer from symptoms associated with malignant ascites.

8 STUDY OBJECTIVES

The **first** primary endpoint consisted of paracentesis-free survival (ParFS) which was calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurred first).

Baseline severity of malignant ascites was assessed by calculating the period (in days) between paracenteses that had been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study.

The **second** endpoint (Best response; BR) was calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU.

The best response value was compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase

Further evaluation of the efficacy, feasibility, and general safety of an intraperitoneal application of Bevacizumab in patients with malignant ascites.

Other measures of efficacy were:

- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites present in the patient as indicated by body weight at each study visit during the treatment period
- Quality of life as assessed by standardized questionnaires (FACIT-AI) filled out by the patient and one questionnaire by the palliative group of the DGHO, which needed to be completed by the medical staff
- Secondary analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events)
- All adverse events
- Changes in laboratory values and vital signs
- Changes in ECOG performance status

Pharmacokinetics of Bevacizumab and VEGF concentrations, correlative analysis of cytokine / chemokine levels in the peripheral blood and malignant ascites:

We have previously shown that VEGF, in addition to its central role in mediating the local accumulation of fluid in the peritoneum of patients with malignant ascites, might also contribute to the establishment of an immunosuppressive and tumor-promoting cytokine/chemokine milieu in the local tumor environment. As part of our prospective clinical study we wanted to examine whether and to what extent bevacizumab contributes to a modification of concentrations of angiogenic and immunologic factors in the immediate tumor environment. To this end, the concentrations of a number of angiogenic factors, cytokines, and chemokines in the serum as well as in the ascites fluid of the patients enrolled were to be determined.

Serum and ascites VEGF and Bevacizumab concentrations were repeatedly analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis with study medication during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis with study medication during the 8-week treatment period, as well as at the time of routine safety follow-up. Measurements were performed at bi-weekly intervals from first paracentesis until EOT and a final sample was collected at the standard follow-up 4 weeks after EOT.

9 INVESTIGATIONAL PLAN

9.1 Overall study design and plan – description

Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancer after conventional therapy have been enrolled in this trial. Patients were treated according to Clinical Study Protocol version 2.0 (07.12.2009), amended in Version 3.0 (07.06.2010) and Version 4.0 (18.04.2012). The treatment duration per patient was 8 weeks.

9.2 Discussion of study design, including the choice of control groups

This trial was a double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy. Before inclusion of the patient into the study, a 4-week screening period allowed for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria, during which at least 1 routine paracentesis for symptom control of malignant ascites must have taken place. The screening visit must not have taken place earlier than 7 days before application of the first paracentesis for study purposes.

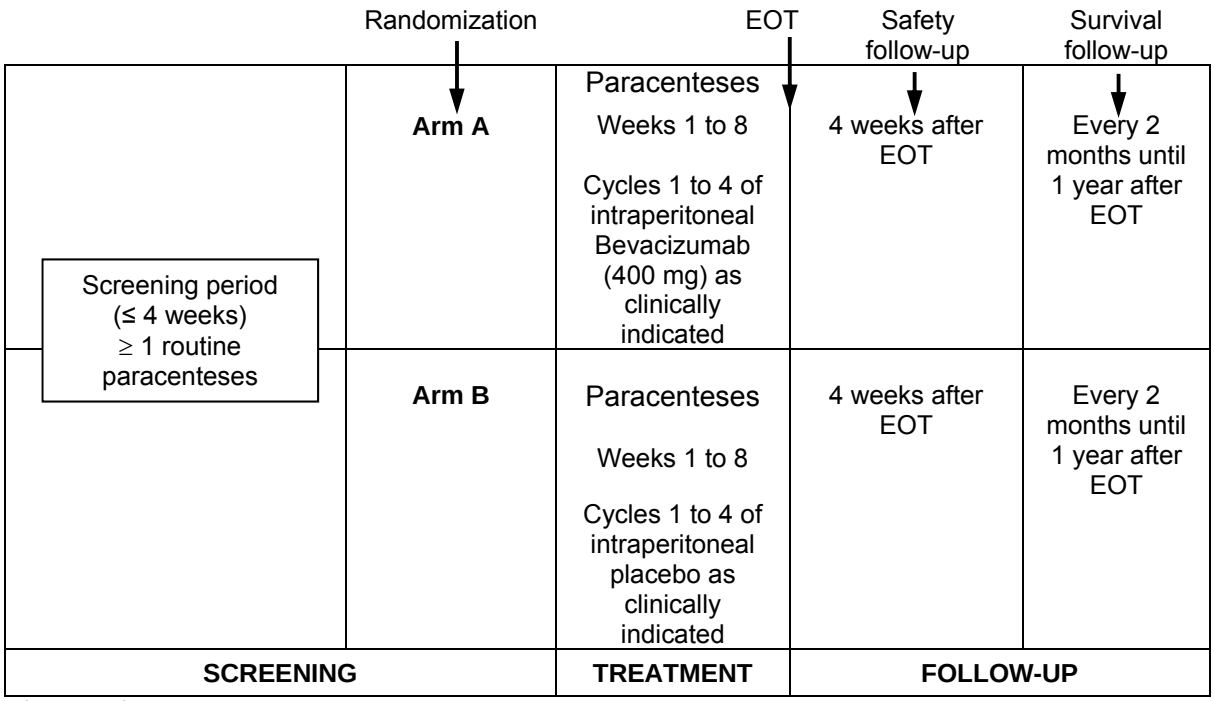
Eligible patients were randomized into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period started with the application of the first paracentesis for study purposes. Patients received up to 4 intraperitoneal administrations of Bevacizumab (400 mg absolute dose) or a placebo depending on clinical necessity of paracentesis for symptom relief. Study drugs

(Bevacizumab, arm A; placebo, arm B) were administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients received a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period was 8 weeks. Minimum interval between applications of the study drug during the treatment period was 14 days. In case of unacceptable toxicity, treatment was prematurely discontinued.

Patients were generally followed regarding response and safety at 4 weeks ("safety follow-up") following EOT. Puncture-free survival follow-up took place every two months for one year after EOT.

A brief overview on the study design is given in Figure 1.

Figure 1 Study Design



EOT= End of treatment

A total of 53 patients were enrolled into the study (37 into treatment arm A, 16 into control arm B). At the baseline visit, each patient received a unique patient number that was given to the investigator by FAX at the time of individual patient enrolment. The number assigned of each patient had to be documented by using a Patient Identification Log and on each patient's Case Report Form.

The rationale for the 2:1 allocation was that the study may gain more information about patient responses to the new intervention, such as toxicity and side effects. Additionally, if the intervention turns out to be beneficial, more study subjects would have benefit than under an equal allocation design. Moreover, from a psychological point of view, the higher chance to receive the intervention rather than placebo may render the trial participation more acceptable to the eligible patients. [78, 79]

9.3 Selection of study population

Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers were eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least once within the 4 weeks prior to treatment start. Under no circumstances were patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

9.3.1 Inclusion criteria

Patients who met all of the following criteria could be enrolled into the study:

1. Age ≥ 18 years
2. Written informed consent has been obtained prior to inclusion into the study
3. Patient is capable and willing to comply with the study
4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma
5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound
6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
7. Ascites clinically judged as not responsive to diuretics
8. At the time of inclusion paracentesis required at least once within past 4 weeks. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase.

9. Before inclusion of the patient into the study, a 4-week screening period allowed for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics were allowed during the 4-week screening period. This also excluded the application of the tri-functional antibody catumaxomab or intraperitoneal chemotherapy.
10. ECOG performance score 0-3
11. Life expectancy > 12 weeks
12. Laboratory parameters:

    Hematology

    – Neutrophils > 1,500/µl
    – Platelets > 100,000/µl
    – Hemoglobin ≥ 9 g/dl or 5.59 mmol/l

    Hemastaseology

    – INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

    Clinical chemistry

    – Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN
    – Serum bilirubin < 3.0 x ULN
    – Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN)

    Urinalysis:

    – Patients with < 2+ proteinuria on dipstick urinalysis.
    – Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.

9.3.2 Exclusion criteria

Patients who met any of the following criteria were not allowed to be enrolled into the study:

1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion

3. Hemorrhagic ascites (ascites hematocrit > 2%)
4. ~~Transudative ascites (total protein in ascites < 30 g/l)~~
5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel treatment with Bevacizumab i.v. is not allowed.
6. Therapy naïve patients
7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
8. Patients with extensive metastases of the liver making up > 70% of the total liver mass
9. Child C cirrhosis of the liver
10. Occlusion or thrombosis of the portal vein.
11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptysis, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II.
13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder)
14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic disease
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.

22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non-approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have previously participated in this study.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[2]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
32. Patients who possibly are dependent on the sponsor or investigator.

9.3.3 Removal of patients from therapy or assessment

Treatment in both arms of the study was discontinued according to the protocol if any of the following applied:

- if any exclusion criteria developed
- Any patient who developed any one of the following toxicities:

[2] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

  - Gastrointestinal perforation
  - Fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
  - Arterial thrombo-embolic events
  - Symptomatic grade 4 thrombosis
  - Grade 3/4 hemorrhagic events
  - Grade 4 hypertension (hypertensive crisis)
  - Grade 4 proteinuria (nephrotic syndrome)
- at the patient's request
- at the investigator's discretion
- Physician's judgment following an adverse event
- Termination by the Sponsor, or a regulatory authority
- Any other reason for withdrawal that the study physician or patient indicates is in the overall best interest of the patient

All patients who prematurely discontinued the treatment period were followed up for safety and survival (exception: patient withdrew consent for further participation or patient was lost to follow-up).

9.4 Treatments

9.4.1 Treatments administered

Patients received up to 4 intraperitoneal administrations of the study drugs depending on clinical necessity of paracentesis for symptom relief. Patients received a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period was 8 weeks. Minimum interval between applications of the study drug during the treatment period was 14 days.

Study drugs (Bevacizumab, arm A; placebo, arm B) were administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control.

Following the application of an 18-22 G intraperitoneal catheter, the largest possible volume of malignant ascites was drained. Thereafter, Bevacizumab or Placebo were applied through the same catheter at a total volume of 100 ml. Bevacizumab was applied at an absolute standardized dosage of 400 mg. The initial dose of the study drug was delivered over 60±15 minutes. If the first infusion was tolerated without infusion-associated adverse events (fever and/or chills) the following infusions may have been delivered over 30±10 minutes. Following the complete

application of the study drug the intraperitoneal distribution was optimized by varying the patient's body position (10 min on the back, 10 minutes on right side, 10 min on left side).

9.4.2 Identity of investigational product(s)

The trial medication was characterised as follows:

	Investigational product
INN:	Bevacizumab
Trade name:	Avastin®
Manufacturer:	Roche Registration Ltd.
Mode of administration:	Bevacizumab was supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contained sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.
Batch no.:	487-6646
Expiry date:	28.02.2014
Storage instructions:	The IMP had to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition had to be documented in a temperature-log. The vials had to be kept in the outer carton in order to protect them from light.

	Placebo
INN:	Placebo
Trade name:	NA
Manufacturer:	Roche Registration Ltd.
Mode of administration:	Placebo was supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contained sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.
Batch no.:	487-6646
Expiry date:	28.02.2014

Storage instructions:	The Placebo has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

Bevacizumab (400 mg, 25 mg/mL) and placebo have been supplied in 20 mL glass vials with a fill of 16 mL. The investigational medicinal product and placebo have been labeled according to § 5 GCP-V and internal requirements for blinding purposes. Bevacizumab infusions were prepared according to the SmPC.

9.4.3 Method of assigning patients to treatment groups

Randomization was performed stratified by center using computer-generated lists consisting of permuted blocks of randomly varying size in order to ensure equal group sizes within strata. The randomization lists were generated by WiSP GmbH and transferred to the facility responsible for blinding, labeling and packaging of the study drugs.

9.4.4 Selection of doses in the study

In the present study, Bevacizumab was administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites, Bevacizumab was administered intraperitoneally [1].

Bevacizumab was administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study

9.4.5 Selection and timing of dose for each patient

Selection of dose for each patient is described in section 9.4.4.

9.4.5.1 Dose modification for Bevacizumab

No dose reduction of Bevacizumab was foreseen for an individual patient. The dose of 400 mg Bevacizumab was proven to be a safe treatment for intravenous treatment. In addition, all studies applying Bevacizumab as an intraperitoneal infusion have used this dosage.

The initial study drug dose was delivered over 60 ± 15 minutes. If the first infusion was tolerated without infusion-associated adverse events (fever and/or chills), the second infusion may have been delivered over 30 ± 10 minutes.

Bevacizumab-specific toxicities:

Any patient who developed any one of the following toxicities should not further receive Bevacizumab. If the Bevacizumab treatment has to be discontinued permanently, the patient had to be withdrawn from the study treatment and followed up for PD and survival only.

Gastrointestinal perforation

Bevacizumab had to be permanently discontinued in patients who developed gastrointestinal perforation.

Fistula formation

Bevacizumab had to be permanently discontinued in patients who developed fistula involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder).

Thrombosis/Embolism

For patients who developed grade 3 or 4 thrombosis/embolism the following action was recommended:

- Arterial thrombo-embolic events: Bevacizumab should be permanently discontinued.
- Grade 3 or 4 venous thrombosis: Bevacizumab should be permanently discontinued.

Hemorrhage

Patients who develop grade 3 or 4 hemorrhage should have permanently discontinued Bevacizumab treatment.

Hypertension

Patients should have been monitored for the development or worsening of hypertension via frequent blood pressure measurement. Blood pressure measurements should have been taken after the patient has been in a resting position for ≥ 5 minutes. Repeat measurements of blood pressure for verification should have been undertaken if the initial reading is ≥ 140 mmHg systolic and/or ≥ 90 mmHg diastolic blood pressures.

- Grade 1 hypertension: Asymptomatic, transient (< 24 h) increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Intervention not indicated.
- Grade 2 hypertension: Recurrent or persistent (> 24 h) or symptomatic increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Monotherapy of anti-hypertensive may be indicated. Once controlled to < 150/100 mmHg, patients may continue Bevacizumab therapy.
- Grade 3 hypertension: Requiring more than one anti-hypertensive or more intensive therapy than previously. Bevacizumab should be withheld for persistent or symptomatic hypertension and should be permanently discontinued if hypertension is not controlled.
- Grade 4 hypertension: Life threatening consequence (e.g. hypertensive crisis). Occurrence of grade 4 hypertension should lead to permanent discontinuation of Bevacizumab. All doses of anti-hypertensive medicines should be recorded at all visits.

Proteinuria

All patients had a dipstick urine analysis performed within 48 h prior to each Bevacizumab dose. All proteinuria toxicity, as determined by 24 h urine collection, was graded according to CTCAE v3.0 classification. Adjustment of Bevacizumab administration for proteinuria of ≥ 2 g/24 h occurred according to the following guidelines, listed below.

First occurrence of proteinuria:

- < 2+ (dipstick): Administer Bevacizumab as scheduled; NO additional evaluation is required.
- ≥ 2+ (dipstick): Administer Bevacizumab as scheduled. Collect 24-hour urine to determine the total protein within 3 days prior to the next scheduled dose:
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24 h total protein.
    - Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

- Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g/24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

Second and subsequent occurrence of proteinuria:

- < 3+ proteinuria (dipstick): administer Bevacizumab as planned. No additional evaluation is required.
- ≥ 3+ proteinuria (dipstick): administer Bevacizumab as planned and collect 24-h urine for determination of total protein within 3 days before the next scheduled Bevacizumab administration.
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24-h total protein.
    - Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
    - Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

Nephrotic syndrome (grade 4, CTCAE v3.0): Discontinue Bevacizumab treatment.

9.4.5.2 Does Modification for Background Medication

Local standard practice and the recommendations provided in the respective SmPCs drove dose reduction or interruption of chemotherapeutic compounds of the background medication.

9.4.6 Blinding

Bevacizumab (400 mg, 25 mg/mL) and placebo were supplied in 20 mL glass vials with a fill of 16 mL. The investigational medicinal product was labeled according to § 5 GCP-V and internal requirements for blinding purposes.

Randomization was performed stratified by center using computer-generated lists consisting of permuted blocks of randomly varying size in order to ensure equal group sizes within strata. The randomization lists were generated by WiSP GmbH and transferred to the facility responsible for blinding, labeling and packaging of the study drugs.

9.4.7 Prior and concomitant therapy

The initiation or continuation of any non-protocol-specific anti-tumor therapy was forbidden from inclusion into the study until EOT. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs were not allowed from start of the screening phase until safety follow-up. This also excluded treatment with the tri-functional antibody catumaxomab or the intraperitoneal application of chemotherapy. Application of such treatments from start of the screening phase until safety follow-up led to the immediate discontinuation of the study for the given patient.

All concomitant medication(s) must have been reported in the Case Report Form (CRF). Any diagnostic, therapeutic, or surgical procedure performed during the study period should have been recorded including the dates, description of the procedure(s) and any clinical findings. Patients should have received full supportive care including transfusion of blood and products, antibiotics, etc. where applicable. The treatment details should have been recorded in the CRF.

With the only exception of full-dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants was allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Prophylactic low-dose aspirin was a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [80] and is supported by an extensive body of literature [81]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus Bevacizumab or placebo. In a retrospective exploratory analysis of patients in the Bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [82]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with Bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event was not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins was permitted before and during study, as was low-dose aspirin (≤ 325 mg/day) and clopidogrel (≤ 75 mg/day).

Note: In patients who experienced thrombo-embolic events during study treatment full dose anti-coagulant was allowed and information on anticoagulant treatment (including doses) was collected and recorded in the CRF.

INR was assessed at baseline for all patients. In patients treated with oral coumarin-derived anticoagulants, INR was checked at least before start of application of Bevacizumab or routine paracentesis, respectively.

In patients treated with full-dose oral anticoagulants due to thrombo-embolic event during study treatment, INR must have been checked at least every second day in the first week of treatment, at least 2 times/week for the following treatment weeks until a stable therapeutic level of INR had been achieved and at least once every 3rd week when the weekly dose had been established and INR was stable with this dose (see also section 9.4.5.1).

The following was recommended regarding the use of concomitant medications:

Oral contraceptives: No dose modifications were required for patients on oral contraceptives.

9.4.8 Treatment compliance

A pre-printed drug dispensing log was provided in the Investigator Site File and had to be kept current and had to identify the patient, and the amount of medication dispensed to each patient at each visit with the corresponding dates.

All medication supplies (empty containers, as well as partly used and unused medication) had to be available for inspection at every monitoring visit. All unused medication, partly-used and empty packages had to be returned by the investigator to Roche at the end of the study.

9.5 Efficacy and safety variables

9.5.1 Efficacy and safety measurements assessed and flow chart

- **Paracentesis-free survival (ParFS)** was calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurred first).
- **Volume of ascites** drained by routine paracentesis was calculated as ascites volume minus lavage volume, if applicable.
- **Total volume of ascites** present in the patient as indicated by body weight at each study visit during the treatment period.

- **Adverse events**: All patients were closely monitored for adverse events (incl. survival) from Day 1 of the first treatment cycle through Week 4 after the last treatment cycle. Thereafter, patients were followed up for progression and survival only. Adverse events were graded according to the Common Terminology Criteria for Adverse Events (CTCAE v3.0).

Table 2 Flowchart: Schedule of assessments during the Study

	Screening[1, 3]		**Baseline** [3,9]	**Treatment Period**[3]				**Safety FU**	**Survival FU**[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT**[19] **+ 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]				X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X[20]	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X[20]	
Paracentesis for symptom control	As indicated			X	As clinically indicated			X[20]	
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

1. The screening visit S1 took place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics were allowed during the 4-week screening period.
2. The treatment period started with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments had to be performed before administration of the study drug
4. The first visit of the survival follow-up period took place two months after the last infusion of the study drug. The last visit took place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.

7. Baseline frequency of paracenteses clinically required was assessed by calculating the mean time frame (in days) between paracenteses which had been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required was individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites were assessed by calculating the mean and total volumes of ascites for paracenteses that had been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed were monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements were performed at the screening visit and on each visit for routine paracentesis. Measurements were performed at biweekly intervals from first paracentesis until EOT. A final measurement was performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) was obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Sera were also collected at 14-day intervals from first paracentesis until EOT and a final sample was collected at safety follow-up.
13. Urinalysis: Dipstick test for protein only. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine was required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) were administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients received a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period was 8 weeks. Minimum interval between applications of the study drug during the treatment period were 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT was set at 8 weeks after first application of the study drug for both arms of the study.
20. When Paracentesis is clinically indicated

9.5.2 Appropriateness of measurements

The efficacy and safety tests used in this trial are routine in oncological clinical trials.

9.5.3 Primary efficacy variable(s)

The **first** primary endpoint was paracentesis-free survival (ParFS) which was calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurred first).

9.5.4 Drug concentration measurements

Serum and ascites VEGF and Bevacizumab concentrations were repeatedly analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

10 ml of blood and 10 ml of ascites fluid were collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum were obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics were obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Heparinized peripheral blood and sera were collected at 14-day intervals from first paracentesis until EOT and a final sample were collected at safety follow-up. Serum concentrations of Bevacizumab are currently analyzed by QP laboratory.

9.5.5 Correlative analysis of cytokine / chemokine levels in the peripheral blood and malignant ascites

Using Luminex technology the levels of different soluble factors in plasma and ascites samples from study patients were determined. All patients with available samples from before initiation of treatment and at least one post-treatment date were included into the analysis. If multiple post-treatment samples were available, the earliest timepoint after start of treatment was chosen. For values below the assay's detection limit half of the lower detection limit was assumed. For values above the detection limit 1.5x of the upper limit was assumed. Student's t test was used for the

comparison of mean cytokine levels and log2 transformations of fold changes. Logistic regression was used to assess the influence of VEGF neutralization on paracentesis-free survival (ParFS).

9.6 Data quality assurance

The centers had to provide site-specific laboratory normal values. The laboratories were validated with routine intra-laboratory tests. Toxicity was assessed using the evaluation criteria from NCI-CTCAE Version 3.0.

9.6.1 Requirement for Investigational sites and staff

The investigator had to be able to demonstrate (e.g. based on retrospective data) a potential for recruiting the required number of suitable subjects within the agreed recruitment period.
The investigator had to have sufficient time to properly conduct and complete the trial within the agreed trial period.
The investigator had to have available an adequate number of qualified staff and adequate facilities for the foreseen duration of the trial to conduct the trial properly and safely.
The investigator had to ensure that all persons assisting with the trial were adequately qualified, informed about the protocol, any amendments to the protocol, the trials treatments, and their trial-related duties and functions.

9.6.2 Direct access to source data/documents

The investigator/institution had to permit trial-related monitoring by the GSO mbH, as well as inspections by the appropriate regulatory authorities and Ethics committees, providing direct access to source data/documents.
The subjects were to be informed that representatives of the sponsor, independent Ethics Committee or regulatory authorities might inspect their medical records to verify the information collected, and that all personal information made available for inspection were handled in strictest confidence and in accordance with local data protection laws.

9.6.3 Investigator site file and archiving

The investigator was provided with an investigator site file (ISF) at the start of the trial. The investigator had to archive all trial data and relevant correspondence in the ISF. The ISF, all source data and all documents were to be kept filed according to the requirements of the ICHGCP guidelines after termination of the trial.
It is the responsibility of the investigator to ensure that the subject-identification sheets are stored for at least 15 years beyond the end of the clinical trial. All original subject files have to be stored

for the longest possible time permitted by the regulations at the hospital, research institute, or practice in question. If archiving can no longer be maintained at the site, the investigator will notify the sponsor.

9.6.4 Monitoring

The trial started with an initiation visit, where a monitor representing the sponsor introduced the study to the investigational site personnel.

During the trial, a monitor representing the sponsor had regular contact with the investigational site to provide information and support the investigator(s). Furthermore, the monitor checked whether the facilities remained acceptable, whether the investigational team was adhering to the protocol, whether data was being accurately recorded in the CRF and whether the study drug accountability checks were correct. The monitor conducted source data verification, which required direct access to all original records for the patient.

9.6.5 Database

Data was entered into a database by GSO Hamburg. The data was filed in digital format and two people entered the data independently. Data was checked for accuracy using range, validity and consistency checks, as well as by cross-checking. Implausible or missing data may have been corrected or completed after discussing with the investigator. The notes of amendment should have been filed together with the case report form (CRF). The validated data was stored in a database and this process shall also be documented.

This database conforms to the requirements of ICH-GCP regarding the following:

- Validation of the system and data
- Presentation of SOPs
- Access and back-up systems
- Traceability and documentation of data amendments (audit trail)

Only authorized persons may access the database. Unauthorized access was prevented via a security system.

9.6.6 Audits

Regulatory authorities might have requested access to all source documents, CRF, and other trial documentation. Direct access to these documents had to be guaranteed by the investigator who had to provide support at all times for these activities.

9.7 Statistical methods planned in the protocol and determination of sample size

9.7.1 Statistical and analytical plans

The primary endpoints of the trial were analyzed confirmatively (within the phase II framework) considering a global level for each hypothesis of $p < 0.05$ as significant.

All other parameters were evaluated in an explorative or descriptive manner, providing means, medians, ranges, standard deviations and/or confidence intervals. If additional p values were calculated (e.g. in subgroup analyses or for secondary endpoints), they are presented explicitly without referring to hypotheses or a significance level. Usually, no error adjustment for multiple testing was performed. Thus the p values reflect the comparison-wise error and not the experiment-wise error. All p values are two-sided if not stated otherwise. The statistical methods described in this section are suited for the data and distributions usually expected in this type of trials. The suitability was checked after data entry. If necessary, the statistical method was modified accordingly and sensitivity analyses performed.

Demographic and prognostic baseline data was checked for homogeneity between treatment groups. In case of relevant imbalances of other important prognostic factors the statistical method were adjusted in order to achieve best possible comparability of the groups, and the results were critically reviewed in comparison to the unadjusted ones.

Primary endpoints: ParFS over one year according to Kaplan-Meier [83], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. If the Peto logrank test [84, 85] was not appropriate because of violation of the proportional hazard assumption [86], Gehan's generalization of the Wilcoxon rank sum test for censored data [87] was applied, preferably in its modification by Peto [84] and Prentice [88]. If necessary or prospectively defined at randomization, prognostic strata were taken into account [85, 89].

Best response was analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison.

Secondary endpoints: Time to first subsequent paracentesis as well as best response were compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Additional response criteria are defined and analyzed as follows: Complete response (CR) was reached if no additional paracentesis needed to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) was reached if less than 3 additional paracenteses were performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions were compared using an exact version of the Cochran-Armitage test for trend.

Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT were calculated and were compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test.

Quality of life as assessed by the standardized questionnaires and analyzed according to the recommendations of the respective developer were compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study was analyzed in a similar way.

In addition, both groups were compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test).

Other analyses that were performed descriptively:

- Overall survival was analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status was displayed by frequency tables
- Essential laboratory values and/or vital signs were compared to baseline and displayed by shift tables.
- For the ECOG performance status, the frequencies of worsening, unchanged and improved status were displayed by frequency tables in each scheduled visit.

The methods mentioned above are likewise suitable for the univariate evaluation of prognostic factors. Multivariate analyses may have been performed by appropriate regression models (proportional hazard regression model [90], logistic regression).

Further details on the analysis are given in a separate Statistical Analysis Plan that had to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to have been decided upon at this time point.

9.7.2 Determination of sample size

Based on the results from Parsons et al. [91] the median ParFS in the untreated control group was expected to be around 14 days. In order to detect a prolongation of ParFS by 100% to a median of 28 days by Bevacizumab (hazard ratio: 0.5), a total number of 60 evaluable patients was required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation was based on the following additional assumptions:

- type I error: 5% (one-sided)
- power: 80%
- observation of all patients until the occurrence of the ParFS event; this assumption was fulfilled due to the extended follow-up period of up to one year [89]
- exponential shape of the Kaplan-Meier [83] curves

In order to allow for non-evaluable cases or drop-outs, a total of 72 patients were randomized.

The sample size calculation concerning the analysis of ParFS was based on methods described by Lachin and Foulkes [92]. Since a group sequential design allowing for interim analyses and early discontinuation was adopted for this trial (see section 9.7.3), the above fixed sample size calculations served only as an orientation for the maximum of patient numbers needed. The expected sample size and/or follow-up duration for reaching a conclusion may have been considerably less than the number given above. This depended on the number and time points of interim looks as well as the actual difference in efficacy and the actual rates of recruitment and treatment failure. In the case of only one “look” (i.e. no interim analysis before completion of recruitment) the sample size coincides with that of the fixed-sample approach given above.

9.7.3 Interim and Final Analysis

In case of longitudinal studies in severe chronic diseases, the study design should allow for interim analyses and, consequently, early stopping of the trial for ethical reasons [93]. A group sequential design was adopted, using the α error spending function methodology by Lan and DeMets [94], implementing a use function according to the O'Brien-Fleming [95] boundary guideline. The design chosen allowed drawing conclusions from interim analyses in the following respect:

- acceptance of superiority of the Bevacizumab arm (rejecting H0)

The additional option of accepting the control arm as non-inferior, when an interim result strongly suggested that the anticipated large difference of HR=0.5 is not detected, was discarded, since a smaller difference might have been discussed as relevant, too, especially if supported by secondary findings. Moreover, there was a less stringent need to stop the trial from an ethical point-of-view, if the data tended to similar results in both arms.

In order to keep an overall type I error of 5%, stopping boundaries were calculated at the respective time points of interim evaluation, using the EaSt software (Cytel Software Corp., Cambridge, USA). This allowed for arbitrary interim analyses, irrespective of time schedules and recruitment number. Moreover, the expected sample size and/or study duration for reaching a conclusion may have been considerably smaller than in a fixed sample design (cf. 8.2), especially if the therapeutic difference was even larger than expected. The extent of "saving" patients mainly depended on the actual difference in efficacy as well as the actual rates of recruitment and failures. However, subsequent interim analyses was not performed, unless an increment of at least 10 further evaluable patients were included in the database.

The final biometrical evaluation of the study and the compilation of the statistical report as part of the integrated clinical/biometrical report was performed after completion and/or correction of all case report forms.

9.8 Changes in the conduct of the study or planned analysis

Protocol version 3.0 (07.06.2010) and patient Informed consent version 1.5 (07.06.2010): Inclusion and exclusion criteria were extended to reflect the clinical situation of the patient better. Cytological confirmation of ascites was adapted by measurement of total protein in the ascites exudate and a morphological diagnosis of MRT was also possible (Inclusion No. 5). For Inclusion criterion 12 the ULN for serum creatinine, alkaline phosphatase and transaminases and protein in the urine were increased. Prior treatment with Bevacizumab for primary malignancy was not exclusionary any more (Exclusion No. 15). With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants was allowed as long as the INR or a PTT was within therapeutic limits (according to the medical standard in the institution) and the patient had been on a stable dose for at least two weeks at the time of randomisation.

Additionally the warning of anaphylactic reactions to Bevacizumab was added in protocol section 7.6 and the patient informed consent. In some clinical trials anaphylactic and anaphylactoid-type reactions were reported more frequently in patients receiving Avastin in combination with chemotherapy than with chemotherapy alone. The incidence of these reactions in some clinical trials of Avastin is common (up to 5% in Bevacizumab treated patients).

Patients may be at risk of developing infusion / hypersensitivity reaction. Close observation of the patient during and following the administration of Bevacizumab was recommended as expected for any infusion of a therapeutic humanized monoclonal antibody. If a reaction occurred, the infusion should have been discontinued and appropriate medical therapies should have been administered. A systematic premedication is not warranted.

Protocol version 4.0 (08.04.2012) and patient Informed consent version 1.6 (15.10.2012):
Inclusion criterion No. 8: 'At the time of inclusion paracentesis required at least twice within past 4 weeks' was extended that the second paracentesis may be done during the study. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase**.** Exclusion criterion No. 4 'Transudative ascites' was deleted.
Due to low recruitment rate, the recruitment period was prolonged by 18 months to December 2014. Administrative changes were included in the protocol. A short version of the patients informed consent (Version 1.0 dated 18.04.2012) for better understanding of the study was additionally provided to the patients informed consent. There were additional administrative changes to protocol V 4.0 and patients informed consent Version 1.6 (dated 15.10.2012) due to change of address of the sponsor.

The trial was prematurely stopped after enrolling 53 of 72 patients on 22.08.2013. Main reason for this decision was the slow recruitment rate caused by the fact that only about one in three pre-screened patients was finally eligible for inclusion into the trial. The high number of pre-screening failures was associated with rapid deterioration of the general condition of the terminally ill patients. To increase recruitment the number of sites was significantly increased from 20 to 29. To counteract the premature worsening of the general conditions leading to the recruitment disability, the protocol was amended on 18.04.2012 allowing to include patients with only one paracentesis during screening phase. However, this reduction of the screening phase did not result in a substantial change.
The situation described above with regard to worsening of the general conditions could already be seen in 2011 and was discussed with the investigators in the framework of an investigators meeting on 08.04.2011.
Due to the conditions mentioned above it was not feasible to randomize the planned number of 72 patients in an acceptable period of time.

Taking into account the established Drop-Out-Rate of 8.9%, 53 patients should be randomized in order to reach a statistical power of approximately 70% in the evaluation of the study and to assure

a reasonable scientific value of the study under the given conditions. After the patient 52nd was included on 18.07.2013, the investigators were informed on 19.07.2013 to finish the recruitment on 22.08.2013. One more patent was randomized on 22.07.2013, so that 53 patients were included into the study as a whole.

10 STUDY PATIENTS

10.1 Disposition of patients

10.1.1 Overview patient disposition

Between June 2010 and July 2013, a total of 53 patients were randomized into the AIO SUP-0108 study. 27 patients were treated with study medication Bevacizumab. A total of 157 had been registered to the trial office, entering the screening phase. However, only about one in three was finally eligible for inclusion into the trial. The most frequent reason for non-inclusion was a deterioration of the general condition during screening, followed by failure to gain informed consent, a too low frequency of paracentesis, a history of thrombotic events, or relevant laboratory abnormalities.

The time of data base closure for the statistical analysis is 2nd of April 2014, with only minor queries on laboratory value assessments clarified thereafter.

A CONSORT-type overview of the status of all study patients is provided in Table 3.

Table 3: Patient disposition and categories of evaluability

Category	Study arm		Total
	Bevacizumab (arm 1)	Placebo (arm 2)	
Screened patients			157
Non-participating			104
Randomised	37	16	53
Non-eligible for analysis due to severe primary violations of study selection criteria*	-	-	-
Non-eligible for analysis (no study treatment received)**	4	-	4

Full analysis set (ITT)	33	16	49
Per-protocol analysis set	33	16	49
Evaluable for baseline parameters	33	16	49
Evaluable for paracentesis therapy	33	16	49
Evaluable for efficacy			
for primary endpoint ParFS	33	16	49
for primary endpoint Best Response	33	16	49
Evaluable for patient reported outcomes	28	10	38
Evaluable for toxicity	33	16	49

10.1.2 Reasons for discontinuation

As shown in Table 4, almost two out of three patients terminated study treatment prematurely. The respective proportions in the randomisation arms are similar. In most of the cases, the reason was death of the patient.

Table 4: End of study treatment

Status / reason	Bevacizumab	Placebo	Total
n	33	16	49
Treatment phase completed regularly after 8 weeks	12 (36%)	6 (37%)	18 (37%)
Treatment phase terminated prematurely	21 (64%)	10 (63%)	31 (63%)
Reasons for early termination:			
Death	13 (39%)	8 (50%)	21 (43%)
Withdrawal of consent	1	--	1
Patient's request	2	--	2
Investigator's decision	2*	1**	3
Non-protocol ascites treatment	--	1	1
Other	3***	--	3

* Worsening of general condition / Clinical progression

** Worsening of general condition

*** Pain in legs due to open wounds / Bacterial peritonitis / Hospitalisation due to bacterial peritonitis

10.2 Protocol deviations

10.2.1 Inclusion and exclusion criteria

Several deviations of inclusion and exclusion criteria occurred: For 16 patients, routine analysis of malignant ascites was not performed according to protocol. Therefore, exclusion criteria No. 02 (neutrophil count > 250 / µl ascites) and / or No. 03 (ascites hematocrit > 2%) as well as exclusion criterion No. 04 (total protein in ascites < 30 g/l, before Amendment 02) were not assessable. One patient showed neutrophils > 250 / µl ascites. Elevated value was supposed to be tumor related and no indication of bacterial peritonitis was suspected according to PI. For one patient, alkaline phosphatase as well as ASAT and for another patient, alkaline phosphatase were evaluated at screening. For one patient, alkaline phosphatase was not measured at screening (Inclusion criterion No. 12: Alkaline phosphatase and transaminases < 3.0 x ULN). For 5 patients urine analysis was not performed at screening and therefore, inclusion criterion No. 12 was not assessable.

One patient had a portal vein thrombosis and pulmonary embolism diagnosed within 6 months before treatment start and exclusion criteria No. 10 and 22 were violated. After confirmation by the PI that events were not clinically relevant and patient's safety was not affected, the patient was eligible for the study. Three patients were included into the study with a history of thrombosis within ≤ 6 months before treatment start (Exclusion criterion No. 22).

Two patients showed a creatinine clearance < 30 mL/min at the screening assessment and inclusion criterion No. 12 was violated, but serum creatinine values were within normal range. After confirmation of the LKP patients were eligible for study.

For two patients histology of cancer was not available, but pancreatic cancer was highly probable. Even though inclusion criterion No. 4 was violated, the eligibility was confirmed by the LKP. For one patient malignity of ascites was not cytologically confirmed (inclusion criterion No. 05), but LKP confirmed eligibility of patient.

10.2.2 Randomisation, treatment allocation and blinding

One deviation of randomization procedure occurred: Start of treatment was 2 days before randomization fax was sent to GSO mbH.

10.2.3 Compliance with time windows

Four deviations of time windows occurred: One patient had only 1 paracentesis within the 4 week screening phase. This deviation occurred before Protocol Version 4.0. For another patient last

administration of study drug was after 9 instead of 8 weeks. As administration of study drug was postponed once, visits were shifted by the site. For two patients the interval between two applications of study medication was once < 14 days (13 days).

10.2.4 Treatment compliance

Two deviations of treatment compliance occurred. One patient received study drug assigned for another patient by error. Patient was allowed to remain in study by LKP and unblinding was not required. For another patient paracentesis was performed without administration of study drug as site had forgotten to order re-supply of medication.

10.2.5 Non-permitted concomitant medication

No protocol violations regarding non-permitted concomitant medication were reported.

10.2.6 Demographic and baseline characteristics

No protocol violations regarding demographic and baseline characteristics were noted.

11 EFFICACY EVALUATION

11.1 Data sets analysed

Four patients did not receive any paracentesis with study drug administration after randomisation, and, thus, were excluded from all quantitative analyses. Table 5 provides details on these cases. According to the protocol these patients are likewise excluded from the safety analysis.

The safety analysis set (SAS) included all patients who had received at least one dose of trial medication and for whom at least one post-baseline safety measurement was available. The SAS consisted of 49 patients. Thus, the safety analysis set is identical to the ITT population.

Table 5: Exclusions due to lack of receiving any study drug administration

Pat. no.	Reason for early drop-out before any study treatment
# 0206	The patient was randomized on 16.01.12. On 19.01.12 an SAE occurred (severe hyponatremia), and the patient died because of the underlying tumor disease on 24.01.12.
# 0301	The patient was randomized on 14.01.11. On the same day, a severe pulmonary infection was diagnosed, precluding any study treatment. The patient died on 07.02.11 due to recurrent aspiration pneumonia.
# 1403	The patient was randomized on 22.06.11. As the general condition of the patient deteriorated following randomization, the patient was withdrawn prior to the first administration of study drug.
# 2603	The patient was randomized on 16.02.11. Start of therapy was planned for 18.02.11, but the patient did not come back to the site for treatment.

11.2 Demographic and other baseline characteristics

11.2.1 Demographic data

53 patients were analysed regarding demographic and baseline characteristics. The median age of patients was 63 years, ranging from 35 to 81 years. 30 patients (61%) were male, 19 patients (39%) were female. Please refer to Table 6 and Table 7 for more details on demographic data. The majority of the patients were more than 60 years of age. Patients over 70 years of age were slightly more frequent in the placebo arm.

The majority of the patients (approximately two thirds) were male (Table 8). Men are somewhat less frequent in the verum group.

Table 6 Age, distribution parameters [years]

	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	60.8 ± 10.5	63.4 ± 9.7	61.7 ± 10.2
Median	62	65.5	63
Quartile	52 - 69	59 - 72.2	52 - 69
Range	35 - 81	46 - 75	35 - 81

Table 7 Age distribution (categories)

Category	Bevacizumab	Placebo	Total
n	33	16	49
< 50 years	5 (15%)	2 (12%)	7 (14%)
50 - 59 years	7 (21%)	2 (12%)	9 (18%)
60 - 69 years	14 (42%)	7 (44%)	21 (43%)
70 - 79 years	6 (18%)	5 (31%)	11 (22%)
>=80 years	1 (3%)	-	1 (2%)

Table 8 Gender

Gender	Bevacizumab	Placebo	Total
n	33	16	49
Female	15 (45%)	4 (25%)	19 (39%)
Male	18 (55%)	12 (75%)	30 (61%)

11.2.2 Disease characteristics

11.2.2.1 Cancer type and histology

More than half of the eligible patients included in the study suffered from pancreatic cancer, followed by gastric carcinoma, which is less common in the placebo group (Table 9). Almost all neoplasms were shown to have an adenocarcinoma histology (Table 10).

Table 9: Primary cancer type

	Bevacizumab	Placebo	Total
n	33	16	49
Cholangiocellular carcinoma	1 (3%)	3 (19%)	4 (8%)

Colorectal carcinoma	5 (15%)	1 (6%)	6 (12%)
Gastric carcinoma	9 (27%)	1 (6%)	10 (20%)
Hepatocellular carcinoma	1 (3%)	-	1 (2%)
Pancreatic carcinoma	17 (52%)	10 (62%)	27 (55%)
Unknown	-	1 (6%)	1 (2%)

Table 10: Histology

	Bevacizumab	**Placebo**	**Total**
n	33	16	49
Adenocarcinoma	31 (94%)	13 (81%)	44 (90%)
Other	-	1 (6%)	1 (2%)
Unknown	2 (6%)	2 (12%)	4 (8%)

11.2.2.2 History of tumor disease

As to be expected, there is a broad heterogeneity of cancer history duration, with a median of almost one year (Table 11). The estimates for the time since initial diagnosis of advanced/metastatic disease are only marginally shorter, indicating the poor prognosis of these patients already at the onset of their disease (Table 12).

Table 11: Time since initial diagnosis of cancer [months]

	Bevacizumab	**Placebo**	**Total**
n	33	16	49
Mean ± SD	12.2 ± 8.9	14.9 ± 11.1	13.1 ± 9.7
Median	9.9	12.1	10.6
Quartile	5.9 - 14.1	6.1 - 22.5	5.9 - 16.4
Range	1.9 - 37.6	2 - 36.7	1.9 - 37.6

Table 12: Time since initial diagnosis of advanced/metastatic disease [months]

	Bevacizumab	**Placebo**	**Total**
n	32	13	45
Mean ± SD	11.2 ± 9.5	15.1 ± 10.5	12.3 ± 9.9
Median	9.1	11.9	9.6
Quartile	4.1 - 13.6	7.7 - 22.3	5.6 - 16.2
Range	1.5 - 37.6	2 - 36.7	1.5 - 37.6

11.2.2.3 Staging

Details on the disease stage according to the TNM system at the first diagnosis of cancer are provided in Tables 13-15. Most patients, namely in the bevacizumab arm, suffered from disseminated disease from the beginning. If UICC staging was reported, it was stage IV in almost each case (Table 16).

Table 13: T stage at initial diagnosis

T stage	Bevacizumab	Placebo	Total
n	33	16	49
T2	3 (9%)	2 (12%)	5 (10%)
T3	5 (15%)	3 (19%)	8 (16%)
T4	10 (30%)	4 (25%)	14 (29%)
TX	6 (18%)	2 (12%)	8 (16%)
Unknown/NA	9 (27%)	5 (31%)	14 (29%)

Table 14: N stage at initial diagnosis

N stage	Bevacizumab	Placebo	Total
n	33	16	49
N0	1 (3%)	2 (12%)	3 (6%)
N1	11 (33%)	4 (25%)	15 (31%)
N2	2 (6%)	--	2 (4%)
N3	5 (15%)	--	5 (10%)
NX	5 (15%)	5 (31%)	10 (20%)
Unknown/NA	9 (27%)	5 (31%)	14 (29%)

Table 15: M stage at initial diagnosis

M stage	Bevacizumab	Placebo	Total
n	33	16	49
M0	2 (6%)	4 (25%)	6 (12%)
M1	22 (67%)	8 (50%)	30 (61%)
MX	1 (3%)	--	1 (2%)
Unknown/NA	8 (24%)	4 (25%)	12 (24%)

Table 16: Disease stage at study entry

Tumor stage	Bevacizumab	Placebo	Total
n	33	16	49
III	--	2 (12%)	2 (4%)
IV	24 (73%)	10 (62%)	34 (69%)
Unknown / NA	9 (27%)	4 (25%)	13 (27%)

11.2.2.4 Antineoplastic pre-treatment

Nine out of ten patients had received at least one previous antineoplastic treatment regimen, being more frequent in the bevacizumab group (Table 17). Table 18 shows, that among pre-treated patients, the median number of regimens was 2 in both trial arms.

Table 17: Antineoplastic pre-treatment

Pre-treatment	Bevacizumab	Placebo	Total
n	32*	16	48
yes	31 (98%)	12 (75%)	43 (90%)
no	1 (2%)	4 (25%)	5 (10%)

* one patient with missing data

Table 18: Number of previous antineoplastic regimens

Number	Bevacizumab	Placebo	Total
n	32	12	44
1	11 (34%)	3 (25%)	14 (32%)
2	7 (22%)	4 (33%)	11 (25%)
3	5 (16%)	2 (17%)	7 (16%)
4	5 (16%)	1 (8%)	6 (14%)
5	3 (9%)	1 (8%)	4 (9%)
6	--	--	--
7	1 (3%)	1 (8%)	2 (5%)
Median	2	2	2

11.2.2.5 Performance status and vital signs

In almost all patients the performance status was more or less impaired. While most patients in the placebo arm had an ECOG score of 2, the verum arm showed a broader distribution (Table 19).

Table 19: Performance status at study entry [ECOG]

ECOG Score	Bevacizumab	Placebo	Total
	33	16	49
0	1 (3%)	--	1 (2%)
1	14 (42%)	4 (25%)	18 (37%)
2	12 (36%)	9 (56%)	21 (43%)
3	5 (15%)	1 (6%)	6 (12%)
4	--	--	--
missing	1 (3%)	2 (12%)	3 (6%)

Tables 20 and 21 show no relevant difference between the groups with respect to blood pressure.

Table 20: Blood pressure at study entry, systolic

Parameter	Bevacizumab	Placebo	Total
n	31	14	45
Mean ± SD	118.8 ± 20.4	113.4 ± 18.2	117.2 ± 19.7
Median	120	112.5	115
Quartile	102.5 - 135	106.2 - 126	105 - 130
Range	83 - 164	80 - 140	80 - 164

Table 21: Blood pressure at study entry, diastolic

Parameter	Bevacizumab	Placebo	Total
n	31	14	45
Mean ± SD	72 ± 12	71.2 ± 12.8	71.8 ± 12.1
Median	70	72.5	70
Quartile	64.5 - 80	62.5 - 79.5	64 - 80
Range	50 - 108	50 - 97	50 - 108

11.3 Measurements of treatment compliance

Since the intraperitoneal infusion was administered in a hospital or in an outpatient setting, compliance could easily be supervised. Bevacizumab/Placebo was administered either by the investigator or under his direct supervision.

11.4 Efficacy results and tabulation of individual patient data

11.4.1 Analysis of efficacy

11.4.1.1 Paracentesis-free survival (1st primary endpoint)

The 1st primary endpoint, paracentesis-free survival (ParFS), i.e. the time from randomisation to either the second on-study paracentesis or to death, whichever occurred first, is shown in Fig. 2 for both study arms and in Fig. 3 for the combined intention-to-treat population. (An additional per-protocol analysis is not applicable as the per-protocol population is identical to the ITT one)

The median overall ParFS of 13 days (95% confidence interval: 10 – 15 days) is well in accordance with the median of 14 days reported by Parsons et al. (2008) for the control group within the non-ovarian cancer subpopulation, which formed the basis for the statistical design calculation of the present study.

As shown in Figure 2, there is no major difference between the Bevacizumab and the control group with ParFS medians of 14.0 (95% confidence interval: 11 – 17 days) and 10.5 (95% confidence interval: 7 – 21 days), with a non-significant logrank test result of p = 0.16 (one-sided, according to the primary study hypothesis as defined in the protocol). The hazard ratio (HR) amounts to 0.74 (95% confidence interval: 0.40 – 1.37). Thus, the latter confidence interval does not exclude the prospectively anticipated target effect size of HR = 0.5, as derived from the catumaxomab trial results.

The ParFS are clearly dominated by paracentesis events, since only in 8 patients (16%) death occurred before a second on-study paracentesis was performed and recorded.

Figure 2 Paracentesis-free survival by study arm

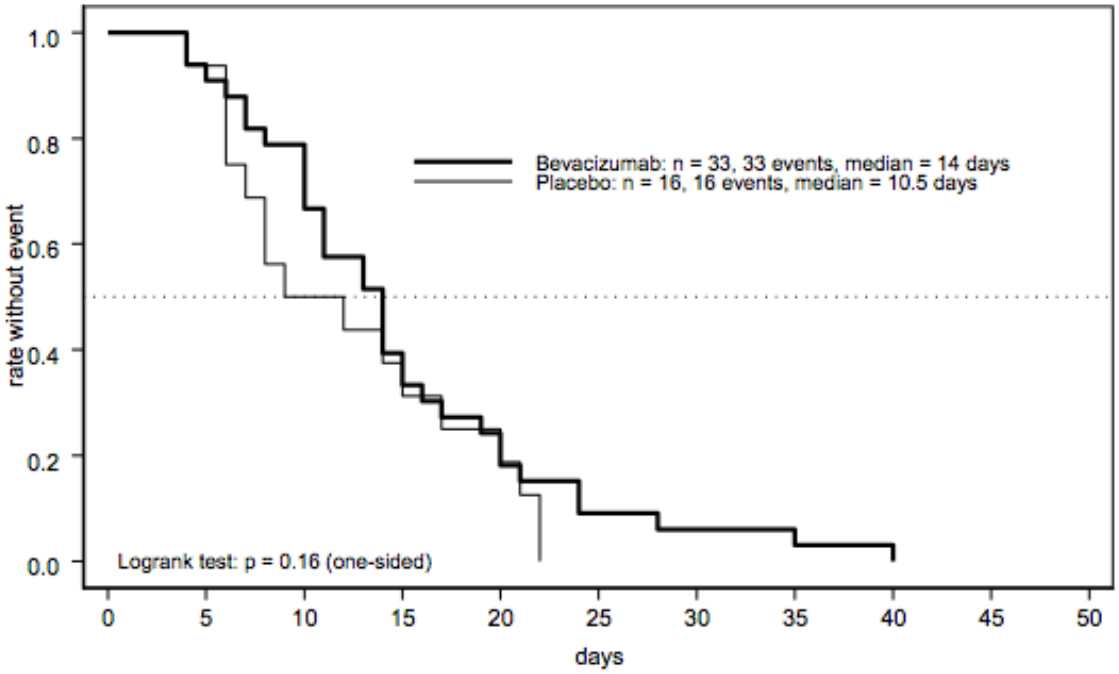
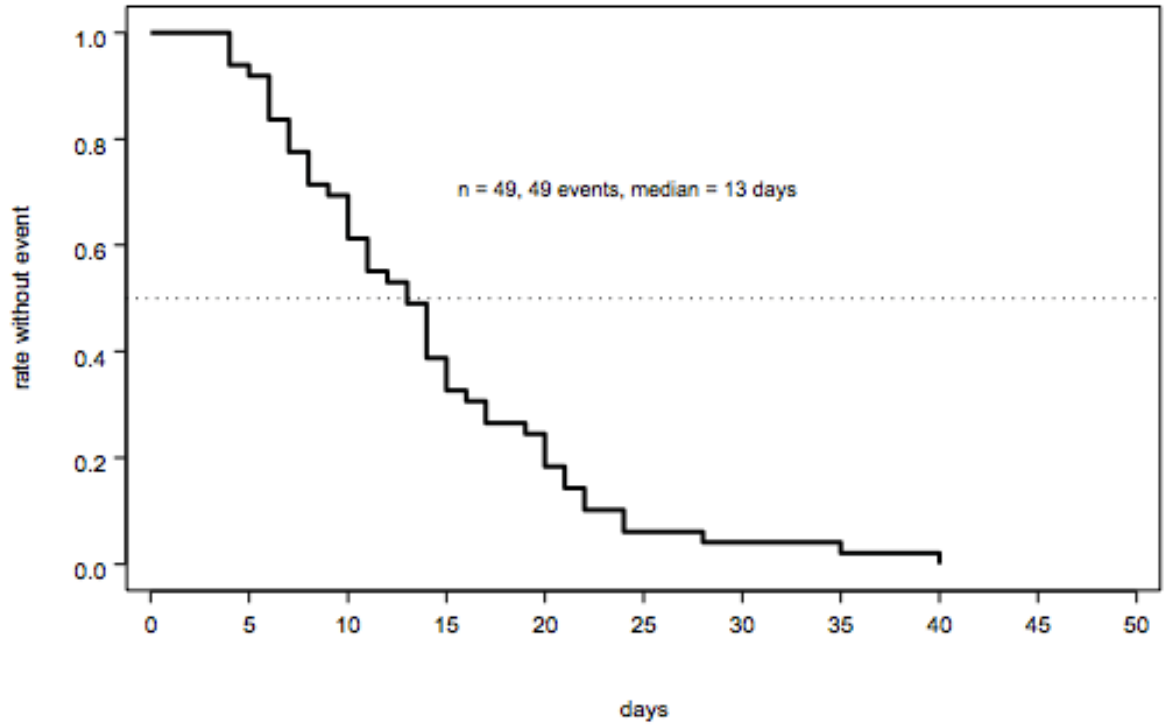


Figure 3 Paracentesis-free survival (total population)



11.4.1.2 Paracenteses

11.4.1.2.1 Paracenteses during the screening period

11.4.1.2.1.1 Number and frequency

Overall, 131 paracenteses were performed and recorded in the screening period of the study, with a median number per patient of two, overall and in both treatment groups (Table 22). As shown in Table 23, the incidence is ranging from 1 to 4 in 96% of the patients, with only two patients in the bevacizumab group with higher counts. (Originally, the protocol required at least two paracenteses performed during the 4 week screening period; however, this requirement was reduced to one after a protocol amendment.)

Table 22: Number of paracenteses during the screening period

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	2.9 ± 1.9	2.3 ± 0.7	2.7 ± 1.6
Median	2	2	2
Quartile	2 - 3	2 - 3	2 - 3
Range	1 - 10	1 - 4	1 - 10

Table 23: Number of paracenteses during the screening period

Number of paracenteses	Bevacizumab	Placebo	Total
n	33	16	49
1	1 (3%)	1 (6%)	2 (4%)
2	18 (55%)	10 (62%)	28 (57%)
3	8 (24%)	4 (25%)	12 (24%)
4	4 (12%)	1 (6%)	5 (10%)
9	1 (3%)	--	1 (2%)
10	1 (3%)	--	1 (2%)

The mean interval between paracenteses during the screening period by patient (based on all recorded screening paracenteses plus the first "therapeutic" one) is a potentially important

prognostic factor. With a median of 8 days, it is equally distributed between the two randomized arms (Table 24). The median of the maximum intervals between paracentesis during the screening period is 10 days in the whole population, and slightly higher in the placebo group (Tables 25 and 26). The Wilcoxon-Mann-Whitney test result for the difference amounts to p = 0.26 (two-sided, exact version).

Table 24: Mean interval between paracenteses during the screening period [days]

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	8.5 ± 4	9.4 ± 3.6	8.8 ± 3.9
Median	8	8	8
Quartile	5.5 - 10.5	7 - 11.6	6 - 11.5
Range	1.1 - 17	3.5 - 16.3	1.1 - 17

Table 25: Maximum interval between paracenteses during the screening period [days]

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	11.2 ± 5.5	13.2 ± 6.5	11.9 ± 5.9
Median	10	11.5	10
Quartile	7 - 15	7.8 - 17.5	7 - 16
Range	2 - 27	6 - 27	2 - 27

Table 26: Maximum interval between paracenteses during the screening period [days]

Number of paracenteses	Bevacizumab	Placebo	Total
n	33	16	49
2	1 (3%)	--	1 (2%)
4	1 (3%)	--	1 (2%)
5	1 (3%)	--	1 (2%)
6	4 (12%)	1 (6%)	5 (10%)
7	5 (15%)	3 (19%)	8 (16%)

8	2 (6%)	2 (12%)	4 (8%)
9	1 (3%)	1 (6%)	2 (4%)
10	2 (6%)	1 (6%)	3 (6%)
11 - 15	8 (24%)	2 (12%)	10 (20%)
16 – 20	6 (18%)	3 (19%)	9 (18%)
> 20	2 (6%)	3 (19%)	5 (10%)

11.4.1.2.1.2 Ascites volume and body weight

As shown in Table 27, the volume of ascites during the screening phase (here calculated as the average of the last two paracenteses) showed a median of three and a half litre with a maximum of more than ten litres, and was rather equally distributed among the randomized groups.

Table 27: Average ascites volume at last two paracenteses during screening [ml]

Parameter	Bevacizumab	Placebo	Total
n	32	16	48
Mean ± SD	3777 ± 1629.7	4614.2 ± 2732.3	4056.1 ± 2072.1
Median	3550	3250	3500
Quartile	2537.5 - 4662.5	2388.1 - 7000	2500 - 4887.5
Range	1425 - 8850	1500 - 10200	1425 - 10200

The body weight before the first paracentesis documented in the screening period is missing in several patients. No major differences can be detected between the treatment arms (Table 28).

Table 28: Body weight before the first paracentesis during screening [kg]

Parameter	Bevacizumab	Placebo	Total
n	22	13	35
Mean ± SD	71.8 ± 19	75.2 ± 12	73.1 ± 16.6
Median	67.3	72.3	70.8
Quartile	59.4 - 80.8	68.4 - 85.2	63.6 - 83.1
Range	39 - 122	54.4 - 100.2	39 - 122

11.4.1.2.2 Paracenteses during the study period

In the 49 evaluable patients, a total of 208 "therapeutic" paracenteses were documented within the trial phase (until and including the 12-week safety follow-up visit), i.e. a mean number of 4.2 paracenteses (Tables 29 and 30). The mean and median number is higher in the bevacizumab group. However, this may be easily explained by the longer median survival time in the active drug arm (c.f. section 11.4.1.5).

Table 29: Number of therapeutic paracenteses during the study period , distribution parameters

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	4.7 ± 3.6	3.4 ± 2.2	4.2 ± 3.3
Median	4	3	4
Quartile	2 - 5	2 - 4.2	2 - 5
Range	1 - 17	1 - 8	1 - 17

Table 30: Number of therapeutic paracenteses during the study period, categories

Number of paracenteses	Bevacizumab	Placebo	Total
n	33	16	49
1	5 (15%)	3 (19%)	8 (16%)
2	4 (12%)	4 (25%)	8 (16%)
3	3 (9%)	3 (19%)	6 (12%)
4	7 (21%)	2 (12%)	9 (18%)
5	6 (18%)	1 (6%)	7 (14%)
6	4 (12%)	1 (6%)	5 (10%)
7	1 (3%)	1 (6%)	2 (4%)
8 - 10	--	1 (6%)	1 (2%)
> 10	3 (9%)	--	3 (6%)

Overall, as shown in Table 31, the average volume of ascites removed during the study period seems to be marginally higher than at the last two paracenteses during the screening period (cf. Table 27). However, the test results provided in Table 32 miss any statistical significance by far.

No differences can be detected between the randomization arms.

Table 31: Average volume of ascites during study period [ml]

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	4083.6 ± 1948.3	4151 ± 1866.5	4105.6 ± 1902.7
Median	4200	4041.7	4200
Quartile	2775 - 5000	2571.9 - 5503.6	2600 - 5250
Range	157.5 - 8040	1500 - 7812.5	157.5 - 8040

Table 32: Average volume of ascites: within-group comparison of screening vs. study period

Group	n	Median, screening period**	Median, study period	p*
Bevacizumab	32	3550 ml	4250 ml	0.22
Placebo	16	3250 ml	4042 ml	0.16
Total	48	3500 ml	4250 ml	0.75

* Wilcoxon signed rank test, exact version

** cf. section 3.1.2

Measurements of body weight during the on study period were performed at only 41 out of 208 paracenteses (in only 22 patients). Therefore, calculations of average body weight and comparisons to the screening period (cf. section 11.4.1.2.1.2) are not possible.

11.4.1.3 Best response (2nd primary endpoint)

"Best response" as the second primary endpoint of this randomized phase II study is defined in the protocol as assessing the longest paracentesis-free period within the 12-week main observation period. Besides comparing these periods between both study arms, within-group comparisons with the respective intervals in the screening phase (Table 25) were to be performed.

The results of the study group comparison show only a minimal numerical advantage for the verum group (medians of 19 vs. 17.5 days) with a one-sided test result of p = 0.85 (Wilcoxon-Mann-Whitney U test, exact version).

When comparing the maximum interval during the treatment period (Table 33) to the respective findings during the screening phase, remarkably longer intervals are calculated for the later period: the total median of 19 days on treatment is significantly longer than the median of 10 days during screening (p < 0.0001, Wilcoxon test for paired observations, exact version, two-sided). The same holds for the respective comparison in the antibody cohort (medians: 19 vs. 10 days, p < 0.0001, Wilcoxon test for paired observations, exact version, two-sided). This difference is somewhat larger than in the placebo group (medians: 17.5 vs. 11.5 days, p = 0.22, Wilcoxon test for paired observations, exact version, two-sided). However, the lower patient number in the placebo arm has to be kept in mind when interpreting these results.

Table 33 Maximum period without paracenteses during the treatment period by patient [days]

Parameter	Bevacizumab	Placebo	Total
n	33	16	49
Mean ± SD	22.9 ± 15.6	18.7 ± 13.6	21.6 ± 15
Median	19	17.5	19
Quartile	11 - 29	7.2 - 28.5	10 - 29
Range	6 - 66	4 - 42	4 - 66

11.4.1.4 Additional response category, based on number of paracenteses

An additional response category was defined by the protocol and the statistical analysis plan:

Complete response (CR) = no paracentesis in the 12 week study period;

Partial response (PR) = 1 – 3 additional paracenteses in the 12 week study period

No response (NR) = otherwise

No complete responses were observed in either group (Table 34). In most of the patients, the 12 week on-study period was truncated by death. No differences were detected between the trial arms (when subsuming NR and death into one category); Cochran-Armitage test for trend, exact version, two-sided: p = 1.0; asymptotic version, two-sided: p = 0.75)

Table 34 Response category during the treatment period

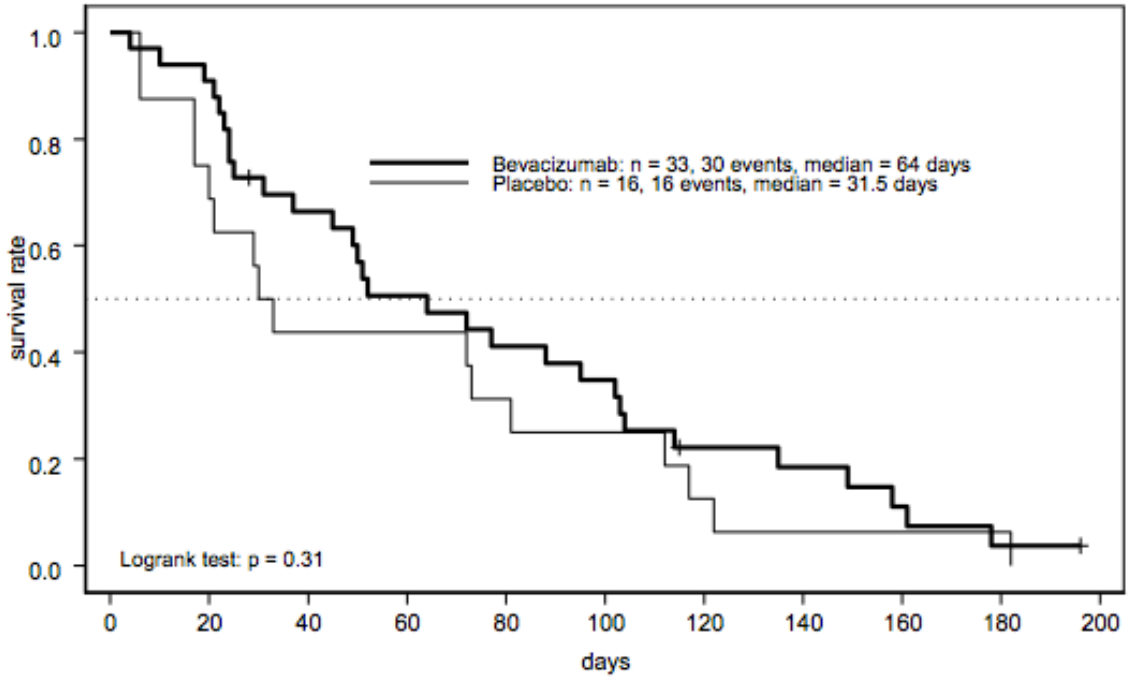
Response	Bevacizumab	Placebo	Total
n	33	16	49
CR	--	--	--
PR	5 (15%)	3 (19%)	8 (16%)
No response	8 (24%)	1 (6%)	9 (18%)
Death	20 (61%)	12 (75%)	32 (65%)

11.4.1.5 Overall survival

The results of the Kaplan-Meier analysis on overall survival (OS) are shown in Figure 4. The median OS is 64 days (95% confidence interval: 45 – 103 days) in the Bevacizumab arm, and 31.5 days (95% confidence interval: 20 – 117 days) under placebo, with a two-sided logrank test result of p = 0.31. The 60-day survival rate amounts to 51% (95% confidence interval: 36% – 71%) and 44% (95% confidence interval: 25% – 76%), respectively. The hazard ratio is 0.73 (95% confidence interval: 0.40 – 1.37).

The median overall survival observed in this study is almost identical to the median OS of about 50 days reported by Parsons et al. (2008) for the non-ovarian cancer subpopulation.

Figure 4 Overall survival by study arm



11.4.1.6 Quality of life by FACIT-AI questionnaire

Quality of life was evaluated using the Functional Assessment of Chronic Illness Therapy (FACIT) – Ascites Index (AI) questionnaire. The 13 items were defined and abbreviated as follows. (Table 35). Except for the first three, all items are reversed in a way that a score of “0” always corresponds to a severely symptomatic patients, while a score of “4” is an asymptomatic patient. All items sum up to the FACIT-AI score ranging from 0 (worst) to 52 (best).

Individual missing items were taken into account by an adapted weighting of the remaining items, such that these patients were assessed on the same overall scale.

Table 35 Items and structure of the FACIT-AI forms

Abbr.		Not at all	A little bit	Some-what	Quite a bit	Very much
C6	I have a good appetite	0	1	2	3	4
GF5	I am sleeping well	0	1	2	3	4
BMT5	I am able to get around by myself	0	1	2	3	4
B1	I have been short of breath	0	1	2	3	4
GP2	I have nausea	0	1	2	3	4
O2	I have been vomiting	0	1	2	3	4
ACT11	I have pain in my stomach area	0	1	2	3	4
O1	I have swelling in my stomach area	0	1	2	3	4
GP1	I have a lack of energy	0	1	2	3	4
ACT10	When I eat, I seem to get full quickly	0	1	2	3	4
BL2	I urinate more frequently than usual	0	1	2	3	4

Cx6	I am bothered by constipation	0	1	2	3	4
	.....					
AI1	I have been emotionally distressed	0	1	2	3	4

Table 36 gives an overview on the number of filled-in FACIT-AI forms at the different time points of the study. While the baseline form was available for the majority of patients, the relative numbers dropped quickly to less than half during the course of the study treatment period (in parallel to the death events, cf. section 11.4.1.5). This lead to the common and inevitable problem of quality of life analysis in highly palliative situations: firstly, comparisons, both between groups or different time points, suffer from an uncontrolled loss process, due to patient's death or inability to perform further visits or fill out the questionnaire. Thus, the remaining data definitely reflect a positive selection. Secondly, this dropout mechanism leads to a loss of power of hypothesis testing.

Table 36 Number of available FACIT-AI forms

Visit	Bevacizumab	Placebo	Total
n (patients)	33	16	49
Screening	20 (61%)	11 (69%)	31 (63%)
Start of study	28 (85%)	10 (63%)	38 (78%)
Week 2	22 (67%)	7 (44%)	29 (59%)
Week 4	13 (39%)	5 (31%)	18 (37%)
Week 6	7 (21%)	4 (25%)	11 (22%)
Week 8	7 (21%)	5 (31%)	12 (24%)
Safety Follow-up	4 (12%)	3 (19%)	7 (14%)
Paracentesis (total number of forms)	4	3	7

Tables 37-39 show the distribution parameters of the FACIT Ascites Summary index over time, both for the therapy groups and for the total population with data available. While there seem to be no relevant differences between the arms, the means and medians in the overall patient group indicate a major impairment of quality of life, but a rather stable condition in the patients remaining alive and completing the forms.

Table 37 FACIT Ascites Summary Index, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	32 ± 5.8	32.6 ± 7.9	31.3 ± 7.1	32.9 ± 5.8	36 ± 8.8	35.7 ± 7.8
Median	32	33	30	34	38	35.9
Quartile	28 - 37	28.2 - 37	27.5 - 36	30 - 36	33 - 39.5	33.3 - 38.2
Range	21 - 44	14.2 - 47	20.6 - 45.5	23 - 41	21 - 48	26 - 45

Table 38 FACIT Ascites Summary Index, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	29.7 ± 4.2	29.6 ± 6.6	25.9 ± 8.4	34.3 ± 4	31.2 ± 9.2	26.7 ± 8.3
Median	29	30	31	32	32	24
Quartile	26 - 33.1	28.5 - 32	17.3 - 32	32 - 35.5	24.8 - 38.5	22 - 30
Range	23 - 36	17.3 - 39	16 - 33	32 - 39	21 - 40	20 - 36

Table 39 FACIT Ascites Summary Index, total group

Week	0	2	4	6	8	Follow-up
n	41	28	16	10	11	7
Mean ± SD	31.3 ± 5.5	31.9 ± 7.6	29.6 ± 7.7	33.3 ± 5.2	34.3 ± 8.8	31.8 ± 8.7
Median	31	30.2	30.5	33	38	35.8
Quartile	27 - 36	28.1 - 36.9	26 - 33.5	30.5 - 36.5	27 - 39.5	25 - 36
Range	21 - 44	14.2 - 47	16 - 45.5	23 - 41	21 - 48	20 - 45

A comparison of the available on-study data to baseline (performed only for week 2 and 4 as numbers are too small at later visits) provided the following results:

- Between baseline and week 2, with corresponding pairs of data available in a total of 28 patients, there is no significant difference (p = 0.59, paired Wilcoxon test, exact). The respective paired Wilcoxon test results are p = 0.54 for 21 patients in arm 1, and p = 0.97 for 7 patients in arm 2.

- When comparing baseline and week 4, with corresponding pairs of data available in a total of 16 patients, there is no significant difference (p = 0.41, paired Wilcoxon test, exact). The respective paired Wilcoxon test results are p = 0.72 for 11 patients in arm 1, and p = 0.31 for 5 patients in arm 2.

Following, all items of the FACIT-AI questionnaires are listed separately, for patients receiving bevacizumab and placebo, respectively:

FACIT-AI item C6: Appetite

Appetite showed a rather distinct impairment with a scale median of 2 ("somewhat"). However, no distinct differences can be detected between the groups or between visits (Tables 40 and 41).

Table 40 FACIT: C6, bevacizumab

Week	0	2	4	6	8	Follow-up
n	28	20	10	7	7	4
Mean ± SD	1.7 ± 1.1	1.8 ± 1.2	1.5 ± 0.7	1.9 ± 1.1	1.7 ± 1	1 ± 1.4
Median	2	2	2	2	2	0.5
Quartile	1 - 2	1 - 3	1 - 2	1.5 - 2.5	1.5 - 2	0 - 1.5
Range	0 - 4	0 - 4	0 - 2	0 - 3	0 - 3	0 - 3

Table 41 FACIT: C6, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	1.2 ± 0.9	1.1 ± 1.1	1 ± 1	1.3 ± 1.2	1.8 ± 1.3	0.7 ± 1.2
Median	1.5	2	1	2	2	0
Quartile	0.8 - 2	0 - 2	0 - 2	1 - 2	1.5 - 2.2	0 - 1
Range	0 - 2	0 - 2	0 - 2	0 - 2	0 - 3	0 - 2

FACIT-AI item GF5: Sleeping

Quality of sleep showed also a rather distinct impairment with a scale median of 2 ("somewhat"). However, no clear differences or trends can be detected between the groups or between visits (Table 42, Table 43).

Table 42 FACIT: GF5, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	2.1 ± 1.3	2.7 ± 1.2	1.8 ± 1.3	2.4 ± 1.5	2.9 ± 1.5	3.2 ± 0.5
Median	2	3	2	3	3	3
Quartile	1 - 3	2 - 4	1 - 3	1.5 - 3.5	2.5 - 4	3 - 3.2
Range	0 - 4	0 - 4	0 - 4	0 - 4	0 - 4	3 - 4

Table 43 FACIT: GF5, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	2.4 ± 0.7	1.9 ± 1.2	2 ± 1.6	2.7 ± 1.2	1.8 ± 1.5	2.3 ± 1.5
Median	2	2	2	2	2	2
Quartile	2 - 3	1.5 - 2	1 - 3	2 - 3	0.8 - 3	1.5 - 3
Range	2 - 4	0 - 4	0 - 4	2 - 4	0 - 3	1 - 4

FACIT-AI item BMT5: Getting around

Ability to get around also showed a scale median of 2 ("somewhat") at baseline. There is a slight trend to improvement in the placebo group, but this may easily be explained by the dropout process (Table 44, Table 45).

Table 44 FACIT: BMT5, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	2.5 ± 1.3	2.5 ± 1.3	2.3 ± 1.6	2.4 ± 1.5	3 ± 1.7	3 ± 0.8
Median	2	2	2	2	4	3
Quartile	2 - 4	1 - 4	1 - 4	1 - 4	2.5 - 4	2.8 - 3.2
Range	0 - 4	1 - 4	0 - 4	1 - 4	0 - 4	2 - 4

Table 45 FACIT: BMT5, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	2.2 ± 1.2	1.3 ± 1.1	1.2 ± 1.1	1.3 ± 1.5	0.8 ± 1	2 ± 1
Median	2	1	1	1	0.5	2
Quartile	1.8 - 3	0.5 - 2	1 - 1	0.5 - 2	0 - 1.2	1.5 - 2.5
Range	0 - 4	0 - 3	0 - 3	0 - 3	0 - 2	1 - 3

FACIT-AI item B1: Shortness of breath

Shortness of breath is somewhat less of a problem with a scale median of 3 ("a little bit") in both groups at almost each time point (Table 46, Table 47).

Table 46 FACIT: B1, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	2.7 ± 1.1	2.6 ± 1.1	2.6 ± 1.1	2.9 ± 1.2	3 ± 1.2	3.2 ± 0.5
Median	3	3	3	3	3	3
Quartile	2 - 4	2 - 3	2 - 3.5	2 - 4	2.5 - 4	3 - 3.2
Range	0 - 4	0 - 4	1 - 4	1 - 4	1 - 4	3 - 4

Table 47 FACIT: B1, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	2.5 ± 1.3	2 ± 1.4	2.2 ± 1.6	2.3 ± 1.5	3.2 ± 1	3 ± 1
Median	3	2	3	2	3.5	3
Quartile	1.8 - 3.2	1 - 3	1 - 3	1.5 - 3	2.8 - 4	2.5 - 3.5
Range	0 - 4	0 - 4	0 - 4	1 - 4	2 - 4	2 - 4

FACIT-AI item GP2: Nausea

Nausea was a minor problem with scale medians of 3 to 4. Again, no clear differences or trends can be detected between the groups or between visits (Table 48, Table 49).

Table 48 FACIT: GP2, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	3 ± 1.1	3.2 ± 1.1	3.1 ± 1	3.1 ± 1.2	3.1 ± 1.1	3 ± 1.4
Median	3	4	3	4	3	3.5
Quartile	3 - 4	3 - 4	2.5 - 4	2.5 - 4	3 - 4	2.5 - 4
Range	0 - 4	1 - 4	1 - 4	1 - 4	1 - 4	1 - 4

Table 49 FACIT: GP2, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	3.2 ± 0.7	3.1 ± 1.5	2.8 ± 1.1	4 ± 0	3.2 ± 1	3 ± 1.7
Median	3	4	3	4	3.5	4
Quartile	3 - 4	3 - 4	3 - 3	4 - 4	2.8 - 4	2.5 - 4
Range	2 - 4	0 - 4	1 - 4	4 - 4	2 - 4	1 - 4

FACIT-AI item O2: Vomiting

Most of the patients had no problems with vomiting before and during the study (Table 50, Table 51).

Table 50 FACIT: O2, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	3.6 ± 0.7	3.3 ± 1.4	3.5 ± 0.9	3.6 ± 0.8	3.6 ± 1.1	3.5 ± 1
Median	4	4	4	4	4	4
Quartile	3 - 4	4 - 4	3 - 4	3.5 - 4	4 - 4	3.5 - 4
Range	2 - 4	0 - 4	1 - 4	2 - 4	1 - 4	2 - 4

Table 51 FACIT: O2, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	3.4 ± 0.5	3.7 ± 0.5	3 ± 1.7	4 ± 0	3.8 ± 0.5	3.3 ± 1.2
Median	3	4	4	4	4	4
Quartile	3 - 4	3.5 - 4	3 - 4	4 - 4	3.8 - 4	3 - 4
Range	3 - 4	3 - 4	0 - 4	4 - 4	3 - 4	2 - 4

FACIT-AI item ACT11: Stomach pain

Stomach pain also rated among the less pronounced symptoms with scale medians of 3 to 4, and no relevant differences with respect to time point or group (Table 52, Table 53).

Table 52 FACIT: ACT11, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	10	7	7	4
Mean ± SD	2.8 ± 1.2	3 ± 0.9	3.3 ± 0.9	3.1 ± 1.1	3.4 ± 1.1	3.5 ± 1
Median	3	3	4	3	4	4
Quartile	2 - 4	2 - 4	2.2 - 4	3 - 4	3.5 - 4	3.5 - 4
Range	1 - 4	1 - 4	2 - 4	1 - 4	1 - 4	2 - 4

Table 53 FACIT: ACT11, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	3.1 ± 1	3.7 ± 0.5	3.2 ± 1.3	4 ± 0	2.8 ± 1.5	3.7 ± 0.6
Median	3	4	4	4	3	4
Quartile	2.8 - 4	3.5 - 4	3 - 4	4 - 4	1.8 - 4	3.5 - 4
Range	1 - 4	3 - 4	1 - 4	4 - 4	1 - 4	3 - 4

FACIT-AI item O1: Swelling

Symptoms of swelling of the stomach area are slightly less pronounced in the bevacizumab group at baseline. A weak trend to improvement can be detected in both groups, but this can be

explained by the fact that the baseline measurement was driven by puncture necessity (Table 54, Table 55).

Table 54 FACIT: O1, bevacizumab

Week	0	2	4	6	8	Follow-up
n	28	20	11	7	6	4
Mean ± SD	2.6 ± 1.5	2.6 ± 1.5	2.2 ± 2.1	2.7 ± 1.4	3.5 ± 0.8	4 ± 0
Median	3	3	4	3	4	4
Quartile	1 - 4	1.8 - 4	0 - 4	2.5 - 3.5	3.2 - 4	4 - 4
Range	0 - 4	0 - 4	0 - 4	0 - 4	2 - 4	4 - 4

Table 55 FACIT: O1, placebo

Week	0	2	4	6	8	Follow-up
n	11	7	4	3	4	3
Mean ± SD	2 ± 1.7	2.9 ± 1.5	3.5 ± 1	4 ± 0	3.2 ± 1.5	3.3 ± 1.2
Median	2	3	4	4	4	4
Quartile	0.5 - 3.5	2.5 - 4	3.5 - 4	4 - 4	3.2 - 4	3 - 4
Range	0 - 4	0 - 4	2 - 4	4 - 4	1 - 4	2 - 4

FACIT-AI item GP1: Lack of energy

Lack of energy was an ubiquitous problem in the ascites patients from the beginning, especially in the placebo group (Table 56, Table 57).

Table 56 FACIT: GP1, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	20	11	7	7	4
Mean ± SD	1.1 ± 1.2	1.2 ± 1.2	1.7 ± 1.3	1 ± 1	1.9 ± 1.9	1.8 ± 1.3
Median	1	1	2	1	2	2
Quartile	0 - 2	0 - 1.2	0.5 - 3	0 - 2	0 - 3.5	1.5 - 2.2
Range	0 - 4	0 - 4	0 - 3	0 - 2	0 - 4	0 - 3

Table 57 FACIT: GP1, placebo

Week	0	2	4	6	8	Follow-up
n	11	7	5	3	4	3
Mean ± SD	0.7 ± 0.8	0.4 ± 0.8	0.2 ± 0.4	0.7 ± 0.6	1.2 ± 1	0.3 ± 0.6
Median	1	0	0	1	1.5	0
Quartile	0 - 1	0 - 0.5	0 - 0	0.5 - 1	0.8 - 2	0 - 0.5
Range	0 - 2	0 - 2	0 - 1	0 - 1	0 - 2	0 - 1

FACIT-AI item ACT10: Getting full quickly

Feeling of fullness was a common symptom, too. Some improvement of mean and median values seems to occur in patients remaining on study for longer periods in the bevacizumab arm (Table 58, Table 59).

Table 58 FACIT: ACT10, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	18	11	7	7	3
Mean ± SD	1.2 ± 1.2	1.3 ± 1.3	1.3 ± 1	1.7 ± 1.4	1.9 ± 1.6	1.7 ± 2.1
Median	1	1	1	2	2	1
Quartile	0 - 2	0 - 2	0.5 - 2	0.5 - 3	0.5 - 3	0.5 - 2.5
Range	0 - 4	0 - 4	0 - 3	0 - 3	0 - 4	0 - 4

Table 59 FACIT: ACT10, placebo

Week	0	2	4	6	8	Follow-up
n	12	6	5	3	4	3
Mean ± SD	1.1 ± 0.9	0.7 ± 0.8	0.4 ± 0.5	1 ± 1.7	1 ± 0.8	0.3 ± 0.6
Median	1	0.5	0	0	1	0
Quartile	0.8 - 1.2	0 - 1	0 - 1	0 - 1.5	0.8 - 1.2	0 - 0.5
Range	0 - 3	0 - 2	0 - 1	0 - 3	0 - 2	0 - 1

FACIT-AI item BL2: Frequent urination Frequent urination seems to be a symptom of minor importance in the study population, with no uniform trend over time (Table 60, Table 61).

Table 60 FACIT: BL2, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	20	11	7	7	4
Mean ± SD	2.7 ± 1.3	2.6 ± 1.1	2.5 ± 1.2	3.3 ± 0.8	3.3 ± 1	2.8 ± 0.5
Median	3	2.5	3	3	4	3
Quartile	2 - 4	2 - 4	2 - 3	3 - 4	2.5 - 4	2.8 - 3
Range	0 - 4	1 - 4	0 - 4	2 - 4	2 - 4	2 - 3

Table 61 FACIT: BL2, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	2.6 ± 1.5	3 ± 1.5	1.8 ± 1.5	4 ± 0	3 ± 1.4	2 ± 2
Median	3	4	2	4	3.5	2
Quartile	1 - 4	2.5 - 4	1 - 2	4 - 4	2.5 - 4	1 - 3
Range	0 - 4	0 - 4	0 - 4	4 - 4	1 - 4	0 - 4

FACIT-AI item Cx6: Constipation

With respect to constipation, severe symptoms are rare in the study population, especially in placebo group patients remaining on study (Table 62, Table 63).

Table 62 FACIT: Cx6, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	20	11	7	7	4
Mean ± SD	3.3 ± 1.1	3.3 ± 1.2	3.2 ± 1.2	1.9 ± 1.8	2.3 ± 1.9	2 ± 2.3
Median	4	4	3	3	3	2
Quartile	3 - 4	3 - 4	3 - 4	0 - 3	0.5 - 4	0 - 4
Range	1 - 4	0 - 4	0 - 4	0 - 4	0 - 4	0 - 4

Table 63 FACIT: Cx6, placebo

Week	0	2	4	6	8	Follow-up
n	11	7	5	3	4	3
Mean ± SD	2.7 ± 1.5	3.7 ± 0.8	3.6 ± 0.5	3 ± 1.7	4 ± 0	1.3 ± 1.2
Median	3	4	4	4	4	2
Quartile	1.5 - 4	4 - 4	3 - 4	2.5 - 4	4 - 4	1 - 2
Range	0 - 4	2 - 4	3 - 4	1 - 4	4 - 4	0 - 2

FACIT-AI item AI1: Emotional distress

Emotional distress was somewhat more frequent at baseline in the placebo group and exhibited a trend to worse scoring, in contrast to the verum group with stable median values (Table 64, Table 65).

Table 64 FACIT: AI1, bevacizumab

Week	0	2	4	6	8	Follow-up
n	29	21	11	7	7	4
Mean ± SD	2.8 ± 1.4	2.4 ± 1.2	2.4 ± 1.7	2.9 ± 1.2	2.6 ± 1.1	2.8 ± 0.5
Median	3	3	3	3	2	3
Quartile	2 - 4	2 - 3	1 - 4	2 - 4	2 - 3.5	2.8 - 3
Range	0 - 4	0 - 4	0 - 4	1 - 4	1 - 4	2 - 3

Table 65 FACIT: AI1, placebo

Week	0	2	4	6	8	Follow-up
n	12	7	5	3	4	3
Mean ± SD	2.3 ± 1.6	2 ± 1.6	1.4 ± 1.7	2 ± 1	1.5 ± 1.3	1.3 ± 1.5
Median	3	1	1	2	1.5	1
Quartile	1 - 4	1 - 3.5	0 - 2	1.5 - 2.5	0.8 - 2.2	0.5 - 2
Range	0 - 4	0 - 4	0 - 4	1 - 3	0 - 3	0 - 3

11.4.1.7 Analysis of the DGHO questionnaires

The questionnaire on symptoms and palliative care, based on a form by the Palliative Group of the DGHO, was completed in 35 out of 49 patients at week 0 (71%) for symptoms, and in 37 patients for concomitant medication (76%). Comparable to the quality of life assessment by FACIT-AI questionnaire (cf. section 11.4.1.6) the number of available forms was distinctly reduced during the course of the treatment period, as to be expected due to the loss processes. At week 8, data from 9 remaining patients only were recorded. Therefore, any comparisons between time points and treatment arms are not reliable.

The first part of the DGHO palliative questionnaire refers to disease symptoms, which were assessed by the clinical personnel at the different time points, using an ordinal scale ranging from 0 to 4 (0 = without symptoms [ohne], 1 = weak [gering], 2 = moderate [mittel], 3 = strong [stark]). The distribution parameters of selected items are displayed in Tables 66-83.
Except for weakness, all other symptoms are only rarely reported, and of mild severity, or even not present at all.

Weakness

Table 66 DGHO: Weakness, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	1.8 ± 1	1.8 ± 0.9	2.1 ± 0.9	1.7 ± 1	2 ± 1
Median	2	2	2	2	2
Quartile	1 - 2.8	1 - 2	2 - 3	1.2 - 2	1 - 3
Range	0 - 3	0 - 3	0 - 3	0 - 3	1 - 3

Table 67 DGHO: Weakness, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	1.8 ± 1	2.3 ± 0.8	2 ± 0.9	2 ± 1	2.5 ± 1
Median	2	2.5	2	2	3
Quartile	1 - 2	2 - 3	1.2 - 2.8	1.5 - 2.5	2.5 - 3
Range	0 - 3	1 - 3	1 - 3	1 - 3	1 - 3

Dysphagia

Table 68 DGHO: Dysphagia, bevacizumab

Week	0	2	4	6	8
n	25	17	11	6	4
Mean ± SD	0.2 ± 0.5	0.1 ± 0.2	0.3 ± 0.6	0 ± 0	0.2 ± 0.5
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0.2
Range	0 - 2	0 - 1	0 - 2	0 - 0	0 - 1

Table 69 DGHO: Dysphagia, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0.1 ± 0.3	0.5 ± 0.8	0.3 ± 0.8	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0.8	0 - 0	0 - 0	0 - 0
Range	0 - 1	0 - 2	0 - 2	0 - 0	0 - 0

Diarrhea

Table 70 DGHO: Diarrhea, bevacizumab

Week	0	2	4	6	8
n	25	17	11	6	5
Mean ± SD	0.1 ± 0.3	0.4 ± 0.9	0 ± 0	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 1	0 - 3	0 - 0	0 - 0	0 - 0

Table 71 DGHO: Diarrhea, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0.1 ± 0.3	0.2 ± 0.4	0.5 ± 1.2	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 1	0 - 1	0 - 3	0 - 0	0 - 0

Cough

Table 72 DGHO: Cough, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	0.2 ± 0.6	0.2 ± 0.5	0.3 ± 0.7	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0.2	0 - 0	0 - 0
Range	0 - 2	0 - 2	0 - 2	0 - 0	0 - 0

Table 73 DGHO: Cough, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0.3 ± 0.7	0.2 ± 0.4	0.5 ± 0.8	0.3 ± 0.6	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0.8	0 - 0.5	0 - 0
Range	0 - 2	0 - 1	0 - 2	0 - 1	0 - 0

Itching

Table 74 DGHO: Itching, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	0.2 ± 0.7	0.2 ± 0.7	0.2 ± 0.4	0 ± 0	0.4 ± 0.9
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 3	0 - 3	0 - 1	0 - 0	0 - 2

Table 75 DGHO: Itching, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0

Range	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0

Decubitus

Table 76 DGHO: Decubitus, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	0.2 ± 0.5	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 2	0 - 0	0 - 0	0 - 0	0 - 0

Table 77 DGHO: Decubitus, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0

Disorientation

Table 78 DGHO: Disorientation, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0

Table 79 DGHO: Disorientation, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0

Median	0	0	0	0	0
Quartile	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0
Range	0 - 0	0 - 0	0 - 0	0 - 0	0 - 0

Anxiety

Table 80 DGHO: Anxiety, bevacizumab

Week	0	2	4	6	8
n	26	18	12	6	5
Mean ± SD	0.5 ± 0.8	0.4 ± 0.7	0.6 ± 0.8	0.3 ± 0.8	0.8 ± 1.1
Median	0	0	0	0	0
Quartile	0 - 1	0 - 1	0 - 1	0 - 0	0 - 2
Range	0 - 2	0 - 2	0 - 2	0 - 2	0 - 2

Table 81 DGHO: Anxiety, placebo

Week	0	2	4	6	8
n	9	6	6	3	4
Mean ± SD	0.4 ± 1	0.5 ± 0.5	0.5 ± 0.8	0.3 ± 0.6	0.8 ± 1
Median	0	0.5	0	0	0.5
Quartile	0 - 0	0 - 1	0 - 0.8	0 - 0.5	0 - 1.2
Range	0 - 3	0 - 1	0 - 2	0 - 1	0 - 2

Depression

Table 82 DGHO: Depression, bevacizumab

Week	0	2	4	6	8
n	26	17	12	6	5
Mean ± SD	0.4 ± 0.7	0.7 ± 1.1	0.7 ± 0.9	0.5 ± 0.8	0.8 ± 1.1
Median	0	0	0	0	0
Quartile	0 - 1	0 - 1	0 - 1.2	0 - 0.8	0 - 2
Range	0 - 2	0 - 3	0 - 2	0 - 2	0 - 2

Table 83 DGHO: Depression, placebo

Week	0	2	4	6	8
n	8	6	6	3	4
Mean ± SD	0.4 ± 0.5	0.7 ± 0.5	1 ± 1.3	0.7 ± 0.6	0.5 ± 0.6
Median	0	1	0.5	1	0.5
Quartile	0 - 1	0.2 - 1	0 - 1.8	0.5 - 1	0 - 1
Range	0 - 1	0 - 1	0 - 3	0 - 1	0 - 1

Tables 84 - 93 display the concomitant medication given during the course of study treatment. While antiemetics were applied quite frequently (41% at week 0), only few patients received antidepressants, corticosteroids, or laxatives. Analgesic treatment including opioids were applied in 27% at study therapy onset.

Antiemetics

Table 84 DGHO: Antiemetics, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	15 (56%)	16 (80%)	9 (69%)	5 (62%)	3 (50%)
yes	12 (44%)	4 (20%)	4 (31%)	3 (38%)	3 (50%)

Table 85 DGHO: Antiemetics, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	7 (70%)	2 (29%)	4 (67%)	3 (75%)	5 (100%)
yes	3 (30%)	5 (71%)	2 (33%)	1 (25%)	0 (0%)

Antidepressants

Table 86 DGHO: Antidepressants, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	25 (93%)	18 (90%)	12 (92%)	7 (88%)	6 (100%)

yes	2 (7%)	2 (10%)	1 (8%)	1 (12%)	0 (0%)

Table 87 DGHO: Antidepressants, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	10 (100%)	7 (100%)	6 (100%)	4 (100%)	5 (100%)

Corticosteroids

Table 88 DGHO: Antidepressants, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	25 (93%)	18 (90%)	12 (92%)	7 (88%)	6 (100%)
yes	2 (7%)	2 (10%)	1 (8%)	1 (12%)	0 (0%)

Table 89 DGHO: Antidepressants, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	10 (100%)	7 (100%)	6 (100%)	4 (100%)	5 (100%)

Laxatives

Table 90 DGHO: Laxatives, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	23 (85%)	17 (85%)	12 (92%)	8 (100%)	5 (83%)
yes	4 (15%)	3 (15%)	1 (8%)	0 (0%)	1 (17%)

Table 91 DGHO: Laxatives, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	10 (100%)	5 (71%)	5 (83%)	4 (100%)	5 (100%)
yes	0 (0%)	2 (29%)	1 (17%)	0 (0%)	0 (0%)

Opioids

Table 92 DGHO: Opioids, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	20 (74%)	14 (70%)	10 (77%)	6 (75%)	6 (100%)
yes	7 (26%)	6 (30%)	3 (23%)	2 (25%)	0 (0%)

Table 93 DGHO: Opioids, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	7 (70%)	5 (71%)	5 (83%)	4 (100%)	5 (100%)
yes	3 (30%)	2 (29%)	1 (17%)	0 (0%)	0 (0%)

The final part of the DGHO palliative questionnaire refers to a variety of further clinical measures. Several patients were recorded to receive additional palliative procedures, as shown in Tables 94-99.

Enteral nutrition

Table 94 DGHO: Enteral nutrition, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	24 (89%)	20 (100%)	13 (100%)	8 (100%)	6 (100%)
yes	3 (11%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)

Table 95 DGHO: Enteral nutrition, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	8 (80%)	5 (71%)	5 (83%)	4 (100%)	5 (100%)
yes	2 (20%)	2 (29%)	1 (17%)	0 (0%)	0 (0%)

Parenteral nutrition

Table 96 DGHO: Parenteral nutrition, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	20 (74%)	13 (65%)	11 (85%)	7 (88%)	5 (83%)
yes	7 (26%)	7 (35%)	2 (15%)	1 (12%)	1 (17%)

Table 97 DGHO: Parenteral nutrition, placebo

Week	0	2	4	6	8
n	10	7	6	4	5
no	8 (80%)	5 (71%)	5 (83%)	4 (100%)	5 (100%)
yes	2 (20%)	2 (29%)	1 (17%)	0 (0%)	0 (0%)

Palliative home care service

Table 98 DGHO: Palliative home care service, bevacizumab

Week	0	2	4	6	8
n	27	20	13	8	6
no	23 (85%)	17 (85%)	13 (100%)	7 (88%)	6 (100%)
yes	4 (15%)	3 (15%)	0 (0%)	1 (12%)	0 (0%)

Table 99 DGHO: Palliative home care service, placebo

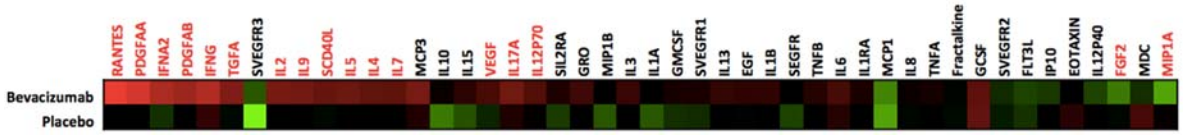
Week	0	2	4	6	8
n	10	7	6	4	5
no	9 (90%)	7 (100%)	6 (100%)	4 (100%)	5 (100%)
yes	1 (10%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)

11.4.1.8 Correlative analysis of cytokine/chemokine levels in the peripheral blood and malignant ascites

In a pilot experiment, the levels of 46 cytokines/chemokines and angiogenic factors were analyzed in pre- and post-treatment ascites samples from the first 8 consecutively enrolled patients (Figure 5). From these data, 17 cytokines showing the highest degree of regulation in the bevacizumab

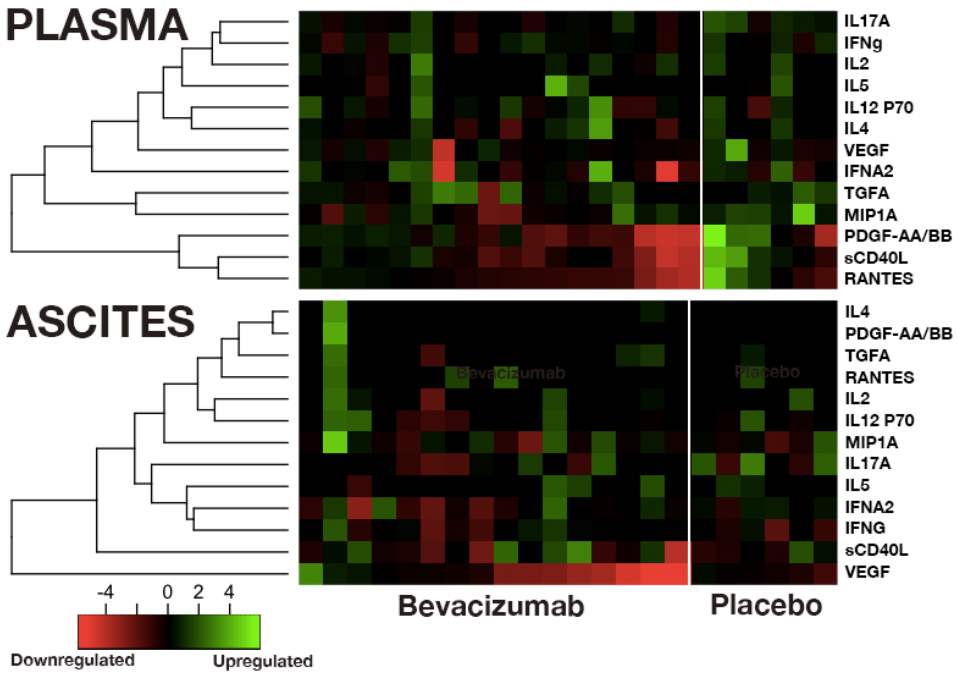
group and little regulation in the placebo group were selected for a comprehensive analysis of all 22 plasma and 24 ascites samples from 12 patients.

Figure 5 **Mean of fold changes in cytokine/chemokine expression levels after treatment with bevacizumab or placebo. *Red = cytokines selected for subsequent analyses*.**



Four cytokine assays (PDGF-AA, FGF-2, IL-7, IL-9) not meeting quality control criteria as a whole or for individual samples were excluded from the final analysis. Analyzing changes in cytokine expression levels for the 13 remaining cytokines/chemokines and angiogenic factors in plasma and ascites samples patterns clearly associated with bevacizumab treatment compared to placebo were observed (Figure 6).

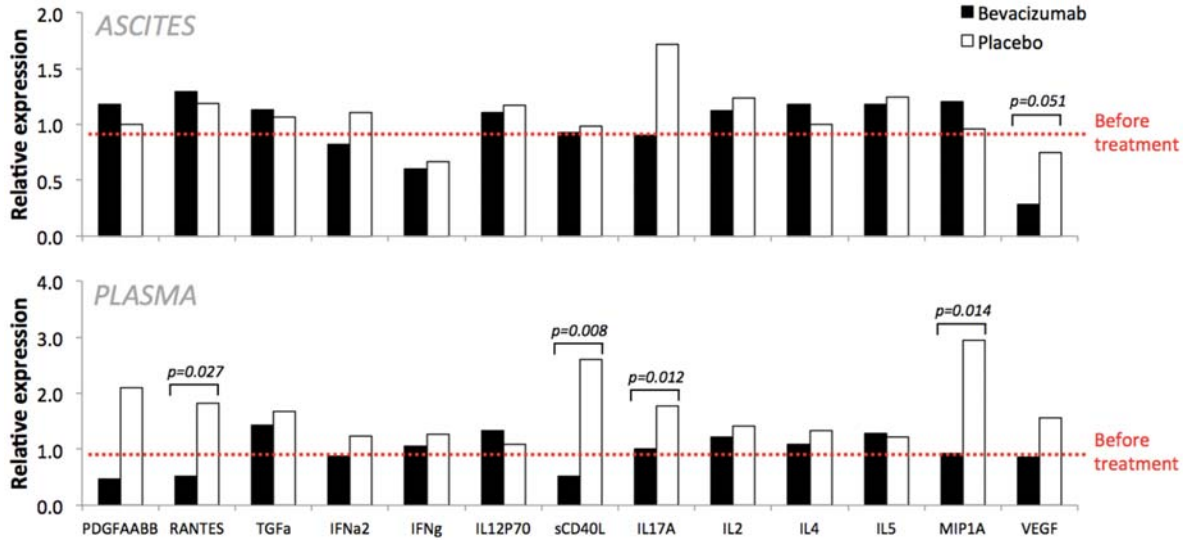
Figure 6 **Heatmap of log2 transformations of fold changes in cytokine levels in patients before and after treatment with bevacizumab of placebo.**



Importantly, a clear and specific reduction in VEGF levels in ascites samples after treatment with bevacizumab was observed (Figure 7), which almost reached statistical significance despite the small placebo cohort. None of the other cytokines/chemokines measured in the malignant ascites

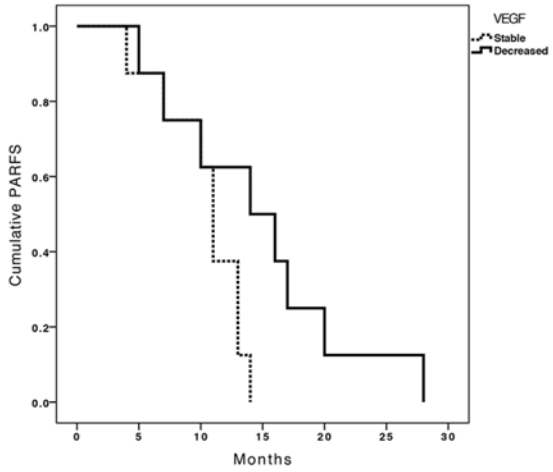
showed a change approaching statistical significance. In contrast to the findings obtained with ascites, no significant changes in VEGF levels were observed in the peripheral blood samples. However, we observed a significant upregulation of cytokines/chemokines RANTES, sCD40L, IL17A, and MIP1a at the later timepoints in the plasma of the placebo group (Figure 7) while cytokine/chemokine levels in the bevacizumab group remained mostly unchanged over time.

Figure 7 Mean expression levels of cytokines relative to the group's mean value before treatment (dotted line).



Finally, within the patient group treated with bevacizumab we observed a prolonged mean estimated paracentesis-free survival (ParFS) in those patients experiencing at least a two-fold reduction in ascites VEGF levels after treatment (14.6 months, 95% CI [9.5, 19.8]) compared to patients with stable VEGF levels (10.4 months, 95% CI [8.0, 12.7]) using logistic regression (Figure 8, p=0.061). We did not find changes in overall survival in patients with reduced compared to stable VEGF levels.

Figure 8 Kaplan-Meyer plot of cumulative ParFS in patients treated with bevacizumab stratified according to the extent of downregulation of VEGF in their ascites samples.



In summary, after intraperitoneal treatment with bevacizumab we observed an efficient downregulation of VEGF in the patients' malignant ascites. We further found that bevacizumab treatment stabilized expression of pro-inflammatory, and possibly tumor-promoting, cytokines/chemokines in the peripheral blood. Finally, patients with strong VEGF neutralization evidenced an improved ParFS.

11.4.2 Statistical/analytical issues

11.4.2.1 Adjustment for covariates

NA

11.4.2.2 Handling of dropouts or missing data

11.4.2.2.1 Protocol violations and early drop-outs: exclusions from analysis

Four patients did not receive any paracentesis with study drug administration after randomisation, and, thus, were excluded from all quantitative analyses (According to section *8.3.1 Intent-to-Treat Population* of the study protocol, Intent-to-treat population (ITT) for ParFS is defined to include all randomized patients with at least one day of follow-up after the initial paracentesis). Please refer to Table 5 for details on the above-mentioned four cases. According to the protocol these patients are likewise excluded from the safety analysis. Thus, the safety analysis set is identical to the ITT population.

11.4.2.2.2 Major protocol violations before reaching the primary endpoint

According to the pre-analysis meeting and blinded decision by the protocol steering group, no patient was withdrawn from the intention-to-treat (ITT) analysis due to a severe protocol violation. Therefore, the per-protocol population is identical to the intention-to-treat population.

11.4.2.3 Interim analysis and data monitoring

NA

11.4.2.4 Multi-centre studies

NA

11.4.2.5 Multiple comparison/multiplicity

NA

11.4.2.6 Use of an "efficacy subset" of patients

NA

11.4.2.7 Active-control studies intended to show equivalence

NA

11.4.2.8 Examination of subgroups

NA

11.4.3 Tabulation of individual response data

Please refer to Appendix 16.2.6 for a listing of individual efficacy response data.

11.4.4 Drug dose, drug concentration, and relationship to response

Only 20% of the patients received the maximum number of four applications of study medication, with almost equal increments of the study population receiving one, two or three applications (Table 100). There are no major differences between the randomization arms in this respect. Table 101 shows the number of paracenteses without study drug per patient.

Overall, 94 paracenteses out of 208 (45%) were not followed by the administration of Bevacizumab (78 out of 154, 51%) or placebo (16 out of 54, 31%). The difference is mainly due to the fact, that the three patients with an extraordinary large number of paracenteses are all located in the verum arm. In the vast majority of cases, study drug was not given due to a time span < 14 days since the last puncture (overall: 66/94, 70%; arm 1: 51/76, 67%; arm 2: 15/18, 83%).

In the Bevacizumab arm, treatment was declared to be temporarily discontinued in 12 cases (16%). The reasons were drug unavailability, pain, patient's wish, bad general condition, or catheter displacement. In the context of another 12 paracenteses, the drug had been permanently discontinued, usually as they took place after the 8 weeks medication period.

In the placebo arm, treatment was declared to be temporarily discontinued in only one case (6%), due to drug unavailability. A permanent discontinuation had happened in two cases (11%), after termination of the study medication period.

Table 100: Number of paracenteses with study medication per patient

Number of paracenteses	Bevacizumab	Placebo	Total
n	33	16	49
1	10 (30%)	6 (38%)	16 (33%)
2	7 (21%)	4 (25%)	11 (22%)
3	10 (30%)	2 (12%)	12 (24%)
4	6 (18%)	4 (25%)	10 (20%)

Table 101: Number of paracenteses without study medication per patient

Number of paracenteses	Bevacizumab	Placebo	Total
n	23	8	31
1	8 (35%)	4 (50%)	12 (39%)
2	5 (22%)	1 (12%)	6 (19%)
3	4 (17%)	1 (12%)	5 (16%)
4	3 (13%)	1 (12%)	4 (13%)
5	--	1 (12%)	1 (3%)
7	1 (4%)	--	1 (3%)
11	1 (4%)	--	1 (3%)
16	1 (4%)	--	1 (3%)

11.4.5 Drug-drug and drug-disease interactions
NA

11.4.6 By-patient displays
NA

11.4.7 Efficacy conclusions
No major difference between the bevacizumab and the control group were observed in paracentesis-free survival (ParFS), i.e. the time from randomization to either the second on-study paracentesis or to death, whichever occurred first. The ParFS median was 14.0 days in the bevacizumab group versus 10.5 days in the control group, the difference was not statistically

significant (p = 0.16). The median overall ParFS was 13.0 days which is in accordance with the median of 14 days reported by Parsons et al. (2008) for the control group within the non-ovarian cancer subpopulation.

Best response, defined as the longest paracentesis-free period within the 12-week main observation period, showed only a minimum advantage for the bevacizumab group with a median of 19 versus 17.5 days (p = 0.85).
A statistically significant difference (p < 0.0001) was observed when comparing the maximum interval during the treatment period (19 days) to the respective findings during the screening phase (10 days) in the bevacizumab group. This difference was larger than in the placebo group (17.5 vs. 11.5 days, p = 0.22). However, when interpreting the test result in the placebo group, one has to keep in mind the lower patient number – and therefore lower power – in the placebo arm.
When discussing the overall difference between the pre-study and on-study periods, one has to consider the fact of different definitions of the respective calculations. While the pre-study period requires the observation of at least two paracenteses, the later limit of the on-study period is often defined by the date of death or the end of the 12-week observation period. Moreover, a selection bias may play a role, since qualification for the study requires at least three paracenteses (later, after the amendment: two paracenteses, including the first therapeutic one). Thus, the recorded within-group difference may rather be a statistical artifact.

The median overall survival was 64 days in the bevacizumab group and 31.5 days in the placebo group (p = 0.31). While the medians seem to indicate a major difference, this may easily be caused by a mere play of chance due to the low patient numbers, as indicated by the widely overlapping confidence intervals.
The median overall survival observed in this study was almost identical to the median overall survival of about 50 days reported by Parsons et al. (2008) for the non-ovarian cancer subpopulation.

Assessing quality of life, the common and inevitable problem of QoL analysis in highly palliative situations occurred: firstly, comparisons, both between groups or different time points, suffer from an uncontrolled loss process, due to patient's death or inability to perform further visits or fill out the questionnaires. Thus the remaining data definitely reflect a positive selection. Secondly, this dropout mechanism leads to a loss of power of hypothesis testing. The quality of life analysis of FACIT-AI revealed no significant differences between baseline and week 2 and baseline and week 4, respectively.

Overall, the intraperitoneal bevacizumab application did not lead to an improvement of ParFS. However, the study was premature stopped after enrolling 53 of 72 patients due to slow recruitment. Therefore, data allow only a limited statement on efficacy.

12 SAFETY EVALUATION

12.1 Extent of exposure

See 11.4.4 “Drug dose, drug concentration and relationship to response”.

12.2 Adverse events (AEs)

12.2.1 Brief summary of adverse events

The adverse events reported according to the NCI Common Toxicity Criteria grading system were primarily analysed presenting the maximum severity grade by patient and SOC occurring during the study period. 103 for the experimental arm, and Table 104 for placebo, respectively, summarize the reported toxicity by body/organ system (cf. 102 for abbreviations) and standardized CTC term in alphabetical order.

Overall, a major increase in severe toxicity cannot be detected for the Bevacizumab arm. The proportion of patients with at least one grade 3 to 5 event occurring is similar with 20/33 (61%) in arm 1 and 11/16 (69%) in arm 2. As one might have expected, fatigue is the most prevalent adverse event: 39% in arm 1, which is somewhat higher than the 3/16 (19%) finding in the placebo group. Nausea (45%) and vomiting (36%) are other frequent adverse events on Bevacizumab, together with pain (33%). The respective rates in the placebo arm are somewhat lower with 31% nausea and 33% pain; remarkably, no cases of vomiting were reported in the control group.

All other CTC symptom categories occurred with a frequency below or equal to 25% only.

No case of hypertension was reported in either treatment arm. Proteinuria was rare with no events of severity grade 3 or 4. (One such event in the Bevacizumab arm was reported without severity grade.) One thromboembolic event of severity grade 3 or worse was reported in each arm (Bevacizumab: 3%; placebo: 6%). Hemorrhagic events of grade 2 or higher were not reported, neither in the antibody nor in the placebo arm.

2 out of the 7 grade 5 events reported (4 in arm 1 and 3 in arm 2) were due to the progressive underlying disease.

Table 102: Organ system according to NCI CTCAE, with abbreviations

BLO	BLOOD/BONE MARROW
COA	COAGULATION
CON	CONSTITUTIONAL SYMPTOMS
DEA	DEATH
DER	DERMATOLOGY/SKIN
GAS	GASTROINTESTINAL
HEM	HEMORRHAGE/BLEEDING
HEP	HEPATOBILIARY/PANCREAS
INF	INFECTION
LYM	LYMPHATICS
MET	METABOLIC/LABORATORY
NEU	NEUROLOGY
OCU	OCULAR/VISUAL
PAI	PAIN
PUL	PULMONARY/UPPER RESPIRATORY
REN	RENAL/GENITOURINARY
SUR	SURGERY/INTRA-OPERATIVE INJURY
VAS	VASCULAR

12.2.2 Display of adverse events

Table 103: Maximum CTC grade by patient and organ system during the study period, Bevacizumab arm (n = 33)

Organ system	Adverse Event	CTC toxicity grading				
		grade 1	grade 2	grade 3	grade 4	grade 5
BLO	Hemoglobin	--	5 (15%)	--	--	--
BLO	Leukocytes (total WBC)	--	--	2 (6%)	--	--
BLO	Platelets	2 (6%)	--	--	--	--
COA	INR (International Normalized Ratio of prothrombin time)	--	--	1 (3%)	--	--

CON	Constitutional Symptoms - Other	--	2 (6%)	4 (12%)	1 (3%)	--
CON	Fatigue (asthenia, lethargy, malaise)	4 (12%)	5 (15%)	4 (12%)	--	--
CON	Insomnia	1 (3%)	1 (3%)	1 (3%)	--	--
CON	Rigors/chills	--	1 (3%)	--	--	--
CON	Weight loss	1 (3%)	2 (6%)	--	--	--
DEA	Death not associated with CTCAE term	--	--	--	--	2 (6%)
DER	Hair loss/alopecia (scalp or body)	1 (3%)	--	--	--	--
DER	Induration/fibrosis (skin and subcutaneous tissue)	1 (3%)	--	--	--	--
DER	Pruritus/itching	--	--	1 (3%)	--	--
DER	Rash/desquamation	1 (3%)	--	--	--	--
DER	Skin breakdown/decubitus ulcer	--	1 (3%)	--	--	--
DER	Ulceration	1 (3%)	--	--	--	--
GAS	Anorexia	3 (9%)	1 (3%)	3 (9%)	--	--
GAS	Ascites (non-malignant)	--	1 (3%)	--	--	--
GAS	Constipation	1 (3%)	4 (12%)	1 (3%)	--	--
GAS	Dehydration	--	1 (3%)	--	--	--
GAS	Diarrhea	3 (9%)	2 (6%)	--	--	--
GAS	Dysphagia (difficulty swallowing)	--	1 (3%)	--	--	--
GAS	Gastrointestinal - Other	1 (3%)	--	3 (9%)	--	--
GAS	Heartburn/dyspepsia	1 (3%)	1 (3%)	--	--	--
GAS	Mucositis/stomatitis (clinical exam)	--	--	1 (3%)	--	--
GAS	Nausea	4 (12%)	3 (9%)	2 (6%)	--	--
GAS	Vomiting	5 (15%)	3 (9%)	3 (9%)	1 (3%)	--
HEM	Hematoma	1 (3%)	--	--	--	--
HEM	Hemorrhage, pulmonary/upper respiratory	1 (3%)	--	--	--	--
INF	Infection - Other	--	1 (3%)	1 (3%)	--	--
INF	Infection with unknown ANC	--	2 (6%)	2 (6%)	--	--
LYM	Edema: limb	4 (12%)	--	--	--	--
LYM	Lymphatics - Other	1 (3%)	--	--	--	--
MET	Calcium, serum-low (hypocalcemia)	--	--	--	1 (3%)	--

MET	Glucose, serum-high (hyperglycemia)	--	--	--	1 (3%)	--
MET	Metabolic/Laboratory - Other	--	--	1 (3%)	--	--
MET	Proteinuria	1 (3%)	--	--	--	--
MET	Sodium, serum-low (hyponatremia)	1 (3%)	--	--	--	--
NEU	CNS cerebrovascular ischemia	--	--	--	--	1 (3%)
NEU	Mood alteration	--	1 (3%)	1 (3%)	--	--
NEU	Neuropathy: sensory	2 (6%)	--	--	--	--
OCU	Keratitis (corneal inflammation /corneal ulceration)	--	1 (3%)	--	--	--
PAI	Pain	4 (12%)	5 (15%)	2 (6%)	--	--
PAI	Pain - Other	--	--	1 (3%)	--	--
PUL	Cough	1 (3%)	--	--	--	--
PUL	Dyspnea (shortness of breath)	2 (6%)	2 (6%)	2 (6%)	1 (3%)	--
PUL	Pleural effusion (non-malignant)	--	1 (3%)	--	--	--
PUL	Pneumothorax	1 (3%)	--	--	--	--
REN	Renal failure	--	--	1 (3%)	--	--
REN	Renal/Genitourinary - Other	1 (3%)	--	--	--	--
REN	Urinary retention (including neurogenic bladder)	1 (3%)	1 (3%)	--	--	--
VAS	Thrombosis/embolism (vascular access-related)	1 (3%)	--	--	--	--
VAS	Thrombosis/thrombus/embolism	--	--	--	--	1 (3%)

Table 104: Maximum CTC grade by patient and SOC during the study period, placebo arm (n = 16)

		CTC toxicity grading				
SOC	**Adverse Event**	**grade 1**	**grade 2**	**grade 3**	**grade 4**	**grade 5**
BLO	Hemoglobin	1 (6%)	3 (19%)	--	--	--
BLO	Platelets	--	--	1 (6%)	--	--
COA	Coagulation - Other	--	1 (6%)	--	--	--
CON	Constitutional Symptoms - Other	--	1 (6%)	1 (6%)	--	--
CON	Fatigue (asthenia, lethargy, malaise)	2 (12%)	1 (6%)	--	--	--
CON	Fever (in the absence of neutropenia, where neutropenia is d	--	--	1 (6%)	--	--

SOC	Adverse Event	CTC toxicity grading				
		grade 1	grade 2	grade 3	grade 4	grade 5
CON	Insomnia	1 (6%)	1 (6%)	--	--	--
DEA	Death not associated with CTCAE term	--	--	--	--	1 (6%)
GAS	Anorexia	1 (6%)	1 (6%)	1 (6%)	--	--
GAS	Constipation	1 (6%)	--	--	--	--
GAS	Diarrhea	--	1 (6%)	1 (6%)	--	--
GAS	Distension/bloating, abdominal	--	1 (6%)	--	--	--
GAS	Gastrointestinal - Other	1 (6%)	--	--	--	--
GAS	Ileus, GI (functional obstruction of bowel, i.e., neuroconst	--	1 (6%)	1 (6%)	--	--
GAS	Mucositis/stomatitis (functional/symptomatic)	1 (6%)	--	--	--	--
GAS	Nausea	3 (19%)	2 (12%)	--	--	--
HEM	Hematoma	1 (6%)	--	--	--	--
HEP	Liver dysfunction/failure (clinical)	--	--	1 (6%)	--	1 (6%)
INF	Infection - Other	--	1 (6%)	--	--	--
INF	Infection with unknown ANC	--	2 (12%)	--	--	--
LYM	Edema: limb	1 (6%)	1 (6%)	--	--	--
LYM	Lymphatics - Other	--	--	--	1 (6%)	--
MET	Potassium, serum-high (hyperkalemia)	1 (6%)	--	--	--	--
MET	Proteinuria	1 (6%)	--	--	--	--
MET	Sodium, serum-low (hyponatremia)	--	--	1 (6%)	--	--
NEU	Confusion	--	1 (6%)	--	--	--
NEU	Mood alteration	--	4 (25%)	--	--	--
NEU	Neuropathy: cranial	--	1 (6%)	--	--	--
NEU	Neuropathy: sensory	--	--	1 (6%)	--	--
NEU	Seizure	--	--	--	1 (6%)	--
PAI	Pain	1 (6%)	1 (6%)	2 (12%)	--	--
PUL	Dyspnea (shortness of breath)	1 (6%)	2 (12%)	--	--	--
PUL	Pleural effusion (non-malignant)	--	--	1 (6%)	--	--
REN	Renal failure	--	--	--	--	1 (6%)
SUR	Intra-operative Injury - Other	1 (6%)	--	--	--	--
VAS	Thrombosis/thrombus/embolism	--	--	1 (6%)	--	--

The following adverse events were reported without a severity grade (Table 105):

Table 105: Adverse events reported without a CTCAE severity grade

Study arm	SOC	Adverse Event
Bevacizumab	GAS	Vomiting
Bevacizumab	NEU	Neuropathy: motor
Bevacizumab	MET	Proteinuria
Bevacizumab	PUL	Dyspnea (shortness of breath)

12.2.3 Analysis of adverse events

No major increase in severe toxicity could be detected for the Bevacizumab arm. The proportion of patients with at least one grade 3 to 5 event occurring was similar with 20/33 (61%) in the Bevacizumab arm and 11/16 (69%) in the placebo arm.

As one might have expected, fatigue was the most prevalent adverse event: 39% in the Bevacizumab arm, which was somewhat higher than the 19% finding in the placebo arm.

Nausea (45%) and vomiting (36%) were other frequent adverse events on Bevacizumab, together with pain (33%). The respective rates in the placebo arm were somewhat lower with 31% nausea and 33% pain; remarkably, no cases of vomiting were reported in the control group.

Altogether, intraperitoneal Bevacizumab was well tolerated.

12.2.4 Listing of adverse events by patient

Please refer to Appendix 16.2.7 for adverse events listings by patient.

12.3 Deaths, other serious adverse events, and other significant adverse events

12.3.1 Listing of deaths, other serious adverse events and other significant adverse events

12.3.1.1 Deaths

In the evaluable patient group receiving at least one application of study drug, the date of death is recorded in 46/49 (94%). The reason for death was progressive tumor disease in 44 out of these (96%), with the cause of demise in the remaining two patients described in Table 106.

Table 106: Causes of death

Pat. no.	Reason for early drop-out before any study treatment
# 1201 (Bevacizumab)	Left sided cerebrovascular ischemia
# 2403 (Bevacizumab)	Multiorgan failure due to bacterial peritonitis

12.3.1.2 Other serious adverse events

A total of 32 SAEs were reported during the course of the study. The most frequent SAE was Constitutional symptoms – other: bad general condition, occurring in 2 patients in the bevacizumab arm and in 1 patient in the placebo arm, followed by vomiting, occurring in 2 patients in the bevacizumab arm and renal failure, Thrombosis/thrombus/embolism and Infection – other: Peritoneum occurring in 1 patient in each arm.

17 SAEs occurred in the bevacizumab arm, and 11 SAEs were reported in patients in the placebo arm.

The following SAEs were reported in patients who were not included in the safety analysis set, as they have not received any study medication: Sodium, serum low (hyponatremia), Infection – other: PULMONARY/UPPER RESPIRATOY – Lung (pneumonia), Muscle weakness – Whole body/generalized. Another SAE was reported as tumor progression which is also not included in the safety analysis set, as this is determined in the study protocol as part of the efficacy assessment and not to be reported as SAE.

These SAEs are not included in Table 107.

Table 107: Serious adverse events by patient and organ system

Organ system	SAE	Bevacizumab (n = 33)	Placebo (n = 16)	Total (n = 49)
CON	Insomnia	1 (3%)	-	1 (2%)
CON	Constitutional symptoms – other: bad general condition	2 (6%)	1 (6%)	3 (6%)
GAS	Ascites	1 (3%)	-	1 (2%)
GAS	Vomiting	2 (6%)	-	2 (4%)
GAS	Gastrointestinal – other: duodenal obstruction	1 (3%)	-	1 (2%)
GAS	Stricture/stenosis (including anastomotic) bile duct	-	1 (6%)	1 (2%)

GAS	Ileus	-	1 (6%)	1 (2%)
HEP	Liver dysfunction	-	1 (6%)	1 (2%)
INF	Infection – other: Blood	1 (3%)	-	1 (2%)
INF	Infection – other: gastrointestinal – Anal/perianal	1 (3%)		1 (2%)
INF	Infection – other: Peritoneum	1 (3%)	1 (6%)	2 (4%)
MET	Calcium, serum low (hypocalcemia)	1 (3%)	-	1 (2%)
MET	Glucose, serum high (hyperglycemia)	1 (3%)	-	1 (2%)
NEU	CNS cerebrovascular ischemia	1 (3%)	-	1 (2%)
NEU	Neuropathy: motor	-	1 (6%)	1 (2%)
NEU	Seizure	-	1 (6%)	1 (2%)
PAI	Pain – unspecified	1 (3%)	-	1 (2%)
PAI	Pain – Gastrointestinal - stomach	-	1 (6%)	1 (2%)
PAI	Pain - peritoneum	1 (3%)	-	1 (2%)
PUL	Pleural effusion	-	1 (6%)	1 (2%)
REN	Renal failure	1 (3%)	1 (6%)	2 (4%)
VAS	Thrombosis/thrombus/embolism	1 (3%)	1 (6%)	2 (4%)

12.3.1.3 Other significant adverse events

3 out of 33 patients in the bevacizumab arm and no patients out of 16 patients in the placebo arm terminated the treatment phase prematurely due to adverse events (cf. section 10.1.2, Table 4). These events were reported as pain in legs due to open wounds in one patient (patient 1407, SAE) and bacterial peritonitis in 2 patients (patient 2402, SAE, and patient 2403, AE).

12.3.2 Narratives of deaths, other serious adverse events and certain other significant adverse events

12.3.2.1 Narratives of deaths

CNS cerebrovascular ischemia (patient 1201):

The 71 year old female patient was hospitalized on 10.02.2011 with a suspected transient ischemic attack (TIA). The first administration of study drug was on 08.02.2011, this was also the last administration prior to the event. As possible cause of the SAE, the (blinded) study medication was named by the investigator. The patient has been treated with bevacizumab. Relevant medical history included coronary heart disease since 2003, left-heart failure since 2003, and arterial hypertension. Concomitant medication included enalapril, ASS, atorvastatin, bisoprolol, and torasemid.

At first, a clinical improvement could be seen, but after one day, the patient showed clinical deterioration, and the cardial computer tomography (CCT) which was normal initially after the hospitalization revealed a large cerebrovascular insult (CVI), which lead to death on 14.02.2011.

Infection other - peritoneum (patient 2403):

On 10.07.2012, a deterioration of the general condition of the 49 year old male patient with confusion and sudden vomiting was noticed and hospitalization was prolonged on 11.07.2012. A bacterial peritonitis was suspected, because the ascites turned turbid and the patient developed progredient leukocytosis. As possible causes of this SAE, the underlying disease peritoneal carcinomatosis, the blinded study medication and the peritoneal drainage since 30.06.2012 were given.

The patient has been treated with bevacizumab. The last administration prior to the SAE has been on 02.07.2012 (this has also been the first administration). On 17.07.2012, a microbiological examination revealed E.coli in the ascites fluid. The patient was treated with systemic antibiotic therapy and lavanid infusions of the peritoneal drainage. After two days, the performance status of the patient improved and the ascites fluid became clear.

However, the peritoneal carcinomatosis caused a complete obstruction of the small intestine and the patient showed clinical symptoms of sepsis, probably due to bacteria penetrating the wall of the small intestine. Anuria occurred on 22.07.2012.

The patient died on 23.07.2012 due to multi-organ failure. No correlation to study medication was seen by the investigator.

12.3.2.2 Narratives of other serious adverse events

Infection – Other: Blood (patient 0101)

The 58 year old male patient was hospitalized on 10.07.2011 due to signs of asthenia and sepsis caused by progression of peritoneal effusion which was assessed by the investigator as suspected tumor progression. Study treatment was first administered on 16.06.2011, the last administration before the SAE was on 30.06.2011. Infection persist under treatment with antibiotic drugs vancomycin, metronidazole, tacobactame and zienam and improved after changing antibiotic therapy to metronidazole, moxifloxacine and ceftibutene. Patient was discharged in improved condition on 19.07.2011.

Calcium, serum-low (patient 0104)

The 51 year old female patient was hospitalized on 18.04.2012 due to symptomatic hypocalcemia, hypoalbuminemia and increased inflammatory markers. The cause of the event was judged by the investigator as result of the underlying tumor disease and due to treatment with diuretic drugs (furosemide). Relevant medical history includes diabetes mellitus type IIb, hypertension, renal failure and peripheral neuropathy. Study treatment was first administered on 21.03.2012, the last administration before the SAE was on 11.04.2012. After i.v. hydration and substitution of calcium, albumin and i.v. antibiotics the outcome was judged as improved.

Neuropathy: motor (patient 0105)

The 66 year old male patient was presented at emergency unit of the hospital on 08.12.2012 with paraparesis. The MRI showed a compression of the spinal canal due to osseous metastasis of BWK 2. After performing a laminectomy and spinal fusion on 11.12.2012 the patient was transferred to intensive-care unit where he developed a hepatorenal-syndrome which resulted in death on 17.12.2012 Study treatment was first and only administered on 04.12.2012. The Investigator judged the SAE as not related to the study drug but due to underlying colorectal carcinoma.

Renal failure (patient 0201)

The 64 year old female patient was hospitalized on 19.07.2010 due to increased CRP and creatinine values and signs of acute renal failure (first shown on 16.07.2010). Previous treatment with oral antibiotic drugs (ciprofloxacine, sultamicilline) showed no significant improvement. The patient received i.v. antibiosis and hydration and the patient’s condition improved. The cause of the SAE was judged as general immunosuppression due to underlying malignancy. Study treatment was first administered on 29.06.2010, the last administration before the SAE was on 16.07.2010. No relation to the study treatment was evaluated by the investigator.

Gastrointestinal other: Duodenal obstruction (patient 0201)

The 64 year old female patient was hospitalized on 19.07.2010 due to increased CRP and creatinine values and signs of acute renal failure. On 20.07.2010 after i.v. antibiosis with ciprofloxacine and sultamicilline and hydration the patient started feculent vomiting. A duodenoscopy on 21.07.2010 showed duodenal obstruction and a decompression probe was implanted. The patient died on 30.07.2010 due to tumor progression of the underlying pancreatic

cancer. Study treatment was first administered on 29.06.2010, the last administration before the SAE was on 16.07.2010. No relation to the study treatment was evaluated by the investigator.

Renal failure (patient 0203):

The 47 year old male patient developed acute renal failure and was hospitalized on 04.10.2011. No concurrent conditions were reported. Medical history included metastatic pancreas carcinoma, alcohol abuse, nicotine abuse, and arterial hypertension. On 28.09.2011, the patient started study therapy. The investigator assessed the SAE as related to (blinded) study medication. On 12.11.2011, the patient died, the cause of death was reported as progression of the underlying disease.

This event was not evaluated as a SUSAR initially, however, based on the review of the Annual Safety Report dated 03.03.2013 by the PEI, the SAE was reported as a SUSAR belated.

After unblinding the study, it appeared that the patient has been treated with placebo.

Glucose, serum-high (hyperglycemia) (patient 0204)

The 62 year old female patient was hospitalized on 18.12.2011 due to hypergycemia caused by pre-existing diabetes mellitus and changeover to parenteral feeding. After application of 12 I.E. insulin, the patient's condition and laboratory values improved and the patient was discharged. The SAE was resolved on 23.12.2011. The first administration of study medication was on 25.11.2011 and the last administration before the SAE was on 09.12.2011. No relation to the study treatment was evaluated by the investigator.

Ileus (patient 0302)

The 50 year old female patient was hospitalized on 19.12.2011 due to relapsing vomiting and diarrhea. The patient's condition improved under treatment with i.v. metoclopramide. The first and only application of study medication was given on 08.12.2011. The causality of the SAE was judged by the investigator as result of the underlying peritoneal carcinomatosis. The outcome of the event was resolved with sequelae on 23.12.2011. No relation to study medication was evaluated by the investigator.

Constitutional symptoms – other: Bad general condition (patient 0502)

The 70 year old male patient was hospitalized on 11.04.2012 due to his bad general health condition. The first administration of study medication was given on 09.03.2012 and the last dose of study medication prior to the SAE on 05.04.2012. The patient was taken off study on 11.04.2012

and died on 24.04.2012 due to tumor progression with massive recurrent ascites of his underlying colon cancer. The Investigator judged the SAE as not related to the study medication.

Constitutional symptoms – other: Bad general condition (patient 0503)

The 53 year old male patient was hospitalized on 02.01.2013 due to his bad general health condition. The first administration of study medication was given on 14.12.2012 and the last dose prior to the SAE was administered on 27.12.2012. The patient was taken off study on 02.01.2013 and died on 12.01.2013 as a result of his underlying pancreatic cancer. No further information are available. The Investigator judged the SAE as not related to the study medication.

Pleural effusion (patient 0702)

The 65 year old male patient was hospitalized for first application of study medication on 25.02.2011. The patient developed recurrent pleural effusion on 26.02.2011. The patient was oxidated and received pleural punction on 07.03.2011. The cause of the event are pre-existing pleural effusions (first discovered 24.02.2011) and judged as not related to study medication by the investigator. The outcome of the event is persisting.

Thrombosis/thrombus/embolism (patient 0705)

The 47 year old male patient was hospitalized on 07.02.2012 due to deterioration of general health condition and progression of his underlying tumor disease. The patient was found dead on 08.02.2012 and all clinically (no diagnostic) symptoms lead to fatal pulmonary embolism since the patient reported shortness of breath on the same date. The first and only administration of study medication was performed on 20.01.2012. No relation to the study medication was evaluated by the investigator.

Constitutional symptoms – other: Bad general condition (patient 0707)

The 52 year old female patient was hospitalized in 19.04.2012 due to deterioration of the patient's general health condition (ECOG 3). The first administration of study medication was performed on 28.03.2012 and the last dose prior to the SAE was given on 11.04.2012. The patient died on 28.04.2012 due to tumor progression of the underlying gastrointestinal cancer. The Investigator judged the SAE as not related to the study medication.

Thrombosis/thrombus/embolism (patient 1001)

The 46 year old male patient was hospitalized on 23.10.2010 after a CT scan from 22.10.2010 showed signs of pulmonary embolism and the patient suffered from dyspnea. Pre-existing

pulmonary embolism was first discovered on 16.07.2010 and the patient received fraxiparine since July 2010. The first administration of study medication was given on 01.09.2010 and the last dose prior to the SAE was administered on 13.10.2010. No further information are available and the outcome is unknown. No relation to the study treatment was evaluated by the investigator.

Vomiting (patient 1405)

The 64 year old male patient was hospitalized on 14.08.2012 due to vomiting grade II. The first and only study medication was administered on 07.08.2012. The patient received parenteral nutrition and was discharged on 24.08.2012 due to his final tumor stage and a general health deterioration. The possible cause of the SAE was judged by the investigator as result of ascites.
Other concomitant medications were spironolactone, dipyrone, omeprazole, furosemide, pancreatine, metaraminol, dalteparine, ceftriaxone, morphine and lorazepam.

Ascites (patient 1407)

The 75 year old male patient was hospitalized on 01.08.2013 due to massive ascites (>7 litres). First paracentesis on 01.08.2013 stopped after 6 litres due to risk of hepato-renal syndrome. The patient received human-albumin and ringer-lactacte solution and second paracentesis on 02.08.2013. The first administration of study medication was also given on 02.08.2013 and the patient was discharged on 03.08.2013. Possible cause of the event is judged as pre-existent disease.

Infection – other: GASTROINTESTINAL Anal/perianal (patient 1407)

The 75 year old male patient was hospitalized on 18.10.2013 due to perineal abscess excision. The first administration of study medication was given on 02.08.2013 and the last dose prior to the SAE was given on 12.09.2013. Study treatment was temporarily interrupted on 12.09.2013. The possible cause of the event is unknown and the SAE was resolved on 20.10.2013.

Pain - unspecified (patient 1407):

The 75 year old male patient was hospitalized on 24.09.2013 due to pain grade 2 in both legs due to open wounds. Study treatment was first administered on 02.08.2013, the last administration before the SAE was on 12.09.2013. Study treatment was stopped on 25.09.2013. The investigator judged the SAE as being possibly related to study drug. The patient has received bevacizumab. The pain situation improved only slowly and the patient was taken off the study.

Pain – GASTROINTESTINAL - Stomach (patient 1902)

The 74 year old female patient was hospitalized on 06.06.2013 1:30 a.m. due to acute epigastric pain. The first administration of study medication was performed on 10.05.2013 and the last dose prior to the SAE was given on 27.05.2013. After consuming hydrotalcit and ibuprofen the pain vanished slowly. The investigator judged the SAE as possibly related to the (blinded) study medication and maybe due to gastric or duodenal ulcer. The patient received bevacizumab.The SAE outcome was judged as resolved on 12.06.2013.

Stricture/stenosis, (including anastomotic) Bile duct (patient 1902)

The 74 year old female patient was hospitalized on 01.08.2013 for the planned paracentesis according to protocol but due to increase of total bilirubin and liver parameters and deterioration of the general health condition a MRI of the abdomen was performed. A bile duct stenosis was discovered and an ERCP with papillotomy with stent implantation was performed on 06.08.2013. The patient was discharged on 07.08.2013 by her own decision and the SAE was evaluated by the investigator as resolved on the same date. The causality of this SAE was judged by the investigator as result of the underlying colorectal cancer with progressive liver metastasis. The first administration of study medication was performed on 10.05.2013 and the last dose prior to the SAE was given on 04.07.2013. No relation to the study treatment was evaluated by the investigator.

Vomiting (patient 2301)

The 44 year old female patient was hospitalized on 13.06.2010 due to worsening of emesis and asthenia. The event became serious on 16.06.2010. The first and only administration of study medication was performed on 02.06.2010. The causality of the SAE was judged by the investigator as result of the underlying gastric cancer. Despite parenteral nutrition the patient's general health condition deteriorated and the patient died on 25.06.2010. No relation to the study treatment was evaluated by the investigator.

Seizure (patient 2401)

The 62 year old male patient developed focal seizures on 18.08.2010. The first and only application of study medication was given on 17.08.2010. The causality of the SAE was judged by the investigator as result of a pre-existing hepato-renal syndrome and ascites. Relevant medical history involved alcoholic liver cirrhosis. No relation to the study treatment was evaluated by the investigator.

Liver dysfunction (patient 2401)

The 62 year old male patient died on 26.08.2010 from liver insufficiency. The first and only application of study medication was given on 17.08.2010. The patient developed a hepato-renal syndrome after release of 4,5 l ascites. Relevant medical history involved alcoholic liver cirrhosis. The causality of the SAE was judged by the investigator as result of a pre-existing hepato-renal syndrome and maybe also due to progression of the underlying tumor disease. The investigator evaluated the relation of the SAE to the study treatment as not likely.

Pain – peritoneum (patient 2402)

The 52 year old female patient was hospitalized on 03.06.2012 for planned paracentesis and application of study medication. The patient suffered from abdominal pain, vomiting and a bad general health condition. The event became serious on 05.06.2012. The causality of this SAE was judged by the investigator as result of pre-existing subileus and peritoneal carcinomatosis as well as probably due to implanted permanent peritoneal drainage. The first and only administration of study medication was performed on 04.06.2012 and temporarily interrupted on 18.06.2012.The outcome of the SAE was evaluated as resolved on 09.07.2012.

Insomnia (patient 2403)

The 49 year old male patient was hospitalized on 09.07.2012 due to insomnia caused by uncontrolled cessation of opioid medication. The first and only administration of study medication was given on 02.07.2012 and the patient's tumor related pain decreases substantially after study treatment that he tried to reduce his opioid medication on 04.07.2012 the first time. The investigator judged the SAE as not related to study medication and the event was resolved on 11.07.2012.

Infection other - peritoneum (patient 2602)

The 61 year old female patient was hospitalized due to signs of fatigue, nausea and anorexia. Ascites punction on 04.10.2012 showed increased WBC and the diagnosis bacterial peritonitis. Patient was treated with antibiotic drugs and the SAE was resolved on 19.10.2012. The causality of the event was judged by the investigator as result of the underlying ascites and tumor disease and probably due to paracentesis procedure. The first administration of study medication was given on 23.08.2012 and the last dose prior to the SAE was given on 04.10.2012. No relation to the study medication was evaluated by the investigator.

12.3.3 Analysis and discussion of deaths, other serious adverse events and other significant adverse events

Due to the severity of the underlying disease, most patients died of disease progression. Death due to (serious) adverse events was rare, even though it occurred only in the bevacizumab arm. Due to a naturally higher toxicity of the verum compared to placebo, this is not unexpected.
As the number of patients in the bevacizumab arm was approximately twice the number of patients in the placebo arm, it is not surprising that more SAEs were reported in the bevacizumab arm.
None of the SAEs was unexpected and therefore not evaluated as being a SUSAR. However, based on the review of the Annual Safety Report dated 03.03.2013 by the PEI, the SAE was reported as a SUSAR belated.
After unblinding the study, it appeared that the patient has been treated with placebo.

12.4 Clinical laboratory evaluation

Introductory remark: All analyses of laboratory measurements are based on a categorical assessment of the investigators, using a three-item score:

- normal
- abnormal, not clinically significant
- abnormal, clinically significant

In fact, the third category was assigned to only very few findings (e.g. only three out of 612 individual measurements at baseline). This may be partly due to a misspelling in the case report forms. Therefore, this sub-categorisation of abnormal findings should be interpreted with caution. The comparative analysis to baseline (in section 11.2) combines the two categories for the initial findings.

12.4.1 Listing of individual laboratory measurements by patient and each abnormal laboratory value

NA

12.4.2 Evaluation of each laboratory parameter

12.4.2.1 Laboratory values over time

12.4.2.1.1 Baseline

12.4.2.1.1.1 Hematology

The baseline values of blood counts, measured before the first application of study drug, are provided in Table 108. While most patients suffer from anemia, white blood and platelet counts are within normal limits in the majority of patients.

Table 108 Assessment of blood counts at baseline

Parameter	Finding	Bevacizumab	Placebo	Total
Hemoglobin	n	32	14	46
	normal	6 (19%)	2 (14%)	8 (17%)
	abnormal, ncs	25 (78%)	11 (79%)	36 (78%)
	abnormal, cs	1 (3%)	1 (7%)	2 (4%)
Platelets	n	32	14	46
	normal	23 (72%)	7 (50%)	30 (65%)
	abnormal, ncs	9 (28%)	7 (50%)	16 (35%)
	abnormal, cs	--	--	--
Leukocytes	n	32	14	46
	normal	19 (59%)	6 (43%)	25 (54%)
	abnormal, ncs	13 (41%)	8 (57%)	21 (46%)
	abnormal, cs	--	--	--
Neutrophils	n	26	14	40
	normal	17 (65%)	8 (57%)	25 (62%)
	abnormal, ncs	9 (35%)	6 (43%)	15 (38%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

12.4.2.1.1.2 Clinical chemistry

The baseline assessments of the ten clinical chemistry items required by the protocol are provided in Table 109, as available. The high rates of abnormal findings correspond to the highly palliative clinical situation of the patients. Worthy of note, total protein and alkaline phosphatase show very high frequencies of abnormality. Some observed baseline differences between the randomization arms are due to the small patient numbers.

Table 109 Assessment of clinical chemistry at baseline

Parameter	Finding	Bevacizumab	Placebo	Total
Creatinine	n	32	14	46
	normal	21 (66%)	5 (36%)	26 (57%)
	abnormal, ncs	11 (34%)	8 (57%)	19 (41%)
	abnormal, cs	--	1 (7%)	1 (2%)
Total bilirubin	n	31	14	45
	normal	23 (74%)	11 (79%)	34 (76%)
	abnormal, ncs	8 (26%)	3 (21%)	11 (24%)
	abnormal, cs	--	--	--
Total protein	n	27	13	40
	normal	5 (19%)	4 (31%)	9 (22%)
	abnormal, ncs	22 (81%)	9 (69%)	31 (78%)
	abnormal, cs	--	--	--
ASAT / GOT	n	31	14	45
	normal	15 (48%)	6 (43%)	21 (47%)
	abnormal, ncs	16 (52%)	8 (57%)	24 (53%)
	abnormal, cs	--	--	--
ALAT / GPT	n	31	14	45
	normal	19 (61%)	11 (79%)	30 (67%)
	abnormal, ncs	12 (39%)	3 (21%)	15 (33%)
	abnormal, cs	--	--	--
LDH	n	27	14	41
	normal	14 (52%)	5 (36%)	19 (46%)
	abnormal, ncs	13 (48%)	9 (64%)	22 (54%)
	abnormal, cs	--	--	--
Alkaline phosphatase	n	30	13	43
	normal	8 (27%)	2 (15%)	10 (23%)
	abnormal, ncs	22 (73%)	11 (85%)	33 (77%)
	abnormal, cs	--	--	--
Creatinine clearance	n	29	14	43
	normal	22 (76%)	6 (43%)	28 (65%)
	abnormal, ncs	7 (24%)	8 (57%)	15 (35%)
	abnormal, cs	--	--	--
INR	n	32	11	43
	normal	27 (84%)	9 (82%)	36 (84%)

	abnormal, ncs	5 (16%)	2 (18%)	7 (16%)
	abnormal, cs	--	--	--
aPTT	n	32	11	43
	normal	22 (69%)	10 (91%)	32 (74%)
	abnormal, ncs	10 (31%)	1 (9%)	11 (26%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

12.4.2.1.2 During the course of therapy

12.4.2.1.2.1 Hematology

All hematology assessments during the course of therapy until week 12 (if reached) (Tables 110 - 121) are analyzed in three ways: firstly, all observations at the respective visits are taken into account. In two additional tables, the respective findings are sub-divided according to the normal/abnormal status of the respective laboratory value of the respective patient at the baseline visit (if available).

Table 110 Assessment of Hb during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	24	9	33
	normal	5 (21%)	1 (11%)	6 (18%)
	abnormal, ncs	17 (71%)	8 (89%)	25 (76%)
	abnormal, cs	2 (8%)	--	2 (6%)
Week 4	n	17	6	23
	normal	4 (24%)	2 (33%)	6 (26%)
	abnormal, ncs	13 (76%)	4 (67%)	17 (74%)
	abnormal, cs	--	--	--
Week 6	n	14	6	20
	normal	4 (29%)	3 (50%)	7 (35%)
	abnormal, ncs	10 (71%)	2 (33%)	12 (60%)
	abnormal, cs	--	1 (17%)	1 (5%)
Week 8	n	11	5	16
	normal	4 (36%)	3 (60%)	7 (44%)
	abnormal, ncs	7 (64%)	2 (40%)	9 (56%)
	abnormal, cs	--	--	--
Week 12	n	11	3	14

	normal	3 (27%)	1 (33%)	4 (29%)
	abnormal, ncs	8 (73%)	2 (67%)	10 (71%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 111 Assessment of Hb during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	5	1	6
	normal	3 (60%)	1 (100%)	4 (67%)
	abnormal, ncs	2 (40%)	--	2 (33%)
Week 4	n	3	1	4
	normal	2 (67%)	1 (100%)	3 (75%)
	abnormal, ncs	1 (33%)	--	1 (25%)
Week 6	n	3	2	5
	normal	3 (100%)	2 (100%)	5 (100%)
	abnormal, ncs	--	--	--
Week 8	n	3	2	5
	normal	3 (100%)	2 (100%)	5 (100%)
	abnormal, ncs	--	--	--
Week 12	n	3	1	4
	normal	3 (100%)	1 (100%)	4 (100%)
	abnormal, ncs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 112 Assessment of Hb during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	18	8	26
	normal	2 (11%)	--	2 (8%)
	abnormal, ncs	14 (78%)	8 (100%)	22 (85%)
	abnormal, cs	2 (11%)	--	2 (8%)

Week 4	n	13	5	18
	normal	2 (15%)	1 (20%)	3 (17%)
	abnormal, ncs	11 (85%)	4 (80%)	15 (83%)
	abnormal, cs	--	--	--
Week 6	n	10	4	14
	normal	1 (10%)	1 (25%)	2 (14%)
	abnormal, ncs	9 (90%)	2 (50%)	11 (79%)
	abnormal, cs	--	1 (25%)	1 (7%)
Week 8	n	7	3	10
	normal	1 (14%)	1 (33%)	2 (20%)
	abnormal, ncs	6 (86%)	2 (67%)	8 (80%)
	abnormal, cs	--	--	--
Week 12	n	7	2	9
	normal	--	--	--
	abnormal, ncs	7 (100%)	2 (100%)	9 (100%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 113 Assessment of platelets during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	24	9	33
	normal	14 (58%)	4 (44%)	18 (55%)
	abnormal, ncs	10 (42%)	5 (56%)	15 (45%)
Week 4	n	17	6	23
	normal	10 (59%)	4 (67%)	14 (61%)
	abnormal, ncs	7 (41%)	2 (33%)	9 (39%)
Week 6	n	14	6	20
	normal	9 (64%)	2 (33%)	11 (55%)
	abnormal, ncs	5 (36%)	4 (67%)	9 (45%)
Week 8	n	11	5	16
	normal	8 (73%)	2 (40%)	10 (62%)
	abnormal, ncs	3 (27%)	3 (60%)	6 (38%)
Week 12	n	11	3	14
	normal	7 (64%)	2 (67%)	9 (64%)
	abnormal, ncs	4 (36%)	1 (33%)	5 (36%)

ncs = not clinically significant, cs = clinically significant

Table 114 Assessment of platelets during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	15	5	20
	normal	13 (87%)	3 (60%)	16 (80%)
	abnormal, ncs	2 (13%)	2 (40%)	4 (20%)
Week 4	n	11	3	14
	normal	8 (73%)	3 (100%)	11 (79%)
	abnormal, ncs	3 (27%)	--	3 (21%)
Week 6	n	9	2	11
	normal	7 (78%)	1 (50%)	8 (73%)
	abnormal, ncs	2 (22%)	1 (50%)	3 (27%)
Week 8	n	7	1	8
	normal	6 (86%)	1 (100%)	7 (88%)
	abnormal, ncs	1 (14%)	--	1 (12%)
Week 12	n	8	2	10
	normal	6 (75%)	2 (100%)	8 (80%)
	abnormal, ncs	2 (25%)	--	2 (20%)

ncs = not clinically significant, cs = clinically significant

Table 115 Assessment of platelets during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	8	4	12
	normal	1 (12%)	1 (25%)	2 (17%)
	abnormal, ncs	7 (88%)	3 (75%)	10 (83%)
Week 4	n	5	3	8
	normal	1 (20%)	1 (33%)	2 (25%)
	abnormal, ncs	4 (80%)	2 (67%)	6 (75%)
Week 6	n	4	4	8
	normal	1 (25%)	1 (25%)	2 (25%)
	abnormal, ncs	3 (75%)	3 (75%)	6 (75%)

Week 8	n	3	4	7
	normal	1 (33%)	1 (25%)	2 (29%)
	abnormal, ncs	2 (67%)	3 (75%)	5 (71%)
Week 12	n	2	1	3
	normal	--	--	--
	abnormal, ncs	2 (100%)	1 (100%)	3 (100%)

ncs = not clinically significant, cs = clinically significant

Table 116 Assessment of white blood count during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	24	9	33
	normal	15 (62%)	3 (33%)	18 (55%)
	abnormal, ncs	9 (38%)	6 (67%)	15 (45%)
	abnormal, cs	--	--	--
Week 4	n	17	6	23
	normal	12 (71%)	4 (67%)	16 (70%)
	abnormal, ncs	5 (29%)	2 (33%)	7 (30%)
	abnormal, cs	--	--	--
Week 6	n	14	6	20
	normal	11 (79%)	4 (67%)	15 (75%)
	abnormal, ncs	2 (14%)	2 (33%)	4 (20%)
	abnormal, cs	1 (7%)	--	1 (5%)
Week 8	n	11	5	16
	normal	9 (82%)	1 (20%)	10 (62%)
	abnormal, ncs	1 (9%)	4 (80%)	5 (31%)
	abnormal, cs	1 (9%)	--	1 (6%)
Week 12	n	11	3	14
	normal	8 (73%)	2 (67%)	10 (71%)
	abnormal, ncs	3 (27%)	1 (33%)	4 (29%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 117 Assessment of white blood count during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	15	5	20
	normal	10 (67%)	3 (60%)	13 (65%)
	abnormal, ncs	5 (33%)	2 (40%)	7 (35%)
	abnormal, cs	--	--	--
Week 4	n	11	2	13
	normal	10 (91%)	2 (100%)	12 (92%)
	abnormal, ncs	1 (9%)	--	1 (8%)
	abnormal, cs	--	--	--
Week 6	n	10	2	12
	normal	8 (80%)	2 (100%)	10 (83%)
	abnormal, ncs	1 (10%)	--	1 (8%)
	abnormal, cs	1 (10%)	--	1 (8%)
Week 8	n	8	2	10
	normal	7 (88%)	1 (50%)	8 (80%)
	abnormal, ncs	--	1 (50%)	1 (10%)
	abnormal, cs	1 (12%)	--	1 (10%)
Week 12	n	10	2	12
	normal	7 (70%)	2 (100%)	9 (75%)
	abnormal, ncs	3 (30%)	--	3 (25%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 118 Assessment of white blood count during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	8	4	12
	normal	4 (50%)	--	4 (33%)
	abnormal, ncs	4 (50%)	4 (100%)	8 (67%)
Week 4	n	5	4	9
	normal	1 (20%)	2 (50%)	3 (33%)
	abnormal, ncs	4 (80%)	2 (50%)	6 (67%)
Week 6	n	3	4	7
	normal	2 (67%)	2 (50%)	4 (57%)
	abnormal, ncs	1 (33%)	2 (50%)	3 (43%)

Week 8	n	2	3	5
	normal	1 (50%)	--	1 (20%)
	abnormal, ncs	1 (50%)	3 (100%)	4 (80%)
Week 12	n	1	--	1
	normal	--	--	--
	abnormal, ncs	1 (100%)	--	1 (100%)

ncs = not clinically significant, cs = clinically significant

Table 119 Assessment of neutrophils during therapy

	Finding	Bevacizumab	Placebo	Total
Week 2	n	22	8	30
	normal	16 (73%)	4 (50%)	20 (67%)
	abnormal, ncs	6 (27%)	4 (50%)	10 (33%)
Week 4	n	14	6	20
	normal	9 (64%)	5 (83%)	14 (70%)
	abnormal, ncs	5 (36%)	1 (17%)	6 (30%)
Week 6	n	11	6	17
	normal	9 (82%)	4 (67%)	13 (76%)
	abnormal, ncs	2 (18%)	2 (33%)	4 (24%)
Week 8	n	10	5	15
	normal	9 (90%)	2 (40%)	11 (73%)
	abnormal, ncs	1 (10%)	3 (60%)	4 (27%)
Week 12	n	8	3	11
	normal	6 (75%)	3 (100%)	9 (82%)
	abnormal, ncs	2 (25%)	--	2 (18%)

ncs = not clinically significant, cs = clinically significant

Table 120 Assessment of neutrophils during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	13	6	19
	normal	11 (85%)	3 (50%)	14 (74%)
	abnormal, ncs	2 (15%)	3 (50%)	5 (26%)
Week 4	n	9	4	13

	normal	7 (78%)	3 (75%)	10 (77%)
	abnormal, ncs	2 (22%)	1 (25%)	3 (23%)
Week 6	n	9	3	12
	normal	7 (78%)	2 (67%)	9 (75%)
	abnormal, ncs	2 (22%)	1 (33%)	3 (25%)
Week 8	n	7	3	10
	normal	6 (86%)	2 (67%)	8 (80%)
	abnormal, ncs	1 (14%)	1 (33%)	2 (20%)
Week 12	n	7	3	10
	normal	5 (71%)	3 (100%)	8 (80%)
	abnormal, ncs	2 (29%)	--	2 (20%)

ncs = not clinically significant, cs = clinically significant

Table 121 Assessment of neutrophils during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	3	2	5
	normal	3 (100%)	1 (50%)	4 (80%)
	abnormal, ncs	--	1 (50%)	1 (20%)
Week 4	n	2	2	4
	normal	1 (50%)	2 (100%)	3 (75%)
	abnormal, ncs	1 (50%)	--	1 (25%)
Week 6	n	1	3	4
	normal	1 (100%)	2 (67%)	3 (75%)
	abnormal, ncs	--	1 (33%)	1 (25%)
Week 8	n	2	2	4
	normal	2 (100%)	--	2 (50%)
	abnormal, ncs	--	2 (100%)	2 (50%)
Week 12	n	--	--	--
	normal	--	--	--
	abnormal, ncs	--	--	--

ncs = not clinically significant, cs = clinically significant

12.4.2.1.2.2 Clinical chemistry

Similar to section 12.4.2.1.2.1, all clinical chemistry assessments during the course of therapy until week 12 (if reached) (Tables 122 - 151) are analyzed in three ways: firstly, all observations at the respective visits are taken into account. In two additional tables, the respective findings are sub-divided according to the normal/abnormal status of the respective laboratory value of the respective patient at the baseline visit (if available).

Table 122 Assessment of creatinine during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	22	9	31
	normal	12 (55%)	5 (56%)	17 (55%)
	abnormal, ncs	10 (45%)	3 (33%)	13 (42%)
	abnormal, cs	--	1 (11%)	1 (3%)
Week 4	n	17	6	23
	normal	12 (71%)	4 (67%)	16 (70%)
	abnormal, ncs	5 (29%)	1 (17%)	6 (26%)
	abnormal, cs	--	1 (17%)	1 (4%)
Week 6	n	14	6	20
	normal	9 (64%)	5 (83%)	14 (70%)
	abnormal, ncs	5 (36%)	1 (17%)	6 (30%)
	abnormal, cs	--	--	--
Week 8	n	11	5	16
	normal	6 (55%)	3 (60%)	9 (56%)
	abnormal, ncs	5 (45%)	2 (40%)	7 (44%)
	abnormal, cs	--	--	--
Week 12	n	10	3	13
	normal	4 (40%)	3 (100%)	7 (54%)
	abnormal, ncs	5 (50%)	--	5 (38%)
	abnormal, cs	1 (10%)	--	1 (8%)

ncs = not clinically significant, cs = clinically significant

Table 123 Assessment of creatinine during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	14	5	19

	normal	11 (79%)	4 (80%)	15 (79%)
	abnormal, ncs	3 (21%)	1 (20%)	4 (21%)
Week 4	n	11	4	15
	normal	10 (91%)	3 (75%)	13 (87%)
	abnormal, ncs	1 (9%)	1 (25%)	2 (13%)
Week 6	n	10	4	14
	normal	8 (80%)	4 (100%)	12 (86%)
	abnormal, ncs	2 (20%)	--	2 (14%)
Week 8	n	7	3	10
	normal	5 (71%)	3 (100%)	8 (80%)
	abnormal, ncs	2 (29%)	--	2 (20%)
Week 12	n	6	3	9
	normal	4 (67%)	3 (100%)	7 (78%)
	abnormal, ncs	2 (33%)	--	2 (22%)

ncs = not clinically significant, cs = clinically significant

Table 124 Assessment of creatinine during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	7	4	11
	normal	1 (14%)	1 (25%)	2 (18%)
	abnormal, ncs	6 (86%)	2 (50%)	8 (73%)
	abnormal, cs	--	1 (25%)	1 (9%)
Week 4	n	5	2	7
	normal	2 (40%)	1 (50%)	3 (43%)
	abnormal, ncs	3 (60%)	--	3 (43%)
	abnormal, cs	--	1 (50%)	1 (14%)
Week 6	n	3	2	5
	normal	1 (33%)	1 (50%)	2 (40%)
	abnormal, ncs	2 (67%)	1 (50%)	3 (60%)
	abnormal, cs	--	--	--
Week 8	n	3	2	5
	normal	1 (33%)	--	1 (20%)
	abnormal, ncs	2 (67%)	2 (100%)	4 (80%)
	abnormal, cs	--	--	--
Week 12	n	3	--	3
	normal	--	--	--
	abnormal, ncs	2 (67%)	--	2 (67%)
	abnormal, cs	1 (33%)	--	1 (33%)

ncs = not clinically significant, cs = clinically significant

Table 125 Assessment of total bilirubin during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	20	9	29
	normal	16 (80%)	7 (78%)	23 (79%)
	abnormal, ncs	4 (20%)	2 (22%)	6 (21%)
Week 4	n	17	6	23
	normal	11 (65%)	5 (83%)	16 (70%)
	abnormal, ncs	6 (35%)	1 (17%)	7 (30%)

Week 6	n	14	6	20
	normal	11 (79%)	4 (67%)	15 (75%)
	abnormal, ncs	3 (21%)	2 (33%)	5 (25%)
Week 8	n	11	5	16
	normal	10 (91%)	4 (80%)	14 (88%)
	abnormal, ncs	1 (9%)	1 (20%)	2 (12%)
Week 12	n	10	3	13
	normal	9 (90%)	2 (67%)	11 (85%)
	abnormal, ncs	1 (10%)	1 (33%)	2 (15%)

ncs = not clinically significant, cs = clinically significant

Table 126 Assessment of total bilirubin during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	13	8	21
	normal	13 (100%)	7 (88%)	20 (95%)
	abnormal, ncs	--	1 (12%)	1 (5%)
Week 4	n	11	5	16
	normal	8 (73%)	5 (100%)	13 (81%)
	abnormal, ncs	3 (27%)	--	3 (19%)
Week 6	n	11	4	15
	normal	9 (82%)	4 (100%)	13 (87%)
	abnormal, ncs	2 (18%)	--	2 (13%)
Week 8	n	8	4	12
	normal	8 (100%)	4 (100%)	12 (100%)
	abnormal, ncs	--	--	--
Week 12	n	7	3	10
	normal	7 (100%)	2 (67%)	9 (90%)
	abnormal, ncs	--	1 (33%)	1 (10%)

ncs = not clinically significant, cs = clinically significant

Table 127 Assessment of total bilirubin during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	5	1	6
	normal	1 (20%)	--	1 (17%)
	abnormal, ncs	4 (80%)	1 (100%)	5 (83%)
Week 4	n	4	1	5
	normal	1 (25%)	--	1 (20%)
	abnormal, ncs	3 (75%)	1 (100%)	4 (80%)
Week 6	n	2	2	4
	normal	1 (50%)	--	1 (25%)
	abnormal, ncs	1 (50%)	2 (100%)	3 (75%)
Week 8	n	2	1	3
	normal	1 (50%)	--	1 (33%)
	abnormal, ncs	1 (50%)	1 (100%)	2 (67%)
Week 12	n	2	--	2
	normal	1 (50%)	--	1 (50%)
	abnormal, ncs	1 (50%)	--	1 (50%)

ncs = not clinically significant, cs = clinically significant

Table 128 Assessment of total protein during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	18	8	26
	normal	4 (22%)	1 (12%)	5 (19%)
	abnormal, ncs	14 (78%)	7 (88%)	21 (81%)
Week 4	n	14	6	20
	normal	4 (29%)	2 (33%)	6 (30%)
	abnormal, ncs	10 (71%)	4 (67%)	14 (70%)
Week 6	n	14	6	20
	normal	6 (43%)	2 (33%)	8 (40%)
	abnormal, ncs	8 (57%)	4 (67%)	12 (60%)
Week 8	n	10	5	15
	normal	4 (40%)	1 (20%)	5 (33%)

	abnormal, ncs	6 (60%)	4 (80%)	10 (67%)
Week 12	n	8	3	11
	normal	2 (25%)	2 (67%)	4 (36%)
	abnormal, ncs	6 (75%)	1 (33%)	7 (64%)

ncs = not clinically significant, cs = clinically significant

Table 129 Assessment of total protein during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	4	2	6
	normal	3 (75%)	1 (50%)	4 (67%)
	abnormal, ncs	1 (25%)	1 (50%)	2 (33%)
Week 4	n	3	1	4
	normal	2 (67%)	1 (100%)	3 (75%)
	abnormal, ncs	1 (33%)	--	1 (25%)
Week 6	n	5	1	6
	normal	4 (80%)	1 (100%)	5 (83%)
	abnormal, ncs	1 (20%)	--	1 (17%)
Week 8	n	3	2	5
	normal	2 (67%)	1 (50%)	3 (60%)
	abnormal, ncs	1 (33%)	1 (50%)	2 (40%)
Week 12	n	1	1	2
	normal	1 (100%)	1 (100%)	2 (100%)
	abnormal, ncs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 130 Assessment of total protein during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	11	5	16
	normal	--	--	--
	abnormal, ncs	11 (100%)	5 (100%)	16 (100%)
Week 4	n	7	5	12

	normal	1 (14%)	1 (20%)	2 (17%)
	abnormal, ncs	6 (86%)	4 (80%)	10 (83%)
Week 6	n	7	4	11
	normal	1 (14%)	--	1 (9%)
	abnormal, ncs	6 (86%)	4 (100%)	10 (91%)
Week 8	n	6	3	9
	normal	1 (17%)	--	1 (11%)
	abnormal, ncs	5 (83%)	3 (100%)	8 (89%)
Week 12	n	6	2	8
	normal	--	1 (50%)	1 (12%)
	abnormal, ncs	6 (100%)	1 (50%)	7 (88%)

ncs = not clinically significant, cs = clinically significant

Table 131 Assessment of ASAT / GOT during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	21	9	30
	normal	12 (57%)	6 (67%)	18 (60%)
	abnormal, ncs	9 (43%)	3 (33%)	12 (40%)
Week 4	n	17	6	23
	normal	9 (53%)	3 (50%)	12 (52%)
	abnormal, ncs	8 (47%)	3 (50%)	11 (48%)
Week 6	n	14	6	20
	normal	9 (64%)	4 (67%)	13 (65%)
	abnormal, ncs	5 (36%)	2 (33%)	7 (35%)
Week 8	n	11	5	16
	normal	7 (64%)	4 (80%)	11 (69%)
	abnormal, ncs	4 (36%)	1 (20%)	5 (31%)
Week 12	n	10	3	13
	normal	8 (80%)	1 (33%)	9 (69%)
	abnormal, ncs	2 (20%)	2 (67%)	4 (31%)

ncs = not clinically significant, cs = clinically significant

Table 132 Assessment of ASAT / GOT during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	10	5	15
	normal	8 (80%)	5 (100%)	13 (87%)
	abnormal, ncs	2 (20%)	--	2 (13%)
	abnormal, cs	7	3	10
Week 4	n	4 (57%)	2 (67%)	6 (60%)
	normal	3 (43%)	1 (33%)	4 (40%)
	abnormal, ncs	7	3	10
	abnormal, cs	5 (71%)	3 (100%)	8 (80%)
Week 6	n	2 (29%)	--	2 (20%)
	normal	6	3	9
	abnormal, ncs	4 (67%)	3 (100%)	7 (78%)
	abnormal, cs	2 (33%)	--	2 (22%)
Week 8	n	7	1	8
	normal	6 (86%)	1 (100%)	7 (88%)
	abnormal, ncs	1 (14%)	--	1 (12%)

ncs = not clinically significant, cs = clinically significant

Table 133 Assessment of ASAT / GOT during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	10	4	14
	normal	3 (30%)	1 (25%)	4 (29%)
	abnormal, ncs	7 (70%)	3 (75%)	10 (71%)
Week 4	n	8	3	11
	normal	3 (38%)	1 (33%)	4 (36%)
	abnormal, ncs	5 (62%)	2 (67%)	7 (64%)
Week 6	n	6	3	9
	normal	3 (50%)	1 (33%)	4 (44%)
	abnormal, ncs	3 (50%)	2 (67%)	5 (56%)
Week 8	n	4	2	6

	normal	2 (50%)	1 (50%)	3 (50%)
	abnormal, ncs	2 (50%)	1 (50%)	3 (50%)
Week 12	n	2	2	4
	normal	1 (50%)	--	1 (25%)
	abnormal, ncs	1 (50%)	2 (100%)	3 (75%)

ncs = not clinically significant, cs = clinically significant

Table 134 Assessment of ALAT / GPT during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	21	9	30
	normal	15 (71%)	7 (78%)	22 (73%)
	abnormal, ncs	6 (29%)	2 (22%)	8 (27%)
Week 4	n	17	6	23
	normal	14 (82%)	5 (83%)	19 (83%)
	abnormal, ncs	3 (18%)	1 (17%)	4 (17%)
Week 6	n	14	6	20
	normal	10 (71%)	5 (83%)	15 (75%)
	abnormal, ncs	4 (29%)	1 (17%)	5 (25%)
Week 8	n	11	5	16
	normal	9 (82%)	5 (100%)	14 (88%)
	abnormal, ncs	2 (18%)	--	2 (12%)
Week 12	n	10	3	13
	normal	7 (70%)	1 (33%)	8 (62%)
	abnormal, ncs	3 (30%)	2 (67%)	5 (38%)

ncs = not clinically significant, cs = clinically significant

Table 135 Assessment of ALAT / GPT during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	12	8	20
	normal	11 (92%)	7 (88%)	18 (90%)
	abnormal, ncs	1 (8%)	1 (12%)	2 (10%)
Week 4	n	10	5	15
	normal	10 (100%)	5 (100%)	15 (100%)

	abnormal, ncs	--	--	--
Week 6	n	8	5	13
	normal	6 (75%)	5 (100%)	11 (85%)
	abnormal, ncs	2 (25%)	--	2 (15%)
Week 8	n	6	4	10
	normal	5 (83%)	4 (100%)	9 (90%)
	abnormal, ncs	1 (17%)	--	1 (10%)
Week 12	n	6	3	9
	normal	4 (67%)	1 (33%)	5 (56%)
	abnormal, ncs	2 (33%)	2 (67%)	4 (44%)

ncs = not clinically significant, cs = clinically significant

Table 136 Assessment of ALAT / GPT during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	8	1	9
	normal	3 (38%)	--	3 (33%)
	abnormal, ncs	5 (62%)	1 (100%)	6 (67%)
Week 4	n	5	1	6
	normal	2 (40%)	--	2 (33%)
	abnormal, ncs	3 (60%)	1 (100%)	4 (67%)
Week 6	n	5	1	6
	normal	3 (60%)	--	3 (50%)
	abnormal, ncs	2 (40%)	1 (100%)	3 (50%)
Week 8	n	4	1	5
	normal	3 (75%)	1 (100%)	4 (80%)
	abnormal, ncs	1 (25%)	--	1 (20%)
Week 12	n	3	--	3
	normal	2 (67%)	--	2 (67%)
	abnormal, ncs	1 (33%)	--	1 (33%)

ncs = not clinically significant, cs = clinically significant

Table 137 Assessment of LDH during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	19	9	28
	normal	11 (58%)	3 (33%)	14 (50%)
	abnormal, ncs	8 (42%)	6 (67%)	14 (50%)
Week 4	n	17	6	23
	normal	12 (71%)	2 (33%)	14 (61%)
	abnormal, ncs	5 (29%)	4 (67%)	9 (39%)
Week 6	n	14	6	20
	normal	8 (57%)	3 (50%)	11 (55%)
	abnormal, ncs	6 (43%)	3 (50%)	9 (45%)
Week 8	n	11	5	16
	normal	5 (45%)	1 (20%)	6 (38%)
	abnormal, ncs	6 (55%)	4 (80%)	10 (62%)
Week 12	n	9	3	12
	normal	3 (33%)	--	3 (25%)
	abnormal, ncs	6 (67%)	3 (100%)	9 (75%)

ncs = not clinically significant, cs = clinically significant

Table 138 Assessment of LDH during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	9	3	12
	normal	7 (78%)	3 (100%)	10 (83%)
	abnormal, ncs	2 (22%)	--	2 (17%)
Week 4	n	8	1	9
	normal	6 (75%)	1 (100%)	7 (78%)
	abnormal, ncs	2 (25%)	--	2 (22%)
Week 6	n	7	2	9
	normal	5 (71%)	2 (100%)	7 (78%)
	abnormal, ncs	2 (29%)	--	2 (22%)
Week 8	n	6	2	8
	normal	2 (33%)	1 (50%)	3 (38%)
	abnormal, ncs	4 (67%)	1 (50%)	5 (62%)

Week 12	n	5	1	6
	normal	1 (20%)	--	1 (17%)
	abnormal, ncs	4 (80%)	1 (100%)	5 (83%)

ncs = not clinically significant, cs = clinically significant

Table 139 Assessment of LDH during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	8	6	14
	normal	3 (38%)	--	3 (21%)
	abnormal, ncs	5 (62%)	6 (100%)	11 (79%)
Week 4	n	5	5	10
	normal	2 (40%)	1 (20%)	3 (30%)
	abnormal, ncs	3 (60%)	4 (80%)	7 (70%)
Week 6	n	5	4	9
	normal	2 (40%)	1 (25%)	3 (33%)
	abnormal, ncs	3 (60%)	3 (75%)	6 (67%)
Week 8	n	4	3	7
	normal	2 (50%)	--	2 (29%)
	abnormal, ncs	2 (50%)	3 (100%)	5 (71%)
Week 12	n	3	2	5
	normal	1 (33%)	--	1 (20%)
	abnormal, ncs	2 (67%)	2 (100%)	4 (80%)

ncs = not clinically significant, cs = clinically significant

Table 140 Assessment of alkaline phosphatase during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	20	8	28
	normal	6 (30%)	1 (12%)	7 (25%)
	abnormal, ncs	14 (70%)	7 (88%)	21 (75%)
Week 4	n	16	5	21
	normal	3 (19%)	--	3 (14%)
	abnormal, ncs	13 (81%)	5 (100%)	18 (86%)
Week 6	n	13	5	18

	normal	4 (31%)	1 (20%)	5 (28%)
	abnormal, ncs	9 (69%)	4 (80%)	13 (72%)
Week 8	n	11	5	16
	normal	5 (45%)	--	5 (31%)
	abnormal, ncs	6 (55%)	5 (100%)	11 (69%)
Week 12	n	9	2	11
	normal	4 (44%)	--	4 (36%)
	abnormal, ncs	5 (56%)	2 (100%)	7 (64%)

ncs = not clinically significant, cs = clinically significant

Table 141 Assessment of alkaline phosphatase during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	4	2	6
	normal	4 (100%)	1 (50%)	5 (83%)
	abnormal, ncs	--	1 (50%)	1 (17%)
Week 4	n	3	2	5
	normal	2 (67%)	--	2 (40%)
	abnormal, ncs	1 (33%)	2 (100%)	3 (60%)
Week 6	n	3	1	4
	normal	2 (67%)	1 (100%)	3 (75%)
	abnormal, ncs	1 (33%)	--	1 (25%)
Week 8	n	3	1	4
	normal	3 (100%)	--	3 (75%)
	abnormal, ncs	--	1 (100%)	1 (25%)
Week 12	n	4	1	5
	normal	2 (50%)	--	2 (40%)
	abnormal, ncs	2 (50%)	1 (100%)	3 (60%)

ncs = not clinically significant, cs = clinically significant

Table 142 Assessment of alkaline phosphatase during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	14	6	20

	normal	2 (14%)	--	2 (10%)
	abnormal, ncs	12 (86%)	6 (100%)	18 (90%)
Week 4	n	11	3	14
	normal	1 (9%)	--	1 (7%)
	abnormal, ncs	10 (91%)	3 (100%)	13 (93%)
Week 6	n	9	4	13
	normal	2 (22%)	--	2 (15%)
	abnormal, ncs	7 (78%)	4 (100%)	11 (85%)
Week 8	n	7	3	10
	normal	1 (14%)	--	1 (10%)
	abnormal, ncs	6 (86%)	3 (100%)	9 (90%)
Week 12	n	4	1	5
	normal	2 (50%)	--	2 (40%)
	abnormal, ncs	2 (50%)	1 (100%)	3 (60%)

ncs = not clinically significant, cs = clinically significant

Table 143 Assessment of creatinine clearance during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	18	9	27
	normal	14 (78%)	4 (44%)	18 (67%)
	abnormal, ncs	4 (22%)	4 (44%)	8 (30%)
	abnormal, cs	--	1 (11%)	1 (4%)
Week 4	n	12	6	18
	normal	8 (67%)	2 (33%)	10 (56%)
	abnormal, ncs	4 (33%)	3 (50%)	7 (39%)
	abnormal, cs	--	1 (17%)	1 (6%)
Week 6	n	11	6	17
	normal	10 (91%)	1 (17%)	11 (65%)
	abnormal, ncs	1 (9%)	5 (83%)	6 (35%)
	abnormal, cs	--	--	--
Week 8	n	7	5	12
	normal	4 (57%)	3 (60%)	7 (58%)
	abnormal, ncs	3 (43%)	2 (40%)	5 (42%)
	abnormal, cs	--	--	--
Week 12	n	6	3	9

	normal	5 (83%)	1 (33%)	6 (67%)
	abnormal, ncs	1 (17%)	2 (67%)	3 (33%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 144 Assessment of creatinine clearance during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	14	4	18
	normal	13 (93%)	4 (100%)	17 (94%)
	abnormal, ncs	1 (7%)	--	1 (6%)
Week 4	n	8	2	10
	normal	8 (100%)	2 (100%)	10 (100%)
	abnormal, ncs	--	--	--
Week 6	n	9	2	11
	normal	9 (100%)	1 (50%)	10 (91%)
	abnormal, ncs	--	1 (50%)	1 (9%)
Week 8	n	5	2	7
	normal	3 (60%)	2 (100%)	5 (71%)
	abnormal, ncs	2 (40%)	--	2 (29%)
Week 12	n	5	2	7
	normal	4 (80%)	1 (50%)	5 (71%)
	abnormal, ncs	1 (20%)	1 (50%)	2 (29%)

ncs = not clinically significant, cs = clinically significant

Table 145 Assessment of creatinine clearance during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	3	5	8
	normal	--	--	--
	abnormal, ncs	3 (100%)	4 (80%)	7 (88%)
	abnormal, cs	--	1 (20%)	1 (12%)
Week 4	n	3	4	7
	normal	--	--	--

	abnormal, ncs	3 (100%)	3 (75%)	6 (86%)
	abnormal, cs	--	1 (25%)	1 (14%)
Week 6	n	1	4	5
	normal	--	--	--
	abnormal, ncs	1 (100%)	4 (100%)	5 (100%)
	abnormal, cs	--	--	--
Week 8	n	1	3	4
	normal	--	1 (33%)	1 (25%)
	abnormal, ncs	1 (100%)	2 (67%)	3 (75%)
	abnormal, cs	--	--	--
Week 12	n	--	1	1
	normal	--	--	--
	abnormal, ncs	--	1 (100%)	1 (100%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 146 Assessment of INR during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	23	9	32
	normal	22 (96%)	6 (67%)	28 (88%)
	abnormal, ncs	1 (4%)	3 (33%)	4 (12%)
	abnormal, cs	--	--	--
Week 4	n	14	6	20
	normal	12 (86%)	5 (83%)	17 (85%)
	abnormal, ncs	1 (7%)	--	1 (5%)
	abnormal, cs	1 (7%)	1 (17%)	2 (10%)
Week 6	n	12	6	18
	normal	10 (83%)	4 (67%)	14 (78%)
	abnormal, ncs	2 (17%)	2 (33%)	4 (22%)
	abnormal, cs	--	--	--
Week 8	n	9	4	13
	normal	8 (89%)	2 (50%)	10 (77%)
	abnormal, ncs	1 (11%)	2 (50%)	3 (23%)
	abnormal, cs	--	--	--
Week 12	n	7	3	10

	normal	7 (100%)	2 (67%)	9 (90%)
	abnormal, ncs	--	1 (33%)	1 (10%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 147 Assessment of INR during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	19	8	27
	normal	19 (100%)	6 (75%)	25 (93%)
	abnormal, ncs	--	2 (25%)	2 (7%)
Week 4	n	10	5	15
	normal	10 (100%)	5 (100%)	15 (100%)
	abnormal, ncs	--	--	--
Week 6	n	8	4	12
	normal	8 (100%)	4 (100%)	12 (100%)
	abnormal, ncs	--	--	--
Week 8	n	8	3	11
	normal	7 (88%)	2 (67%)	9 (82%)
	abnormal, ncs	1 (12%)	1 (33%)	2 (18%)
Week 12	n	5	3	8
	normal	5 (100%)	2 (67%)	7 (88%)
	abnormal, ncs	--	1 (33%)	1 (12%)

ncs = not clinically significant, cs = clinically significant

Table 148 Assessment of INR during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	3	1	4
	normal	2 (67%)	--	2 (50%)
	abnormal, ncs	1 (33%)	1 (100%)	2 (50%)
	abnormal, cs	--	--	--
Week 4	n	3	1	4
	normal	1 (33%)	--	1 (25%)
	abnormal, ncs	1 (33%)	--	1 (25%)
	abnormal, cs	1 (33%)	1 (100%)	2 (50%)

Week 6	n	3	2	5
	normal	1 (33%)	--	1 (20%)
	abnormal, ncs	2 (67%)	2 (100%)	4 (80%)
	abnormal, cs	--	--	--
Week 8	n	--	1	1
	normal	--	--	--
	abnormal, ncs	--	1 (100%)	1 (100%)
	abnormal, cs	--	--	--
Week 12	n	1	--	1
	normal	1 (100%)	--	1 (100%)
	abnormal, ncs	--	--	--
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 149 Assessment of aPTT during therapy

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	23	9	32
	normal	17 (74%)	6 (67%)	23 (72%)
	abnormal, ncs	6 (26%)	3 (33%)	9 (28%)
	abnormal, cs	--	--	--
Week 4	n	14	6	20
	normal	9 (64%)	2 (33%)	11 (55%)
	abnormal, ncs	4 (29%)	3 (50%)	7 (35%)
	abnormal, cs	1 (7%)	1 (17%)	2 (10%)
Week 6	n	12	6	18
	normal	10 (83%)	4 (67%)	14 (78%)
	abnormal, ncs	2 (17%)	2 (33%)	4 (22%)
	abnormal, cs	--	--	--
Week 8	n	9	4	13
	normal	5 (56%)	3 (75%)	8 (62%)
	abnormal, ncs	4 (44%)	1 (25%)	5 (38%)
	abnormal, cs	--	--	--
Week 12	n	7	3	10
	normal	6 (86%)	3 (100%)	9 (90%)
	abnormal, ncs	1 (14%)	--	1 (10%)

	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 150 Assessment of aPTT during therapy, subgroup: normal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	15	8	23
	normal	14 (93%)	5 (62%)	19 (83%)
	abnormal, ncs	1 (7%)	3 (38%)	4 (17%)
	abnormal, cs	--	--	--
Week 4	n	9	5	14
	normal	8 (89%)	2 (40%)	10 (71%)
	abnormal, ncs	--	2 (40%)	2 (14%)
	abnormal, cs	1 (11%)	1 (20%)	2 (14%)
Week 6	n	8	5	13
	normal	8 (100%)	4 (80%)	12 (92%)
	abnormal, ncs	--	1 (20%)	1 (8%)
	abnormal, cs	--	--	--
Week 8	n	6	3	9
	normal	3 (50%)	3 (100%)	6 (67%)
	abnormal, ncs	3 (50%)	--	3 (33%)
	abnormal, cs	--	--	--
Week 12	n	5	2	7
	normal	4 (80%)	2 (100%)	6 (86%)
	abnormal, ncs	1 (20%)	--	1 (14%)
	abnormal, cs	--	--	--

ncs = not clinically significant, cs = clinically significant

Table 151 Assessment of aPTT during therapy, subgroup: abnormal at baseline

Visit	Finding	Bevacizumab	Placebo	Total
Week 2	n	7	1	8
	normal	2 (29%)	1 (100%)	3 (38%)
	abnormal, ncs	5 (71%)	--	5 (62%)
Week 4	n	4	1	5

	normal	--	--	--
	abnormal, ncs	4 (100%)	1 (100%)	5 (100%)
Week 6	n	3	1	4
	normal	1 (33%)	--	1 (25%)
	abnormal, ncs	2 (67%)	1 (100%)	3 (75%)
Week 8	n	2	1	3
	normal	1 (50%)	--	1 (33%)
	abnormal, ncs	1 (50%)	1 (100%)	2 (67%)
Week 12	n	1	1	2
	normal	1 (100%)	1 (100%)	2 (100%)
	abnormal, ncs	--	--	--

ncs = not clinically significant, cs = clinically significant

12.4.2.2 Individual patient changes

NA

12.4.2.3 Individual clinically significant abnormalities

NA

12.5 Vital signs, physical findings and other observations related to safety

12.5.1 Performance status

As for most of the parameters observed during the on study period, the analysis of the course of the ECOG performance status suffers from the continuous "loss" of patients (Tables 152 - 156). In the respective remaining patients, no consisted time trend can be detected except for an unequivocal deterioration at week 12.

Table 152 ECOG performance status, week 2

ECOG score	Bevacizumab	Placebo	Total
n	22	9	31
0	1 (5%)	--	1 (3%)
1	10 (45%)	4 (44%)	14 (45%)
2	11 (50%)	2 (22%)	13 (42%)
3	--	3 (33%)	3 (10%)
4	--	--	--

Table 153 ECOG performance status, week 4

ECOG score	Bevacizumab	Placebo	Total
n	16	6	22
0	1 (6%)	--	1 (5%)
1	8 (50%)	2 (33%)	10 (45%)
2	4 (25%)	3 (50%)	7 (32%)
3	3 (19%)	1 (17%)	4 (18%)
4	--	--	--

Table 154 ECOG performance status, week 6

ECOG score	Bevacizumab	Placebo	Total
n	14	6	20
0	1 (7%)	--	1 (5%)
1	7 (50%)	3 (50%)	10 (50%)
2	5 (36%)	2 (33%)	7 (35%)
3	1 (7%)	1 (17%)	2 (10%)
4	--	--	--

Table 155 ECOG performance status, week 8

ECOG score	Bevacizumab	Placebo	Total
n	9	4	13
1	6 (67%)	2 (50%)	8 (62%)
2	3 (33%)	1 (25%)	4 (31%)
3	--	1 (25%)	1 (8%)
4	--	--	--

Table 156 ECOG performance status, week 12

ECOG score	Bevacizumab	Placebo	Total
n	9	3	12
1	4 (44%)	1 (33%)	5 (42%)
2	1 (11%)	--	1 (8%)

3	3 (33%)	2 (67%)	5 (42%)
4	1 (11%)	--	1 (8%)

A more sophisticated analysis of the performance status development is depicted in Tables 157 - 161, based on the respective difference to baseline in each patient under observation at both respective visits. (Negative values indicate an improvement versus baseline.) Again, apart from the final visit, most surviving patients show a stable performance compared to the initial assessment.

Table 157 ECOG performance status, week 2 vs. baseline

ECOG score difference vs. baseline	Bevacizumab	Placebo	Total
n	22	8	30
-2	1 (5%)	-	1 (3%)
-1	-	1 (12%)	1 (3%)
0	20 (91%)	4 (50%)	24 (80%)
1	1 (5%)	3 (38%)	4 (13%)

Table 158 ECOG performance status, week 4 vs. baseline

ECOG score difference vs. baseline	Bevacizumab	Placebo	Total
n	16	5	21
0	13 (81%)	3 (60%)	16 (76%)
1	3 (19%)	2 (40%)	5 (24%)

Table 159 ECOG performance status, week 6 vs. baseline

ECOG score difference vs. baseline	Bevacizumab	Placebo	Total
n	14	5	19
-2	1 (7%)	-	1 (5%)
0	11 (79%)	3 (60%)	14 (74%)
1	2 (14%)	2 (40%)	4 (21%)

Table 160 ECOG performance status, week 8 vs. baseline

ECOG score difference vs. baseline	Bevacizumab	Placebo	Total
n	9	4	13
-2	1 (11%)	-	1 (8%)

0	6 (67%)	2 (50%)	8 (62%)
1	2 (22%)	2 (50%)	4 (31%)

Table 161 ECOG performance status, week 12 vs. baseline

ECOG score difference vs. baseline	Bevacizumab	Placebo	Total
n	8	3	11
-2	1 (12%)	-	1 (9%)
0	2 (25%)	1 (33%)	3 (27%)
1	3 (38%)	2 (67%)	5 (45%)
2	1 (12%)	-	1 (9%)
3	1 (12%)	-	1 (9%)

12.5.2 Blood pressure

There is no, or at most a marginal hint for a systemic effect of intraperitoneal Bevacizumab treatment, based on blood pressure measurements between week 2 and 12 (Tables 162 - 171). The distribution parameters show neither major differences from baseline nor a consistent trend over time.

Table 162 Blood pressure at week 2, systolic

Parameter	Bevacizumab	Placebo	Total
n	22	9	31
Mean ± SD	119.3 ± 20.1	119.8 ± 16.4	119.4 ± 18.8
Median	116.5	115	115
Quartile	106.2 - 130	108 - 135	106.5 - 130
Range	89 - 160	100 - 145	89 - 160

Table 163 Blood pressure at week 4, systolic

Parameter	Bevacizumab	Placebo	Total
n	14	6	20
Mean ± SD	118.5 ± 18.2	108.3 ± 11.7	115.4 ± 16.9
Median	120	110	118
Quartile	106.2 - 127.5	102.5 - 117.5	104 - 120
Range	90 - 153	90 - 120	90 - 153

Table 164 Blood pressure at week 6, systolic

Parameter	Bevacizumab	Placebo	Total
n	13	6	19
Mean ± SD	117.8 ± 17.5	115.8 ± 24.2	117.2 ± 19.2
Median	110	115	110
Quartile	105 - 128	110 - 135	107.5 - 129
Range	95 - 155	75 - 140	75 - 155

Table 165 Blood pressure at week 8, systolic

Parameter	Bevacizumab	Placebo	Total
n	8	4	12
Mean ± SD	117.1 ± 16.9	110 ± 18.3	114.8 ± 16.9
Median	112.5	110	112.5
Quartile	109.8 - 127.8	97.5 - 122.5	106.8 - 127.8
Range	90 - 146	90 - 130	90 - 146

Table 166 Blood pressure at week 12, systolic

Parameter	Bevacizumab	Placebo	Total
n	8	3	11
Mean ± SD	117.1 ± 22.1	103.3 ± 11.5	113.4 ± 20.3
Median	110	110	110
Quartile	107.5 - 124.2	100 - 110	105 - 115
Range	90 - 160	90 - 110	90 - 160

Table 167 Blood pressure at week 2, diastolic

Parameter	Bevacizumab	Placebo	Total
n	22	9	31
Mean ± SD	73 ± 10.4	74.6 ± 10.9	73.4 ± 10.4
Median	77.5	80	80
Quartile	60.2 - 80	65 - 83	60.5 - 80
Range	60 - 90	60 - 88	60 - 90

Table 168 Blood pressure at week 4, diastolic

Parameter	Bevacizumab	Placebo	Total
n	14	6	20
Mean ± SD	70.1 ± 9.9	72.5 ± 7.6	70.8 ± 9.2
Median	68.5	72.5	70
Quartile	60 - 78.8	70 - 78.8	60 - 80
Range	60 - 90	60 - 80	60 - 90

Table 169 Blood pressure at week 6, diastolic

Parameter	Bevacizumab	Placebo	Total
n	13	6	19
Mean ± SD	72.9 ± 11.9	71.7 ± 20.4	72.5 ± 14.5
Median	78	75	78
Quartile	60 - 80	62.5 - 80	60 - 80
Range	55 - 92	40 - 100	40 - 100

Table 170 Blood pressure at week 8, diastolic

Parameter	Bevacizumab	Placebo	Total
n	8	4	12
Mean ± SD	71.6 ± 4.7	70 ± 16.3	71.1 ± 9.4
Median	70	70	70
Quartile	69.5 - 75	65 - 75	69.5 - 75
Range	65 - 80	50 - 90	50 - 90

Table 171 Blood pressure at week 12, diastolic

Parameter	Bevacizumab	Placebo	Total
n	8	3	11
Mean ± SD	77.8 ± 14	63.3 ± 5.8	73.8 ± 13.8
Median	80	60	70
Quartile	67.5 - 83.8	60 - 65	60 - 80
Range	60 - 97	60 - 70	60 - 97

12.6 Safety conclusions

No new safety concerns occurred during the course of the study.

Due to the terminal illness of the patients included, the main cause of death was progression of disease.

A major increase in severe toxicity cannot be detected for the Bevacizumab arm. The proportion of patients with at least one grade 3 to 5 event occurring is similar with 20/33 (61%) in arm 1 and 11/16 (69%) in arm 2. As one might have expected, fatigue is the most prevalent adverse event: 39% in arm 1, which is somewhat higher than the 3/16 (19%) finding in the placebo group. Nausea (45%) and vomiting (36%) are other frequent adverse events on Bevacizumab, together with pain (33%). The respective rates in the placebo arm are somewhat lower with 31% nausea and 33% pain; remarkably, no cases of vomiting were reported in the control group. Altogether, Bevacizumab was well tolerated.

13 DISCUSSION AND OVERALL CONCLUSION

Malignant ascites is a debilitating and unpleasant complication for patients with several types of advanced cancer that negatively impacts the quality of life (QoL). Furthermore, accumulation of massive amounts of malignant ascites is associated with a poor prognosis and a significant cause of morbidity and mortality in patients with intra-abdominal tumors (1). Causative treatment approaches are still limited. Animal and laboratory studies have shown that tumor cell production and/or increases in the amount of Vascular Endothelial Growth Factor (VEGF) are a major cause of the formation of malignant ascites. Therefore, giving patients with malignant ascites a drug that targets and neutralizes VEGF should prevent the recurrence of malignant ascites following paracentesis (a procedure to remove fluid from the abdominal cavity).

Malignant ascites represents a serious medical condition in the clinical routine. So far, there is no standardized and evidence-based treatment for malignant ascites and therapies which are commonly being used, such as paracentesis, are only temporarily effective.

To date, the tri-specific antibody catumaxomab is the only effective and approved treatment option for malignant ascites. Trials exist for patients suffering of ovarian and gastric cancer and show a significant symptom relief after catumaxomab application (96). Puncture-free survival was significantly longer in the catumaxomab group than in the control group (median 46 vs. 11 days).

Furthermore, a phase II/III trial has assessed catumaxomab in the treatment of malignant ascites with significant results regarding to puncture free survival and quality of life. Another phase II/III study by Heiss et al. showed a significantly improved overall survival rate after catumaxomab application vs. paracentesis alone (6-month survival rate: 28.9% vs. 6.7%) in patients with malignant ascites (97). However, catumaxomab is associated with significant side effects and can only be administered to patients in good general condition. Therefore, there is still an urgent need for more effective and less toxic treatment options for peritoneal effusion caused by gastrointestinal cancers.

Preclinical data strongly suggest that Bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large part caused by the hyperpermeability-promoting factor VEGF. Aoyagi et al. conducted research in the high potential peritoneal gastric cancer cell line MKN-45P showing elevated levels of VEGF in the supernatant, suggesting that therapies targeting VEGF could lower ascites (98). In mice injected with the same gastric cancer cell line, intraperitoneal administration of Bevacizumab reduced malignant ascites (99).

High ascites VEGF levels were shown to be a risk factor for survival and VEGF expression was observed in 70% of the patients with ascites in a recent study by Fushida et al. (100). Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. It has previously been shown that tumor cell production and/or increase of Vascular Endothelial Growth Factor (VEGF) might be a major cause of the formation of malignant ascites (100). However, no clinical studies have investigated the effect of Bevacizumab in this clinical setting so far. In the literature, only two case reports of women with refractory peritoneal dissemination of gastric cancer and severe symptomatic ascites are present. The ascites was resistant to systemic and intraperitoneal chemotherapy, but showed a dramatic improvement when treated with intravenous Bevacizumab.

In the present study, Bevacizumab was administered as an intraperitoneal infusion at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.

In this trial, the intraperitoneal Bevacizumab application did not lead to an improvement of paracentesis-free survival. However, the study was prematurely stopped after enrolling 53 of 72 patients due to slow recruitment. Therefore, data allow only a limited statement on efficacy. Out of the 53 patients randomized, 49 received at least one application of the study drug. In the full analysis set, there is no major difference between the Bevacizumab and the control group with paracentesis free survival (ParFS) medians of 14.0 (95% confidence interval: 11 – 17 days) and 10.5 (95% confidence interval: 7 – 21 days), with a non-significant logrank test result of p = 0.16 (one-sided, according to the primary study hypothesis as defined in the protocol). This might be because patients were in decreased general condition and terminally ill.

The hazard ratio (HR) amounts to 0.74 (95% confidence interval: 0.40 – 1.37). Thus, the latter confidence interval does not exclude the prospectively anticipated target effect size of HR = 0.5, as derived from the catumaxomab trial results.

The ParFS are clearly dominated by paracentesis events, since only in 8 patients (16%) death occurred before a second on-study paracentesis was performed and recorded. The median overall survival is 64 days (95% confidence interval: 45 – 103 days) in the Bevacizumab arm, and 31.5 days (95% confidence interval: 20 – 117 days) under placebo, with a two-sided logrank test result of p = 0.31. While the medians seem to indicate a major difference, this may easily be caused by a mere play of chance due to the low patient numbers, as indicated by the widely overlapping confidence intervals.

Assessing quality of life, the common and inevitable problem of QoL analysis in highly palliative situations occurred: firstly, comparisons, both between groups or different time points, suffer from an uncontrolled loss process, due to patient's death or inability to perform further visits or fill out the questionnaires. Thus the remaining data definitely reflect a positive selection. Secondly, this dropout mechanism leads to a loss of power of hypothesis testing. The quality of life analysis of FACIT-AI revealed no significant differences between baseline and week 2 and baseline and week 4, respectively.

In the correlative analysis of cytokine / chemokine levels in the peripheral blood and malignant ascites, an efficient downregulation of VEGF in the patients' malignant ascites was observed after intraperitoneal treatment with Bevacizumab. Furthermore, Bevacizumab treatment stabilized the expression of pro-inflammatory, and possibly tumor-promoting, cytokines / chemokines in the peripheral blood. Finally, patients with strong VEGF neutralization evidenced an improved ParFS.

No new safety concerns occurred during the course of the study. Due to the terminal illness of the patients included, the main cause of death was progression of disease. A major increase in severe toxicity cannot be detected for the Bevacizumab arm. The proportion of patients with at least one

grade 3 to 5 event occurring is similar with 20/33 (61%) in arm 1 and 11/16 (69%) in arm 2. As one might have expected, fatigue is the most prevalent adverse symptom: 39% in arm 1, which is somewhat higher than the 3/16 (19%) finding in the placebo group. Nausea (45%) and vomiting (36%) are other frequent adverse events on Bevacizumab, together with pain (33%). The respective rates in the placebo arm are somewhat lower with 31% nausea and 33% pain; remarkably, no cases of vomiting were reported in the control group.

Altogether, intraperitoneal Bevacizumab was well tolerated but did not result in a significantly better symptom control of malignant ascites compared to the placebo group.

14 TABLES, FIGURES AND GRAPHS REFERRED TO BUT NOT INCLUDED IN THE TEXT

14.1 Demographic data

NA

14.2 Efficacy data

NA

14.3 Safety data

NA

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16 APPENDICES

16.1 Study information

16.1.1 Protocol and protocol amendments

AIO- Trial SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Short title: AIO-SUP-0108

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EudraCT Nr.: 2009-014725-16

Protocol identification number: AIO-SUP-0108

Protocol version: 2.0 (07/12/2009)

Confidentiality

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Drug Supply	Roche Pharma AG Deutschland

APPROVAL OF THE PROTOCOL

PD Dr. Ullrich Graeven – Representative of the Sponsor

Signature

07.12.2009
Date (DD Month YYYY)

Dr. Karin Jordan – Principal Investigator

Signature

Date (DD Month YYYY)

Dr. Axel Hinke - Statistician

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10.12.2009
Date (DD Month YYYY)

Dr. Anne L. Kranich – for the CRO

Signature

Date (DD Month YYYY)

APPROVAL OF THE PROTOCOL

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08. 12. 2009
Date (DD Month YYYY)

Dr. Axel Hinke - Statistician

Signature

Date (DD Month YYYY)

Dr. Anne L. Kranich – for the CRO

Signature

09.12.2009
Date (DD Month YYYY)

INVESTIGATOR'S AGREEMENT

I have read the attached protocol entitled

"Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers", dated 13. August 2009,

and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonisation Tripartite Guideline on Good Clinical Practice.

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the study sponsor.

Signature

Date (DD Month YYYY)

Investigator

Investigator's Institution

SYNOPSIS

Protocol No.	AIO-SUP-0108
Protocol Version (Date)	Version 2.0 (07/12/2009)
Title	Bevacizumab as a palliative treatment for patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers
Detailed Title	Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers
EudraCT No.	2009-014725-16
Principle Investigator	Dr. Karin Jordan Supportive Care Study Group, (for the Arbeitsgemeinschaft Internistische Onkologie, DKG) Clinic for Internal Medicine IV Department of Oncology/Hematology Martin Luther University Halle-Wittenberg
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Sponsor	AIO-Studien-gGmbH Straße des 17. Juni 106–108 “Tiergartentower” 10623 Berlin Phone: 030-322932933 Fax: 030-322932943
Study design	• Double-blind, placebo-controlled, randomized, multi-center, phase II • Patients will receive repeated intraperitoneal application of Bevacizumab/Placebo in a 2:1 ratio
Anticipated start date	09/2009
Duration of study	Approx. 2 years
Total number of centers	Approx. 20 centers
Study population	Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy

Rationale	Malignant ascites represents a severe clinical problem for physicians and patients. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites, and therapies that are commonly being used are only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers. Preclinical data strongly suggest that Bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites treated with Bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1]. Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.
Objectives	
Primary objective	• To evaluate the paracentesis-free survival (ParFS) following intraperitoneal application of Bevacizumab/Placebo
Secondary objectives	• To measure the frequency of paracenteses required for symptom control following intraperitoneal application of

	Bevacizumab/Placebo, by assessing the longest paracentesis-free period within the 12-week main observation period ("best response") • To measure the volume of ascites following intraperitoneal application of Bevacizumab/Placebo • To measure the effect of study treatment on the quality of life • To assess feasibility and safety of intraperitoneal application of Bevacizumab including pharmakocinetic of Bevacizumab • To evaluate the effect of an intraperitoneal application of Bevacizumab/Placebo on serum and ascites VEGF concentrations
Planned sample size	• In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized • Number of evaluable patients: 40 arm A (treatment), 20 in arm B (control)
Inclusion criteria	1. Age ≥ 18 years 2. Written informed consent has been obtained prior to inclusion into the study 3. Patient is capable and willing to comply with the study 4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma 5. Cytologically confirmed ascites OR diagnosis of an exsudate (serum albumin – ascites albumin < 1.1 g/dl) clinically suggestive for malignant ascites 6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy 7. Ascites clinically judged as not responsive to diuretics 8. At the time of inclusion paracentesis required at least twice within past 4 weeks. 9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. <u>Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.</u> 10. ECOG performance score 0-3 11. Life expectancy > 12 weeks 12. Laboratory parameters: <u>Hematology</u> • Neutrophils > 1,500/µl • Platelets > 100,000/µl • Hemoglobin ≥ 9 g/dl or 5.59 mmol/l <u>Hemastasiology</u> • INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

	Clinical chemistry • Creatinine clearance > 30 ml/min, serum creatinine < 1.5 x ULN • Serum bilirubin < 3.0 x ULN • Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 5 x ULN) Urinalysis: • Patients with < 2+ proteinuria on dipstick urinalysis. • Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 1.0 g of protein/24 h on 24-h urine collection.
Exclusion criteria	Patients with any of the following will not be eligible for participation: 1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible). 2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion 3. Hemorrhagic ascites (ascites hematocrit > 2%) 4. Transudative ascites (Serum albumin – ascites albumin > 1.1 g/dl) 5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed. 6. Therapy naïve patients 7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up. 8. Patients with extensive metastases of the liver making up > 70% of the total liver mass 9. Child C cirrhosis of the liver 10. Occlusion or thrombosis of the portal vein. 11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression. 12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, arrhythmia requiring medication, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, myocardial infarction within the last year before treatment start, peripheral arterial disease stage ≥ II. 13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder) 14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or

anticipation of the need for major surgical procedure during the course of the study.

15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic diseases.
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contra-indicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.
25. As the following medication(s) can have interactive effects and may interfere with the patient's ability to meet the study requirements, they cannot be administered during the clinical study:
    - Current or recent (within 10 days of first dose of study treatment) treatment with full-dose oral or parenteral anticoagulants or thrombolytic agents (e.g., marcumar therapy) for therapeutic purposes.
    - Current or recent (within 10 days of first dose of study treatment) chronic use of aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day).
26. Patients who participate currently in another clinical trial or patients who participated in another clinical trial during the last 30 days prior to enrolment.
27. Patients who have participated in this study before.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[1]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).

[1] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

	30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG). 31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study. 32. Patients who possibly are dependent on the sponsor or investigator.
Duration of study	Recruitment: 2 years Treatment: Patients will receive paracentesis as needed for symptom control. In addition, patients will receive up to 4 intraperitoneal administrations of Bevacizumab/Placebo after paracentesis has been performed. During the 8-week treatment period, a minimum interval of 14 days will be kept between applications of the study medication. Follow-up: End of treatment (EOT) is set at eight weeks after application of the first paracentesis within the treatment period for both arms of the study. Follow-up regarding response and safety in both arms of the study will be conducted at week 4 after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-month intervals for a total of 12 months.
Arms of study	• 48 patients randomized into arm A will receive repeated intraperitoneal application of Bevacizumab • 26 patients randomized into arm B will receive repeated intraperitoneal application of Placebo
Data Safety Monitoring Board	A Data Safety Monitoring Board (DSMB) will be established prior to the start of the study and will be responsible for reviewing safety data on a regular basis. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT).
Primary parameter	Efficacy: • Paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first)
Secondary parameters	Efficacy • Best Response (BR) representing the longest period of time

(in days) from
  - o one paracentesis until next paracentesis within the treatment period
  - o or, if longer, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up)
  - o or, if longer, from the last paracentesis performed within the treatment period until death (before end of the standard 4 week follow-up)
  - o or, if longer, from the last paracentesis performed within the treatment period until 4 week follow-up
- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites as indicated by body weight
- Quality of life as assessed by standardized questionnaires
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:
- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5.
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events).
- All adverse events.
- Changes in laboratory values and vital signs.
- Changes in ECOG performance status.

Pharmacokinetics of Bevacizumab and VEGF concentrations:
- Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.
- Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at bi-weekly intervals from last paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

Study procedure	After the initial screening procedure, eligible patients will enter the treatment part of the study. Patients will receive up to 4 intraperitoneal administrations of bevacizumab/placebo depending on clinical necessity of routine paracentesis for symptom relief. In case of unacceptable toxicity, treatment will be prematurely discontinued. A final follow-up regarding response and safety will performed for both arms at 4 weeks after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-months intervals for a total of 12 months.
Randomization procedure	Permuted block randomization will be applied to guarantee balanced group numbers.
Statistical considerations	
Sample size calculation	The primary variable is efficacy as indicated by the paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death, whichever occurs first). Under the assumption of an expected median ParFS in the control group of 14 days [3] and a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions: • type I error: 5% (one-sided) • power: 80% • observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year (Parsons et al., 2007) [3] • exponential shape of the Kaplan-Meier [4] curves In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized. As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death or end of the standard 4-week follow-up) within in the 12 week observation period. However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data.

Analysis plan	Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison. Secondary analyses: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend. Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test. In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test). Quality of life as assessed by the standardized questionnaires (FACIT-AI) will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a comparable manner.

Other analyses to be performed descriptively:

- Overall survival will be analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

INFORMATION TO BE GIVEN ON SAE / PREGNANCY

In the case of a serious adverse event (SAE) or pregnancy the following person must be contacted within one working day by fax:

Clinical Trial Manager	**GSO mbH**
Address:	**Johnsallee 30** **D-20148 Hamburg**
Phone:	**+49 40 44 19 54 60**
Fax:	**+49 40 44 19 54 78**

FLOW CHART: SCHEDULE OF ASSESSMENTS DURING THE STUDY

	Screening[1, 3]		Baseline[3,9]	Treatment Period[3]				Safety FU	Survival FU[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT[19] + 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]	X			X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X	
Paracentesis for symptom control	As indicated			X	As clinically indicated				
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

Notes

1. The screening visit S1 will take place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments have to be performed before administration of the study drug
4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.
7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals from last paracentesis until EOT. A final measurement will be performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis during the treatment period, sera will be collected at 14-day intervals from last paracentesis until EOT and a final sample will be collected at safety follow-up.
13. Urinalysis: Dipstick test for protein/albumin/erythrocytes at screening, thereafter for protein only. In case of protein ≥ 1+ with dipstick: Quantitative determination in 24 h urine is required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.

GLOSSARY OF ABBREVIATIONS

β-HCG	Beta human chorionic gonadotropin
AE	Adverse event
AIO	Arbeitsgemeinschaft Internistische Onkologie
ALT (SGPT)	Alanine aminotransferase
AMG	Arzneimittelgesetz (German Drug Law)
ANC	Absolute neutrophil count
aPPT	Activated partial thromboplastin time
AST (SGOT)	Aspartate aminotransferase
BR	Best Response
CHF	Congestive heart failure
CHO	Chinese hamster ovary
CNS	Central nervous system
CR	Complete response
CRF	Case report form
CT	Computed tomography
CTCAE	Common terminology criteria for adverse events
DGHO	Deutsche Gesellschaft für Hämatologie/Onkologie
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDTA	
ELISA	Enzyme-linked Immuno-absorbant Assay
EOT	End of treatment
ESF	Eligibility screening form
FACIT-AI	Functional Assessment of Chronic Illness Therapy - Ascites Index
GCP-V	GCP-Verordnung
h	Hour
IC	Informed consent
ICH	International Conference on Harmonization
IMP	Investigational medicinal product
INR	International normalized ratio
ITT	Intent to treat
iv	intravenous
LD	Longest diameter
LDH	Lactate dehydrogenase
LKP	Leiter der klinischen Prüfung (Co-ordinating Investigator)
m2	Square meter (body surface area)
mg	Milligram
min	Minute
mL	Milliliter
MRI	Magnetic resonance imaging
NB	Nota bene (please note)
NCI	National Cancer Institute
NCT	National Center for Tumor Diseases
NSAIDS	Non-steroidal anti-inflammatory drugs
NYHA	New York Heart Association
PVS	Peritovenous shunting
PD	Progressive disease
PR	Partial response
RECIST	Response Evaluation Criteria in Solid Tumors
rHuMAb	Recombinant humanized monoclonal antibody

rpm	Rounds per minute
SAE	Serious adverse event
SD	Stable disease
SmPC	Summary of product characteristics
UICC	International Union Against Cancer
ULN	Upper limit of normal
VEGF	Vascular Endothelial Growth Factor
VPF	Vascular Permeability Factor
w/wo	With or without

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PART I – STUDY DESIGN AND CONDUCT

1 BACKGROUND AND RATIONALE

1.1 Background

1.1.1 Malignant ascites

Growth of tumors in serous cavities such as the peritoneum is often accompanied by the accumulation of a protein-rich exudates and formation of malignant effusions is a common problem for patients with advanced-stage cancer. Malignant ascites is defined as an abnormal accumulation of fluid in the peritoneal cavity as a consequence of cancer occurring in association with a wide variety of neoplasms such as colorectal, stomach, pancreatic, ovarian, breast, and lung cancer [5].

Accumulation of massive amounts of malignant ascites is a significant cause of morbidity and mortality in patients with intra-abdominal tumors [6]. Accordingly, mean survival is only 20 weeks after ascites has been discovered [7]. Local fluid accumulations provoke discomfort and distress in many patients in advanced stages of their disease. By increasing abdominal pressure malignant ascites causes severe symptoms such as abdominal pain, bowel obstruction, shortness of breath, loss of appetite, nausea, cachexia, anorexia, reduced mobility, and fatigue [6]. Palliation of the symptomatic patient is the foremost goal and elimination of fluid accumulation in a patient with these symptoms will certainly improve the patient's quality of life and may even prolong survival [8]. Unfortunately, treatment of malignant ascites is problematic and challenging. No single method has been developed that works satisfactorily for the majority of patients and, accordingly, effective management of malignant ascites has been a frustrating problem for many physicians and their patients [8].

Currently, treatment modalities commonly applied for patients with malignant ascites include diuresis, salt restriction, paracentesis, and peritoneo-venous shunts, however, evidence for each of these treatment options is weak, and there are no randomized controlled trials evaluating their safety and efficacy. In addition, results achieved by the application of these conventional methods are variable, only occasionally provide prolonged relief from symptoms, and do not improve survival [9]. Accordingly, in contrast to treatments for the underlying cancer, there is no generally accepted and evidence-based guideline for the management of malignant ascites [10].

Reduced sodium intake together with diuretics are often used to treat malignant ascites but there is no consensus regarding effectiveness [11]. As in the case of other treatment modali-

ties for malignant ascites, there are no randomized controlled trials assessing the efficacy of diuretic therapy in malignant ascites. Available data are controversial and there are no clear predictors to identify which patients would benefit from diuretics. The use of diuretics should, therefore, be evaluated individually [10] and some authors have even concluded that medical therapies such as diuretics, and sodium and fluid restriction are not effective in ascites caused by malignancies [8].

Paracentesis has been the most common treatment option offering the advantage of a quick, simple, and relatively low-risk procedure with immediate symptom relief. However, as the patient's disease progresses, the frequency of hospital visits for the procedure increase as well. Patients are left with the choice between frequent hospital visits or waiting as long as possible between procedures until the ascites symptoms are no longer tolerable [8]. In addition, repeated paracenteses subject the patient to risks such as bleeding, infection, visceral perforation, and hypotension associated with invasive fluid depletion, and renal impairment [5]. Finally, the procedure itself is painful, inconvenient, and, most importantly, only temporarily effective [8].

To avoid repeated paracenteses, a peritoneo-venous shunting (PVS) may theoretically be considered. However, PVS requires hospitalization and surgery and is associated with significant risks both in shunt placement as well as in the ordinary function of the shunt [12]. Accordingly, major complications such as pulmonary edema, pulmonary emboli, clinically relevant disseminated intravascular coagulation, and infection, have to be expected in a significant number of patients undergoing PVS [8, 10]. Moreover, survival and quality are not improved in patients who have received PVS in comparison with patients treated with serial paracentesis [12]. Therefore, it is agreed that shunt insertion is contraindicated in patients with gastrointestinal cancer and malignant ascites due to relatively poor prognosis and limited survival [10, 13, 14].

Recently, the tri-specific antibody catumaxomab has been introduced to the treatment of malignant ascites [15, 16]. This CD3- and EpCAM-specific antibody is thought to stimulate the T cellular immune system as well as to induce MHC-unrestricted cytotoxicity and phagocytosis of tumor cells [17, 18]. Cohorts of patients suffering of ovarian cancer and gastric cancer have experienced symptom relieve after catumaxomab application and, more recently, a phase II/III trial has assessed catumaxomab in the treatment of malignant ascites, with significant results regarding to puncture-free survival and quality of life [19]. In conclusion, although these data need to be confirmed in daily clinical practice, catumaxomab might represent a new approach for the therapy of malignant effusions. However, the need for

placement of an intraperitoneal catheter for several days and the prolonged hospital admission required for the treatment severely limit its potential use in patients with end-stage cancer who require palliative treatment [16]. In addition, EpCAM is known to be expressed on a variety of healthy tissues including hepatocytes [20]. Accordingly, significant grade III-IV hepatic toxicity has been observed in patients treated with intravenous antibody, sometimes even associated with an impaired hepatic function [21]. Similar side effects have been described when catumaxomab was applied intraperitoneally [16]. Therefore, catumaxomab seems to be better suited for patients with a relatively good performance state, able to tolerate both the continuous presence of an intraperitoneal catheter in an inpatient setting as well as hepatic toxicity [21]. Most patients with malignant ascites, however, are unlikely to be candidates for this particular mode of treatment.

In conclusion, effective palliation of malignant ascites remains a difficult management issue. As patients are expected to survive only for a very limited period of time, a desirable treatment should (1) effectively alleviate associated symptoms, (2) be minimally invasive, (3) allow for rapid discharge from the hospital, (4) be relatively simple with low associated risk of complications, and (5) be of tolerable cost to the patient and his/her family [22].

1.1.2 Vascular Endothelial Growth Factor

A better understanding of the molecular mechanisms that regulate the formation of malignant effusions may offer ways to design novel and more effective modes of therapy for this severe cancer-related clinical problem. The etiology of malignant pleural effusions and ascites had traditionally been attributed to lymphatic obstruction caused by tumor spread into draining lymph vessels [23, 24, 25, 26]. It had also been suggested that tumor-induced angiogenesis might contribute to the development of ascites [27, 28]. In 1983, however, Senger et al. suggested an alternative possibility [29]. They isolated vascular permeability factor (VPF) from ascites of tumor-bearing animals and hypothesized that this factor secreted by tumor cells in a paracrine fashion was responsible for the cancer-related fluid accumulations [30]. A few years later, vascular endothelial growth factor (VEGF) was discovered as a potent stimulator of angiogenesis and was subsequently recognized to be identical to VPF [31, 32].

VEGF is a highly conserved 34-42 kD glycoprotein secreted by a large variety of human tumors [30, 33, 34]. In addition, peritoneal mesothelial cells [35], monocytes/macrophages infiltrating malignant effusions [36], and even tumor-infiltrating T cells [37] are capable of producing VEGF.

By interacting with two high affinity tyrosine kinase receptors (Flt-1 and KDR/Flk-1), which are selectively expressed in vascular endothelium [38], VEGF acts on endothelium both normal and newly induced by tumor angiogenesis [39].

Angiogenesis, the development of new blood vessels from pre-existing vasculature, is an essential component of solid tumor growth and metastasis [40, 41, 42, 43]. It is now generally accepted that solid tumor growth must be accompanied by angiogenesis to provide the vascular support necessary for the expanding tumor mass. However, not only does neovascularization permit further tumor growth of the primary tumor, but it also provides a pathway for migrating tumor cells to gain access to the systemic circulation and to establish distant metastases. Tumors express a variety of angiogenic factors in order to promote their own vascularization by activating the host endothelium. One angiogenic factor that is thought to play a decisive role in the vascularization of neoplastic tissue is VEGF which is a potent and specific mitogen for endothelial cells [44] and stimulates the full cascade of events required for angiogenesis *in vitro* and *in vivo* [45]. However, in addition to its ability to promote angiogenesis, VEGF is also capable of markedly augmenting the permeability of pre-existing microvasculature [29, 39, 46].

1.1.3 VEGF and malignant ascites

VEGF is over-expressed in a variety of tumors causing malignant ascites [47, 48, 49] and intratumoral VEGF expression correlates with an increased metastatic potential [49, 50] and poorer survival rates, among others, in gastrointestinal tumors, ovarian, breast, and lung cancer [48, 49, 51, 52, 53, 54, 55, 56]. Accordingly, serum concentrations of soluble VEGF have often been shown to be increased in patients with various solid tumors [47, 51, 57, 58, 59, 60, 61, 62] when compared to normal controls. Serum levels of VEGF correlate positively with the stage of the disease [58, 60, 61, 63], and elevated concentrations of VEGF in the peripheral blood of cancer patients might also be associated with a poorer overall and progression-free survival [58, 59, 61].

Initial studies had already indicated that the accumulation of malignant ascites results in large parts from an increased permeability of peritoneal lining vessels [64, 65]. However, until the identification of VEGF in malignant ascites, the molecular basis of peritoneal vascular hyperpermeability had not been deciphered. It was later shown that that malignant effusions derived from tumor-bearing mice and guinea pigs contain high concentrations of soluble VEGF [36, 66, 67, 68, 69] and that in mice injected with tumor cells increases in microvascular permeability of pre-existing small vessels located in tissues lining the peritoneal cavity as

well as the total volume of the peritoneal fluid correlated closely with the appearance of VEGF within the ascites [36, 67, 70]. Furthermore, it was shown that VEGF protein accumulated in the leaky blood vessels that line the peritoneal cavities of mice bearing ascites tumors [38, 39, 70] and that in low nanomolar or picomolar concentrations VEGF increased the permeability of venules and small veins for plasma proteins with a potency 10.000 times higher than histamine [29]. Finally, the expression level of VEGF by cancer cells has been shown to directly correlate with the tumor cell-induced production of ascites in the animal model [71, 72, 73]. Accordingly, transfection of renal cancer cells with VEGF cDNA or viral vectors encoding for VEGF increased the capacity of these cells to induce hyperpermeability of peritoneal blood vessels and ascites following implantation into mice [74, 75, 76, 77]. Even direct transfection of mouse peritoneum with VEGF was sufficient to cause an accumulation of ascites [75]. In contrast, transfection of tumor cell lines with VEGF antisense oligonucleotides resulted in a reduced formation of malignant effusions in the mouse [72, 76, 78]. Altogether, these collected findings allowed for the firm conclusion that local VEGF secretion is responsible in large parts for initiating and maintaining the ascites pattern of tumor growth.

In numerous human studies, markedly increased concentrations of VEGF have been found in malignant pleural effusions and ascites derived from patients with a large variety of solid tumors, such as ovarian cancer, gastric cancer, colorectal cancer, pancreatic cancer, breast cancer, and lung cancer. Generally, much lower concentrations were detected in non-malignant effusions caused by congestive heart failure, liver cirrhosis, or infections [34, 35, 38, 57, 62, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95]. In a very recent study by Atanackovic et al. concentrations of 21 different cytokines/chemokines were simultaneously analyzed in malignant and non-malignant ascites and it was clearly shown that VEGF is the one cytokine most strongly over-expressed in ascites related to cancer [96]. VEGF concentrations within the effusions are always higher than in the corresponding sera from the respective patients [47, 57, 92, 97] and the volume of malignant ascites correlates with VEGF concentrations within the effusion [98] and with the intensity of VEGF expression in abdominal tumors removed from the same patient [99], indicating a significant local release within the peritoneal or pleural cavity. Furthermore, concentrations of VEGF within human malignant effusions correlate with their capability to induce vascular leakage in an experimental model, an effect that can be blocked by treatment with an antibody directed against VEGF receptor Flk-1 [82, 83]. Most importantly, concentrations of VEGF in malignant ascites have recently been shown to correlate with chemosensitivity and represent an independent predictor of progression-free and overall survival of cancer patients [94, 97, 98].

1.1.4 Inhibition of VEGF activity as a potential therapy for malignant effusions

If VEGF is responsible for fluid accumulation in the environment of solid tumors then anti-VEGF therapies should be able to directly influence the development of malignant effusions in addition to possessing an immediate anti-tumor effect. Importantly, it has repeatedly been shown in *in vitro* experiments that the capacity of VEGF, present in the supernatant of tumor cell lines or in malignant ascites, to induce vascular hyperpermeability can indeed completely be neutralized using an antibody directed against VEGF [29, 30, 38, 67, 100]. Furthermore, it was already shown in an initial animal study by Senger et al. that an anti-VEGF antibody is able to block the increased peritoneal influx associated with the intra-abdominal presence of VEGF-secreting tumor cells *in vivo*. Since then, a number of studies have clearly demonstrated that the intraperitoneal application of anti-VEGF antibodies is safe and leads to impressive and often complete remissions of the local fluid accumulations in mice following inoculation with different carcinoma or sarcoma cell lines [67, 68, 73, 101, 102]. Consistent with these findings, the vascular permeability of microvessels lining the peritoneal cavity of tumor-bearing mice decreased significantly in the anti-VEGF antibody-treated animals compared with controls [67]. Antibody treatment to a lesser degree also inhibited tumor growth [67, 68, 101, 102] and prolonged the survival of mice inoculated with tumor cells [67]. A comparable preclinical efficacy was seen with tyrosine kinase inhibitors targeting VEGF receptors [103, 104] or with a soluble VEGF decoy receptor inhibiting VEGF [75, 105], and after intraperitoneal infusion of a VEGF antisense oligonucleotide [106], but not with conventional chemotherapy applied intraperitoneally alone [73]. Interestingly, the delay in tumor growth induced by the anti-VEGF antibody was mainly attributed to the blockage of ascites development and vascular permeability and not to the inhibition of VEGF-induced angiogenesis [67]. In this context, it has been hypothesized that increased vascular permeability within the peritoneum leading to the development of ascites might indeed result in increased shedding of mesothelial cells into the abdominal cavity facilitating indirectly peritoneal tumor dissemination [107].

Despite the very strong preclinical evidence for an obligatory role of VEGF in the formation of malignant ascites and for a possible therapeutic efficacy of anti-VEGF therapies in the setting of malignant effusions, there are currently no reports from clinical studies addressing this point in cancer patients. However, recently a number of articles reporting on small series of patients with malignant effusions treated off-label with bevacizumab have presented impressing results. It was first reported by Pichelmayer et al. that Bevacizumab might be active in malignant ascites [108]. Following their observation of a marked responses to treatment

with Bevacizumab in a patient with benign pleural effusion [109], they decided to apply a single dose of Bevacizumab intravenously at 15 mg/kg to two patients with malignant ascites due to colorectal cancer and adenocarcinoma of unknown origin, respectively. They found that both patients, in whom paracentesis was previously required at least every second week, treatment with bevacizumab was safe and highly successful. They observed significant reductions in ascites volume resulting in a discontinuation of repeat paracentesis. Moreover, both patients had a marked decrease in their VEGF plasma levels after treatment. [109]. In agreement with these early observations, Numnum et al. reported the intravenous application of bevacizumab (15 mg/kg every 3 weeks) to 4 heavily pretreated patients with end-stage ovarian cancer with the intent to palliate symptomatic ascites. In all 4 patients repeatedly applied paracenteses could be discontinued because of dramatically reduced levels of ascites after initiation of therapy with bevacizumab [110].

In a very recent publication, Hamilton et al. reported on the treatment of an 88-year-old patient receiving home hospice care with refractory ovarian cancer, a very poor functional status, and severe symptomatic ascites. They performed paracentesis and treated the patient with two subsequent doses (5 mg/kg) of intraperitoneal bevacizumab with dramatic improvement in her ascites and the quality of her final weeks of life [2]. The largest series of patients treated with intraperitoneal Bevacizumab has recently been presented by El-Shami et al. who evaluated the safety and efficacy if intraperitoneal administration of bevacizumab (5 mg/kg every 4 weeks) to a total of 9 patients with refractory ascites due to colorectal, breast, uterine, or ovarian cancer. Impressingly, malignant ascites resolved after a single intraperitoneal dose in every single patient without reaccumulation or repeat paracentesis over a median observation period of more than two months. Moreover, no grade 2-5 adverse events were observed [1].

1.1.5 Study Drug Bevacizumab

Bevacizumab (Avastin®) is a recombinant humanized monoclonal antibody (rHuMAb) to VEGF composed of human IgG1 framework regions and antigen-binding complementarity-determining regions from a murine antibody (A.4.6.1) that blocks the binding of human VEGF to its receptors [111] (further details can be found in the Investigator's Brochure). By neutralizing the biologic activity of VEGF produced by solid tumors, Bevacizumab has the capability to reduce the vascularization of tumors, thereby inhibiting the growth of the malignancy. In numerous mouse models, bevacizumab has clearly demonstrated an inhibition of human tumor growth. The administration of bevacizumab in hetero-transplant models of

colon carcinoma has shown to result in a reduction in microvessel formation and number of metastases.

A number of properties make some agents more favorable than others for intraperitoneal therapy. One important characteristic might be that the drug has activity in the malignancy to be treated. The efficacy of bevacizumab when used in combination with chemotherapy has been demonstrated in several prospective, randomized phase III studies [112, 113, 114, 115]. For example, in a phase III trial in patients with metastatic colon cancer, bevacizumab in combination with standard chemotherapy was found to increase overall and progression-free survival and response rates when compared with chemotherapy plus placebo [113]. Because of this and the results from other trials [116], bevacizumab has become widely used for the treatment of colorectal cancer and non-small-cell lung cancer and is being studied as part of the treatment regimen in a wide range of malignancies [117, 118, 119, 120]. In Germany, bevacizumab has been approved for first-line treatment of metastatic colorectal carcinoma (in combination with 5-FU/folic acid or 5-FU/folic acid/irinotecan [FOLFIRI]) in January 2005. Since January 2008, bevacizumab has been approved for the treatment of mCRC in combination with any fluoropyrimidin-based chemotherapy.

Bevacizumab is generally well tolerated and has an acceptable toxicity profile consisting primarily of hypertension and proteinuria. Other rare but important adverse effects, however, include delayed wound healing, arterial thrombosis, and bleeding [121]. Another potentially serious adverse effect of bevacizumab is gastrointestinal (GI) perforation and, although comparably infrequent, this potentially life-threatening complication has generated significant clinical interest. Overall, GI perforation was found to be an uncommon but well-documented side-effect of treatment in the phase III trials of bevacizumab, as well as in subsequent surveillance trials, with a reported incidence of 1% to 2% [113, 114, 117]. Accordingly, in an observational study by Hedrick et al. 1968 patients with unresectable colorectal cancer received bevacizumab and first-line chemotherapy and GI perforation was observed in 1.7% of patients [122]. Recently, a retrospective analysis was published examining adverse events in 1442 cancer patients who had received bevacizumab at M.D. Anderson Cancer Center over a 2-year period. Bowel perforation or fistula occurred in 1.7% of patients with a variety of malignancies including gastrointestinal cancers. In patients with colorectal cancer, for example, such adverse events were only observed in 6 of 478 cases (1.3%) Median time to perforation after the initiation of bevacizumab treatment was 71 days. Only five of all 1442 patients ultimately underwent surgical exploration and overall 30-day mortality rate was only 12.5% in these patients [123].

Though strong evidence identifying specific risk factors is lacking, investigators have urged caution when treating patients with known bowel implants or large tumor burden, prior radiation, and recent surgery or bowel obstruction [124]. Accordingly, in their phase II study with colorectal cancer patients Hurwitz et al. [113] identified colon surgery within 2 months as a risk factor, they also found a history of peptic ulcer disease and a partial or complete response to therapy as potential risk factors for perforation. Sugrue et al. [125] analyzed the same registry as Hedrick et al. and found no statistically significant associations between specific patient characteristics and an increased risk of GI perforations were identified, however, 67% of the patients with GI perforation showed at least one of the following findings: tumor at the site of perforation, obstruction, intra-abdominal abscess, intraabdominal carcinomatosis, acute diverticulitis, or prior abdominal or pelvic radiation therapy. Importantly, these potential risk factors were similar to those observed in the study by Badgwell et al. [123] as was the median time until first event of approximately two months. Finally, in a case report series of patients treated with bevacizumab for colorectal, lung, renal cell, and unknown primary cancer, ischemic bowel complications were more frequent in patients with a history of pelvic irradiation [126]. Although this series consisted of only 33 patients, bowel complications occurred in those three patients who had received infradiaphragmatic irradiation but in none of the 30 remaining patients who had not received this mode of treatment.

Further details on non-clinical and clinical data for bevacizumab are provided in the Investigator's Brochure.

1.2 Rationale

1.2.1 Rationale for the Study, Relevance of the Study and Study Design

Malignant ascites represents a severe clinical problem for physicians and patients being confronted with this common symptom of advanced-stage gastrointestinal cancer. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites and therapies which are currently being used are, if anything, only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for more effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers.

Preclinical data strongly suggest that bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites

treated with bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. While there are, so far, no reports on GI perforation resulting from intraperitoneal application of bevacizumab, there might still be a significant risk of such adverse reactions. However, we believe that palliative intraperitoneal treatment with bevacizumab is still indicated in these patients with advanced-stage gastrointestinal cancer patients who are capable of providing informed consent and who often severely suffer from symptoms associated with malignant ascites.

1.2.2 Rationale for the route of application and dosage selection

In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1].

Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.

2 OBJECTIVES OF THE STUDY

2.1 Primary Objectives

The **first** primary endpoint will consist of paracentesis-free survival (ParFS) which will be calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first).

2.2 Secondary Objectives

Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study.

The **second** endpoint (Best response; BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU.

The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase

Further evaluation of the efficacy, feasibility, and general safety of an intraperitoneal application of Bevacizumab in patients with malignant ascites.

Other measures of efficacy will be:

- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites present in the patient as indicated by body weight at each study visit during the treatment period
- Quality of life as assessed by standardized questionnaires (FACIT-AI) filled out by the patient and one questionnaire by the palliative group of the DGHO, which needs to be completed by the medical staff
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events)
- All adverse events
- Changes in laboratory values and vital signs
- Changes in ECOG performance status

Pharmacokinetics of Bevacizumab and VEGF concentrations:

Serum and ascites VEGF and Bevacizumab concentrations will repeatedly be analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at bi-weekly intervals from last paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

3 STUDY DESIGN

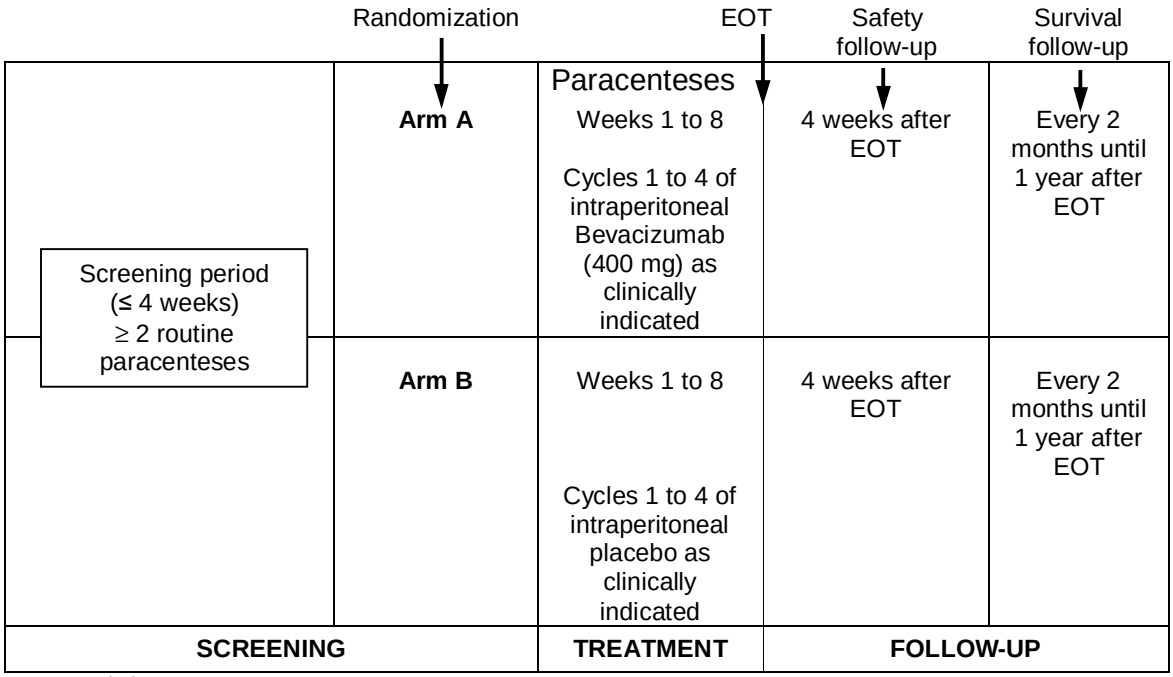
3.1 Overview of Study Design and Dosing Regimen

Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the tri-functional antibody catumaxomab or intraperitoneal chemotherapy. At the end of a screening phase of up to 4 weeks during which at least 2 routine paracenteses for symptom control of malignant ascites must have taken place, the screening visit S1 will take place. The screening visit must not take place earlier than 7 days before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. Eligible patients will be randomized into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period starts with the application of the first paracentesis for study purposes. Patients will receive up to 4 intraperitoneal administrations of Bevacizumab (400 mg absolute dose) or a placebo depending on clinical necessity of paracentesis for symptom relief. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days. In case of unacceptable toxicity, treatment will be prematurely discontinued.

At End of Safety FU, which is set at week 12 after the first paracentesis within the treatment period for both arms of the study, the study will allow for unblinding of the result of randomization in case of a missing clinical response of the ascites to study treatment. Unblinding has to be requested and approved by the sponsor. Patients will generally be followed regarding response and safety at 4 weeks (“safety follow-up”) following EOT. Puncture-free survival follow-up will take place every two months for one year after EOT.

A brief overview on the study design is given in Figure 1.

Figure 1. Study Design



EOT= End of treatment

3.2 Number of Patients/Assignment to Treatment

A total of 72 patients will be enrolled into the study (48 into treatment arm A, 26 into control arm B). At the baseline visit, each patient will receive an unique patient number that will be given to the investigator by FAX at the time of individual patient enrolment. The number assigned of each patient has to be documented by using a Patient Identification Log and on each patient's Case Report Form.

The rational for the 2:1 allocation is that the study may gain more information about patient responses to the new intervention, such as toxicity and side effects. Additionally, if the intervention turns out to be beneficial, more study subjects would benefit than under an equal allocation design. Moreover, from a psychological point of view, the higher chance to receive the intervention rather than placebo may render the trial participation more acceptable to the eligible patients. [155, 158]

3.3 Centers

An approximate number of 20 centers will participate in the study. Each center is expected to recruit at least 4 patients until the planned total number of 72 patients is reached. A list of all participating investigational sites including information regarding names of the principal investigators and contact details (address, phone, fax email) will be handled separately.

3.4 Study Duration

The study is planned to start in January 2010 with respect to first patient in (FPI) including a recruitment period of two years. The follow-up periods will be approximately 6 months in total. The total study duration is approximately 2 and a half year and the last patient will probably complete the study (last patient out; LPO) in June 2012.

Submission to EC/CA: September 2009

First Patient in (FPI): January 2010

Recruitment Phase: 2.0 years

FU phase: 0.5 year

Last Patient out (LPO): June 2012

4 STUDY POPULATION

4.1 Target Population

Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers will be eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least twice within the 4 weeks prior to inclusion into the study. Under no circumstances are patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

4.2 Inclusion Criteria

To be eligible for this trial, patients must fulfill the following criteria:

1. Age ≥ 18 years
2. Written informed consent has been obtained prior to inclusion into the study
3. Patient is capable and willing to comply with the study
4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma
5. Cytologically confirmed ascites OR diagnosis of an exsudate (serum albumin – ascites albumin < 1.1 g/dl) clinically suggestive for malignant ascites
6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
7. Ascites clinically judged as not responsive to diuretics
8. At the time of inclusion paracentesis required at least twice within past 4 weeks
9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the trifunctional antibody catumaxomab or intraperitoneal chemotherapy.
10. ECOG performance score 0-3
11. Life expectancy > 12 weeks
12. Laboratory parameters:

Hematology

– Neutrophils > 1,500/µl
– Platelets > 100,000/µl
– Hemoglobin ≥ 9 g/dl or 5.59 mmol/l

Hemastasiology

– INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

Clinical chemistry

– Creatinine clearance > 30 ml/min, serum creatinine < 1.5 x ULN
– Serum bilirubin < 3.0 x ULN
– Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 5 x ULN)

Urinalysis:

– Patients with < 2+ proteinuria on dipstick urinalysis.
– Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 1.0 g of protein/24 h on 24-h urine collection.

4.3 Exclusion Criteria

Patients with any of the following will not be eligible for the study:

1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion
3. Hemorrhagic ascites (ascites hematocrit > 2%)
4. Transudative ascites (Serum albumin – ascites albumin > 1.1 g/dl)
5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed.
6. Therapy naïve patients

7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
8. Patients with extensive metastases of the liver making up > 70% of the total liver mass
9. Child C cirrhosis of the liver
10. Occlusion or thrombosis of the portal vein.
11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, arrhythmia requiring medication, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, myocardial infarction within the last year before treatment start, peripheral arterial disease stage ≥ II.
13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder)
14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic disease
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.

25. As the following medication(s) can have interactive effects and may interfere with the patient's ability to meet the study requirements, they cannot be administered during the clinical study:
    a. Current or recent (within 10 days of first dose of study treatment) treatment with full-dose oral or parenteral anticoagulants or thrombolytic agents (e.g., marcumar therapy) for therapeutic purposes.
    b. Current or recent (within 10 days of first dose of study treatment) chronic use of aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day).
26. Patients who participate currently in another clinical trial or patients who participated in another clinical trial during the last 30 days prior to enrolment.
27. Patients who have previously participated in this study.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[2]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
32. Patients who possibly are dependent on the sponsor or investigator.

4.4 Concomitant Medication and Treatment

The initiation or continuation of any non-protocol-specific anti-tumor therapy is forbidden from inclusion into the study until EOT. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs will not be allowed from start of the screening phase until safety follow-up. This also excludes treatment with the tri-functional antibody catumaxomab or the intraperitoneal application of chemotherapy. Application of such treatments from start of the screening phase until safety follow-up will lead to the immediate discontinuation of the study for the given patient.

[2] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

All concomitant medication(s) must be reported in the Case Report Form (CRF). Any diagnostic, therapeutic, or surgical procedure performed during the study period should be recorded including the dates, description of the procedure(s) and any clinical findings. Patients should receive full supportive care including transfusion of blood and products, antibiotics, etc. where applicable. The treatment details should be recorded in the CRF.

The following concomitant medications for purposes other than malignant ascites are forbidden in this study (see also Section 4.3):

Full-dose Oral Coumarin-Derived Anticoagulants, Heparin, Aspirin:

Full-dose oral coumarin-derived anticoagulants (INR > 1.5), heparin, thrombolytic agents, or chronic, daily treatment with aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day), other NSAIDs or COX-2 inhibitors, are not allowed at entry into the study.

Prophylactic low-dose aspirin is a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [127] and is supported by an extensive body of literature [128]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus bevacizumab or placebo. In a retrospective exploratory analysis of patients in the bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [129]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event is not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins is permitted before and during study, as is low-dose aspirin (≤325 mg/day) and clopidogrel (≤ 75 mg/day).

Note: In patients who experience thrombo-embolic events during study treatment full dose anticoagulant are allowed and information on anticoagulant treatment (including doses) will be collected and recorded in the CRF.

INR will be assessed at baseline for all patients. In patients treated with oral coumarin-derived anticoagulants INR will be checked at least before start of application of Bevacizumab or routine paracentesis, respectively.

In patients treated with full-dose oral anticoagulants due to thrombo-embolic event during study treatment, INR must be checked at least every second day the first week of treatment, at least 2 times/week for the following treatment weeks until a stable therapeutic level of INR has been achieved and at least once every 3rd week when the weekly dose has been established and INR is stable with this dose (see also section 7.3.2).

The following is recommended regarding the use of concomitant medications:

Oral contraceptives: No dose modifications are required for patients on oral contraceptives.

5 SCHEDULE OF ASSESSMENT AND PROCEDURE

5.1 Screening Examination and Eligibility Screening Form

At the end of a screening phase the screening visit S1 will take place. The screening visit must not take place earlier than 7 days respectively 3 days (see Flow Chart) before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. After detailed oral and written information about the study, all patients willing to participate in this study have to provide a written Informed Consent (IC) before any study-specific assessment is performed.

An eligibility screening form (ESF) documenting the patient's fulfillment of the entry criteria for all patients considered for the study and subsequently included or excluded, is to be completed by the investigator and forwarded to the GSO mbH, Johnsallee 30, D-20148 Hamburg. All patients undergoing screening activities (documented by completion of an ESF for each patient) must be listed in the Patient Screening Log.

Patients who are considered for study entry, but who fail to meet the eligibility requirements, should also have an ESF completed with the reason for lack of eligibility given, since this provides information on the selected trial population. These patients will not be entered on the clinical trials database. The ESFs for patients who fail to meet the eligibility requirements should be kept in the study files at the sites. The same applies to patients who fulfill the entry criteria at the screening visit, but no longer at the baseline visit. For patients who did not sign the IC in the first, the ESF have not to be filled in. The patients will only be present in the pre-screening log.

A CRF should be filled out only for patients fulfilling the entry criteria both at screening and at baseline visits.

5.2 Study Assessments

5.2.1 Clinical Assessments

5.2.1.1 Assessment of Severity of Ascites

Baseline severity of malignant ascites will be assessed by calculating the mean time frame (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before start of the treatment phase

(screening phase). In addition, mean volumes of ascites (minus the volume of lavage fluid, if applicable) as well as body weight will be calculated for paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within 4 weeks of screening.

Between first paracentesis performed within the treatment period and until safety follow-up, the frequency of paracenteses clinically required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses with (arm A) or without (arm B) application of Bevacizumab. The Best response (BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or (if longer) from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or (if longer) from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU. Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study. The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase.

Between first paracentesis within the treatment period and until EOT, volume of ascites (minus the volume of lavage fluid, if applicable) drained during routine paracentesis for symptom relief with (arm B) or without (arm B) subsequent application of Bevacizumab will be recorded. Mean volumes and total volumes of ascites removed will be compared between groups and to the respective baseline values (mean and total volumes of paracentesis performed for symptom relief and not only diagnostic purposes during screening period) within the same group.

5.2.1.2 ECOG Performance Status

To be eligible for study entry, patients must have an ECOG performance score between 0 and 3 (see also section 4.2, Item 10). The patients' ECOG performance status (see section 18.3) will be assessed at the screening visit, before every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, assessment of ECOG performance status will be performed at least every 2 weeks from last paracentesis until EOT and at safety follow-up.

5.2.1.3 Assessment of Quality of Life

In order to assess quality of life, utilities will be generated using a standardized and validated instrument of quality of life questionnaire. Quality of life will be evaluated using the FACIT-AI questionnaire, which needs to be filled out by the patient. Another questionnaire developed by the DGHO palliative care group has to be completed by the medical staff. Quality of life (see section 18.4) will be assessed at the screening visit, at every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, assessment of QoL performance status will be performed at biweekly intervals from last paracentesis until EOT and at safety follow-up. FACIT-AI will be applied at the same time point during the study as the other quality of life questionnaire. Analyses will reveal the typical severity of each symptom during the different study phases to further assess burden of disease and treatment benefit. Additionally, a total ascites score derive from all symptoms will be calculated for each point in regard to the FACIT-AI.

5.2.2 Safety Assessments

5.2.2.1 Assessment of Toxicity

Throughout the treatment period and the 4-week safety follow-up period, patients will be assessed for toxicities attributable to therapy. Common terminology criteria for adverse events (CTCAE v3.0; see Investigator's File) will be used for grading. If necessary, the patient may be withdrawn from the study treatment. For details, see Section 7.3.

- **Medical history** including cancer and treatment history will be reviewed and recorded at the screening visit.
- **Concomitant medications** will be documented throughout treatment phase and the 4-week safety follow-up period. During the survival follow-up period, only anti-tumor drugs will be documented.
- A **physical examination** will be performed at the screening visit, before start of each treatment cycle and at the safety follow-up visit 4 weeks after the last treatment cycle (safety follow-up).
- **ECG** will be measured at screening and if clinically indicated.

- **Vital signs** (blood pressure, heart rate, body temperature and body weight), will be measured at the screening visit, before start of each treatment cycle and 4 weeks after the last treatment cycle (safety follow-up). Body height will be measured at screening only. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals form last paracentesis until EOT.
- **Adverse events** (see also Section 7.1): All patients will be closely monitored for adverse events (incl. survival) from Day 1 of the first treatment cycle through Week 4 after the last treatment cycle. Thereafter, patients will be followed up for progression and survival only. Adverse events will be graded according to the Common Terminology Criteria for Adverse Events (CTCAE v3.0).

5.2.3 Laboratory Assessments

5.2.3.1 Routine Laboratory Assessments

Blood samples will be taken for hematological and serum chemistry monitoring at each scheduled visit. The local laboratory will perform the analyses and provide reference ranges. All Assessments must be performed at the screening visit. Thereafter, assessments (with the exception of urinalysis and pregnancy test) will be repeated before each treatment cycle and 4 weeks after EOT (8 weeks after first paracentesis within the treatment period).

- **Hematology** - Hemoglobin, platelets, leukocytes, neutrophils,
- **Hemostasiology** - INR and aPTT.
- **Clinical Chemistry** - Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance, total protein.
- **Urinalysis -** Dipstick test for protein/albumin/erythrocytes will only be performed at the screening visit. Thereafter at every visit within the treatment period for protein only (see Flow Chart).. In case of protein ≥ 1+ with dipstick: Quantitative determination in 24 h urine is required. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals form last paracentesis until EOT.
- **Pregnancy test -** A serum β-HCG pregnancy test will be performed at the screening visit, if childbearing potential cannot be ruled out. Additional pregnancy test will be done as clinically indicated.

5.2.3.2 Routine analysis of malignant ascites

Samples of malignant ascites will be taken for hematological analysis and routine chemistry at screening and at each visit scheduled for paracentesis during the treatment period. Analyses will be performed in the local laboratory.

- **Hematology (2-5 ml EDTA-anticoagulated ascites)** – Hemoglobin, hematocrit, absolute numbers of total leukocytes and neutrophils.
- **Chemistry (5 ml heparinized ascites)** – Concentration of total protein and of albumin.

5.2.3.3 Pharmacokinetics of Bevacizumab and investigational analyses

10 ml of blood and 10 ml of ascites fluid will be collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum will be obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics will be obtained before each routine paracentesis performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis during the treatment period, heparinized peripheral blood and sera will be collected at 14-day intervals from last paracentesis until EOT and a final sample will be collected at safety follow-up. Serum concentrations of Bevacizumab will be analyzed by Xendo laboratory.

In addition to its role as a vascular permeability and as an angiogenic factor, VEGF also exerts a number of suppressive effects on the T cell-mediated immune system. First, VEGF functions as a chemo-attractant for CD34+ progenitor cells mobilizing these T cell-inhibiting cells into the tumor tissue where they inhibit the function of tumor-infiltrating T cells [130]. In addition, VEGF has repeatedly shown to reduce the phagocytic capacity and to inhibit the functional maturation of antigen-presenting dendritic cells (DC) *in vitro* and *in vivo* [131, 132, 133, 134, 135] and increased levels of VEGF are associated with reduced numbers of DC within the peripheral blood [136] and the tumor tissue [132]. In gastric cancer, for example, multivariate analysis showed that infiltration by DC was an independent prognostic indicator and there was an inverse correlation between the intratumoral density of DC and the expression of VEGF [132]. Accordingly, treatment with anti-VEGF antibody significantly improved the number and function of lymph node and spleen DC in tumor-bearing mice. Moreover, the efficacy of tumor vaccination with peptide-pulsed DC was dramatically enhanced by combining it with anti-VEGF antibody treatment [134, 137].

On the other hand, VEGF may also directly suppress immune effector cells by reducing cytokine-induced leukocyte-endothelial interactions *in vivo* [138] and by decreasing transendothelial migration of leukocytes [139]. Moreover, VEGF has recently been shown to exert an immediate effect on T cell immunity by inhibiting the thymic development of T cells [140] and enhancing Th2-type immunity [136, 141]. Accordingly, cytokines like IL-1β [107], IL-6 [142], and TGF-β [143, 144, 145] as well as a number of Th2-type cytokines (IL-4, IL-5, IL13) [145] might promote the production of intraperitoneal VEGF. In contrast, Th1-type cytokines, such as IFN-γ, inhibit VEGF production [145].

Finally findings for our group and others have recently indicated that the typical immune environment present within malignant ascites, which is marked by a dramatically elevated concentration of VEGF, might also contribute to the accumulation of immunosuppressive T regulatory cells (Tregs) within the effusion [96, 146]. These combined results suggest that expression of VEGF might be associated with tumor progression and poor prognosis not only because VEGF stimulates angiogenesis and vascular permeability, but also because it allows tumors to escape from attack by the immune system in patients with cancer.

In order to assess the effect of Bevacizumab-induced effects on angiogenic/vascular permeability factors as well as the acquired immune system 10 ml of serum and 10 ml of ascites fluid will be obtained from each patient included at each visit performed for routine paracentesis. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, sera will be collected at bi-weekly intervals from last paracentesis until EOT. A final serum (and ascites sample, if possible) will be obtained at safety follow-up.

As soon as possible after removal, heparinized blood and ascites samples, and sera will be centrifuged for 10 min at 1000g and supernatants will be divided into 3 x 1 ml volumes per sample type (3 ml total volume for each sample type, plasma and ascites fluid) and will immediately be frozen at a minimum of -20°C. After completion of the study by a given patient, the collective Plasma, ascites and serum samples of the respective patients will be picked up by TNT express who will also provide complete packaging and dry ice for the transport to the central investigational laboratory in Hamburg. For each participating center, four pick-up dates will be determined by the sponsor of the study.

All plasma and ascites samples in regard to the investigational as well as Serum and ascites samples for pharmacokinetic analyses will be firstly transported to the Laboratory for Tumor Immunology (Hamburg), Dr. Djordje Atanackovic. The Laboratory for Tumor Immunology (Hamburg) will afterwards forward the pharmacokinetic samples to the Xendo Laboratory.

5.2.3.3.1 Central laboratory for pharmacokinetics

Xendo Laboratory
LE I D E N - N E T H E R L A N D S
Bio Science Park, Archimedesweg 17, 2333 CM Leiden
P.O. Box 255, 2300 AG Leiden, The Netherlands
Tel +31 (0)71 524 40 00 Fax +31 (0)71 524 40 01
E-Mail office.pharmaservices@xendo.com

5.2.3.3.2 Central laboratory for investigational analyses

Dr. Djordje Atanackovic
University Medical Center Hamburg-Eppendorf
Center of Oncology
Laboratory for Tumor Immunology
Building N27, 4th floor, Room 04.083
Phone: 0049-40-7410-55032
Mobile: 0049-177-7329398
Fax: 0049-40-7410-55735
E-Mail: D.Atanackovic@uke.uni-hamburg.de

Samples can only be sent from Monday to Wednesday of each week. The central laboratory needs to be informed by email at least one week before the samples are sent. In addition, the courier needs to be informed of the pick-up date at least 24 hours before the samples are expected to be sent:

TNT Express Hotline (01805-633725)

To be reached on working days until 4.00 p.m.

In a first step, serum and ascites samples derived from 5 representative patients will be analyzed by antibody arrays for immunomodulatory cytokines and chemokines as well as a broad variety of angiogenic factors. Results derived from these analyses will be confirmed by analyzing serum and ascites samples of the whole patient collective for the respective cytokines/chemokines/angiogenic factors by ELISA. Sera of 50 anonymized blood donors will serve as controls.

In addition, 20 ml of fresh heparinized blood will be collected at each appointment for collection of serum for investigational analyses and the complete volume of malignant ascites removed at a given paracentesis performed during the treatment period and at safety follow-up will be collected from each patient treated in centers being in close proximity to the laboratory performing the investigational analysis (Laboratory for Tumor Immunology, Center of Oncology, II. Medical Clinic, University Medical Center Hamburg-Eppendorf). From the ascites material, tumor cells as well as endothelial cells and T cells will be separated in order to analyze by real-time PCR-based arrays which cells produce the angiogenic and/or immune factors creating the typical immune environment within malignant effusions. The same patient material as well as the peripheral blood will also be analyzed by flow cytometry for the presence of Tregs and other immunomodulatory cell types

5.2.4 Additional Assessments

Additional assessments will be required in the case of hypertension, proteinuria, thrombosis and hemorrhagic events as specified below. These additional assessments will be recorded on specific CRF forms in addition to the completion of the adverse events form.

- **Hypertension:** In the case of grade 3/4 hypertension, additional blood pressure measurements should be performed on a weekly basis (for the duration of trial therapy) until resolution of the event. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Proteinuria:** A 24-h urine collection is required in case of a protein ≥ 1+ dipstick result before the subsequent treatment cycle. Further study treatment should be in line with the recommendations as defined in Section 7.3.2. 24-h urine protein will be measured on a 2-weekly basis (for the duration of trial therapy) until level drops below 0.5 g/24 h.

- **Thrombosis:** In case of grade 3/4 thrombosis a blood sample for the following laboratory values should be taken prior to initiation of treatment for the event: INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Hemorrhagic events:** In case of a hemorrhagic event of grade 2 or higher, a blood sample for the following laboratory values should be taken prior to treatment for the event: Platelet count, INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.

6 INVESTIGATIONAL PRODUCT

6.1 Investigational Medicinal Product (IMP)

According to § 3 (3) GCP-V an investigational medicinal product is a pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial, including products already with a marketing authorization but used or assembled (formulated or packaged) in a way different from the authorized form, or when used for an unauthorized indication, or when used to gain further information about the authorized form.

The IMP in this study is Bevacizumab and its corresponding placebo.

6.2 Background Medication

Background medication for treatment of the primary malignancy or other medical problems of the patient will be applied at the discretion of the investigator. Background medication will not be supplied or reimbursed by Roche. The investigator needs to observe the summary of product characteristics of the different component of the background medication with special attention to contraindications.

6.3 Dose and Schedule of Test Drug Bevacizumab and comparator drug

Patients will receive up to 4 intraperitoneal administrations of the study drugs depending on clinical necessity of paracentesis for symptom relief. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.

Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control.

Following the application of an 18-22 G intraperitoneal catheter, the largest possible volume of malignant ascites will be drained. Thereafter, Bevacizumab or the comparator drug will be applied through the same catheter at a total volume of 100 ml. Bevacizumab will be applied at an absolute standardized dosage of 400 mg. The initial dose of the study drug will be delivered over 60±15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills) the following infusions may be delivered over 30±10

minutes. Following the complete application of the study drug the intraperitoneal distribution will be optimized by varying the patient's body position (10 min on the back, 10 minutes on right side, 10 min on left side).

6.4 Preparation and Administration of Bevacizumab

6.4.1 Drug Name, Formulation and Storage

Drug name:

INN: Bevacizumab
Trade name: Avastin®
Manufacturer: Roche Registration Ltd.

Formulation:

Bevacizumab is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The IMP has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

Drug name:

INN: Placebo
Trade name: NA
Manufacturer: Roche Registration Ltd.

Formulation:

Placebo is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The Placebo has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

6.4.2 Packaging and Labeling

Bevacizumab (400 mg, 25 mg/mL) and placebo will be supplied in 20 mL glass vials with a fill of 16 mL. The investigational medicinal product will be labeled according to § 5 GCP-V and internal requirements for blinding purposes.

6.4.3 Preparation of Study Drug

Bevacizumab infusions will be prepared according to the SmPC.

6.4.4 Route of Administration

Bevacizumab will be administered as an intraperitoneal infusion. See also 6.3.

6.4.5 Blinding and Randomization

Randomization will be performed stratified by center using computer-generated lists consisting of permuted blocks of randomly varying size in order to ensure equal group sizes within strata. The randomization lists will be generated by WiSP GmbH and transferred to the facility responsible for blinding, labeling and packaging of the study drugs.

6.4.6 Compliance

A pre-printed drug dispensing log is provided in the Investigator Site File and must be kept current and must identify the patient, and the amount of medication dispensed to each patient at each visit with the corresponding dates.

All medication supplies (empty containers, as well as partly used and unused medication) must be available for inspection at every monitoring visit. All unused medication, partly-used and empty packages must be returned by the investigator to Roche at the end of the study.

7 SAFETY ISSUES

The Investigator's Brochure will be used as reference document for Bevacizumab and will be provided to the investigators in the Investigator's File.

7.1 Adverse Events and Laboratory Abnormalities

It is the responsibility of the investigator(s) to report all adverse events in the case report form. Any serious adverse event (SAE) must be reported to GSO within one working day. GSO will forward the SAEs to the sponsor and Roche within one working day.

7.1.1 Clinical Adverse Events

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. Pre-existing conditions that worsen during a study are to be reported as Adverse Events. They can become Serious Adverse Events if they fulfill one of the seriousness criteria described in section 18.2.

All clinical adverse events (AEs) encountered during the clinical study (treatment period and the 4-week safety follow-up) will be reported on the AE page of the CRF.

Intensity of adverse events will be graded using the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE), version 3.0 (see Investigator's File) and reported in detail as indicated on the CRF. If an adverse event occurs which is not contained in the CTCAE v3.0, the four-point scale below will be used.

Mild:	Discomfort noticed but no disruption of normal daily activity.
Moderate:	Discomfort sufficient to reduce or affect daily activity.
Severe:	Inability to work or perform normal daily activity
Life-threatening:	Represents an immediate threat to life

Relationship of the adverse event to the treatment should also be assessed. Description of scales can be found in section 18.1.

Progression or deterioration of the malignancy under study (including new metastatic lesions and death due to disease progression) will be part of the efficacy assessment and should NOT be reported as AE or SAE.

Signs and symptoms of the malignancy under study should only be reported if:

1. Newly emergent (i.e. not present at baseline) and the association with the underlying malignancy and old/new metastatic lesions is unclear and/or
2. The investigator attributed deterioration of malignancy-associated signs and symptoms directly to the study drug.

Should there be any uncertainty regarding the attribution of the malignancy under study to an AE, it should be reported as an AE or a SAE accordingly.

7.1.2 Laboratory Test Abnormalities

Laboratory test results will be recorded on the laboratory results pages of the Case Report Form, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable. Laboratory-test-value abnormalities as such should not be reported on the AE page of the CRF as adverse events, unless they are treatment-emergent and they satisfy one or more of the following conditions for clinical significance:

1. Accompanied by clinical symptoms.
2. Leading to a change in study medication (e.g. dose modification, interruption or permanent discontinuation).
3. Requiring a change in concomitant therapy (e.g. addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).

Please note: any laboratory result abnormality fulfilling the criteria for a serious adverse event (SAE) should be reported as such, in addition to being recorded as an adverse event in the CRF.

7.1.3 Adverse Events of Special Interest

Adverse events of special interest are any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events. Adverse events of special interest are to be processed like serious adverse events.

7.2 Handling of Safety Parameters

7.2.1 Serious Adverse Events or Adverse Events of Special Interest (Immediately Reportable to GSO)

Any clinical adverse event or abnormal laboratory test value that is serious or of special interest occurring during the course of the study, irrespective of the treatment received by the patient, must be reported to the GSO within one working day of knowledge (expedited reporting). For each patient, all serious adverse events should be reported until 8 weeks since last Bevacizumab infusion. SAEs considered to have a causal relationship to the Investigational Product should be reported regardless of time elapsed since last Bevacizumab dose.

The definition and reporting requirements according to German Drug Law, GCP-V and ICH Guideline for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2 will be adhered (for details refer to section 18.2).

7.2.2 Treatment and Follow-up of Adverse Events

Adverse events, especially those for which the relationship to test drug is not "unrelated", should be followed up until they have returned to baseline status or stabilized. If a clear explanation is established it should be recorded on the CRF.

7.2.3 Follow-up of Abnormal Laboratory Test Values

In the event of unexplained abnormal laboratory test values, the tests should be repeated immediately and followed up until they have returned to the normal range and/or an adequate explanation of the abnormality is found. If a clear explanation is established it should be recorded on the CRF.

7.2.4 Pregnancy

A female patient must be instructed to immediately inform the investigator if she becomes pregnant during the study. The study treatment must immediately be stopped and the patient must be withdrawn from the study. Pregnancies occurring up to 90 days after the completion of the last treatment cycle must also be reported to the investigator. The investigator must report all pregnancies within one working day to the sponsor and the CRO. The investigator should counsel the patient, discuss the risks of continuing the pregnancy, and possible effects on the fetus. Monitoring of the patient should continue until conclusion of the pregnancy.

Pregnancy occurring in the partner of a patient participating in the study should also be reported to the investigator, the sponsor and the CRO. The partner should be counseled and followed as described above.

7.3 Dose Modifications for Toxicity

The observed toxicity will be graded according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events. Dose modifications will be implemented according to the observed toxicity grade as specified below.

7.3.1 General Notes Regarding Dose Modifications

For adverse events which are considered by the Investigator unlikely to develop into serious events and which do not result in a delay or interruption of therapy, treatment will be continued at the same dose without reduction or interruption. If the toxicity is attributable to a certain drug, dose modifications should only be made for this drug. In case of several toxicities occurring simultaneously, the highest dose reduction should be applied.

7.3.2 Dose Modifications for Bevacizumab

No dose reduction of Bevacizumab is foreseen for an individual patient. The dose of 400 mg Bevacizumab was proven to be a safe treatment for intravenous treatment. In addition, all studies applying Bevacizumab as an intraperitoneal infusion have used this dosage.

The initial study drug dose will be delivered over 60 ± 15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills), the second infusion may be delivered over 30 ± 10 minutes.

Bevacizumab-specific toxicities:

Any patient who develops any one of the following toxicities should not further receive Bevacizumab. (for details see below):

- Gastrointestinal perforation
- Fistula Formation involving internal organ
- Arterial thrombo-embolic events
- Symptomatic grade 4 thrombosis
- Grade 3/4 hemorrhagic events
- Grade 4 hypertension (hypertensive crisis)
- Grade 4 proteinuria (nephrotic syndrome)

If the Bevacizumab treatment has to be discontinued permanently, the patient must be withdrawn from the study treatment and followed up for PD and survival only.

Gastrointestinal perforation

Bevacizumab should be permanently discontinued in patients who develop gastrointestinal perforation.

Fistula formation

Bevacizumab should be permanently discontinued in patients who develop fistula involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder).

Thrombosis/Embolism

For patients who develop grade 3 or 4 thrombosis/embolism the following action is recommended:

- Arterial thrombo-embolic events: Bevacizumab should be permanently discontinued.
- Grade 3 or 4 venous thrombosis: Bevacizumab should be permanently discontinued.

Hemorrhage

Patients who develop grade 3 or 4 hemorrhage should permanently discontinue Bevacizumab treatment.

Hypertension

Patients should be monitored for the development or worsening of hypertension via frequent blood pressure measurement. Blood pressure measurements should be taken after the patient has been in a resting position for ≥ 5 minutes. Repeat measurements of blood pressure for verification should be undertaken if the initial reading is ≥ 140 mmHg systolic and/or ≥ 90 mmHg diastolic blood pressure.

- Grade 1 hypertension: Asymptomatic, transient (< 24 h) increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Intervention not indicated.
- Grade 2 hypertension: Recurrent or persistent (> 24 h) or symptomatic increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Monotherapy of anti-hypertensive may be indicated. Once controlled to < 150/100 mmHg, patients may continue Bevacizumab therapy.
- Grade 3 hypertension: Requiring more than one anti-hypertensive or more intensive therapy than previously. Bevacizumab should be withheld for persistent or symptomatic hypertension and should be permanently discontinued if hypertension is not controlled.

- Grade 4 hypertension: Life threatening consequence (e.g. hypertensive crisis). Occurrence of grade 4 hypertension should lead to permanent discontinuation of Bevacizumab. All doses of anti-hypertensive medicines should be recorded at all visits.

Proteinuria

All patients will have a dipstick urinalysis performed within 48 h prior to each Bevacizumab dose. All proteinuria toxicity, as determined by 24 h urine collection, will be graded according to CTCAE v3.0 classification. Adjustment of Bevacizumab administration for proteinuria of ≥ 2 g/24 h will occur according to the following guidelines, listed below.

First occurrence of proteinuria:

- < 2+ (dipstick): Administer Bevacizumab as scheduled; NO additional evaluation is required.
- ≥ 2+ (dipstick): Administer Bevacizumab as scheduled. Collect 24-hour urine to determine the total protein within 3 days prior to the next scheduled dose:
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24 h total protein.
    - Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
    - Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g/24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

Second and subsequent occurrence of proteinuria:

- < 3+ proteinuria (dipstick): administer Bevacizumab as planned. No additional evaluation is required.
- ≥3+ proteinuria (dipstick): administer Bevacizumab as planned and collect 24-h urine for determination of total protein within 3 days before the next scheduled Bevacizumab administration.
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24-h total protein.

- Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
- Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g. 24-h protein should be further monitored prior to each administration of bevacizumab until it has decreased to ≤ 1 g/24 h.

Nephrotic syndrome (grade 4, CTCAE v3.0): Discontinue Bevacizumab treatment.

7.3.3 Dose Modifications for Background Medication

Local standard practice and the recommendations provided in the respective SmPCs will drive dose reduction or interruption of chemotherapeutic compounds of the background medication.

7.4 Criteria for Discontinuation or Termination of the Study

7.4.1 Criteria for Discontinuation of the Treatment or Premature Withdrawal of the Patient

Treatment in both arms of the study will discontinue according to the protocol if any of the following apply:

- if any exclusion criteria develop
- Any patient who develops any one of the following toxicities:
  - Gastrointestinal perforation
  - Fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
  - Arterial thrombo-embolic events
  - Symptomatic grade 4 thrombosis
  - Grade 3/4 hemorrhagic events
  - Grade 4 hypertension (hypertensive crisis)
  - Grade 4 proteinuria (nephrotic syndrome)
- at the patient's request
- at the investigator's discretion
- Physician's judgment following an adverse event
- Termination by the Sponsor, or a regulatory authority
- Any other reason for withdrawal that the study physician or patient indicates is in the overall best interest of the patient

All patients who prematurely discontinue the treatment period will be followed up for safety and survival (exception: patient withdraws consent for further participation or patient is lost to follow-up).

7.5 Treatment after Discontinuation or Termination of the Study

After discontinuation or termination of the study, patients will be treated at the discretion of the investigator and according to medical routine. Further treatment modalities include continuing diuresis, salt restriction and repeated paracentesis. This would also include the possibility of applying the tri-functional antibody catumaxomab or intraperitoneal chemotherapy.

7.6 Warnings and Precautions

No evidence available at the time of the approval of this study protocol indicated that special warnings or precautions were appropriate, other than those noted in the IB (see Investigator File) for Bevacizumab.

8 BIOSTATISTICAL ASPECTS

8.1 Trial design and hypotheses

The trial is designed as a two-arm parallel group phase II study with a 2:1 randomization. Its primary objective is to obtain evidence, that bevacizumab treatment is effective in the symptom control of patients with malignant ascites. The primary endpoint is defined as paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or death, whichever occurs first). Thus, the following hypotheses will be tested:

H_0: ParFS (bevacizumab) ≤ ParFS (placebo)

H_1: ParFS (bevacizumab) > ParFS (placebo)

ParFS = paracentesis-free survival

As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis or death or end of the standard 4-week follow-up, whichever occurs first) within in the 12 week observation period.

H_0: BR (bevacizumab) ≤ BR (placebo)

H_1: BR (bevacizumab) > BR (placebo)

However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data. According to these hypotheses, the tests concerning the primary endpoints will be performed one-sided. In accordance with the phase II character of the trial, no type I error adjustment for multiplicity is performed.

8.2 Sample Size Calculation

Based on the results from Parsons et al. [3] the median ParFS in the untreated control group is expected to be around 14 days. In order to detect a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions:

- type I error: 5% (one-sided)
- power: 80%
- observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year [3]
- exponential shape of the Kaplan-Meier [4] curves

In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized.

The sample size calculation concerning the analysis of ParFS is based on methods described by Lachin and Foulkes [147]. Since a group sequential design allowing for interim analyses and early discontinuation will be adopted for this trial (see section 8.5), the above fixed sample size calculations serve only as an orientation for the maximum of patient numbers needed. The expected sample size and/or follow-up duration for reaching a conclusion may be considerably less than the number given above. This depends on the number and time points of interim looks as well as the actual difference in efficacy and the actual rates of recruitment and treatment failure. In the case of only one “look” (i.e. no interim analysis before completion of recruitment) the sample size coincides with that of the fixed-sample approach given above.

8.3 Evaluation categories of the patients

8.3.1 Intent-to-Treat Population

Intent-to-treat population (ITT) for ParFS is defined to include all randomized patients with at least one day of follow-up after the initial paracentesis. With respect to BR, this population consists of all patients with at least one documented paracentesis after the initial one or reaching the observation point at 4 weeks after EOT without any second paracentesis.

8.3.2 Per Protocol Population

The per protocol population (PP) is defined to include the intent-to-treat population excluding those patients with major protocol violations. As major protocol violations are considered those that may have an influence to the primary variables (e.g. not obtaining at least one application of study medication according to the protocol and randomization; receiving unauthorized antineoplastic or ascites treatment before observation of a second paracentesis or EOT + 4 weeks). Criteria that are assumed to have such an influence will be defined in a review meeting before data base lock.

8.3.3 Safety Population

According to the definition of the safety population all patients who received at least one dose of the trial medication and a safety follow-up, whether prematurely withdrawn or not, will be included in the safety analysis.

8.4 Methods of Statistical Analysis

The primary endpoints of the trial will be analyzed confirmatively (within the phase II framework) considering a global level for each hypothesis of $p < 0.05$ as significant.

All other parameters will be evaluated in an explorative or descriptive manner, providing means, medians, ranges, standard deviations and/or confidence intervals. If additional p values are calculated (e.g. in subgroup analyses or for secondary endpoints), they will be presented explicitly without referring to hypotheses or a significance level. Usually, no error adjustment for multiple testing will be performed. Thus the p values will reflect the comparison-wise error and not the experiment-wise error. All p values will be two-sided if not stated otherwise. The statistical methods described in this section are suited for the data and distributions usually expected in this type of trials. The suitability will be checked after data entry. If necessary, the statistical method will be modified accordingly and sensitivity analyses performed.

Demographic and prognostic baseline data will be checked for homogeneity between treatment groups. In case of relevant imbalances of other important prognostic factors the statistical method will be adjusted in order to achieve best possible comparability of the groups, and the results will be critically reviewed in comparison to the unadjusted ones.

Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. If the Peto logrank test [148, 149] is not appropriate because of violation of the proportional hazard assumption [150], Gehan's generalization of the Wilcoxon rank sum test for censored data [151] will be applied, preferably in its modification by Peto [148] and Prentice [152]. If necessary or prospectively defined at randomization, prognostic strata will be taken into account [149, 153].

Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison.

Secondary endpoints: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend.

Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test.

Quality of life as assessed by the standardized questionnaires and analyzed according to the recommendations of the respective developer, will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a similar way.

In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test).

Other analyses to be performed descriptively:

- Overall survival will be analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.
- For the ECOG performance status, the frequency of worsening, unchanged and improved status will be displayed by frequency tables in each scheduled visit.

The methods mentioned above are likewise suitable for the univariate evaluation of prognostic factors. Multivariate analyses may be performed by appropriate regression models (proportional hazard regression model [154], logistic regression).

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

8.5 Interim and Final Analysis

In case of longitudinal studies in severe chronic diseases, the study design should allow for interim analyses and, consequently, early stopping of the trial for ethical reasons [155]. A group sequential design will be adopted, using the α error spending function methodology by Lan and DeMets [156], implementing a use function according to the O'Brien-Fleming [157] boundary guideline. The design chosen will allow drawing conclusions from interim analyses in the following respect:

- acceptance of superiority of the bevacizumab arm (rejecting H0)

The additional option of accepting the control arm as non-inferior, when an interim result strongly suggests that the anticipated large difference of HR=0.5 will not be detected, is discarded, since a smaller difference might be discussed as relevant, too, especially if supported by secondary findings. Moreover, there is a less stringent need to stop the trial from an ethical point-of-view, if the data tend to similar results in both arms.

In order to keep an overall type I error of 5%, stopping boundaries will be calculated at the respective time points of interim evaluation, using the EaSt software (Cytel Software Corp., Cambridge, USA). This allows for arbitrary interim analyses, irrespective of time schedules and recruitment number. Moreover, the expected sample size and/or study duration for reaching a conclusion may be considerably smaller than in a fixed sample design (cf. 8.2), especially if the therapeutic difference is even larger than expected. The extent of "saving" patients mainly depends on the actual difference in efficacy as well as the actual rates of recruitment and failures. However, subsequent interim analyses will not be performed, unless an increment of at least 10 further evaluable patients are included in the database.

The final biometrical evaluation of the study and the compilation of the statistical report as part of the integrated clinical/biometrical report will be performed after completion and/or correction of all case report forms.

9 DATA QUALITY ASSURANCE

Data will be entered into a database by GSO Hamburg. The data will be filed in digital format and two people will enter the data independently. Data will be checked for accuracy using range, validity and consistency checks, as well as by cross-checking. Implausible or missing data may be corrected or completed after discussing with the investigator. The notes of amendment shall be filed together with the case report form (CRF). The validated data will be stored in a database and this process shall also be documented.

This database conforms to the requirements of ICH-GCP regarding the following:

- Validation of the system and data
- Presentation of SOPs
- Access and back-up systems
- Traceability and documentation of data amendments (audit trail)

Only authorised persons may access the database. Unauthorised access will be prevented via a security system.

10 STUDY COMMITTEES – DATA SAFETY MONITORING BOARD

A Protocol Committee consisting of experienced oncologists will be emplaced and will ensure the development of a clinically appropriate protocol. The Protocol Committee will also organize collaboration with reviewing statisticians and will make suggestions regarding centers to participate in the study.

The Data Safety Monitoring Board (DSMB) will be an independent board consisting of a group of 3 physicians with experience in oncology. A physician is not allowed to participate in this clinical trial while serving on the DSMB. The DSMB will be supported by an independent statistician, if necessary. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT). Safety variables (laboratory data, adverse events and serious adverse events) collected in the study will be reviewed every month during the first year. Later the DSMB can extend the regular intervals to e.g. 3 months. The collection and summary of these data will be prepared by the independent statistician. The members of the board will be primarily responsible for the clinical interpretation of the safety results. The board members can recommend a premature discontinuation of the trial or any other changes in the study conduct at any time, if required.

PART II - ETHICS AND GENERAL STUDY ADMINISTRATION

11 ETHICAL ASPECTS

11.1 Declaration of Helsinki/Good Clinical Practice

The Declaration of Helsinki is the accepted basis for clinical study ethics, and must be fully followed and respected by all engaged in research on human beings. Any exceptions must be justified and stated in the protocol. The latest version of the Declaration of Helsinki is available under http://www.wma.net/e/policy/b3.htm.

Additionally it is the responsibility of all engaged in research on human beings to ensure that the study is performed in accordance with the international Good Clinical Practice standards and according to all local laws and regulations concerning clinical studies.

11.2 Patient Information and Informed Consent

It is the responsibility of the investigator to obtain written informed consent from each patient participating in this study, after adequate explanation of aim, importance, anticipated benefits, and potential hazards and consequences of the study according to § 40 Abs. 2 and § 40 Abs. 2a AMG. Written informed consent must be obtained before any study specific procedures are performed. It must be also explained to the patient that they are completely free to refuse to enter the study or to withdraw from it at any time for any reason without incurring any penalty or withholding of treatment on the part of the investigator.

By signing the consent form, the subject/patient agrees with the "unwiderrufliche datenschutzrechtliche Einwilligung" according to § 40 Abs. 2a AMG. The subject/patient also agrees to allow the monitor/auditor/health authorities to verify the collected patient data against the subject's/patient's original medical records for the purpose of source data verification.

The informed consent form personally signed and dated by the patient must be kept on file by the investigator(s), and documented in the CRF and the subject's medical records. The investigator confirms obtaining the written informed consent to the sponsor.

If new safety information results in significant changes in the risk/benefit assessment, the consent form should be reviewed and updated if necessary. All patients (including those already being treated) should be informed of the new information and must give their written informed consent to continue in the study.

If the family doctors are informed of their patients' participation in the clinical study, this should be mentioned in the consent form.

11.3 Independent Ethics Committees and Regulatory Authorities

11.3.1 Approval of the Study by the Federal Regulatory Authority and Independent Ethics Committees

According to §§ 40-42 of the German drug law (AMG) it is the responsibility of the sponsor to obtain and maintain independent approval from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees to conduct the study.

The insurance coverage (study subject insurance) led down in § 40 AMG is in force. For each patient, the sponsor has provided insurance with HDI-Gerling Industrie Versicherung AG, Märkische Straße 23-33, 44141 Dortmund, contract number 48 158388 03055 390.

The sponsor names the "Leiter der klinischen Prüfung" (LKP) who has to be a physician with at least 2 years experience in the conduct of clinical trials of drugs according to § 4 (25) and § 40 (1) No. 5 AMG..

11.3.2 Notification of the Study

According to § 67 German drug law (AMG) the sponsor is responsible to notify competent regional authority about the study and all principal investigators of the participating investigational sites. If no other agreements are made, the sponsor will take over responsibility for investigator's obligation to report (§ 12 (3) GCP-V).

11.3.3 Report and Documentation Obligation

The sponsor is responsible to comply with the report and documentation obligation according to § 13 GCP-V.

The investigator is responsible to comply with the report and documentation obligation according to § 12 GCP-V.

12 CONDITIONS FOR MODIFYING THE PROTOCOL

Protocol modifications to ongoing studies must be made via amendment. The sponsor is responsible to obtain independent approval for the amendment from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees if required according to § 10 GCP-V. According to § 67 AMG competent regional authorities and the federal regulatory authority must be notified about the amendment, if they concern items according to § 12 Abs. 1 GCP-V.

13 DISCONTINUATION OR EARLY TERMINATION OF THE STUDY

13.1 Discontinuation of the Treatment or Premature Withdrawal of the Patient

All study specific withdrawal criteria are described in Section 7.4.1.

In addition, all patients have the right to withdraw from the study at any time for any reason. The investigator also has the right to withdraw patients from the study in the event of inter-current illness, adverse events and treatment failure after a prescribed procedure, protocol violations, cure, administrative reasons or other reasons.

The study has to be terminated when the patient starts a new tumor therapy.

An excessive rate of withdrawals can render the study uninterpretable; therefore, unnecessary withdrawal of patients should be avoided.

Should a patient decide to withdraw, all efforts will be made to complete and report the observations as thoroughly as possible.

The investigator should contact the patient to determine as completely as possible the reason for the withdrawal. A complete final evaluation at the time of the patient's withdrawal should be made with an explanation of why the patient is withdrawing from the study. If the reason for removal of a patient from the study is an adverse event or an abnormal laboratory test result, the principal specific event or test will be recorded on the Case Report Form.

13.2 Discontinuation or Early Termination of the Study

Both the sponsor and the investigator reserve the right to terminate the study at any time. Should this be necessary, both parties will arrange the procedures on an individual study basis after review and consultation. In terminating the study, the sponsor and the investigator will assure that adequate consideration is given to the protection of the patient's interests.

Following criteria could lead to a discontinuation or early termination of the study:

- Safety reason regarding patients' safety
- Negative benefit/risk assessment due to new information

In case of premature termination of the study all collected data have to be analyzed and a report has to be written. The sponsor has to inform the federal regulatory authority and the ethics committees within 15 days, giving detailed reason for the premature termination.

14 STUDY DOCUMENTATION, CRFS AND RECORD-KEEPING

14.1 Investigator's Files / Retention of Documents

The Investigator must maintain adequate and accurate records to enable the conduct of the study to be fully documented and the study data to be subsequently verified. These documents should be classified into two different separate categories Investigator's Study File, and subject/patient data.

The Investigator's Study File will contain all essential documents as the protocol/amendments, Case Report and Query Forms, patient information and informed consent form, Ethics Committee and federal regulatory authority approval, notification of the federal regulatory authority and competent regional authorities, drug records, staff curriculum vitae and authorization forms and other appropriate documents/correspondence etc.

Patient data include patient hospital/clinic records (medical reports, OP reports appointment book, medical records, pathology and laboratory reports, ECG, EEG, X-ray, etc.) and signed informed consent forms and patient screening and eligibility screening forms.

The investigator must keep these two categories of documents on file for at least 15 years (or more as legally required) after completion or discontinuation of the study. The documents must be archived in a secure place and treated as confidential material.

Should the investigator wish to assign the study records to another party or move them to another location, the sponsor must be notified in advance.

If the investigator can not guarantee this archiving requirement at the investigational site for any or all of the documents, special arrangements must be made between the investigator and the sponsor to store these in a sealed container(s) outside of the site so that they can be returned sealed to the Investigator in case of a regulatory audit. Where source documents are required for the continued care of the patient, appropriate copies should be made for storing outside of the site.

The sponsor must archive the protocol, documentation, approvals and all other essential documents related to the study, including certificates that satisfactory audit and inspection procedures have been carried out, as long as the test product(s) remains on the market.

All documents must be archived in a secure place and treated as confidential material.

14.2 Source Documents and Background Data

The investigator shall supply the sponsor on request with any required background data from the study documentation or clinic records. This is particularly important when Case Report Forms are illegible or when errors in data transcription are suspected. In case of special problems and/or governmental queries or requests for audit inspections, it is also necessary to have access to the complete study records, provided that patient confidentiality is protected. According to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy.

14.3 Audits and Inspections

This study may be audited by the sponsor, any person authorized by the sponsor or the competent health authority in order to determine the authenticity of the recorded data and compliance with the study protocol.

The investigator should understand that source documents for this trial should be made available to appropriately qualified personnel from sponsor/monitors/auditor/health authority inspectors after appropriate notification needed for source data verification and proper review of the study progress. The verification of the Case Report Form data must be by direct inspection of source documents. The investigator agrees to comply with the sponsor and regulatory authority requirements regarding the auditing of the study.

All material used in clinical studies are subjected to quality control.

14.4 Case Report Forms

For each patient enrolled, a Case Report Form must be completed and signed by the principal investigator or authorized delegate from the study staff. This also applies to records for those patients who fail to complete the study (even during a screening period if a Case Report Form was initiated). If a patient withdraws from the study, the reason must be noted on the Case Report Form. If a patient is withdrawn from the study because of a treatment-limiting adverse event, thorough efforts should be made to clearly document the outcome.

All forms should be typed or filled out using indelible ink, and must be legible. Errors should be crossed out but not obliterated, the correction inserted, and the change initialed and dated by the investigator or his/her authorized delegate. The investigator should ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the CRFs and in all required reports.

15 MONITORING THE STUDY

The monitor has the responsibility to familiarize the investigator(s) and the entire center staff involved in the study with all study procedures including the administration of study drug.

The GSOmbH must provide a trained monitor to assist the investigator(s) in conducting the clinical study. The monitor must visit the clinical study center on a regular basis and at least before the first patient has been enrolled, once during the course of the study, and at study completion. The monitor has the responsibility of reviewing the ongoing study with the investigator(s) to verify adherence to the protocol and to deal with any problems that arise. At all times the GSOmbH must maintain the confidentiality of the study documents. It is the responsibility of the study monitor to verify the study documents against the patient's original medical records.

The investigator (or his/her deputy) agrees to cooperate with the monitor to ensure that any problems detected in the course of these monitoring visits are resolved.

16 CONFIDENTIALITY OF TRIAL DOCUMENTS AND PATIENT RECORDS

The investigator and the sponsor (or designee) must assure that according to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy. CRFs or other documents should be submitted to the sponsor in a pseudonymous manner. The investigator should keep a patient identification log showing codes and names. The investigator should maintain documents not for submission to Sponsor, e.g., patients' written consent forms, in strict confidence.

17 PUBLICATION OF DATA

This study will be entered into the clinical trial protocol registry and clinical results database.

The sponsor is responsible for the timely reporting of study data. An integrated clinical study report (CSR) has to be completed one year after end of the study (whether completed or prematurely terminated). The report has to be approved by the responsible specialist chosen by the sponsor, the project manager of the CRO, the statistician and the principal investigator/LKP (for multicenter studies) by provision of their signatures.

The results of this study may be published or presented at scientific meetings as soon as after completion of the study. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor prior to submission.

In a multicenter study, it must be ensured that the data from one center are not published before the publication of the whole study. Roche reserves the right to review the manuscript(s) before their submission for publication or presentation. This is not intended to restrict or hinder publication or presentation, but is to allow the sponsor to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the investigator(s).

In accord with standard editorial and ethical practice, Roche will generally support publication of multicenter trials only in their entirety and not as individual center data. In this case, a coordinating investigator will be designated by mutual agreement.

Any formal publication of the study in which input of Roche personnel exceeded that of conventional monitoring will be considered as a joint publication by the investigator and the appropriate Roche personnel. Authorship will be determined by mutual agreement.

18 APPENDICES

Appendix 1: Adverse Events Categories for Determining Relationship to Test Drug

Appendix 2: Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

Appendix 3: ECOG Performance Status

Appendix 4: Quality of Life Questionnaires

18.1 Appendix 1 – Adverse Events Categories for Determining Relationship to Test Drug

(a) Probable (must have first three)

This category applies to those adverse events that are considered, with a high degree of certainty, to be related to the test drug. An adverse event may be considered probable, if:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It cannot be reasonably explained by the known characteristics of the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It disappears or decreases on cessation or reduction in dose. (There are important exceptions when an adverse event does not disappear upon discontinuation of the drug, yet drug-relatedness clearly exists: e. g. (1) bone marrow depression, (2) tardive dyskinesias).
4. It follows a known pattern of response to the suspected drug.
5. It reappears upon rechallenge.

(b) Possible (must have first two)

This category applies to those adverse events in which the connection with the test drug administration appears unlikely but cannot be ruled out with certainty. An adverse event may be considered possible if, or when:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It may have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It follows a known pattern of response to the suspected drug.

(c) Remote (must have first two)

In general, this category is applicable to an adverse event that meets the following criteria:

1. It does not follow a reasonable temporal sequence from administration of the drug.
2. It may readily have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.

3. It does not follow a known pattern of response to the suspected drug.
4. It does not reappear or worsen when the drug is readministered.

(d) Unrelated

This category is applicable to those adverse events which are judged to be clearly and incontrovertibly due only to extraneous causes (disease, environment, etc.) and do not meet the criteria for drug relationship listed under remote, possible, or probable.

	Probable	**Possible**	**Remote**	**Unrelated**
Clearly due to extraneous causes	–	–	–	+
Reasonable temporal association with drug administration	+	+	–	–
May be produced by subject clinical state, etc.	–	+	+	+
Known response pattern to suspected drug	+	+	–	–
Disappears or decreases on cessation or reduction in dose	+	–	–	–
Reappears on rechallenge	+	–	–	–

18.2 Appendix 2 – Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject, administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

Adverse reactions are all untoward and unintended responses to an investigational medicinal product related to any dose administered.

A serious adverse event or serious adverse reaction is any experience that suggests a significant hazard, contraindication, side effect or precaution. It is any Adverse Event that at any dose fulfils at least one of the following criteria:

- is fatal (results in death) *(NOTE: death is an outcome, not an event)*
- is life-threatening *(NOTE: the term "life-threatening" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not refer to an event which could hypothetically have caused a death had it been more severe.)*
- required in-patient hospitalization or prolongation of existing hospitalization
- results in persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- is medically significant or requires intervention to prevent one or other of the outcomes listed above

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the outcomes listed in the definitions above. These situations should also usually be considered serious.

Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

An unexpected Adverse Event is one, the nature or severity of which is not consistent with the applicable product information.

Causality is initially assessed by the investigator. With respect to report and documentation obligation (regulatory authorities, ethics committees and other investigators) for Serious Adverse Events, causality can be one of 2 possibilities:

- No (unrelated; equals not drug related).
- Yes (remotely, possibly, probably or definitely drug related).

All adverse events not assessed as definitive "not drug related" by either the investigator or Roche will be considered as adverse drug reaction.

A suspected unexpected serious adverse reaction (SUSAR) is a serious adverse reaction, the nature, or severity of which is not consistent with the applicable product information.

It is important that the severity of an adverse event is not confounded with the seriousness of the event. For example, vomiting which persists for many hours may be severe, but is not necessarily a serious adverse event. On the other hand, stroke which results in only a limited degree of disability may be considered a mild stroke, but would be a serious adverse event.

A serious adverse event occurring during the study or which comes to the attention of the investigator within 8 weeks after stopping the treatment or during the protocol-defined follow-up period, if this is longer, whether considered treatment-related or not, must be reported. In addition, a serious adverse event that occurs after this time, if considered related to test "drug", should be reported.

Such preliminary reports will be followed by detailed descriptions later that will include copies of hospital case reports, autopsy reports and other documents.

For serious adverse events, the following must be assessed and recorded on the adverse events page of the Case Report Form: intensity, relationship to test substance, action taken, and outcome to date.

Document and report obligation have to be adhered according to the national and international laws and regulations.

Contact details and Fax No. for SAE and pregnancy reporting refer to page 17.

18.3 Appendix 3 – ECOG Performance Status

GRADE	SCALE
0	Fully active, able to carry out all pre-disease performance without restriction. (Karnofsky 100%)
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light housework, office work. (Karnofsky 80-90%)
2	Ambulatory and capable of all self-care, but unable to carry out work activities. Up and about more than 50% of waking hours (Karnofsky 60-70%)
3	Capable of only limited self-care, confined to bed or chair more than 50% of waking hours. (Karnofsky 40-50%)
4	Completely disabled. Cannot carry out any self-care. Totally confined to bed or chair. (Karnofsky 10-30%)
5	Dead (Karnofsky 0%)

18.4 Appendix 4 – Quality of Life Questionnaires

FACIT ASCITES INDEX (Patientenfragebogen)

Nachfolgend finden Sie eine Liste von Aussagen, die von anderen Personen mit Ihrer Krankheit für wichtig befunden wurden. **Bitte geben Sie jeweils an, wie sehr jede der folgenden Aussagen im Laufe der letzten 7 Tage auf Sie zugetroffen hat, indem Sie die entsprechende Zahl ankreuzen.**

		Überhaupt nicht	Ein wenig	Mäßig	Ziemlich	Sehr
C6	Ich habe einen guten Appetit	0	1	2	3	4
GF5	Ich schlafe gut	0	1	2	3	4
BMT5	Ich bin in der Lage, mich alleine fortzubewegen	0	1	2	3	4
B1	Ich leide unter Atemnot	0	1	2	3	4
GP2	Mir ist übel	0	1	2	3	4
O2	Ich habe mich übergeben	0	1	2	3	4
ACT11	Ich habe Schmerzen in der Magengegend	0	1	2	3	4
O1	Ich habe Schwellungen im Magenbereich	0	1	2	3	4
GP1	Mir fehlt es an Energie	0	1	2	3	4
ACT10	Wenn ich esse, fühle ich mich rasch satt	0	1	2	3	4
BL2	Ich muss häufiger Wasserlassen als üblich	0	1	2	3	4
Cx6	Ich leide an Verstopfung	0	1	2	3	4
AI1	Ich bin bekümmert	0	1	2	3	4

Patientenlebensqualitätsfragebogen

(modifiziert nach der Palliativgruppe der DGHO)

Der Fragebogen ist durch das medizinische Personal zu ergänzen

Betreuungsverlauf

Datum des ersten Besuchs ☐☐☐☐☐☐

Ort des ersten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Datum des letzten Besuchs ☐☐☐☐☐☐

Ort des letzten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Begründung des Abschlusses der Home Care-Versorgung ★
- ● Tod des Patienten
- ● Änderung des Wohnorts des Patienten
- ● unerwartete Verbesserung des Gesundheitszustands des Pat.
- ● Fortsetzung der tumorspezifischen Therapie
- ● Krankenhauseinweisung
- ● Wechsel des HC-Arztes
- ● sonstiges: _____

Sterbedatum ☐☐☐☐☐☐

Sterbeort
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Palliativstation
- ● andere Krkhs.-Station
- ● sonstiges

Ausprägung von Symptomen zum Zeitpunkt der Aufnahme und nach erfolgter Einstellung

alles Zutreffende nach Schweregrad ausfüllen (Schweregrad: 0-ohne, 1-gering, 2-mittel, 3-stark) ■ vor Intervention ☐ nach Intervention

■	Schmerzen ☐	■	Dysphagie ☐	■	(Lymph-)Ödem ☐	■	Krampfanfälle ☐
■	Schwäche ☐	■	Obstipation ☐	■	Juckreiz ☐	■	motor. Unruhe ☐
■	Appetitlosigkeit ☐	■	Diarrhoe ☐	■	Dekubitus ☐	■	Verwirrtheit ☐
■	Übelkeit ☐	■	Ascites ☐	■	exulc. Wunde ☐	■	Schlafstörung ☐
■	Erbrechen ☐	■	Dyspnoe ☐	■	Harnverhalt ☐	■	Angst ☐
■	(Tumor-)Blutung ☐	■	Husten ☐	■	Lähmungen ☐	■	Depression ☐

Therapie bei Aufnahme und im Verlauf / zum Ende

Schmerztherapie WHO
- ● keine ○
- ● nur bei Bedarf ○
- ● WHO Stufe I ○
- ● WHO Stufe II ○
- ● WHO Stufe III ○

Koanalgetika / Begleitmedikation ★
- ● Nichtopioide Analget. ○
- ● Antiemetika ○
- ● Antidepressiva ○
- ● Antikonvulsiva ○
- ● Kortikosteroide ○
- ● Laxanzien ○
- ● Neuroleptika ○
- ● Sedativa / Hypnotika ○

Opioidtherapie ★

Substanz(en) ★
- ● Morphin ○
- ● Hydromorphon ○
- ● Fentanyl ○
- ● Oxycodon ○
- ● Buprenorphin ○
- ● Levomethadon ○
- ● Piritramid ○

Applikationsform(en) ★
- ● oral / PEG / rektal ○
- ● transdermal ○
- ● s.c. Injektion ○
- ● s.c. Dauerinfusion ○
- ● i.v. Dauerinfusion ○
- ● pendural ○

sonstige Palliativmaßnahmen ★
- ● PEG / transnasale Sonde ○
- ● zentraler venöser Zugang ○
- ● enterale Ernährung ○
- ● parenterale Ernährung ○
- ● i.v. Flüssigkeitssubstitution ○
- ● s.c. Flüssigkeitssubstitution ○
- ● Ascitespunktion(en) ○
- ● Pleurapunktion(en) ○
- ● palliative Chirurgie ○
- ● palliative Radiatio ○
- ● palliative Chemotherapie ○
- ● palliative endoskop. Eingriffe ○
- ● nichtspezialisierter Pflegedienst ○
- ● Palliativpflegedienst ○

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AIO- Trial SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Amendment 1

dated 07.06.2010

Study Code: AIO SUP 0108

EudraCT-No: 2009-014725-16

Approval of the Amendment:

I hereby approve the Amendment 1 to the protocol.

Priv. Doz. Dr. Ulrich Graeven
Sponsor

10-06-10
Date

Priv. Doz. Dr. Karin Jordan
Coordinating Investigator

14-06-2010
Date

1. Rationale

1.1 Change of Inclusion/Exclusion Criteria

The aim of this Amendment is to adapt the inclusion and exclusion criteria to the real clinical situation of patients with ascites due to gastrointestinal cancers. The criteria should be rendered as feasible as possible without compromising patients' safety. Therefore, the following criteria will be changed:

Inclusion Criterion No. 5 - Diagnosis: To simplify the diagnosis of an exsudate, the amount of total protein in ascites will be measured instead of the serum albumin – ascites albumin gradient. In addition, patients with a peritoneal carcinosis confirmed by CT, MRT or ultrasound should also be allowed to enter the study.
Therefore the inclusion criterion will be changed to:

Cytologically confirmed ascites OR diagnosis of an exsudate **(total protein in ascites > 30 g/l**) clinically suggestive for malignant ascites **OR morphological diagnosis of peritoneal carcinosis by CT, MRT or ultrasound**

Inclusion Criterion No. 12 – Laboratory Parameters:
The upper limits for creatinine, alkaline phosphatase and transaminases should be adapted to the clinical situation of the patient population. To allow participation of patients with proteinuria due to concomitant diseases such as diabetes, patients with ≥ 2 + in dipstick urinalysis are considered eligible for the study if they demonstrate < 2.0 g of protein/24 h on 24-h urine collection.
Therefore, the criterion will be changed to:

Clinical chemistry
- Creatinine clearance > 30 ml/min, serum creatinine < **2.5 x ULN**
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < **7 x ULN**)

Urinalysis:
- Patients with < 2+ proteinuria on dipstick urinalysis.
- Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate **< 2.0 g** of protein/24 h on 24-h urine collection.

Exclusion Criterion No. 12 – Cardiovascular Diseases: only patients with uncontrolled arrhythmia should be excluded and myocardial infarction within the last year will be deleted. Therefore the criterion will be changed to:

Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, **uncontrolled arrhythmia**, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II.

Exclusion Criterion No. 15 – Treatment with bevacizumab: It will be clarified that prior treatment with bevacizumab is not exclusionary. Therefore, the criterion will be changed to:

Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. **Prior treatment with Bevacizumab for primary malignancy is not exclusionary.**

Exclusion Criterion No. 25 – Anticoagulant Therapy: Anticoagulant therapy in combination with bevacizumab should be allowed as long as the INR or PTT is within therapeutic limits, as this is currently the standard in other clinical trials with bevacizumab as well. Therefore, the exclusion criterion will be changed to:

With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Exclusion Criterion No. 26 – Participation in other clinical trials. It will be clarified that this criterion refers to clinical trials with non-approved investigational drugs and does not include the follow-up phase of such trials. Therefore, the criterion will be changed to:

Patients who participate currently in another clinical trial or patients who participated in another clinical trial **and received a non approved investigational drug** during the last 30 days prior to enrolment **(e.g. follow up within the trial is not exclusionary).**

1.2 Additional Safety Information

In addition, the Amendment will include recent information from a Dear Health Care Professional Letter issued by Roche regarding allergic reactions observed with bevacizumab.

1.3 Administrative Changes

In addition, some minor corrections and clarifications have been performed throughout the protocol to synchronize the wording.

2. Tracking of Changes

2.1 Protocol

The following text was changed from protocol Version 2.0, 07.12.09 to Version 3.0, 07.06.10:

Section Synopsis, Page 8, Inclusion Criteria

Old text:
5. Cytologically confirmed ascites OR diagnosis of an exsudate (serum albumin – ascites albumin < 1.1 g/dl) clinically suggestive for malignant ascites

New text:
5. Cytologically confirmed ascites OR diagnosis of an exsudate **(total protein in ascites > 30 g/l)** clinically suggestive for malignant ascites **OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound**

Section Synopsis, Page 9, Inclusion Criteria

Old text:
12. Laboratory parameters

Clinical chemistry

- Creatinine clearance > 30 ml/min, serum creatinine < 1.5 x ULN
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 5 x ULN)

Urinalysis:

- Patients with < 2+ proteinuria on dipstick urinalysis.

Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 1.0 g of protein/24 h on 24-h urine collection.

New text:
12. Laboratory parameters

Clinical chemistry

- Creatinine clearance > 30 ml/min, serum creatinine < **2.5 x ULN**
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < **7 x ULN**)

Urinalysis:

- Patients with < 2+ proteinuria on dipstick urinalysis.
- Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate **< 2.0 g** of protein/24 h on 24-h urine collection.

Section Synopsis, page 9, Exclusion Criteria

Old text:
4. Transudative ascites (Serum albumin – ascites albumin > 1.1 g/dl)

New text:
4. Transudative ascites (**total protein in ascites < 30 g/l**)

Section Synopsis, page 9, Exclusion Criteria

Old text:
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, arrhythmia requiring medication, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, myocardial infarction within the last year before treatment start, peripheral arterial disease stage ≥ II.

New text:
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, **uncontrolled arrhythmia**, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, ~~**myocardial infarction within the last year before treatment start**~~, peripheral arterial disease stage ≥ II.

Section Synopsis, page 10, Exclusion Criteria

Old text:
15: Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up

New text:
15: Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. **Prior treatment with Bevacizumab for primary malignancy is not exclusionary**

Section Synopsis, page 10, Exclusion Criteria

Old text:
25. As the following medication(s) can have interactive effects and may interfere with the patient's ability to meet the study requirements, they cannot be administered during the clinical study:

- Current or recent (within 10 days of first dose of study treatment) treatment with full-dose oral or parenteral anticoagulants or thrombolytic agents (e.g., marcumar therapy) for therapeutic purposes.
- Current or recent (within 10 days of first dose of study treatment) chronic use of aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day).

New text:
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Section Synopsis, page 10, Exclusion Criteria

Old text:
26. Patients who participate currently in another clinical trial or patients who participated in another clinical trial during the last 30 days prior to enrolment.

New text:

26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).

Section Synopsis, page 17, Flow Chart

Old text :

	Screening[1, 3]		**Baseline**[3,9]	**Treatment Period**[3]				**Safety FU**	**Survival FU**[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT**[19] **+ 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]	X			X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X	
Paracentesis for symptom control	As indicated			X	As clinically indicated				
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

New text:

	Screening[1, 3]		**Baseline**[3,9]	**Treatment Period**[3]				**Safety FU**	**Survival FU**[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT**[19] **+ 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]	~~X~~			X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X[20]	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X[20]	
Paracentesis for symptom control	As indicated			X	As clinically indicated			X[20]	
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

Old text:

Notes

1. The screening visit S1 will take place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments have to be performed before administration of the study drug
4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.
7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals from last paracentesis until EOT. A final measurement will be performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis during the treatment period, sera will be collected at 14-day intervals from last paracentesis until EOT and a final sample will be collected at safety follow-up.
13. Urinalysis: Dipstick test for protein/albumin/erythrocytes at screening, thereafter for protein only. In case of protein ≥ 1+ with dipstick: Quantitative determination in 24 h urine is required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.

New text:

Notes

1. The screening visit S1 will take place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments have to be performed before administration of the study drug
4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.
7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. ~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ **M**easurements will be performed at biweekly intervals from **first** paracentesis until EOT. A final measurement will be performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis **with study medication** performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up~~**. In case a second paracentesis is not clinically required after the first paracentesis during the treatment period,**~~ **S**era will **also** be collected at 14-day intervals from **first** paracentesis until EOT and a final sample will be collected at safety follow-up.
13. Urinalysis: Dipstick test for ~~**protein/albumin/erythrocytes at screening, thereafter for**~~ protein only. In case of protein **>** 1+ with dipstick: Quantitative determination in 24 h urine is required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.
20. **When Paracentesis is clinically indicated**

Section 2.2 Secondary Objectives, page 36

Old text:

Pharmacokinetics of Bevacizumab and VEGF concentrations:

Serum and ascites VEGF and Bevacizumab concentrations will repeatedly be analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at bi-weekly intervals from last paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

New text:

Pharmacokinetics of Bevacizumab and VEGF concentrations:

Serum and ascites VEGF and Bevacizumab concentrations will repeatedly be analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis **with study medication** during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis **with study medication** during the 8-week treatment period, as well as at the time of routine safety follow-up. ~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ Measurements will be performed at bi-weekly intervals from **first** paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

Section 3.1 Overview of Study Design and Dosing Regimen, page 37

Old text:

At End of Safety FU, which is set at week 12 after the first paracentesis within the treatment period for both arms of the study, the study will allow for unblinding of the result of randomization in case of a missing clinical response of the ascites to study treatment. Unblinding has to be requested and approved by the sponsor. Patients will generally be followed regarding response and safety at 4 weeks (“safety follow-up”) following EOT. Puncture-free survival follow-up will take place every two months for one year after EOT.

New text:

~~**At End of Safety FU, which is set at week 12 after the first paracentesis within the treatment period for both arms of the study, the study will allow for unblinding of the result of randomization in case of a missing clinical response of the ascites to study treatment. Unblinding has to be requested and approved by the sponsor.**~~ Patients will generally be followed regarding response and safety at 4 weeks (“safety follow-up”) following EOT. Puncture-free survival follow-up will take place every two months for one year after EOT.

Section 4.2 Inclusion Criteria, page 40

Old text:
5. Cytologically confirmed ascites OR diagnosis of an exsudate (serum albumin – ascites albumin < 1.1 g/dl) clinically suggestive for malignant ascites

New text:
5. Cytologically confirmed ascites OR diagnosis of an exsudate **(total protein in ascites > 30 g/l**) clinically suggestive for malignant ascites **OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound**

Section 4.2 Inclusion Criteria, page 41

Old text:
12. Laboratory parameters

Clinical chemistry

- Creatinine clearance > 30 ml/min, serum creatinine < 1.5 x ULN
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 5 x ULN)

Urinalysis:

- Patients with < 2+ proteinuria on dipstick urinalysis.

Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 1.0 g of protein/24 h on 24-h urine collection.

New text:
12. Laboratory parameters

Clinical chemistry

- Creatinine clearance > 30 ml/min, serum creatinine < **2.5 x ULN**
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < **7 x ULN**)

Urinalysis:

- Patients with < 2+ proteinuria on dipstick urinalysis.
- Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate **< 2.0 g** of protein/24 h on 24-h urine collection.

Section 4.3 Exclusion Criteria, page 41

Old text:
4. Transudative ascites (Serum albumin – ascites albumin > 1.1 g/dl)

New text:
4. Transudative ascites (**total protein in ascites < 30 g/l**)

Section 4.3 Exclusion Criteria, page 42

Old text:
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, arrhythmia requiring medication, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic

coronary heart disease, myocardial infarction within the last year before treatment start, peripheral arterial disease stage ≥ II.

New text:
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, **uncontrolled arrhythmia**, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, ~~**myocardial infarction within the last year before treatment start**~~, peripheral arterial disease stage ≥ II.

Section 4.3 Exclusion Criteria, page 42

Old text:
15: Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up

New text:
15: Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. **Prior treatment with Bevacizumab for primary malignancy is not exclusionary**

Section 4.3 Exclusion Criteria, page 43

Old text:
25. As the following medication(s) can have interactive effects and may interfere with the patient's ability to meet the study requirements, they cannot be administered during the clinical study:

- Current or recent (within 10 days of first dose of study treatment) treatment with full-dose oral or parenteral anticoagulants or thrombolytic agents (e.g., marcumar therapy) for therapeutic purposes.
- Current or recent (within 10 days of first dose of study treatment) chronic use of aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day).

New text:
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Section 4.3 Exclusion Criteria, page 43

Old text:
26. Patients who participate currently in another clinical trial or patients who participated in another clinical trial during the last 30 days prior to enrolment.

New text:
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).

Section 4.4 Concomitant Medication and Treatment, page 44

Old text:

The following concomitant medications for purposes other than malignant ascites are forbidden in this study (see also Section 4.3):

Full-dose Oral Coumarin-Derived Anticoagulants, Heparin, Aspirin:

Full-dose oral coumarin-derived anticoagulants (INR > 1.5), heparin, thrombolytic agents, or chronic, daily treatment with aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day), other NSAIDs or COX-2 inhibitors, are not allowed at entry into the study.

Prophylactic low-dose aspirin is a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [127] and is supported by an extensive body of literature [128]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus bevacizumab or placebo. In a retrospective exploratory analysis of patients in the bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [129]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event is not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins is permitted before and during study, as is low-dose aspirin (≤ 325 mg/day) and clopidogrel (≤ 75 mg/day).

New text:

~~**The following concomitant medications for purposes other than malignant ascites are forbidden in this study (see also Section 4.3):**~~

~~**Full-dose Oral Coumarin-Derived Anticoagulants, Heparin, Aspirin:**~~

~~**Full-dose oral coumarin-derived anticoagulants (INR > 1.5), heparin, thrombolytic agents, or chronic, daily treatment with aspirin (> 325 mg/day) or clopidogrel (> 75 mg/day), other NSAIDs or COX-2 inhibitors, are not allowed at entry into the study.**~~

With the only exception of full-dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Prophylactic low-dose aspirin is a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [127] and is supported by an extensive body of literature [128]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus bevacizumab or placebo. In a retrospective exploratory analysis of patients in the bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [129]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event is not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins is permitted before and during study, as is low-dose aspirin (≤ 325 mg/day) and clopidogrel (≤ 75 mg/day).

Section 5.2.1.2 ECOG Performance Status, page 47

Old text:
In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, assessment of ECOG performance status will be performed at least every 2 weeks from last paracentesis until EOT and at safety follow-up.

New text:
~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ **In addition,** assessment of ECOG performance status will be performed ~~**at least**~~ every 2 weeks from **first** paracentesis until EOT and at safety follow-up.

Section 5.2.1.3 Assessment of Quality of Life, page 48

Old text:
In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, assessment of QoL performance status will be performed at biweekly intervals from last paracentesis until EOT and at safety follow-up.

New text:
~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ **In addition,** assessment of QoL performance status will be performed at biweekly intervals from **first** paracentesis until EOT and at safety follow-up.

Section 5.2.2.1 Assessment of Toxicity, page 48

Old text:
A **physical examination** will be performed at the screening visit, before start of each treatment cycle and at the safety follow-up visit 4 weeks after the last treatment cycle (safety follow-up).

New text:
A **physical examination** will be performed at the screening visit, before start of ~~**each treatment cycle**~~ **each paracentesis**, **biweekly** and at the safety follow-up visit 4 weeks after the last treatment cycle (safety follow-up).

Section 5.2.2.1 Assessment of Toxicity, page 48

Old text:
- **Vital signs** (blood pressure, heart rate, body temperature and body weight), will be measured at the screening visit, before start of each treatment cycle and 4 weeks after the last treatment cycle (safety follow-up). Body height will be measured at screening only. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals form last paracentesis until EOT.

New text:

- **Vital signs** (blood pressure, heart rate, body temperature and body weight), will be measured at the screening visit, before start of each ~~**treatment cycle**~~ **paracentesis** and 4 weeks after the last treatment cycle (safety follow-up). Body height will be measured at screening only. ~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ **In addition,** measurements will be performed at biweekly intervals form **first** paracentesis until EOT.

Section 5.2.3.1 Routine Laboratory Assessments, page 49

Old text:

- **Urinalysis -** Dipstick test for protein/albumin/erythrocytes will only be performed at the screening visit. Thereafter at every visit within the treatment period for protein only (see Flow Chart).. In case of protein ≥ 1+ with dipstick: Quantitative determination in 24 h urine is required. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at biweekly intervals form last paracentesis until EOT.

New text:

- **Urinalysis -** Dipstick test ~~**for protein/albumin/erythrocytes**~~ will only be performed ~~**at the screening visit. Thereafter at every visit within the treatment period**~~ for protein ~~**only**~~ (see Flow Chart).. In case of protein **>** 1+ with dipstick: Quantitative determination in 24 h urine is required. ~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ **Measurements will be performed at screening, prior to each paracentesis within the treatment period and at biweekly intervals form first paracentesis until EOT.**

Section 5.2.3.3 Pharmacokinetics of Bevacizumab and investigational analyses, page 50-52

Old text:

10 ml of blood and 10 ml of ascites fluid will be collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum will be obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics will be obtained before each routine paracentesis performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis during the treatment period, heparinized peripheral blood and sera will be collected at 14-day intervals from last paracentesis until EOT and a final sample will be collected at safety follow-up. Serum concentrations of Bevacizumab will be analyzed by Xendo laboratory.

New text:

10 ml of blood and 10 ml of ascites fluid will be collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum will be obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics will be obtained before each routine paracentesis **with study medication** performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. ~~**In case a second paracentesis is not clinically required after the first paracentesis during the treatment period,**~~ Heparinized peripheral blood and sera will be collected at 14-day intervals from **first** paracentesis until EOT and a final sample will be

collected at safety follow-up. Serum concentrations of Bevacizumab will be analyzed by Xendo laboratory.

Old text:
In order to assess the effect of Bevacizumab-induced effects on angiogenic/vascular permeability factors as well as the acquired immune system 10 ml of serum and 10 ml of ascites fluid will be obtained from each patient included at each visit performed for routine paracentesis. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, sera will be collected at bi-weekly intervals from last paracentesis until EOT. A final serum (and ascites sample, if possible) will be obtained at safety follow-up.

New text:
In order to assess the effect of Bevacizumab-induced effects on angiogenic/vascular permeability factors as well as the acquired immune system 10 ml of serum and 10 ml of ascites fluid will be obtained from each patient included at each visit performed for routine paracentesis **with study medication**. ~~**In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study,**~~ Sera will **also** be collected at bi-weekly intervals from **first** paracentesis until EOT. A final serum (and ascites sample, if possible) will be obtained at safety follow-up.

Old text:
All plasma and ascites samples in regard to the investigational as well as Serum and ascites samples for pharmacokinetic analyses will be firstly transported to the Laboratory for Tumor Immunology (Hamburg), Dr. Djordje Atanackovic. The Laboratory for Tumor Immunology (Hamburg) will afterwards forward the pharmacokinetic samples to the Xendo Laboratory.

New text:
All plasma and ascites samples in regard to the investigational ~~**as well as Serum and ascites samples for pharmacokinetic**~~ analyses will be ~~**firstly**~~ transported to the Laboratory for Tumor Immunology (Hamburg), Dr. Djordje Atanackovic. ~~**The Laboratory for Tumor Immunology (Hamburg) will afterwards forward**~~ The pharmacokinetic samples **will be sent** to the Xendo Laboratory.

Section 5.2.4 Additional Assessments, page 53

Old text:
- **Proteinuria:** A 24-h urine collection is required in case of a protein ≥ 1+ dipstick result before the subsequent treatment cycle. Further study treatment should be in line with the recommendations as defined in Section 7.3.2. 24-h urine protein will be measured on a 2-weekly basis (for the duration of trial therapy) until level drops below 0.5 g/24 h.

New text:
- **Proteinuria:** A 24-h urine collection is required in case of a protein **>** 1+ dipstick result before the subsequent treatment cycle. Further study treatment should be in line with the recommendations as defined in Section 7.3.2. ~~**24-h urine protein will be measured on a 2-weekly basis (for the duration of trial therapy) until level drops below 0.5 g/24 h.**~~

Section 7.6 Warning and Precautions, page 65

Old text:
No evidence available at the time of the approval of this study protocol indicated that special warnings or precautions were appropriate, other than those noted in the IB (see Investigator File) for Bevacizumab.

New text:
In some clinical trials anaphylactic and anaphylactoid-type reactions were reported more frequently in patients receiving Avastin in combination with chemotherapy than with chemotherapy alone. The incidence of these reactions in some clinical trials of Avastin is common (up to 5% in bevacizumab treated patients).
Patients may be at risk of developing infusion / hypersensitivity reaction. Close observation of the patient during and following the administration of bevacizumab is recommended as expected for any infusion of a therapeutic humanized monoclonal antibody. If a reaction occurs, the infusion should be discontinued and appropriate medical therapies should be administered. A systematic premedication is not warranted.

2.2 Patient informed consent form

The following text was changed from PIC Version 1.4, 08.12.2009 to Version 1.5, 17.05.2010

Section 6 Mögliche Belastungen und Risiken, page 8

Old text:
Häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten), geordnet nach Organsystemen:

New text:
Häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten), geordnet nach Organsystemen:
Sonstige Erkrankungen: In einigen klinischen Studien wurden anaphylaktische und anaphylaktoide Reationen (allergische Allgemeinreaktion) häufiger bei Patienten berichtet, die Bevacizumab in Kombination mit Chemotherapie erhielten als bei alleiniger Chemotherapie (bis zu 5%).

AIO- Trial SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Short title: AIO-SUP-0108

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EudraCT Nr.: 2009-014725-16

Protocol identification number: AIO-SUP-0108

Protocol version: 3.0 (07/06/2010)

Confidentiality

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Data Safety Monitoring Board

For contact details see DSMB Charta

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Drug Supply

Roche Pharma AG Deutschland

APPROVAL OF THE PROTOCOL

PD Dr. Ullrich Graeven – Representative of the Sponsor

Signature _____ Date (DD Month YYYY) _____

Dr. Karin Jordan – Principal Investigator

Signature _____ Date (DD Month YYYY) 21-06-2010

Dr. Axel Hinke - Statistician

Signature _____ Date (DD Month YYYY) _____

Dr. Anne L. Kranich – for the CRO

Signature _____ Date (DD Month YYYY) _____

APPROVAL OF THE PROTOCOL

PD Dr. Ullrich Graeven – Representative of the Sponsor

Signature	10. JUNE. 2010 Date (DD Month YYYY)

Dr. Karin Jordan – Principal Investigator

Signature	Date (DD Month YYYY)

Dr. Axel Hinke – Statistician

Signature	16.06.2010 Date (DD Month YYYY)

Dr. Anne L. Kranich – for the CRO

Signature	16.06.2010 Date (DD Month YYYY)

INVESTIGATOR'S AGREEMENT

I have read the attached protocol entitled

"Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers", dated 07. June 2010,

and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonisation Tripartite Guideline on Good Clinical Practice.

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the study sponsor.

Signature	Date (DD Month YYYY)

Investigator

Investigator's Institution

SYNOPSIS

Protocol No.	AIO-SUP-0108
Protocol Version (Date)	Version 3.0 (07/06/2010)
Title	Bevacizumab as a palliative treatment for patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers
Detailed Title	Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers
EudraCT No.	2009-014725-16
Principle Investigator	Dr. Karin Jordan Supportive Care Study Group, (for the Arbeitsgemeinschaft Internistische Onkologie, DKG) Clinic for Internal Medicine IV Department of Oncology/Hematology Martin Luther University Halle-Wittenberg
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Sponsor	AIO-Studien-gGmbH Straße des 17. Juni 106–108 “Tiergartentower” 10623 Berlin Phone: 030-322932933 Fax: 030-322932943
Study design	• Double-blind, placebo-controlled, randomized, multi-center, phase II • Patients will receive repeated intraperitoneal application of Bevacizumab/Placebo in a 2:1 ratio
Anticipated start date	09/2009
Duration of study	Approx. 2 years
Total number of centers	Approx. 20 centers
Study population	Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy

Rationale	Malignant ascites represents a severe clinical problem for physicians and patients. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites, and therapies that are commonly being used are only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers. Preclinical data strongly suggest that Bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites treated with Bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1]. Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.
Objectives	
Primary objective	• To evaluate the paracentesis-free survival (ParFS) following intraperitoneal application of Bevacizumab/Placebo
Secondary objectives	• To measure the frequency of paracenteses required for symptom control following intraperitoneal application of

	Bevacizumab/Placebo, by assessing the longest paracentesis-free period within the 12-week main observation period ("best response") • To measure the volume of ascites following intraperitoneal application of Bevacizumab/Placebo • To measure the effect of study treatment on the quality of life • To assess feasibility and safety of intraperitoneal application of Bevacizumab including pharmakocinetic of Bevacizumab • To evaluate the effect of an intraperitoneal application of Bevacizumab/Placebo on serum and ascites VEGF concentrations
Planned sample size	• In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized • Number of evaluable patients: 40 arm A (treatment), 20 in arm B (control)
Inclusion criteria	1. Age ≥ 18 years 2. Written informed consent has been obtained prior to inclusion into the study 3. Patient is capable and willing to comply with the study 4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma 5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound 6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy 7. Ascites clinically judged as not responsive to diuretics 8. At the time of inclusion paracentesis required at least twice within past 4 weeks. 9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. <u>Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.</u> 10. ECOG performance score 0-3 11. Life expectancy > 12 weeks 12. Laboratory parameters: <u>Hematology</u> • Neutrophils > 1,500/µl • Platelets > 100,000/µl • Hemoglobin ≥ 9 g/dl or 5.59 mmol/l <u>Hemastasiology</u> • INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

	Clinical chemistry • Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN • Serum bilirubin < 3.0 x ULN • Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN) Urinalysis: • Patients with < 2+ proteinuria on dipstick urinalysis. • Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.
Exclusion criteria	Patients with any of the following will not be eligible for participation: 1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible). 2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion 3. Hemorrhagic ascites (ascites hematocrit > 2%) 4. Transudative ascites (total protein in ascites < 30 g/l) 5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed. 6. Therapy naïve patients 7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up. 8. Patients with extensive metastases of the liver making up > 70% of the total liver mass 9. Child C cirrhosis of the liver 10. Occlusion or thrombosis of the portal vein. 11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression. 12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II. 13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder) 14. Major surgical procedure, open biopsy, or significant trau-

matic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic diseases.
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contra-indicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have participated in this study before.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[1]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of

[1] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

	the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG). 31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study. 32. Patients who possibly are dependent on the sponsor or investigator.
Duration of study	Recruitment: 2 years Treatment: Patients will receive paracentesis as needed for symptom control. In addition, patients will receive up to 4 intraperitoneal administrations of Bevacizumab/Placebo after paracentesis has been performed. During the 8-week treatment period, a minimum interval of 14 days will be kept between applications of the study medication. Follow-up: End of treatment (EOT) is set at eight weeks after application of the first paracentesis within the treatment period for both arms of the study. Follow-up regarding response and safety in both arms of the study will be conducted at week 4 after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-month intervals for a total of 12 months.
Arms of study	• 48 patients randomized into arm A will receive repeated intraperitoneal application of Bevacizumab • 26 patients randomized into arm B will receive repeated intraperitoneal application of Placebo
Data Safety Monitoring Board	A Data Safety Monitoring Board (DSMB) will be established prior to the start of the study and will be responsible for reviewing safety data on a regular basis. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT).
Primary parameter	Efficacy: • Paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first)
Secondary parameters	Efficacy • Best Response (BR) representing the longest period of time (in days) from o one paracentesis until next paracentesis within the

treatment period
  - or, if longer, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up)
  - or, if longer, from the last paracentesis performed within the treatment period until death (before end of the standard 4 week follow-up)
  - or, if longer, from the last paracentesis performed within the treatment period until 4 week follow-up
- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites as indicated by body weight
- Quality of life as assessed by standardized questionnaires
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:
- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5.
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events).
- All adverse events.
- Changes in laboratory values and vital signs.
- Changes in ECOG performance status.

Pharmacokinetics of Bevacizumab and VEGF concentrations:
- Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.
- Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at bi-weekly intervals from last paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

Study procedure	After the initial screening procedure, eligible patients will enter the treatment part of the study. Patients will receive up to 4 intraperitoneal administrations of bevacizumab/placebo depending on clinical necessity of routine paracentesis for symptom relief. In case of unacceptable toxicity, treatment will be prematurely discontinued. A final follow-up regarding response and safety will performed for both arms at 4 weeks after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-months intervals for a total of 12 months.
Randomization procedure	Permuted block randomization will be applied to guarantee balanced group numbers.
Statistical considerations	
Sample size calculation	The primary variable is efficacy as indicated by the paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death, whichever occurs first). Under the assumption of an expected median ParFS in the control group of 14 days [3] and a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions: • type I error: 5% (one-sided) • power: 80% • observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year (Parsons et al., 2007) [3] • exponential shape of the Kaplan-Meier [4] curves In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized. As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death or end of the standard 4-week follow-up) within in the 12 week observation period. However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data.

Analysis plan	Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison. Secondary analyses: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend. Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test. In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test). Quality of life as assessed by the standardized questionnaires (FACIT-AI) will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a comparable manner.

Other analyses to be performed descriptively:

- Overall survival will be analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

INFORMATION TO BE GIVEN ON SAE / PREGNANCY

In the case of a serious adverse event (SAE) or pregnancy the following person must be contacted within one working day by fax:

Clinical Trial Manager	**GSO mbH**
Address:	**Johnsallee 30** **D-20148 Hamburg**
Phone:	**+49 40 44 19 54 60**
Fax:	**+49 40 44 19 54 78**

FLOW CHART: SCHEDULE OF ASSESSMENTS DURING THE STUDY

	Screening[1, 3]		Baseline[3,9]	Treatment Period[3]				Safety FU	Survival FU[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT[19] + 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]				X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X[20]	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X[20]	
Paracentesis for symptom control	As indicated			X	As clinically indicated			X[20]	
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

Notes

1. The screening visit S1 will take place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments have to be performed before administration of the study drug
4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.
7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. Measurements will be performed at biweekly intervals from first paracentesis until EOT. A final measurement will be performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Sera will also be collected at 14-day intervals from first paracentesis until EOT and a final sample will be collected at safety follow-up.
13. Urinalysis: Dipstick test for protein only. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine is required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.
20. When Paracentesis is clinically indicated

GLOSSARY OF ABBREVIATIONS

β-HCG	Beta human chorionic gonadotropin
AE	Adverse event
AIO	Arbeitsgemeinschaft Internistische Onkologie
ALT (SGPT)	Alanine aminotransferase
AMG	Arzneimittelgesetz (German Drug Law)
ANC	Absolute neutrophil count
aPPT	Activated partial thromboplastin time
AST (SGOT)	Aspartate aminotransferase
BR	Best Response
CHF	Congestive heart failure
CHO	Chinese hamster ovary
CNS	Central nervous system
CR	Complete response
CRF	Case report form
CT	Computed tomography
CTCAE	Common terminology criteria for adverse events
DGHO	Deutsche Gesellschaft für Hämatologie/Onkologie
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDTA	
ELISA	Enzyme-linked Immuno-absorbant Assay
EOT	End of treatment
ESF	Eligibility screening form
FACIT-AI	Functional Assessment of Chronic Illness Therapy - Ascites Index
GCP-V	GCP-Verordnung
h	Hour
IC	Informed consent
ICH	International Conference on Harmonization
IMP	Investigational medicinal product
INR	International normalized ratio
ITT	Intent to treat
iv	intravenous
LD	Longest diameter
LDH	Lactate dehydrogenase
LKP	Leiter der klinischen Prüfung (Co-ordinating Investigator)
m2	Square meter (body surface area)
mg	Milligram
min	Minute
mL	Milliliter
MRI	Magnetic resonance imaging
NB	Nota bene (please note)
NCI	National Cancer Institute
NCT	National Center for Tumor Diseases
NSAIDS	Non-steroidal anti-inflammatory drugs
NYHA	New York Heart Association
PVS	Peritovenous shunting
PD	Progressive disease
PR	Partial response
RECIST	Response Evaluation Criteria in Solid Tumors
rHuMAb	Recombinant humanized monoclonal antibody

rpm	Rounds per minute
SAE	Serious adverse event
SD	Stable disease
SmPC	Summary of product characteristics
UICC	International Union Against Cancer
ULN	Upper limit of normal
VEGF	Vascular Endothelial Growth Factor
VPF	Vascular Permeability Factor
w/wo	With or without

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PART I – STUDY DESIGN AND CONDUCT

1 BACKGROUND AND RATIONALE

1.1 Background

1.1.1 Malignant ascites

Growth of tumors in serous cavities such as the peritoneum is often accompanied by the accumulation of a protein-rich exudates and formation of malignant effusions is a common problem for patients with advanced-stage cancer. Malignant ascites is defined as an abnormal accumulation of fluid in the peritoneal cavity as a consequence of cancer occurring in association with a wide variety of neoplasms such as colorectal, stomach, pancreatic, ovarian, breast, and lung cancer [5].

Accumulation of massive amounts of malignant ascites is a significant cause of morbidity and mortality in patients with intra-abdominal tumors [6]. Accordingly, mean survival is only 20 weeks after ascites has been discovered [7]. Local fluid accumulations provoke discomfort and distress in many patients in advanced stages of their disease. By increasing abdominal pressure malignant ascites causes severe symptoms such as abdominal pain, bowel obstruction, shortness of breath, loss of appetite, nausea, cachexia, anorexia, reduced mobility, and fatigue [6]. Palliation of the symptomatic patient is the foremost goal and elimination of fluid accumulation in a patient with these symptoms will certainly improve the patient's quality of life and may even prolong survival [8]. Unfortunately, treatment of malignant ascites is problematic and challenging. No single method has been developed that works satisfactorily for the majority of patients and, accordingly, effective management of malignant ascites has been a frustrating problem for many physicians and their patients [8].

Currently, treatment modalities commonly applied for patients with malignant ascites include diuresis, salt restriction, paracentesis, and peritoneo-venous shunts, however, evidence for each of these treatment options is weak, and there are no randomized controlled trials evaluating their safety and efficacy. In addition, results achieved by the application of these conventional methods are variable, only occasionally provide prolonged relief from symptoms, and do not improve survival [9]. Accordingly, in contrast to treatments for the underlying cancer, there is no generally accepted and evidence-based guideline for the management of malignant ascites [10].

Reduced sodium intake together with diuretics are often used to treat malignant ascites but there is no consensus regarding effectiveness [11]. As in the case of other treatment modali-

ties for malignant ascites, there are no randomized controlled trials assessing the efficacy of diuretic therapy in malignant ascites. Available data are controversial and there are no clear predictors to identify which patients would benefit from diuretics. The use of diuretics should, therefore, be evaluated individually [10] and some authors have even concluded that medical therapies such as diuretics, and sodium and fluid restriction are not effective in ascites caused by malignancies [8].

Paracentesis has been the most common treatment option offering the advantage of a quick, simple, and relatively low-risk procedure with immediate symptom relief. However, as the patient's disease progresses, the frequency of hospital visits for the procedure increase as well. Patients are left with the choice between frequent hospital visits or waiting as long as possible between procedures until the ascites symptoms are no longer tolerable [8]. In addition, repeated paracenteses subject the patient to risks such as bleeding, infection, visceral perforation, and hypotension associated with invasive fluid depletion, and renal impairment [5]. Finally, the procedure itself is painful, inconvenient, and, most importantly, only temporarily effective [8].

To avoid repeated paracenteses, a peritoneo-venous shunting (PVS) may theoretically be considered. However, PVS requires hospitalization and surgery and is associated with significant risks both in shunt placement as well as in the ordinary function of the shunt [12]. Accordingly, major complications such as pulmonary edema, pulmonary emboli, clinically relevant disseminated intravascular coagulation, and infection, have to be expected in a significant number of patients undergoing PVS [8, 10]. Moreover, survival and quality are not improved in patients who have received PVS in comparison with patients treated with serial paracentesis [12]. Therefore, it is agreed that shunt insertion is contraindicated in patients with gastrointestinal cancer and malignant ascites due to relatively poor prognosis and limited survival [10, 13, 14].

Recently, the tri-specific antibody catumaxomab has been introduced to the treatment of malignant ascites [15, 16]. This CD3- and EpCAM-specific antibody is thought to stimulate the T cellular immune system as well as to induce MHC-unrestricted cytotoxicity and phagocytosis of tumor cells [17, 18]. Cohorts of patients suffering of ovarian cancer and gastric cancer have experienced symptom relieve after catumaxomab application and, more recently, a phase II/III trial has assessed catumaxomab in the treatment of malignant ascites, with significant results regarding to puncture-free survival and quality of life [19]. In conclusion, although these data need to be confirmed in daily clinical practice, catumaxomab might represent a new approach for the therapy of malignant effusions. However, the need for

placement of an intraperitoneal catheter for several days and the prolonged hospital admission required for the treatment severely limit its potential use in patients with end-stage cancer who require palliative treatment [16]. In addition, EpCAM is known to be expressed on a variety of healthy tissues including hepatocytes [20]. Accordingly, significant grade III-IV hepatic toxicity has been observed in patients treated with intravenous antibody, sometimes even associated with an impaired hepatic function [21]. Similar side effects have been described when catumaxomab was applied intraperitoneally [16]. Therefore, catumaxomab seems to be better suited for patients with a relatively good performance state, able to tolerate both the continuous presence of an intraperitoneal catheter in an inpatient setting as well as hepatic toxicity [21]. Most patients with malignant ascites, however, are unlikely to be candidates for this particular mode of treatment.

In conclusion, effective palliation of malignant ascites remains a difficult management issue. As patients are expected to survive only for a very limited period of time, a desirable treatment should (1) effectively alleviate associated symptoms, (2) be minimally invasive, (3) allow for rapid discharge from the hospital, (4) be relatively simple with low associated risk of complications, and (5) be of tolerable cost to the patient and his/her family [22].

1.1.2 Vascular Endothelial Growth Factor

A better understanding of the molecular mechanisms that regulate the formation of malignant effusions may offer ways to design novel and more effective modes of therapy for this severe cancer-related clinical problem. The etiology of malignant pleural effusions and ascites had traditionally been attributed to lymphatic obstruction caused by tumor spread into draining lymph vessels [23, 24, 25, 26]. It had also been suggested that tumor-induced angiogenesis might contribute to the development of ascites [27, 28]. In 1983, however, Senger et al. suggested an alternative possibility [29]. They isolated vascular permeability factor (VPF) from ascites of tumor-bearing animals and hypothesized that this factor secreted by tumor cells in a paracrine fashion was responsible for the cancer-related fluid accumulations [30]. A few years later, vascular endothelial growth factor (VEGF) was discovered as a potent stimulator of angiogenesis and was subsequently recognized to be identical to VPF [31, 32].

VEGF is a highly conserved 34-42 kD glycoprotein secreted by a large variety of human tumors [30, 33, 34]. In addition, peritoneal mesothelial cells [35], monocytes/macrophages infiltrating malignant effusions [36], and even tumor-infiltrating T cells [37] are capable of producing VEGF.

By interacting with two high affinity tyrosine kinase receptors (Flt-1 and KDR/Flk-1), which are selectively expressed in vascular endothelium [38], VEGF acts on endothelium both normal and newly induced by tumor angiogenesis [39].

Angiogenesis, the development of new blood vessels from pre-existing vasculature, is an essential component of solid tumor growth and metastasis [40, 41, 42, 43]. It is now generally accepted that solid tumor growth must be accompanied by angiogenesis to provide the vascular support necessary for the expanding tumor mass. However, not only does neovascularization permit further tumor growth of the primary tumor, but it also provides a pathway for migrating tumor cells to gain access to the systemic circulation and to establish distant metastases. Tumors express a variety of angiogenic factors in order to promote their own vascularization by activating the host endothelium. One angiogenic factor that is thought to play a decisive role in the vascularization of neoplastic tissue is VEGF which is a potent and specific mitogen for endothelial cells [44] and stimulates the full cascade of events required for angiogenesis *in vitro* and *in vivo* [45]. However, in addition to its ability to promote angiogenesis, VEGF is also capable of markedly augmenting the permeability of pre-existing microvasculature [29, 39, 46].

1.1.3 VEGF and malignant ascites

VEGF is over-expressed in a variety of tumors causing malignant ascites [47, 48, 49] and intratumoral VEGF expression correlates with an increased metastatic potential [49, 50] and poorer survival rates, among others, in gastrointestinal tumors, ovarian, breast, and lung cancer [48, 49, 51, 52, 53, 54, 55, 56]. Accordingly, serum concentrations of soluble VEGF have often been shown to be increased in patients with various solid tumors [47, 51, 57, 58, 59, 60, 61, 62] when compared to normal controls. Serum levels of VEGF correlate positively with the stage of the disease [58, 60, 61, 63], and elevated concentrations of VEGF in the peripheral blood of cancer patients might also be associated with a poorer overall and progression-free survival [58, 59, 61].

Initial studies had already indicated that the accumulation of malignant ascites results in large parts from an increased permeability of peritoneal lining vessels [64, 65]. However, until the identification of VEGF in malignant ascites, the molecular basis of peritoneal vascular hyperpermeability had not been deciphered. It was later shown that that malignant effusions derived from tumor-bearing mice and guinea pigs contain high concentrations of soluble VEGF [36, 66, 67, 68, 69] and that in mice injected with tumor cells increases in microvascular permeability of pre-existing small vessels located in tissues lining the peritoneal cavity as

well as the total volume of the peritoneal fluid correlated closely with the appearance of VEGF within the ascites [36, 67, 70]. Furthermore, it was shown that VEGF protein accumulated in the leaky blood vessels that line the peritoneal cavities of mice bearing ascites tumors [38, 39, 70] and that in low nanomolar or picomolar concentrations VEGF increased the permeability of venules and small veins for plasma proteins with a potency 10.000 times higher than histamine [29]. Finally, the expression level of VEGF by cancer cells has been shown to directly correlate with the tumor cell-induced production of ascites in the animal model [71, 72, 73]. Accordingly, transfection of renal cancer cells with VEGF cDNA or viral vectors encoding for VEGF increased the capacity of these cells to induce hyperpermeability of peritoneal blood vessels and ascites following implantation into mice [74, 75, 76, 77]. Even direct transfection of mouse peritoneum with VEGF was sufficient to cause an accumulation of ascites [75]. In contrast, transfection of tumor cell lines with VEGF antisense oligonucleotides resulted in a reduced formation of malignant effusions in the mouse [72, 76, 78]. Altogether, these collected findings allowed for the firm conclusion that local VEGF secretion is responsible in large parts for initiating and maintaining the ascites pattern of tumor growth.

In numerous human studies, markedly increased concentrations of VEGF have been found in malignant pleural effusions and ascites derived from patients with a large variety of solid tumors, such as ovarian cancer, gastric cancer, colorectal cancer, pancreatic cancer, breast cancer, and lung cancer. Generally, much lower concentrations were detected in non-malignant effusions caused by congestive heart failure, liver cirrhosis, or infections [34, 35, 38, 57, 62, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95]. In a very recent study by Atanackovic et al. concentrations of 21 different cytokines/chemokines were simultaneously analyzed in malignant and non-malignant ascites and it was clearly shown that VEGF is the one cytokine most strongly over-expressed in ascites related to cancer [96]. VEGF concentrations within the effusions are always higher than in the corresponding sera from the respective patients [47, 57, 92, 97] and the volume of malignant ascites correlates with VEGF concentrations within the effusion [98] and with the intensity of VEGF expression in abdominal tumors removed from the same patient [99], indicating a significant local release within the peritoneal or pleural cavity. Furthermore, concentrations of VEGF within human malignant effusions correlate with their capability to induce vascular leakage in an experimental model, an effect that can be blocked by treatment with an antibody directed against VEGF receptor Flk-1 [82, 83]. Most importantly, concentrations of VEGF in malignant ascites have recently been shown to correlate with chemosensitivity and represent an independent predictor of progression-free and overall survival of cancer patients [94, 97, 98].

1.1.4 Inhibition of VEGF activity as a potential therapy for malignant effusions

If VEGF is responsible for fluid accumulation in the environment of solid tumors then anti-VEGF therapies should be able to directly influence the development of malignant effusions in addition to possessing an immediate anti-tumor effect. Importantly, it has repeatedly been shown in *in vitro* experiments that the capacity of VEGF, present in the supernatant of tumor cell lines or in malignant ascites, to induce vascular hyperpermeability can indeed completely be neutralized using an antibody directed against VEGF [29, 30, 38, 67, 100]. Furthermore, it was already shown in an initial animal study by Senger et al. that an anti-VEGF antibody is able to block the increased peritoneal influx associated with the intra-abdominal presence of VEGF-secreting tumor cells *in vivo*. Since then, a number of studies have clearly demonstrated that the intraperitoneal application of anti-VEGF antibodies is safe and leads to impressive and often complete remissions of the local fluid accumulations in mice following inoculation with different carcinoma or sarcoma cell lines [67, 68, 73, 101, 102]. Consistent with these findings, the vascular permeability of microvessels lining the peritoneal cavity of tumor-bearing mice decreased significantly in the anti-VEGF antibody-treated animals compared with controls [67]. Antibody treatment to a lesser degree also inhibited tumor growth [67, 68, 101, 102] and prolonged the survival of mice inoculated with tumor cells [67]. A comparable preclinical efficacy was seen with tyrosine kinase inhibitors targeting VEGF receptors [103, 104] or with a soluble VEGF decoy receptor inhibiting VEGF [75, 105], and after intraperitoneal infusion of a VEGF antisense oligonucleotide [106], but not with conventional chemotherapy applied intraperitoneally alone [73]. Interestingly, the delay in tumor growth induced by the anti-VEGF antibody was mainly attributed to the blockage of ascites development and vascular permeability and not to the inhibition of VEGF-induced angiogenesis [67]. In this context, it has been hypothesized that increased vascular permeability within the peritoneum leading to the development of ascites might indeed result in increased shedding of mesothelial cells into the abdominal cavity facilitating indirectly peritoneal tumor dissemination [107].

Despite the very strong preclinical evidence for an obligatory role of VEGF in the formation of malignant ascites and for a possible therapeutic efficacy of anti-VEGF therapies in the setting of malignant effusions, there are currently no reports from clinical studies addressing this point in cancer patients. However, recently a number of articles reporting on small series of patients with malignant effusions treated off-label with bevacizumab have presented impressing results. It was first reported by Pichelmayer et al. that Bevacizumab might be active in malignant ascites [108]. Following their observation of a marked responses to treatment

with Bevacizumab in a patient with benign pleural effusion [109], they decided to apply a single dose of Bevacizumab intravenously at 15 mg/kg to two patients with malignant ascites due to colorectal cancer and adenocarcinoma of unknown origin, respectively. They found that both patients, in whom paracentesis was previously required at least every second week, treatment with bevacizumab was safe and highly successful. They observed significant reductions in ascites volume resulting in a discontinuation of repeat paracentesis. Moreover, both patients had a marked decrease in their VEGF plasma levels after treatment. [109]. In agreement with these early observations, Numnum et al. reported the intravenous application of bevacizumab (15 mg/kg every 3 weeks) to 4 heavily pretreated patients with end-stage ovarian cancer with the intent to palliate symptomatic ascites. In all 4 patients repeatedly applied paracenteses could be discontinued because of dramatically reduced levels of ascites after initiation of therapy with bevacizumab [110].

In a very recent publication, Hamilton et al. reported on the treatment of an 88-year-old patient receiving home hospice care with refractory ovarian cancer, a very poor functional status, and severe symptomatic ascites. They performed paracentesis and treated the patient with two subsequent doses (5 mg/kg) of intraperitoneal bevacizumab with dramatic improvement in her ascites and the quality of her final weeks of life [2]. The largest series of patients treated with intraperitoneal Bevacizumab has recently been presented by El-Shami et al. who evaluated the safety and efficacy if intraperitoneal administration of bevacizumab (5 mg/kg every 4 weeks) to a total of 9 patients with refractory ascites due to colorectal, breast, uterine, or ovarian cancer. Impressingly, malignant ascites resolved after a single intraperitoneal dose in every single patient without reaccumulation or repeat paracentesis over a median observation period of more than two months. Moreover, no grade 2-5 adverse events were observed [1].

1.1.5 Study Drug Bevacizumab

Bevacizumab (Avastin®) is a recombinant humanized monoclonal antibody (rHuMAb) to VEGF composed of human IgG1 framework regions and antigen-binding complementarity-determining regions from a murine antibody (A.4.6.1) that blocks the binding of human VEGF to its receptors [111] (further details can be found in the Investigator's Brochure). By neutralizing the biologic activity of VEGF produced by solid tumors, Bevacizumab has the capability to reduce the vascularization of tumors, thereby inhibiting the growth of the malignancy. In numerous mouse models, bevacizumab has clearly demonstrated an inhibition of human tumor growth. The administration of bevacizumab in hetero-transplant models of

colon carcinoma has shown to result in a reduction in microvessel formation and number of metastases.

A number of properties make some agents more favorable than others for intraperitoneal therapy. One important characteristic might be that the drug has activity in the malignancy to be treated. The efficacy of bevacizumab when used in combination with chemotherapy has been demonstrated in several prospective, randomized phase III studies [112, 113, 114, 115]. For example, in a phase III trial in patients with metastatic colon cancer, bevacizumab in combination with standard chemotherapy was found to increase overall and progression-free survival and response rates when compared with chemotherapy plus placebo [113]. Because of this and the results from other trials [116], bevacizumab has become widely used for the treatment of colorectal cancer and non-small-cell lung cancer and is being studied as part of the treatment regimen in a wide range of malignancies [117, 118, 119, 120]. In Germany, bevacizumab has been approved for first-line treatment of metastatic colorectal carcinoma (in combination with 5-FU/folic acid or 5-FU/folic acid/irinotecan [FOLFIRI]) in January 2005. Since January 2008, bevacizumab has been approved for the treatment of mCRC in combination with any fluoropyrimidin-based chemotherapy.

Bevacizumab is generally well tolerated and has an acceptable toxicity profile consisting primarily of hypertension and proteinuria. Other rare but important adverse effects, however, include delayed wound healing, arterial thrombosis, and bleeding [121]. Another potentially serious adverse effect of bevacizumab is gastrointestinal (GI) perforation and, although comparably infrequent, this potentially life-threatening complication has generated significant clinical interest. Overall, GI perforation was found to be an uncommon but well-documented side-effect of treatment in the phase III trials of bevacizumab, as well as in subsequent surveillance trials, with a reported incidence of 1% to 2% [113, 114, 117]. Accordingly, in an observational study by Hedrick et al. 1968 patients with unresectable colorectal cancer received bevacizumab and first-line chemotherapy and GI perforation was observed in 1.7% of patients [122]. Recently, a retrospective analysis was published examining adverse events in 1442 cancer patients who had received bevacizumab at M.D. Anderson Cancer Center over a 2-year period. Bowel perforation or fistula occurred in 1.7% of patients with a variety of malignancies including gastrointestinal cancers. In patients with colorectal cancer, for example, such adverse events were only observed in 6 of 478 cases (1.3%) Median time to perforation after the initiation of bevacizumab treatment was 71 days. Only five of all 1442 patients ultimately underwent surgical exploration and overall 30-day mortality rate was only 12.5% in these patients [123].

Though strong evidence identifying specific risk factors is lacking, investigators have urged caution when treating patients with known bowel implants or large tumor burden, prior radiation, and recent surgery or bowel obstruction [124]. Accordingly, in their phase II study with colorectal cancer patients Hurwitz et al. [113] identified colon surgery within 2 months as a risk factor, they also found a history of peptic ulcer disease and a partial or complete response to therapy as potential risk factors for perforation. Sugrue et al. [125] analyzed the same registry as Hedrick et al. and found no statistically significant associations between specific patient characteristics and an increased risk of GI perforations were identified, however, 67% of the patients with GI perforation showed at least one of the following findings: tumor at the site of perforation, obstruction, intra-abdominal abscess, intraabdominal carcinomatosis, acute diverticulitis, or prior abdominal or pelvic radiation therapy. Importantly, these potential risk factors were similar to those observed in the study by Badgwell et al. [123] as was the median time until first event of approximately two months. Finally, in a case report series of patients treated with bevacizumab for colorectal, lung, renal cell, and unknown primary cancer, ischemic bowel complications were more frequent in patients with a history of pelvic irradiation [126]. Although this series consisted of only 33 patients, bowel complications occurred in those three patients who had received infradiaphragmatic irradiation but in none of the 30 remaining patients who had not received this mode of treatment.

Further details on non-clinical and clinical data for bevacizumab are provided in the Investigator's Brochure.

1.2 Rationale

1.2.1 Rationale for the Study, Relevance of the Study and Study Design

Malignant ascites represents a severe clinical problem for physicians and patients being confronted with this common symptom of advanced-stage gastrointestinal cancer. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites and therapies which are currently being used are, if anything, only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for more effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers.

Preclinical data strongly suggest that bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites

treated with bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. While there are, so far, no reports on GI perforation resulting from intraperitoneal application of bevacizumab, there might still be a significant risk of such adverse reactions. However, we believe that palliative intraperitoneal treatment with bevacizumab is still indicated in these patients with advanced-stage gastrointestinal cancer patients who are capable of providing informed consent and who often severely suffer from symptoms associated with malignant ascites.

1.2.2 Rationale for the route of application and dosage selection

In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1].

Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.

2 OBJECTIVES OF THE STUDY

2.1 Primary Objectives

The **first** primary endpoint will consist of paracentesis-free survival (ParFS) which will be calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first).

2.2 Secondary Objectives

Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study.

The **second** endpoint (Best response; BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU.

The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase

Further evaluation of the efficacy, feasibility, and general safety of an intraperitoneal application of Bevacizumab in patients with malignant ascites.

Other measures of efficacy will be:

- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites present in the patient as indicated by body weight at each study visit during the treatment period
- Quality of life as assessed by standardized questionnaires (FACIT-AI) filled out by the patient and one questionnaire by the palliative group of the DGHO, which needs to be completed by the medical staff
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events)
- All adverse events
- Changes in laboratory values and vital signs
- Changes in ECOG performance status

Pharmacokinetics of Bevacizumab and VEGF concentrations:

Serum and ascites VEGF and Bevacizumab concentrations will repeatedly be analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis with study medication during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis with study medication during the 8-week treatment period, as well as at the time of routine safety follow-up. Measurements will be performed at bi-weekly intervals from first paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

3 STUDY DESIGN

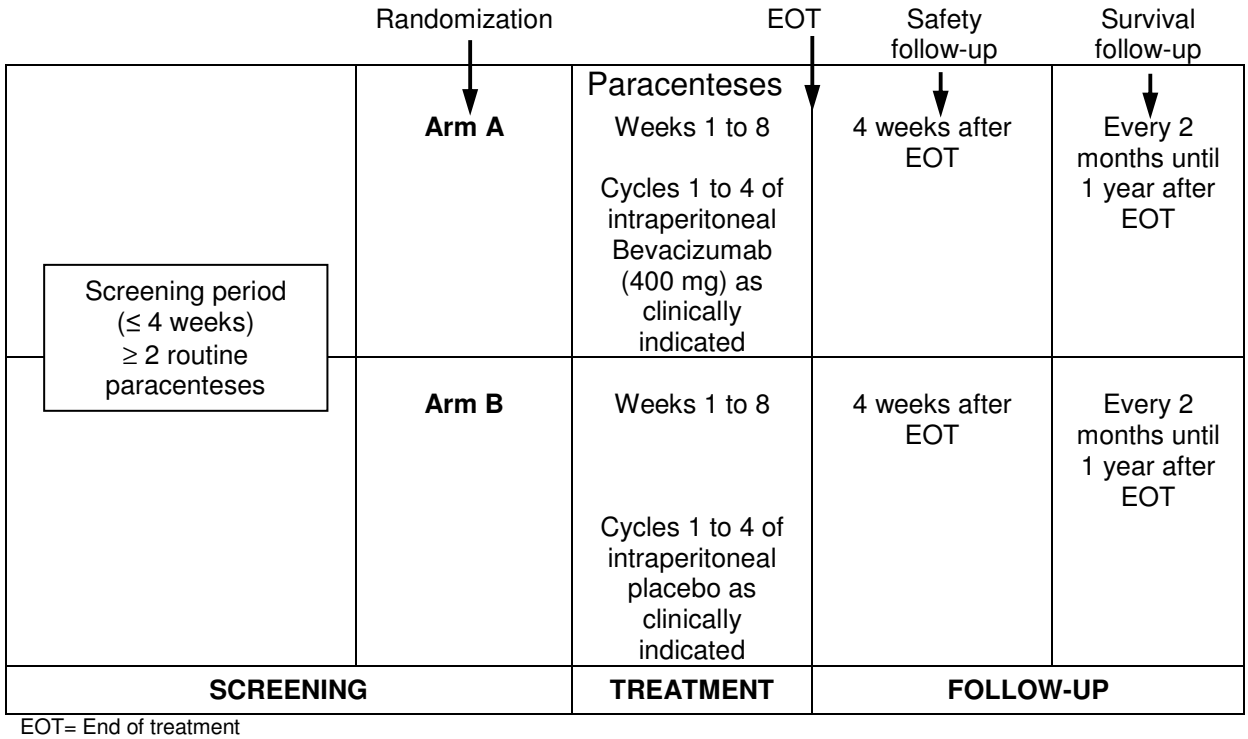
3.1 Overview of Study Design and Dosing Regimen

Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the tri-functional antibody catumaxomab or intraperitoneal chemotherapy. At the end of a screening phase of up to 4 weeks during which at least 2 routine paracenteses for symptom control of malignant ascites must have taken place, the screening visit S1 will take place. The screening visit must not take place earlier than 7 days before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. Eligible patients will be randomized into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period starts with the application of the first paracentesis for study purposes. Patients will receive up to 4 intraperitoneal administrations of Bevacizumab (400 mg absolute dose) or a placebo depending on clinical necessity of paracentesis for symptom relief. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days. In case of unacceptable toxicity, treatment will be prematurely discontinued.

Patients will generally be followed regarding response and safety at 4 weeks (“safety follow-up”) following EOT. Puncture-free survival follow-up will take place every two months for one year after EOT.

A brief overview on the study design is given in Figure 1.

Figure 1. Study Design



EOT= End of treatment

3.2 Number of Patients/Assignment to Treatment

A total of 72 patients will be enrolled into the study (48 into treatment arm A, 26 into control arm B). At the baseline visit, each patient will receive an unique patient number that will be given to the investigator by FAX at the time of individual patient enrolment. The number assigned of each patient has to be documented by using a Patient Identification Log and on each patient's Case Report Form.

The rational for the 2:1 allocation is that the study may gain more information about patient responses to the new intervention, such as toxicity and side effects. Additionally, if the intervention turns out to be beneficial, more study subjects would benefit than under an equal allocation design. Moreover, from a psychological point of view, the higher chance to receive the intervention rather than placebo may render the trial participation more acceptable to the eligible patients. [155, 158]

3.3 Centers

An approximate number of 20 centers will participate in the study. Each center is expected to recruit at least 4 patients until the planned total number of 72 patients is reached. A list of all participating investigational sites including information regarding names of the principal investigators and contact details (address, phone, fax email) will be handled separately.

3.4 Study Duration

The study is planned to start in January 2010 with respect to first patient in (FPI) including a recruitment period of two years. The follow-up periods will be approximately 6 months in total. The total study duration is approximately 2 and a half year and the last patient will probably complete the study (last patient out; LPO) in June 2012.

Submission to EC/CA: September 2009

First Patient in (FPI): January 2010

Recruitment Phase: 2.0 years

FU phase: 0.5 year

Last Patient out (LPO): June 2012

4 STUDY POPULATION

4.1 Target Population

Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers will be eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least twice within the 4 weeks prior to inclusion into the study. Under no circumstances are patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

4.2 Inclusion Criteria

To be eligible for this trial, patients must fulfill the following criteria:

1. Age ≥ 18 years
2. Written informed consent has been obtained prior to inclusion into the study
3. Patient is capable and willing to comply with the study
4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma
5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound
6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
7. Ascites clinically judged as not responsive to diuretics
8. At the time of inclusion paracentesis required at least twice within past 4 weeks
9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the trifunctional antibody catumaxomab or intraperitoneal chemotherapy.
10. ECOG performance score 0-3
11. Life expectancy > 12 weeks
12. Laboratory parameters:

Hematology

- Neutrophils > 1,500/µl
- Platelets > 100,000/µl
- Hemoglobin ≥ 9 g/dl or 5.59 mmol/l

Hemastasiology

- INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

Clinical chemistry

- Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN
- Serum bilirubin < 3.0 x ULN
- Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN)

Urinalysis:

- Patients with < 2+ proteinuria on dipstick urinalysis.
- Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.

4.3 Exclusion Criteria

Patients with any of the following will not be eligible for the study:

1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion
3. Hemorrhagic ascites (ascites hematocrit > 2%)
4. Transudative ascites (total protein in ascites < 30 g/l)
5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed.
6. Therapy naïve patients

7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
8. Patients with extensive metastases of the liver making up > 70% of the total liver mass
9. Child C cirrhosis of the liver
10. Occlusion or thrombosis of the portal vein.
11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II.
13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder)
14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic disease
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.

25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have previously participated in this study.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[2]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
32. Patients who possibly are dependent on the sponsor or investigator.

4.4 Concomitant Medication and Treatment

The initiation or continuation of any non-protocol-specific anti-tumor therapy is forbidden from inclusion into the study until EOT. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs will not be allowed from start of the screening phase until safety follow-up. This also excludes treatment with the tri-functional antibody catumaxomab or the intraperitoneal application of chemotherapy. Application of such treatments from start of the screening phase until safety follow-up will lead to the immediate discontinuation of the study for the given patient.

[2] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

All concomitant medication(s) must be reported in the Case Report Form (CRF). Any diagnostic, therapeutic, or surgical procedure performed during the study period should be recorded including the dates, description of the procedure(s) and any clinical findings. Patients should receive full supportive care including transfusion of blood and products, antibiotics, etc. where applicable. The treatment details should be recorded in the CRF.

With the only exception of full-dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Prophylactic low-dose aspirin is a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [127] and is supported by an extensive body of literature [128]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus bevacizumab or placebo. In a retrospective exploratory analysis of patients in the bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [129]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event is not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins is permitted before and during study, as is low-dose aspirin (≦325 mg/day) and clopidogrel (≤ 75 mg/day).

Note: In patients who experience thrombo-embolic events during study treatment full dose anticoagulant are allowed and information on anticoagulant treatment (including doses) will be collected and recorded in the CRF.

INR will be assessed at baseline for all patients. In patients treated with oral coumarin-derived anticoagulants INR will be checked at least before start of application of Bevacizumab or routine paracentesis, respectively.

In patients treated with full-dose oral anticoagulants due to thrombo-embolic event during study treatment, INR must be checked at least every second day the first week of treatment, at least 2 times/week for the following treatment weeks until a stable therapeutic level of INR

has been achieved and at least once every 3rd week when the weekly dose has been established and INR is stable with this dose (see also section 7.3.2).

The following is recommended regarding the use of concomitant medications:

Oral contraceptives: No dose modifications are required for patients on oral contraceptives.

5 SCHEDULE OF ASSESSMENT AND PROCEDURE

5.1 Screening Examination and Eligibility Screening Form

At the end of a screening phase the screening visit S1 will take place. The screening visit must not take place earlier than 7 days respectively 3 days (see Flow Chart) before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. After detailed oral and written information about the study, all patients willing to participate in this study have to provide a written Informed Consent (IC) before any study-specific assessment is performed.

An eligibility screening form (ESF) documenting the patient's fulfillment of the entry criteria for all patients considered for the study and subsequently included or excluded, is to be completed by the investigator and forwarded to the GSO mbH, Johnsallee 30, D-20148 Hamburg. All patients undergoing screening activities (documented by completion of an ESF for each patient) must be listed in the Patient Screening Log.

Patients who are considered for study entry, but who fail to meet the eligibility requirements, should also have an ESF completed with the reason for lack of eligibility given, since this provides information on the selected trial population. These patients will not be entered on the clinical trials database. The ESFs for patients who fail to meet the eligibility requirements should be kept in the study files at the sites. The same applies to patients who fulfill the entry criteria at the screening visit, but no longer at the baseline visit. For patients who did not sign the IC in the first, the ESF have not to be filled in. The patients will only be present in the pre-screening log.

A CRF should be filled out only for patients fulfilling the entry criteria both at screening and at baseline visits.

5.2 Study Assessments

5.2.1 Clinical Assessments

5.2.1.1 Assessment of Severity of Ascites

Baseline severity of malignant ascites will be assessed by calculating the mean time frame (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before start of the treatment phase

(screening phase). In addition, mean volumes of ascites (minus the volume of lavage fluid, if applicable) as well as body weight will be calculated for paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within 4 weeks of screening.

Between first paracentesis performed within the treatment period and until safety follow-up, the frequency of paracenteses clinically required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses with (arm A) or without (arm B) application of Bevacizumab. The Best response (BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or (if longer) from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or (if longer) from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU. Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study. The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase.

Between first paracentesis within the treatment period and until EOT, volume of ascites (minus the volume of lavage fluid, if applicable) drained during routine paracentesis for symptom relief with (arm B) or without (arm B) subsequent application of Bevacizumab will be recorded. Mean volumes and total volumes of ascites removed will be compared between groups and to the respective baseline values (mean and total volumes of paracentesis performed for symptom relief and not only diagnostic purposes during screening period) within the same group.

5.2.1.2 ECOG Performance Status

To be eligible for study entry, patients must have an ECOG performance score between 0 and 3 (see also section 4.2, Item 10). The patients' ECOG performance status (see section 18.3) will be assessed at the screening visit, before every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In addition, assessment of ECOG performance status will be performed every 2 weeks from first paracentesis until EOT and at safety follow-up.

5.2.1.3 Assessment of Quality of Life

In order to assess quality of life, utilities will be generated using a standardized and validated instrument of quality of life questionnaire. Quality of life will be evaluated using the FACIT-AI questionnaire, which needs to be filled out by the patient. Another questionnaire developed by the DGHO palliative care group has to be completed by the medical staff. Quality of life (see section 18.4) will be assessed at the screening visit, at every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In addition, assessment of QoL performance status will be performed at biweekly intervals from first paracentesis until EOT and at safety follow-up. FACIT-AI will be applied at the same time point during the study as the other quality of life questionnaire. Analyses will reveal the typical severity of each symptom during the different study phases to further assess burden of disease and treatment benefit. Additionally, a total ascites score derive from all symptoms will be calculated for each point in regard to the FACIT-AI.

5.2.2 Safety Assessments

5.2.2.1 Assessment of Toxicity

Throughout the treatment period and the 4-week safety follow-up period, patients will be assessed for toxicities attributable to therapy. Common terminology criteria for adverse events (CTCAE v3.0; see Investigator's File) will be used for grading. If necessary, the patient may be withdrawn from the study treatment. For details, see Section 7.3.

- **Medical history** including cancer and treatment history will be reviewed and recorded at the screening visit.
- **Concomitant medications** will be documented throughout treatment phase and the 4-week safety follow-up period. During the survival follow-up period, only anti-tumor drugs will be documented.
- A **physical examination** will be performed at the screening visit, before start each paracentesis, biweekly and at the safety follow-up visit 4 weeks after the last treatment cycle (safety follow-up).
- **ECG** will be measured at screening and if clinically indicated.
- **Vital signs** (blood pressure, heart rate, body temperature and body weight), will be measured at the screening visit, before start of each paracentesis and 4 weeks after the last treatment cycle (safety follow-up). Body height will be measured at screening only. In addition, measurements will be performed at biweekly intervals form first paracentesis until EOT.

- **Adverse events** (see also Section 7.1): All patients will be closely monitored for adverse events (incl. survival) from Day 1 of the first treatment cycle through Week 4 after the last treatment cycle. Thereafter, patients will be followed up for progression and survival only. Adverse events will be graded according to the Common Terminology Criteria for Adverse Events (CTCAE v3.0).

5.2.3 Laboratory Assessments

5.2.3.1 Routine Laboratory Assessments

Blood samples will be taken for hematological and serum chemistry monitoring at each scheduled visit. The local laboratory will perform the analyses and provide reference ranges. All Assessments must be performed at the screening visit. Thereafter, assessments (with the exception of urinalysis and pregnancy test) will be repeated before each treatment cycle and 4 weeks after EOT (8 weeks after first paracentesis within the treatment period).

- **Hematology** - Hemoglobin, platelets, leukocytes, neutrophils,
- **Hemostasiology** - INR and aPTT.
- **Clinical Chemistry** - Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance, total protein.
- **Urinalysis -** Dipstick test will only be performed for protein (see Flow Chart).. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine is required. Measurements will be performed at screening, prior to each paracentesis within the treatment period and at biweekly intervals form first paracentesis until EOT.
- **Pregnancy test -** A serum β-HCG pregnancy test will be performed at the screening visit, if childbearing potential cannot be ruled out. Additional pregnancy test will be done as clinically indicated.

5.2.3.2 Routine analysis of malignant ascites

Samples of malignant ascites will be taken for hematological analysis and routine chemistry at screening and at each visit scheduled for paracentesis during the treatment period. Analyses will be performed in the local laboratory.

- **Hematology (2-5 ml EDTA-anticoagulated ascites)** – Hemoglobin, hematocrit, absolute numbers of total leukocytes and neutrophils.
- **Chemistry (5 ml heparinized ascites)** – Concentration of total protein and of albumin.

5.2.3.3 Pharmacokinetics of Bevacizumab and investigational analyses

10 ml of blood and 10 ml of ascites fluid will be collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum will be obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics will be obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Heparinized peripheral blood and sera will be collected at 14-day intervals from first paracentesis until EOT and a final sample will be collected at safety follow-up. Serum concentrations of Bevacizumab will be analyzed by Xendo laboratory.

In addition to its role as a vascular permeability and as an angiogenic factor, VEGF also exerts a number of suppressive effects on the T cell-mediated immune system. First, VEGF functions as a chemo-attractant for CD34+ progenitor cells mobilizing these T cell-inhibiting cells into the tumor tissue where they inhibit the function of tumor-infiltrating T cells [130]. In addition, VEGF has repeatedly shown to reduce the phagocytic capacity and to inhibit the functional maturation of antigen-presenting dendritic cells (DC) *in vitro* and *in vivo* [131, 132, 133, 134, 135] and increased levels of VEGF are associated with reduced numbers of DC within the peripheral blood [136] and the tumor tissue [132]. In gastric cancer, for example, multivariate analysis showed that infiltration by DC was an independent prognostic indicator and there was an inverse correlation between the intratumoral density of DC and the expression of VEGF [132]. Accordingly, treatment with anti-VEGF antibody significantly improved the number and function of lymph node and spleen DC in tumor-bearing mice. Moreover, the efficacy of tumor vaccination with peptide-pulsed DC was dramatically enhanced by combining it with anti-VEGF antibody treatment [134, 137].

On the other hand, VEGF may also directly suppress immune effector cells by reducing cytokine-induced leukocyte-endothelial interactions *in vivo* [138] and by decreasing transendothelial migration of leukocytes [139]. Moreover, VEGF has recently been shown to exert an immediate effect on T cell immunity by inhibiting the thymic development of T cells [140] and enhancing Th2-type immunity [136, 141]. Accordingly, cytokines like IL-1β [107], IL-6 [142], and TGF-β [143, 144, 145] as well as a number of Th2-type cytokines (IL-4, IL-5, IL13) [145] might promote the production of intraperitoneal VEGF. In contrast, Th1-type cytokines, such as IFN-γ, inhibit VEGF production [145].

Finally findings for our group and others have recently indicated that the typical immune environment present within malignant ascites, which is marked by a dramatically elevated concentration of VEGF, might also contribute to the accumulation of immunosuppressive T regulatory cells (Tregs) within the effusion [96, 146]. These combined results suggest that expression of VEGF might be associated with tumor progression and poor prognosis not only because VEGF stimulates angiogenesis and vascular permeability, but also because it allows tumors to escape from attack by the immune system in patients with cancer.

In order to assess the effect of Bevacizumab-induced effects on angiogenic/vascular permeability factors as well as the acquired immune system 10 ml of serum and 10 ml of ascites fluid will be obtained from each patient included at each visit performed for routine paracentesis with study medication. Sera will also be collected at bi-weekly intervals from first paracentesis until EOT. A final serum (and ascites sample, if possible) will be obtained at safety follow-up.

As soon as possible after removal, heparinized blood and ascites samples, and sera will be centrifuged for 10 min at 1000g and supernatants will be divided into 3 x 1 ml volumes per sample type (3 ml total volume for each sample type, plasma and ascites fluid) and will immediately be frozen at a minimum of -20°C. After completion of the study by a given patient, the collective Plasma, ascites and serum samples of the respective patients will be picked up by TNT express who will also provide complete packaging and dry ice for the transport to the central investigational laboratory in Hamburg. For each participating center, four pick-up dates will be determined by the sponsor of the study.

All plasma and ascites samples in regard to the investigational analyses will be transported to the Laboratory for Tumor Immunology (Hamburg), Dr. Djordje Atanackovic. The pharmacokinetic samples will be sent to the Xendo Laboratory.

5.2.3.3.1 Central laboratory for pharmacokinetics

Xendo Laboratory
LE I D E N - N E T H E R L A N D S
Bio Science Park, Archimedesweg 17, 2333 CM Leiden
P.O. Box 255, 2300 AG Leiden, The Netherlands
Tel +31 (0)71 524 40 00 Fax +31 (0)71 524 40 01
E-Mail office.pharmaservices@xendo.com

5.2.3.3.2 Central laboratory for investigational analyses

Dr. Djordje Atanackovic
University Medical Center Hamburg-Eppendorf
Center of Oncology
Laboratory for Tumor Immunology
Building N27, 4th floor, Room 04.083
Phone: 0049-40-7410-55032
Mobile: 0049-177-7329398
Fax: 0049-40-7410-55735
E-Mail: D.Atanackovic@uke.uni-hamburg.de

Samples can only be sent from Monday to Wednesday of each week. The central laboratory needs to be informed by email at least one week before the samples are sent. In addition, the courier needs to be informed of the pick-up date at least 24 hours before the samples are expected to be sent:

TNT Express Hotline (01805-633725)

To be reached on working days until 4.00 p.m.

In a first step, serum and ascites samples derived from 5 representative patients will be analyzed by antibody arrays for immunomodulatory cytokines and chemokines as well as a broad variety of angiogenic factors. Results derived from these analyses will be confirmed by analyzing serum and ascites samples of the whole patient collective for the respective cytokines/chemokines/angiogenic factors by ELISA. Sera of 50 anonymized blood donors will serve as controls.

In addition, 20 ml of fresh heparinized blood will be collected at each appointment for collection of serum for investigational analyses and the complete volume of malignant ascites removed at a given paracentesis performed during the treatment period and at safety follow-up will be collected from each patient treated in centers being in close proximity to the laboratory performing the investigational analysis (Laboratory for Tumor Immunology, Center of Oncology, II. Medical Clinic, University Medical Center Hamburg-Eppendorf). From the ascites material, tumor cells as well as endothelial cells and T cells will be separated in order to analyze by real-time PCR-based arrays which cells produce the angiogenic and/or immune factors creating the typical immune environment within malignant effusions. The same patient material as well as the peripheral blood will also be analyzed by flow cytometry for the presence of Tregs and other immunomodulatory cell types

5.2.4 Additional Assessments

Additional assessments will be required in the case of hypertension, proteinuria, thrombosis and hemorrhagic events as specified below. These additional assessments will be recorded on specific CRF forms in addition to the completion of the adverse events form.

- **Hypertension:** In the case of grade 3/4 hypertension, additional blood pressure measurements should be performed on a weekly basis (for the duration of trial therapy) until resolution of the event. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Proteinuria:** A 24-h urine collection is required in case of a protein > 1+ dipstick result before the subsequent treatment cycle. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Thrombosis:** In case of grade 3/4 thrombosis a blood sample for the following laboratory values should be taken prior to initiation of treatment for the event: INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Hemorrhagic events:** In case of a hemorrhagic event of grade 2 or higher, a blood sample for the following laboratory values should be taken prior to treatment for the event: Platelet count, INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.

6 INVESTIGATIONAL PRODUCT

6.1 Investigational Medicinal Product (IMP)

According to § 3 (3) GCP-V an investigational medicinal product is a pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial, including products already with a marketing authorization but used or assembled (formulated or packaged) in a way different from the authorized form, or when used for an unauthorized indication, or when used to gain further information about the authorized form.

The IMP in this study is Bevacizumab and its corresponding placebo.

6.2 Background Medication

Background medication for treatment of the primary malignancy or other medical problems of the patient will be applied at the discretion of the investigator. Background medication will not be supplied or reimbursed by Roche. The investigator needs to observe the summary of product characteristics of the different component of the background medication with special attention to contraindications.

6.3 Dose and Schedule of Test Drug Bevacizumab and comparator drug

Patients will receive up to 4 intraperitoneal administrations of the study drugs depending on clinical necessity of paracentesis for symptom relief. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.

Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control.

Following the application of an 18-22 G intraperitoneal catheter, the largest possible volume of malignant ascites will be drained. Thereafter, Bevacizumab or the comparator drug will be applied through the same catheter at a total volume of 100 ml. Bevacizumab will be applied at an absolute standardized dosage of 400 mg. The initial dose of the study drug will be delivered over 60±15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills) the following infusions may be delivered over 30±10

minutes. Following the complete application of the study drug the intraperitoneal distribution will be optimized by varying the patient's body position (10 min on the back, 10 minutes on right side, 10 min on left side).

6.4 Preparation and Administration of Bevacizumab

6.4.1 Drug Name, Formulation and Storage

Drug name:

INN: Bevacizumab
Trade name: Avastin®
Manufacturer: Roche Registration Ltd.

Formulation:

Bevacizumab is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The IMP has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

Drug name:

INN: Placebo
Trade name: NA
Manufacturer: Roche Registration Ltd.

Formulation:

Placebo is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The Placebo has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

6.4.2 Packaging and Labeling

Bevacizumab (400 mg, 25 mg/mL) and placebo will be supplied in 20 mL glass vials with a fill of 16 mL. The investigational medicinal product will be labeled according to § 5 GCP-V and internal requirements for blinding purposes.

6.4.3 Preparation of Study Drug

Bevacizumab infusions will be prepared according to the SmPC.

6.4.4 Route of Administration

Bevacizumab will be administered as an intraperitoneal infusion. See also 6.3.

6.4.5 Blinding and Randomization

Randomization will be performed stratified by center using computer-generated lists consisting of permuted blocks of randomly varying size in order to ensure equal group sizes within strata. The randomization lists will be generated by WiSP GmbH and transferred to the facility responsible for blinding, labeling and packaging of the study drugs.

6.4.6 Compliance

A pre-printed drug dispensing log is provided in the Investigator Site File and must be kept current and must identify the patient, and the amount of medication dispensed to each patient at each visit with the corresponding dates.

All medication supplies (empty containers, as well as partly used and unused medication) must be available for inspection at every monitoring visit. All unused medication, partly-used and empty packages must be returned by the investigator to Roche at the end of the study.

7 SAFETY ISSUES

The Investigator's Brochure will be used as reference document for Bevacizumab and will be provided to the investigators in the Investigator's File.

7.1 Adverse Events and Laboratory Abnormalities

It is the responsibility of the investigator(s) to report all adverse events in the case report form. Any serious adverse event (SAE) must be reported to GSO within one working day. GSO will forward the SAEs to the sponsor and Roche within one working day.

7.1.1 Clinical Adverse Events

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. Pre-existing conditions that worsen during a study are to be reported as Adverse Events. They can become Serious Adverse Events if they fulfill one of the seriousness criteria described in section 18.2.

All clinical adverse events (AEs) encountered during the clinical study (treatment period and the 4-week safety follow-up) will be reported on the AE page of the CRF.

Intensity of adverse events will be graded using the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE), version 3.0 (see Investigator's File) and reported in detail as indicated on the CRF. If an adverse event occurs which is not contained in the CTCAE v3.0, the four-point scale below will be used.

Mild:	Discomfort noticed but no disruption of normal daily activity.
Moderate:	Discomfort sufficient to reduce or affect daily activity.
Severe:	Inability to work or perform normal daily activity
Life-threatening:	Represents an immediate threat to life

Relationship of the adverse event to the treatment should also be assessed. Description of scales can be found in section 18.1.

Progression or deterioration of the malignancy under study (including new metastatic lesions and death due to disease progression) will be part of the efficacy assessment and should NOT be reported as AE or SAE.

Signs and symptoms of the malignancy under study should only be reported if:

1. Newly emergent (i.e. not present at baseline) and the association with the underlying malignancy and old/new metastatic lesions is unclear and/or
2. The investigator attributed deterioration of malignancy-associated signs and symptoms directly to the study drug.

Should there be any uncertainty regarding the attribution of the malignancy under study to an AE, it should be reported as an AE or a SAE accordingly.

7.1.2 Laboratory Test Abnormalities

Laboratory test results will be recorded on the laboratory results pages of the Case Report Form, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable. Laboratory-test-value abnormalities as such should not be reported on the AE page of the CRF as adverse events, unless they are treatment-emergent and they satisfy one or more of the following conditions for clinical significance:

1. Accompanied by clinical symptoms.
2. Leading to a change in study medication (e.g. dose modification, interruption or permanent discontinuation).
3. Requiring a change in concomitant therapy (e.g. addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).

Please note: any laboratory result abnormality fulfilling the criteria for a serious adverse event (SAE) should be reported as such, in addition to being recorded as an adverse event in the CRF.

7.1.3 Adverse Events of Special Interest

Adverse events of special interest are any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events. Adverse events of special interest are to be processed like serious adverse events.

7.2 Handling of Safety Parameters

7.2.1 Serious Adverse Events or Adverse Events of Special Interest (Immediately Reportable to GSO)

Any clinical adverse event or abnormal laboratory test value that is serious or of special interest occurring during the course of the study, irrespective of the treatment received by the patient, must be reported to the GSO within one working day of knowledge (expedited reporting). For each patient, all serious adverse events should be reported until 8 weeks since last Bevacizumab infusion. SAEs considered to have a causal relationship to the Investigational Product should be reported regardless of time elapsed since last Bevacizumab dose.

The definition and reporting requirements according to German Drug Law, GCP-V and ICH Guideline for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2 will be adhered (for details refer to section 18.2).

7.2.2 Treatment and Follow-up of Adverse Events

Adverse events, especially those for which the relationship to test drug is not "unrelated", should be followed up until they have returned to baseline status or stabilized. If a clear explanation is established it should be recorded on the CRF.

7.2.3 Follow-up of Abnormal Laboratory Test Values

In the event of unexplained abnormal laboratory test values, the tests should be repeated immediately and followed up until they have returned to the normal range and/or an adequate explanation of the abnormality is found. If a clear explanation is established it should be recorded on the CRF.

7.2.4 Pregnancy

A female patient must be instructed to immediately inform the investigator if she becomes pregnant during the study. The study treatment must immediately be stopped and the patient must be withdrawn from the study. Pregnancies occurring up to 90 days after the completion of the last treatment cycle must also be reported to the investigator. The investigator must report all pregnancies within one working day to the sponsor and the CRO. The investigator should counsel the patient, discuss the risks of continuing the pregnancy, and possible effects on the fetus. Monitoring of the patient should continue until conclusion of the pregnancy.

Pregnancy occurring in the partner of a patient participating in the study should also be reported to the investigator, the sponsor and the CRO. The partner should be counseled and followed as described above.

7.3 Dose Modifications for Toxicity

The observed toxicity will be graded according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events. Dose modifications will be implemented according to the observed toxicity grade as specified below.

7.3.1 General Notes Regarding Dose Modifications

For adverse events which are considered by the Investigator unlikely to develop into serious events and which do not result in a delay or interruption of therapy, treatment will be continued at the same dose without reduction or interruption. If the toxicity is attributable to a certain drug, dose modifications should only be made for this drug. In case of several toxicities occurring simultaneously, the highest dose reduction should be applied.

7.3.2 Dose Modifications for Bevacizumab

No dose reduction of Bevacizumab is foreseen for an individual patient. The dose of 400 mg Bevacizumab was proven to be a safe treatment for intravenous treatment. In addition, all studies applying Bevacizumab as an intraperitoneal infusion have used this dosage.

The initial study drug dose will be delivered over 60 ± 15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills), the second infusion may be delivered over 30 ± 10 minutes.

Bevacizumab-specific toxicities:

Any patient who develops any one of the following toxicities should not further receive Bevacizumab. (for details see below):

- Gastrointestinal perforation
- Fistula Formation involving internal organ
- Arterial thrombo-embolic events
- Symptomatic grade 4 thrombosis
- Grade 3/4 hemorrhagic events
- Grade 4 hypertension (hypertensive crisis)
- Grade 4 proteinuria (nephrotic syndrome)

If the Bevacizumab treatment has to be discontinued permanently, the patient must be withdrawn from the study treatment and followed up for PD and survival only.

Gastrointestinal perforation

Bevacizumab should be permanently discontinued in patients who develop gastrointestinal perforation.

Fistula formation

Bevacizumab should be permanently discontinued in patients who develop fistula involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder).

Thrombosis/Embolism

For patients who develop grade 3 or 4 thrombosis/embolism the following action is recommended:

- Arterial thrombo-embolic events: Bevacizumab should be permanently discontinued.
- Grade 3 or 4 venous thrombosis: Bevacizumab should be permanently discontinued.

Hemorrhage

Patients who develop grade 3 or 4 hemorrhage should permanently discontinue Bevacizumab treatment.

Hypertension

Patients should be monitored for the development or worsening of hypertension via frequent blood pressure measurement. Blood pressure measurements should be taken after the patient has been in a resting position for ≥ 5 minutes. Repeat measurements of blood pressure for verification should be undertaken if the initial reading is ≥ 140 mmHg systolic and/or ≥ 90 mmHg diastolic blood pressure.

- Grade 1 hypertension: Asymptomatic, transient (< 24 h) increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Intervention not indicated.
- Grade 2 hypertension: Recurrent or persistent (> 24 h) or symptomatic increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Monotherapy of anti-hypertensive may be indicated. Once controlled to < 150/100 mmHg, patients may continue Bevacizumab therapy.
- Grade 3 hypertension: Requiring more than one anti-hypertensive or more intensive therapy than previously. Bevacizumab should be withheld for persistent or symptomatic hypertension and should be permanently discontinued if hypertension is not controlled.

- Grade 4 hypertension: Life threatening consequence (e.g. hypertensive crisis). Occurrence of grade 4 hypertension should lead to permanent discontinuation of Bevacizumab. All doses of anti-hypertensive medicines should be recorded at all visits.

Proteinuria

All patients will have a dipstick urinalysis performed within 48 h prior to each Bevacizumab dose. All proteinuria toxicity, as determined by 24 h urine collection, will be graded according to CTCAE v3.0 classification. Adjustment of Bevacizumab administration for proteinuria of ≥ 2 g/24 h will occur according to the following guidelines, listed below.

First occurrence of proteinuria:

- < 2+ (dipstick): Administer Bevacizumab as scheduled; NO additional evaluation is required.
- ≥ 2+ (dipstick): Administer Bevacizumab as scheduled. Collect 24-hour urine to determine the total protein within 3 days prior to the next scheduled dose:
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24 h total protein.
    - Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
    - Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g/24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

Second and subsequent occurrence of proteinuria:

- < 3+ proteinuria (dipstick): administer Bevacizumab as planned. No additional evaluation is required.
- ≥3+ proteinuria (dipstick): administer Bevacizumab as planned and collect 24-h urine for determination of total protein within 3 days before the next scheduled Bevacizumab administration.
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24-h total protein.

- Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
- Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g. 24-h protein should be further monitored prior to each administration of bevacizumab until it has decreased to ≤ 1 g/24 h.

Nephrotic syndrome (grade 4, CTCAE v3.0): Discontinue Bevacizumab treatment.

7.3.3 Dose Modifications for Background Medication

Local standard practice and the recommendations provided in the respective SmPCs will drive dose reduction or interruption of chemotherapeutic compounds of the background medication.

7.4 Criteria for Discontinuation or Termination of the Study

7.4.1 Criteria for Discontinuation of the Treatment or Premature Withdrawal of the Patient

Treatment in both arms of the study will discontinue according to the protocol if any of the following apply:

- if any exclusion criteria develop
- Any patient who develops any one of the following toxicities:
  - Gastrointestinal perforation
  - Fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
  - Arterial thrombo-embolic events
  - Symptomatic grade 4 thrombosis
  - Grade 3/4 hemorrhagic events
  - Grade 4 hypertension (hypertensive crisis)
  - Grade 4 proteinuria (nephrotic syndrome)
- at the patient's request
- at the investigator's discretion
- Physician's judgment following an adverse event
- Termination by the Sponsor, or a regulatory authority

- Any other reason for withdrawal that the study physician or patient indicates is in the overall best interest of the patient

All patients who prematurely discontinue the treatment period will be followed up for safety and survival (exception: patient withdraws consent for further participation or patient is lost to follow-up).

7.5 Treatment after Discontinuation or Termination of the Study

After discontinuation or termination of the study, patients will be treated at the discretion of the investigator and according to medical routine. Further treatment modalities include continuing diuresis, salt restriction and repeated paracentesis. This would also include the possibility of applying the tri-functional antibody catumaxomab or intraperitoneal chemotherapy.

7.6 Warnings and Precautions

In some clinical trials anaphylactic and anaphylactoid-type reactions were reported more frequently in patients receiving Avastin in combination with chemotherapy than with chemotherapy alone. The incidence of these reactions in some clinical trials of Avastin is common (up to 5% in bevacizumab treated patients).
Patients may be at risk of developing infusion / hypersensitivity reaction. Close observation of the patient during and following the administration of bevacizumab is recommended as expected for any infusion of a therapeutic humanized monoclonal antibody. If a reaction occurs, the infusion should be discontinued and appropriate medical therapies should be administered. A systematic premedication is not warranted.

8 BIOSTATISTICAL ASPECTS

8.1 Trial design and hypotheses

The trial is designed as a two-arm parallel group phase II study with a 2:1 randomization. Its primary objective is to obtain evidence, that bevacizumab treatment is effective in the symptom control of patients with malignant ascites. The primary endpoint is defined as paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or death, whichever occurs first). Thus, the following hypotheses will be tested:

H_0: ParFS (bevacizumab) ≤ ParFS (placebo)

H_1: ParFS (bevacizumab) > ParFS (placebo)

ParFS = paracentesis-free survival

As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis or death or end of the standard 4-week follow-up, whichever occurs first) within in the 12 week observation period.

H_0: BR (bevacizumab) ≤ BR (placebo)

H_1: BR (bevacizumab) > BR (placebo)

However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data. According to these hypotheses, the tests concerning the primary endpoints will be performed one-sided. In accordance with the phase II character of the trial, no type I error adjustment for multiplicity is performed.

8.2 Sample Size Calculation

Based on the results from Parsons et al. [3] the median ParFS in the untreated control group is expected to be around 14 days. In order to detect a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions:

- type I error: 5% (one-sided)
- power: 80%
- observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year [3]
- exponential shape of the Kaplan-Meier [4] curves

In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized.

The sample size calculation concerning the analysis of ParFS is based on methods described by Lachin and Foulkes [147]. Since a group sequential design allowing for interim analyses and early discontinuation will be adopted for this trial (see section 8.5), the above fixed sample size calculations serve only as an orientation for the maximum of patient numbers needed. The expected sample size and/or follow-up duration for reaching a conclusion may be considerably less than the number given above. This depends on the number and time points of interim looks as well as the actual difference in efficacy and the actual rates of recruitment and treatment failure. In the case of only one “look” (i.e. no interim analysis before completion of recruitment) the sample size coincides with that of the fixed-sample approach given above.

8.3 Evaluation categories of the patients

8.3.1 Intent-to-Treat Population

Intent-to-treat population (ITT) for ParFS is defined to include all randomized patients with at least one day of follow-up after the initial paracentesis. With respect to BR, this population consists of all patients with at least one documented paracentesis after the initial one or reaching the observation point at 4 weeks after EOT without any second paracentesis.

8.3.2 Per Protocol Population

The per protocol population (PP) is defined to include the intent-to-treat population excluding those patients with major protocol violations. As major protocol violations are considered those that may have an influence to the primary variables (e.g. not obtaining at least one application of study medication according to the protocol and randomization; receiving unauthorized antineoplastic or ascites treatment before observation of a second paracentesis or EOT + 4 weeks). Criteria that are assumed to have such an influence will be defined in a review meeting before data base lock.

8.3.3 Safety Population

According to the definition of the safety population all patients who received at least one dose of the trial medication and a safety follow-up, whether prematurely withdrawn or not, will be included in the safety analysis.

8.4 Methods of Statistical Analysis

The primary endpoints of the trial will be analyzed confirmatively (within the phase II framework) considering a global level for each hypothesis of $p < 0.05$ as significant.

All other parameters will be evaluated in an explorative or descriptive manner, providing means, medians, ranges, standard deviations and/or confidence intervals. If additional p values are calculated (e.g. in subgroup analyses or for secondary endpoints), they will be presented explicitly without referring to hypotheses or a significance level. Usually, no error adjustment for multiple testing will be performed. Thus the p values will reflect the comparison-wise error and not the experiment-wise error. All p values will be two-sided if not stated otherwise. The statistical methods described in this section are suited for the data and distributions usually expected in this type of trials. The suitability will be checked after data entry. If necessary, the statistical method will be modified accordingly and sensitivity analyses performed.

Demographic and prognostic baseline data will be checked for homogeneity between treatment groups. In case of relevant imbalances of other important prognostic factors the statistical method will be adjusted in order to achieve best possible comparability of the groups, and the results will be critically reviewed in comparison to the unadjusted ones.

Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. If the Peto logrank test [148, 149] is not appropriate because of violation of the proportional hazard assumption [150], Gehan's generalization of the Wilcoxon rank sum test for censored data [151] will be applied, preferably in its modification by Peto [148] and Prentice [152]. If necessary or prospectively defined at randomization, prognostic strata will be taken into account [149, 153].

Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison.

Secondary endpoints: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend.

Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test.

Quality of life as assessed by the standardized questionnaires and analyzed according to the recommendations of the respective developer, will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a similar way.

In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test).

Other analyses to be performed descriptively:

- Overall survival will be analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.
- For the ECOG performance status, the frequency of worsening, unchanged and improved status will be displayed by frequency tables in each scheduled visit.

The methods mentioned above are likewise suitable for the univariate evaluation of prognostic factors. Multivariate analyses may be performed by appropriate regression models (proportional hazard regression model [154], logistic regression).

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

8.5 Interim and Final Analysis

In case of longitudinal studies in severe chronic diseases, the study design should allow for interim analyses and, consequently, early stopping of the trial for ethical reasons [155]. A group sequential design will be adopted, using the α error spending function methodology by Lan and DeMets [156], implementing a use function according to the O'Brien-Fleming [157] boundary guideline. The design chosen will allow drawing conclusions from interim analyses in the following respect:

- acceptance of superiority of the bevacizumab arm (rejecting H0)

The additional option of accepting the control arm as non-inferior, when an interim result strongly suggests that the anticipated large difference of HR=0.5 will not be detected, is discarded, since a smaller difference might be discussed as relevant, too, especially if supported by secondary findings. Moreover, there is a less stringent need to stop the trial from an ethical point-of-view, if the data tend to similar results in both arms.

In order to keep an overall type I error of 5%, stopping boundaries will be calculated at the respective time points of interim evaluation, using the EaSt software (Cytel Software Corp., Cambridge, USA). This allows for arbitrary interim analyses, irrespective of time schedules and recruitment number. Moreover, the expected sample size and/or study duration for reaching a conclusion may be considerably smaller than in a fixed sample design (cf. 8.2), especially if the therapeutic difference is even larger than expected. The extent of "saving" patients mainly depends on the actual difference in efficacy as well as the actual rates of recruitment and failures. However, subsequent interim analyses will not be performed, unless an increment of at least 10 further evaluable patients are included in the database.

The final biometrical evaluation of the study and the compilation of the statistical report as part of the integrated clinical/biometrical report will be performed after completion and/or correction of all case report forms.

9 DATA QUALITY ASSURANCE

Data will be entered into a database by GSO Hamburg. The data will be filed in digital format and two people will enter the data independently. Data will be checked for accuracy using range, validity and consistency checks, as well as by cross-checking. Implausible or missing data may be corrected or completed after discussing with the investigator. The notes of amendment shall be filed together with the case report form (CRF). The validated data will be stored in a database and this process shall also be documented.

This database conforms to the requirements of ICH-GCP regarding the following:

- Validation of the system and data
- Presentation of SOPs
- Access and back-up systems
- Traceability and documentation of data amendments (audit trail)

Only authorised persons may access the database. Unauthorised access will be prevented via a security system.

10 STUDY COMMITTEES – DATA SAFETY MONITORING BOARD

A Protocol Committee consisting of experienced oncologists will be emplaced and will ensure the development of a clinically appropriate protocol. The Protocol Committee will also organize collaboration with reviewing statisticians and will make suggestions regarding centers to participate in the study.

The Data Safety Monitoring Board (DSMB) will be an independent board consisting of a group of 3 physicians with experience in oncology. A physician is not allowed to participate in this clinical trial while serving on the DSMB. The DSMB will be supported by an independent statistician, if necessary. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT). Safety variables (laboratory data, adverse events and serious adverse events) collected in the study will be reviewed every month during the first year. Later the DSMB can extend the regular intervals to e.g. 3 months. The collection and summary of these data will be prepared by the independent statistician. The members of the board will be primarily responsible for the clinical interpretation of the safety results. The board members can recommend a premature discontinuation of the trial or any other changes in the study conduct at any time, if required.

PART II - ETHICS AND GENERAL STUDY ADMINISTRATION

11 ETHICAL ASPECTS

11.1 Declaration of Helsinki/Good Clinical Practice

The Declaration of Helsinki is the accepted basis for clinical study ethics, and must be fully followed and respected by all engaged in research on human beings. Any exceptions must be justified and stated in the protocol. The latest version of the Declaration of Helsinki is available under http://www.wma.net/e/policy/b3.htm.

Additionally it is the responsibility of all engaged in research on human beings to ensure that the study is performed in accordance with the international Good Clinical Practice standards and according to all local laws and regulations concerning clinical studies.

11.2 Patient Information and Informed Consent

It is the responsibility of the investigator to obtain written informed consent from each patient participating in this study, after adequate explanation of aim, importance, anticipated benefits, and potential hazards and consequences of the study according to § 40 Abs. 2 and § 40 Abs. 2a AMG. Written informed consent must be obtained before any study specific procedures are performed. It must be also explained to the patient that they are completely free to refuse to enter the study or to withdraw from it at any time for any reason without incurring any penalty or withholding of treatment on the part of the investigator.

By signing the consent form, the subject/patient agrees with the "unwiderrufliche datenschutzrechtliche Einwilligung" according to § 40 Abs. 2a AMG. The subject/patient also agrees to allow the monitor/auditor/health authorities to verify the collected patient data against the subject's/patient's original medical records for the purpose of source data verification.

The informed consent form personally signed and dated by the patient must be kept on file by the investigator(s), and documented in the CRF and the subject's medical records. The investigator confirms obtaining the written informed consent to the sponsor.

If new safety information results in significant changes in the risk/benefit assessment, the consent form should be reviewed and updated if necessary. All patients (including those already being treated) should be informed of the new information and must give their written informed consent to continue in the study.

If the family doctors are informed of their patients' participation in the clinical study, this should be mentioned in the consent form.

11.3 Independent Ethics Committees and Regulatory Authorities

11.3.1 Approval of the Study by the Federal Regulatory Authority and Independent Ethics Committees

According to §§ 40-42 of the German drug law (AMG) it is the responsibility of the sponsor to obtain and maintain independent approval from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees to conduct the study.

The insurance coverage (study subject insurance) led down in § 40 AMG is in force. For each patient, the sponsor has provided insurance with HDI-Gerling Industrie Versicherung AG, Märkische Straße 23-33, 44141 Dortmund, contract number 48 158388 03055 390.

The sponsor names the "Leiter der klinischen Prüfung" (LKP) who has to be a physician with at least 2 years experience in the conduct of clinical trials of drugs according to § 4 (25) and § 40 (1) No. 5 AMG..

11.3.2 Notification of the Study

According to § 67 German drug law (AMG) the sponsor is responsible to notify competent regional authority about the study and all principal investigators of the participating investigational sites. If no other agreements are made, the sponsor will take over responsibility for investigator's obligation to report (§ 12 (3) GCP-V).

11.3.3 Report and Documentation Obligation

The sponsor is responsible to comply with the report and documentation obligation according to § 13 GCP-V.

The investigator is responsible to comply with the report and documentation obligation according to § 12 GCP-V.

12 CONDITIONS FOR MODIFYING THE PROTOCOL

Protocol modifications to ongoing studies must be made via amendment. The sponsor is responsible to obtain independent approval for the amendment from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees if required according to § 10 GCP-V. According to § 67 AMG competent regional authorities and the federal regulatory authority must be notified about the amendment, if they concern items according to § 12 Abs. 1 GCP-V.

13 DISCONTINUATION OR EARLY TERMINATION OF THE STUDY

13.1 Discontinuation of the Treatment or Premature Withdrawal of the Patient

All study specific withdrawal criteria are described in Section 7.4.1.

In addition, all patients have the right to withdraw from the study at any time for any reason. The investigator also has the right to withdraw patients from the study in the event of inter-current illness, adverse events and treatment failure after a prescribed procedure, protocol violations, cure, administrative reasons or other reasons.

The study has to be terminated when the patient starts a new tumor therapy.

An excessive rate of withdrawals can render the study uninterpretable; therefore, unnecessary withdrawal of patients should be avoided.

Should a patient decide to withdraw, all efforts will be made to complete and report the observations as thoroughly as possible.

The investigator should contact the patient to determine as completely as possible the reason for the withdrawal. A complete final evaluation at the time of the patient's withdrawal should be made with an explanation of why the patient is withdrawing from the study. If the reason for removal of a patient from the study is an adverse event or an abnormal laboratory test result, the principal specific event or test will be recorded on the Case Report Form.

13.2 Discontinuation or Early Termination of the Study

Both the sponsor and the investigator reserve the right to terminate the study at any time. Should this be necessary, both parties will arrange the procedures on an individual study basis after review and consultation. In terminating the study, the sponsor and the investigator will assure that adequate consideration is given to the protection of the patient's interests.

Following criteria could lead to a discontinuation or early termination of the study:

- Safety reason regarding patients' safety
- Negative benefit/risk assessment due to new information

In case of premature termination of the study all collected data have to be analyzed and a report has to be written. The sponsor has to inform the federal regulatory authority and the ethics committees within 15 days, giving detailed reason for the premature termination.

14 STUDY DOCUMENTATION, CRFS AND RECORD-KEEPING

14.1 Investigator's Files / Retention of Documents

The Investigator must maintain adequate and accurate records to enable the conduct of the study to be fully documented and the study data to be subsequently verified. These documents should be classified into two different separate categories Investigator's Study File, and subject/patient data.

The Investigator's Study File will contain all essential documents as the protocol/amendments, Case Report and Query Forms, patient information and informed consent form, Ethics Committee and federal regulatory authority approval, notification of the federal regulatory authority and competent regional authorities, drug records, staff curriculum vitae and authorization forms and other appropriate documents/correspondence etc.

Patient data include patient hospital/clinic records (medical reports, OP reports appointment book, medical records, pathology and laboratory reports, ECG, EEG, X-ray, etc.) and signed informed consent forms and patient screening and eligibility screening forms.

The investigator must keep these two categories of documents on file for at least 15 years (or more as legally required) after completion or discontinuation of the study. The documents must be archived in a secure place and treated as confidential material.

Should the investigator wish to assign the study records to another party or move them to another location, the sponsor must be notified in advance.

If the investigator can not guarantee this archiving requirement at the investigational site for any or all of the documents, special arrangements must be made between the investigator and the sponsor to store these in a sealed container(s) outside of the site so that they can be returned sealed to the Investigator in case of a regulatory audit. Where source documents are required for the continued care of the patient, appropriate copies should be made for storing outside of the site.

The sponsor must archive the protocol, documentation, approvals and all other essential documents related to the study, including certificates that satisfactory audit and inspection procedures have been carried out, as long as the test product(s) remains on the market.

All documents must be archived in a secure place and treated as confidential material.

14.2 Source Documents and Background Data

The investigator shall supply the sponsor on request with any required background data from the study documentation or clinic records. This is particularly important when Case Report Forms are illegible or when errors in data transcription are suspected. In case of special problems and/or governmental queries or requests for audit inspections, it is also necessary to have access to the complete study records, provided that patient confidentiality is protected. According to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy.

14.3 Audits and Inspections

This study may be audited by the sponsor, any person authorized by the sponsor or the competent health authority in order to determine the authenticity of the recorded data and compliance with the study protocol.

The investigator should understand that source documents for this trial should be made available to appropriately qualified personnel from sponsor/monitors/auditor/health authority inspectors after appropriate notification needed for source data verification and proper review of the study progress. The verification of the Case Report Form data must be by direct inspection of source documents. The investigator agrees to comply with the sponsor and regulatory authority requirements regarding the auditing of the study.

All material used in clinical studies are subjected to quality control.

14.4 Case Report Forms

For each patient enrolled, a Case Report Form must be completed and signed by the principal investigator or authorized delegate from the study staff. This also applies to records for those patients who fail to complete the study (even during a screening period if a Case Report Form was initiated). If a patient withdraws from the study, the reason must be noted on the Case Report Form. If a patient is withdrawn from the study because of a treatment-limiting adverse event, thorough efforts should be made to clearly document the outcome.

All forms should be typed or filled out using indelible ink, and must be legible. Errors should be crossed out but not obliterated, the correction inserted, and the change initialed and dated by the investigator or his/her authorized delegate. The investigator should ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the CRFs and in all required reports.

15 MONITORING THE STUDY

The monitor has the responsibility to familiarize the investigator(s) and the entire center staff involved in the study with all study procedures including the administration of study drug.

The GSOmbH must provide a trained monitor to assist the investigator(s) in conducting the clinical study. The monitor must visit the clinical study center on a regular basis and at least before the first patient has been enrolled, once during the course of the study, and at study completion. The monitor has the responsibility of reviewing the ongoing study with the investigator(s) to verify adherence to the protocol and to deal with any problems that arise. At all times the GSOmbH must maintain the confidentiality of the study documents. It is the responsibility of the study monitor to verify the study documents against the patient's original medical records.

The investigator (or his/her deputy) agrees to cooperate with the monitor to ensure that any problems detected in the course of these monitoring visits are resolved.

16 CONFIDENTIALITY OF TRIAL DOCUMENTS AND PATIENT RECORDS

The investigator and the sponsor (or designee) must assure that according to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy. CRFs or other documents should be submitted to the sponsor in a pseudonymous manner. The investigator should keep a patient identification log showing codes and names. The investigator should maintain documents not for submission to Sponsor, e.g., patients' written consent forms, in strict confidence.

17 PUBLICATION OF DATA

This study will be entered into the clinical trial protocol registry and clinical results database.

The sponsor is responsible for the timely reporting of study data. An integrated clinical study report (CSR) has to be completed one year after end of the study (whether completed or prematurely terminated). The report has to be approved by the responsible specialist chosen by the sponsor, the project manager of the CRO, the statistician and the principal investigator/LKP (for multicenter studies) by provision of their signatures.

The results of this study may be published or presented at scientific meetings as soon as after completion of the study. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor prior to submission.

In a multicenter study, it must be ensured that the data from one center are not published before the publication of the whole study. Roche reserves the right to review the manuscript(s) before their submission for publication or presentation. This is not intended to restrict or hinder publication or presentation, but is to allow the sponsor to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the investigator(s).

In accord with standard editorial and ethical practice, Roche will generally support publication of multicenter trials only in their entirety and not as individual center data. In this case, a coordinating investigator will be designated by mutual agreement.

Any formal publication of the study in which input of Roche personnel exceeded that of conventional monitoring will be considered as a joint publication by the investigator and the appropriate Roche personnel. Authorship will be determined by mutual agreement.

18 APPENDICES

Appendix 1: Adverse Events Categories for Determining Relationship to Test Drug

Appendix 2: Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

Appendix 3: ECOG Performance Status

Appendix 4: Quality of Life Questionnaires

18.1 Appendix 1 – Adverse Events Categories for Determining Relationship to Test Drug

(a) Probable (must have first three)

This category applies to those adverse events that are considered, with a high degree of certainty, to be related to the test drug. An adverse event may be considered probable, if:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It cannot be reasonably explained by the known characteristics of the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It disappears or decreases on cessation or reduction in dose. (There are important exceptions when an adverse event does not disappear upon discontinuation of the drug, yet drug-relatedness clearly exists: e. g. (1) bone marrow depression, (2) tardive dyskinesias).
4. It follows a known pattern of response to the suspected drug.
5. It reappears upon rechallenge.

(b) Possible (must have first two)

This category applies to those adverse events in which the connection with the test drug administration appears unlikely but cannot be ruled out with certainty. An adverse event may be considered possible if, or when:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It may have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It follows a known pattern of response to the suspected drug.

(c) Remote (must have first two)

In general, this category is applicable to an adverse event that meets the following criteria:

1. It does not follow a reasonable temporal sequence from administration of the drug.
2. It may readily have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.

3. It does not follow a known pattern of response to the suspected drug.
4. It does not reappear or worsen when the drug is readministered.

(d) Unrelated

This category is applicable to those adverse events which are judged to be clearly and incontrovertibly due only to extraneous causes (disease, environment, etc.) and do not meet the criteria for drug relationship listed under remote, possible, or probable.

	Probable	**Possible**	**Remote**	**Unrelated**
Clearly due to extraneous causes	–	–	–	+
Reasonable temporal association with drug administration	+	+	–	–
May be produced by subject clinical state, etc.	–	+	+	+
Known response pattern to suspected drug	+	+	–	–
Disappears or decreases on cessation or reduction in dose	+	–	–	–
Reappears on rechallenge	+	–	–	–

18.2 Appendix 2 – Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject, administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

Adverse reactions are all untoward and unintended responses to an investigational medicinal product related to any dose administered.

A serious adverse event or serious adverse reaction is any experience that suggests a significant hazard, contraindication, side effect or precaution. It is any Adverse Event that at any dose fulfils at least one of the following criteria:

- is fatal (results in death) *(NOTE: death is an outcome, not an event)*
- is life-threatening *(NOTE: the term "life-threatening" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not refer to an event which could hypothetically have caused a death had it been more severe.)*
- required in-patient hospitalization or prolongation of existing hospitalization
- results in persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- is medically significant or requires intervention to prevent one or other of the outcomes listed above

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the outcomes listed in the definitions above. These situations should also usually be considered serious.

Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

An unexpected Adverse Event is one, the nature or severity of which is not consistent with the applicable product information.

Causality is initially assessed by the investigator. With respect to report and documentation obligation (regulatory authorities, ethics committees and other investigators) for Serious Adverse Events, causality can be one of 2 possibilities:

- No (unrelated; equals not drug related).
- Yes (remotely, possibly, probably or definitely drug related).

All adverse events not assessed as definitive "not drug related" by either the investigator or Roche will be considered as adverse drug reaction.

A suspected unexpected serious adverse reaction (SUSAR) is a serious adverse reaction, the nature, or severity of which is not consistent with the applicable product information.

It is important that the severity of an adverse event is not confounded with the seriousness of the event. For example, vomiting which persists for many hours may be severe, but is not necessarily a serious adverse event. On the other hand, stroke which results in only a limited degree of disability may be considered a mild stroke, but would be a serious adverse event.

A serious adverse event occurring during the study or which comes to the attention of the investigator within 8 weeks after stopping the treatment or during the protocol-defined follow-up period, if this is longer, whether considered treatment-related or not, must be reported. In addition, a serious adverse event that occurs after this time, if considered related to test "drug", should be reported.

Such preliminary reports will be followed by detailed descriptions later that will include copies of hospital case reports, autopsy reports and other documents.

For serious adverse events, the following must be assessed and recorded on the adverse events page of the Case Report Form: intensity, relationship to test substance, action taken, and outcome to date.

Document and report obligation have to be adhered according to the national and international laws and regulations.

Contact details and Fax No. for SAE and pregnancy reporting refer to page 17.

18.3 Appendix 3 – ECOG Performance Status

GRADE	SCALE
0	Fully active, able to carry out all pre-disease performance without restriction. (Karnofsky 100%)
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light housework, office work. (Karnofsky 80-90%)
2	Ambulatory and capable of all self-care, but unable to carry out work activities. Up and about more than 50% of waking hours (Karnofsky 60-70%)
3	Capable of only limited self-care, confined to bed or chair more than 50% of waking hours. (Karnofsky 40-50%)
4	Completely disabled. Cannot carry out any self-care. Totally confined to bed or chair. (Karnofsky 10-30%)
5	Dead (Karnofsky 0%)

18.4 Appendix 4 – Quality of Life Questionnaires

FACIT ASCITES INDEX (Patientenfragebogen)

Nachfolgend finden Sie eine Liste von Aussagen, die von anderen Personen mit Ihrer Krankheit für wichtig befunden wurden. **Bitte geben Sie jeweils an, wie sehr jede der folgenden Aussagen im Laufe der letzten 7 Tage auf Sie zugetroffen hat, indem Sie die entsprechende Zahl ankreuzen.**

		Überhaupt nicht	Ein wenig	Mäßig	Ziemlich	Sehr
C6	Ich habe einen guten Appetit	0	1	2	3	4
GF5	Ich schlafe gut	0	1	2	3	4
BMT5	Ich bin in der Lage, mich alleine fortzubewegen	0	1	2	3	4
B1	Ich leide unter Atemnot	0	1	2	3	4
GP2	Mir ist übel	0	1	2	3	4
O2	Ich habe mich übergeben	0	1	2	3	4
ACT11	Ich habe Schmerzen in der Magengegend	0	1	2	3	4
O1	Ich habe Schwellungen im Magenbereich	0	1	2	3	4
GP1	Mir fehlt es an Energie	0	1	2	3	4
ACT10	Wenn ich esse, fühle ich mich rasch satt	0	1	2	3	4
BL2	Ich muss häufiger Wasserlassen als üblich	0	1	2	3	4
Cx6	Ich leide an Verstopfung	0	1	2	3	4
AI1	Ich bin bekümmert	0	1	2	3	4

Patientenlebensqualitätsfragebogen

(modifiziert nach der Palliativgruppe der DGHO)

Der Fragebogen ist durch das medizinische Personal zu ergänzen

Betreuungsverlauf

Datum des ersten Besuchs ☐☐☐☐☐☐

Ort des ersten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Datum des letzten Besuchs ☐☐☐☐☐☐

Ort des letzten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Begründung des Abschlusses der Home Care-Versorgung ★
- ● Tod des Patienten
- ● Änderung des Wohnorts des Patienten
- ● unerwartete Verbesserung des Gesundheitszustands des Pat.
- ● Fortsetzung der tumorspezifischen Therapie
- ● Krankenhauseinweisung
- ● Wechsel des HC-Arztes
- ● sonstiges:_____

Sterbedatum ☐☐☐☐☐☐

Sterbeort
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Palliativstation
- ● andere Krkhs.-Station
- ● sonstiges

Ausprägung von Symptomen zum Zeitpunkt der Aufnahme und nach erfolgter Einstellung

alles Zutreffende nach Schweregrad ausfüllen (Schweregrad: 0-ohne, 1-gering, 2-mittel, 3-stark) ■ vor Intervention ☐ nach Intervention

■	Schmerzen ☐	■	Dysphagie ☐	■	(Lymph-)Ödem ☐	■	Krampfanfälle ☐
■	Schwäche ☐	■	Obstipation ☐	■	Juckreiz ☐	■	motor. Unruhe ☐
■	Appetitlosigkeit ☐	■	Diarrhoe ☐	■	Dekubitus ☐	■	Verwirrtheit ☐
■	Übelkeit ☐	■	Ascites ☐	■	exulc. Wunde ☐	■	Schlafstörung ☐
■	Erbrechen ☐	■	Dyspnoe ☐	■	Harnverhalt ☐	■	Angst ☐
■	(Tumor-)Blutung ☐	■	Husten ☐	■	Lähmungen ☐	■	Depression ☐

Therapie bei Aufnahme und im Verlauf / zum Ende

Schmerztherapie WHO
- ● keine ○
- ● nur bei Bedarf ○
- ● WHO Stufe I ○
- ● WHO Stufe II ○
- ● WHO Stufe III ○

Koanalgetika / Begleitmedikation ★
- ● Nichtopioide Analget. ○
- ● Antiemetika ○
- ● Antidepressiva ○
- ● Antikonvulsiva ○
- ● Kortikosteroide ○
- ● Laxanzien ○
- ● Neuroleptika ○
- ● Sedativa / Hypnotika ○

Opioidtherapie ★

Substanz(en) ★
- ● Morphin ○
- ● Hydromorphon ○
- ● Fentanyl ○
- ● Oxycodon ○
- ● Buprenorphin ○
- ● Levomethadon ○
- ● Piritramid ○

Applikationsform(en) ★
- ● oral / PEG / rektal ○
- ● transdermal ○
- ● s.c. Injektion ○
- ● s.c. Dauerinfusion ○
- ● i.v. Dauerinfusion ○
- ● peridural ○

sonstige Palliativmaßnahmen ★
- ● PEG / transnasale Sonde ○
- ● zentraler venöser Zugang ○
- ● enterale Ernährung ○
- ● parenterale Ernährung ○
- ● i.v. Flüssigkeitssubstitution ○
- ● s.c. Flüssigkeitssubstitution ○
- ● Ascitespunktion(en) ○
- ● Pleurapunktion(en) ○
- ● palliative Chirurgie ○
- ● palliative Radiatio ○
- ● palliative Chemotherapie ○
- ● palliative endoskop. Eingriffe ○
- ● nichtspezialisierter Pflegedienst ○
- ● Palliativpflegedienst ○

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AIO- Trial SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Amendment 2

dated 18.04.2012

Study Code: AIO-SUP-0108

EudraCT-No: 2009-014725-16

Approval of the Amendment:

I hereby approve the Amendment 2 to the protocol.

Priv. Doz. Dr. Ulrich Graeven
Sponsor

Date

Priv. Doz. Dr. Karin Jordan
Coordinating Investigator

Date

AIO- Trial SUP-0108

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Priv. Doz. Dr. Ulrich Graeven **Sponsor**	**Date**
Priv. Doz. Dr. Karin Jordan **Coordinating Investigator**	**Date** Halle 24.4.2012

1. Rationale

1.1 Change of Inclusion/Exclusion Criteria

The aim of this amendment is to modify the inclusion criterion No. 8: “At the time of inclusion paracentesis required at least twice within past 4 weeks”.
In patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers, the general condition of the patients may worsen rather quickly. Therefore, study treatment should start as early as possible to allow patients to enter the study in good condition so that they could benefit from study treatment as early as possible. Patients will therefore be allowed to enter the study already after one paracentesis within the 4-week screening phase, provided that the first paracentesis within the treatment phase is performed within four weeks after the preceding paracentesis in the screening phase.

Therefore, the respective inclusion criterion will be changed as follows:

Inclusion criterion No. 8:
At the time of inclusion paracentesis required at least **once** within past 4 weeks**. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase.**

1.2 Administrative Changes

In addition, administrative changes such as the new address of the sponsor and the CRO as well as adapted study duration and prolonged recruitment period are amended in the protocol. Exclusion criterion No. 4 (transudative ascites (total protein in ascites < 30 g/l) was deleted as it is obsolete due to inclusion criterion No. 5 (Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound).

2. Tracking of Changes

2.1 Protocol

The following text was changed from protocol Version 3.0, 07.06.2010 to Version 4.0, 18.04.2012:

Title page:

Old text:
Data Monitoring: GSO mbH,
Johnsallee 30
20148 Hamburg

New text:
Data Monitoring: GSO mbH
Harvestehuder Weg 21
20148 Hamburg

Contact addresses, Page 2:

Old text:
CRO: GSO mbH,
Johnsallee 30
20148 Hamburg

New text:
CRO: GSO mbH
Harvestehuder Weg 21
20148 Hamburg

Synopsis, Page 6:

Old text:

Anticipated start date: 09/2009

Duration of study : Approx. 2 years

New text :

Start date (FPI): 06/2010

Duration of study : Approx. **4** years

Synopsis, Page 8:

Old text:
Inclusion Criteria:
8. At the time of inclusion paracentesis required at least twice within past 4 weeks

New text:

Inclusion Criteria:
8. At the time of inclusion paracentesis required at least **once** within past 4 weeks**. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase.**

Synopsis, Page 9:

Old text:
Exclusion Criteria:
4. Transudative ascites (total protein in ascites < 30 g/l)

New text:
Exclusion Criteria:
~~4. Transudative ascites (total protein in ascites < 30 g/l)~~

Synopsis, Page 11:

Old text:
Duration of study: Recruitment: 2 years

New text:
Duration of study: Recruitment: **3.5 years**

Information to be given on SAE / Pregnancy, Page 16

Old text:

Clinical Trial Manager:	GSO mbH
Address:	Johnsallee 30
	D-20148 Hamburg

New text:

Clinical Trial Manager:	**Dr. Anne L. Kranich**
Address:	GSO mbH
	Harvestehuder Weg 21
	D-20148 Hamburg

3.1 Overview of study design and dosing regimen, Page 37

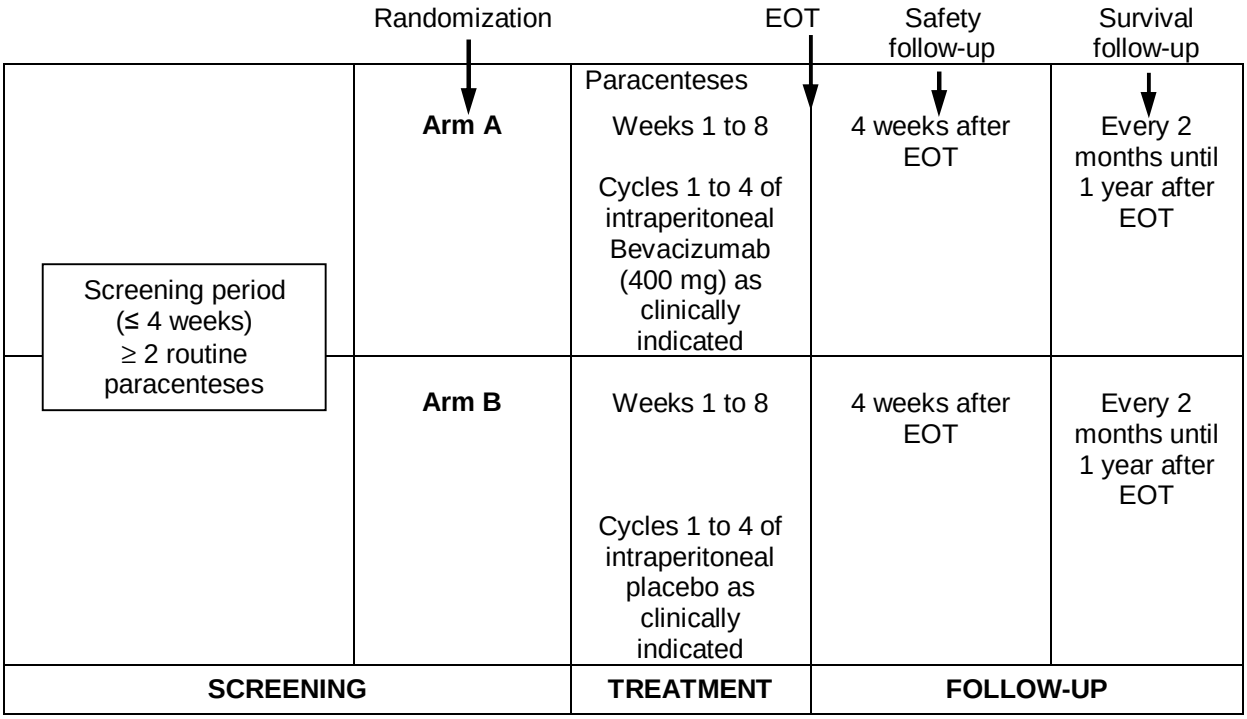
Old text:
At the end of a screening phase of up to 4 weeks during which at least 2 routine paracenteses for symptom control of malignant ascites must have taken place, the screening visit S1 will take place. The screening visit must not take place earlier than 7 days before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. Eligible patients will be randomized into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period starts with the application of the first paracentesis for study purposes.

New text:
At the end of a screening phase of up to 4 weeks during which **at least 1 routine paracentesis** for symptom control of malignant ascites must have taken place, the screening visit S1 will take place. The screening visit must not take place earlier than 7 days before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. Eligible patients will be randomized

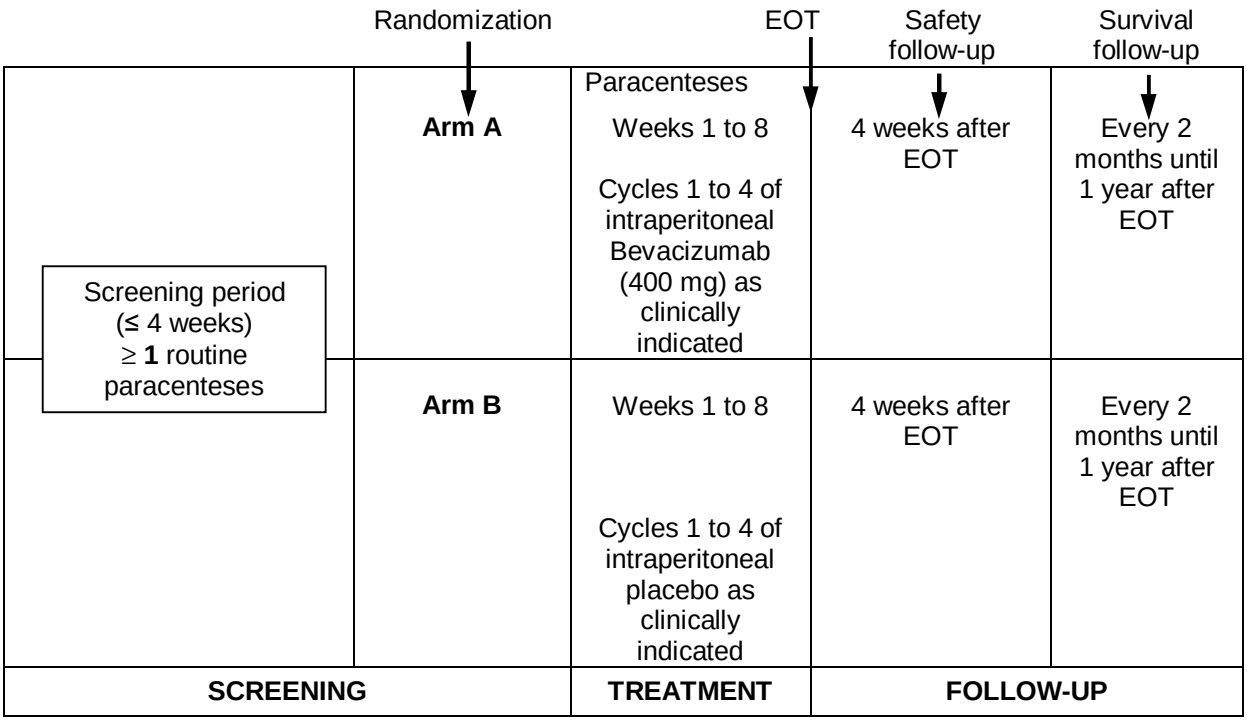
into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period starts with the application of the first paracentesis for study purposes **and must not take place later than 4 weeks after the preceding paracentesis in screening phase**.

3.1 Figure 1, Study Design, Page 38

Old text:



New text:



3.4 Study Duration, Page 39

Old text:
The study is planned to start in January 2010 with respect to first patient in (FPI) including a recruitment period of two years. The follow-up periods will be approximately 6 months in total. The total study duration is approximately 2 and a half year and the last patient will probably complete the study (last patient out; LPO) in June 2012.

Submission to EC/CA:	September 2009
First Patient in (FPI):	January 2010
Recruitment Phase:	2.0 years
FU phase:	0.5 year
Last Patient out (LPO):	June 2012

New text:
The study **started** in **June 2010** with respect to first patient in (FPI) **and a recruitment period of approximately 3 and a half year is planned.** The follow-up periods will be approximately 6 months in total. The total study duration is approximately **4 years** and the last patient will probably complete the study (last patient out; LPO) in **December 2013**.

Submission to EC/CA:	September 2009
First Patient in (FPI):	**June 2010**
Recruitment Phase:	**3.5** years
FU phase:	**1 year**
Last Patient out (LPO):	**December 2014**

4.1 Target Population, Page 40

Old text:
Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers will be eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least twice within the 4 weeks prior to inclusion into the study. Under no circumstances are patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

New text:
Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers will be eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least **once** within the 4 weeks prior to **treatment start**. Under no circumstances are patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

4.2 Inclusion Criteria, Page 40

Old text:
8. At the time of inclusion paracentesis required at least twice within past 4 weeks

New text:
8. At the time of inclusion paracentesis required at least **once** within past 4 weeks**. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase.**

4.3 Exclusion Criteria, Page 41

Old text:

4. Transudative ascites (total protein in ascites < 30 g/l)

New text:
Exclusion Criteria:
~~4. Transudative ascites (total protein in ascites < 30 g/l)~~

5.1 Screening Examination and Eligibility Screening Form, Page 46

Old text:
An eligibility screening form (ESF) documenting the patient's fulfillment of the entry criteria for all patients considered for the study and subsequently included or excluded, is to be completed by the investigator and forwarded to the GSO mbH, Johnsallee 30, D-20148 Hamburg. All patients undergoing screening activities (documented by completion of an ESF for each patient) must be listed in the Patient Screening Log.

New text:
An eligibility screening form (ESF) documenting the patient's fulfillment of the entry criteria for all patients considered for the study and subsequently included or excluded, is to be completed by the investigator and forwarded to the GSO mbH, **Harvestehuder Weg 21**, D-20148 Hamburg. All patients undergoing screening activities (documented by completion of an ESF for each patient) must be listed in the Patient Screening Log.

AIO- Trial SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Short title: AIO-SUP-0108

Sponsor: AIO-Studien-gGmbH
Straße des 17. Juni 106-108
10623 Berlin
Phone: 030-322932933
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Study Coordinator (LKP)
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Dr. Karin Jordan

Expanded Protocol committee
Dr. Axel Hinke
Prof. Dr. Norbert Frickhofen
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EudraCT Nr.: 2009-014725-16

Protocol identification number: AIO-SUP-0108

Protocol version: 4.0 (18.04.2012)

Confidentiality

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Signature	Date (DD Month YYYY)

INVESTIGATOR'S AGREEMENT

I have read the attached protocol entitled

"Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers", dated 18. April, 2012,

and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonisation Tripartite Guideline on Good Clinical Practice.

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the study sponsor.

Signature	Date (DD Month YYYY)
Investigator	
Investigator's Institution	

SYNOPSIS

Protocol No.	AIO-SUP-0108
Protocol Version (Date)	Version 4.0 (18.04.2012)
Title	Bevacizumab as a palliative treatment for patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers
Detailed Title	Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers
EudraCT No.	2009-014725-16
Principle Investigator	PD. Karin Jordan Supportive Care Study Group, (for the Arbeitsgemeinschaft Internistische Onkologie, DKG) Clinic for Internal Medicine IV Department of Oncology/Hematology Martin Luther University Halle-Wittenberg
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Sponsor	AIO-Studien-gGmbH Straße des 17. Juni 106-108 10623 Berlin Phone: 030-322932933 Fax: 030-322932943
Study design	• Double-blind, placebo-controlled, randomized, multi-center, phase II • Patients will receive repeated intraperitoneal application of Bevacizumab/Placebo in a 2:1 ratio
Start date (FPI)	06/2010
Duration of study	Approx. 4 years
Total number of centers	Approx. 20 centers
Study population	Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy

Rationale	Malignant ascites represents a severe clinical problem for physicians and patients. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites, and therapies that are commonly being used are only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers. Preclinical data strongly suggest that Bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites treated with Bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1]. Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.
Objectives	
Primary objective	• To evaluate the paracentesis-free survival (ParFS) following intraperitoneal application of Bevacizumab/Placebo
Secondary objectives	• To measure the frequency of paracenteses required for symptom control following intraperitoneal application of

	Bevacizumab/Placebo, by assessing the longest paracentesis-free period within the 12-week main observation period ("best response") • To measure the volume of ascites following intraperitoneal application of Bevacizumab/Placebo • To measure the effect of study treatment on the quality of life • To assess feasibility and safety of intraperitoneal application of Bevacizumab including pharmakocinetic of Bevacizumab • To evaluate the effect of an intraperitoneal application of Bevacizumab/Placebo on serum and ascites VEGF concentrations
Planned sample size	• In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized • Number of evaluable patients: 40 arm A (treatment), 20 in arm B (control)
Inclusion criteria	1. Age ≥ 18 years 2. Written informed consent has been obtained prior to inclusion into the study 3. Patient is capable and willing to comply with the study 4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma 5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound 6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy 7. Ascites clinically judged as not responsive to diuretics 8. At the time of inclusion paracentesis required at least once within past 4 weeks. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase**.** 9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. <u>Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.</u> 10. ECOG performance score 0-3 11. Life expectancy > 12 weeks 12. Laboratory parameters: <u>Hematology</u> • Neutrophils > 1,500/µl • Platelets > 100,000/µl • Hemoglobin ≥ 9 g/dl or 5.59 mmol/l <u>Hemastaseology</u>

	• INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d Clinical chemistry • Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN • Serum bilirubin < 3.0 x ULN • Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN) Urinalysis: • Patients with < 2+ proteinuria on dipstick urinalysis. • Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.
Exclusion criteria	Patients with any of the following will not be eligible for participation: 1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible). 2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion 3. Hemorrhagic ascites (ascites hematocrit > 2%) ~~4. Transudative ascites (total protein in ascites < 30 g/l)~~ 5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed. 6. Therapy naïve patients 7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up. 8. Patients with extensive metastases of the liver making up > 70% of the total liver mass 9. Child C cirrhosis of the liver 10. Occlusion or thrombosis of the portal vein. 11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression. 12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II. 13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder) 14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or

anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic diseases.
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contra-indicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have participated in this study before.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[1]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to

[1] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

	§ 40 (4) and § 41 (2) and (3) AMG). 31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study. 32. Patients who possibly are dependent on the sponsor or investigator.
Duration of study	Recruitment: 3.5 years Treatment: Patients will receive paracentesis as needed for symptom control. In addition, patients will receive up to 4 intraperitoneal administrations of Bevacizumab/Placebo after paracentesis has been performed. During the 8-week treatment period, a minimum interval of 14 days will be kept between applications of the study medication. Follow-up: End of treatment (EOT) is set at eight weeks after application of the first paracentesis within the treatment period for both arms of the study. Follow-up regarding response and safety in both arms of the study will be conducted at week 4 after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-month intervals for a total of 12 months.
Arms of study	• 48 patients randomized into arm A will receive repeated intraperitoneal application of Bevacizumab • 26 patients randomized into arm B will receive repeated intraperitoneal application of Placebo
Data Safety Monitoring Board	A Data Safety Monitoring Board (DSMB) will be established prior to the start of the study and will be responsible for reviewing safety data on a regular basis. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT).
Primary parameter	Efficacy: • Paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first)
Secondary parameters	Efficacy • Best Response (BR) representing the longest period of time (in days) from o one paracentesis until next paracentesis within the treatment period

- or, if longer, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up)
- or, if longer, from the last paracentesis performed within the treatment period until death (before end of the standard 4 week follow-up)
- or, if longer, from the last paracentesis performed within the treatment period until 4 week follow-up

- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites as indicated by body weight
- Quality of life as assessed by standardized questionnaires
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5.
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events).
- All adverse events.
- Changes in laboratory values and vital signs.
- Changes in ECOG performance status.

Pharmacokinetics of Bevacizumab and VEGF concentrations:

- Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis during the 8-week treatment period and, if possible, at safety follow-up.
- Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis during the 8-week treatment period, as well as at the time of routine safety follow-up. In case a second paracentesis is not clinically required after the first paracentesis (with or without Bevacizumab) following inclusion into the study, measurements will be performed at bi-weekly intervals from last paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

Study procedure	After the initial screening procedure, eligible patients will enter the treatment part of the study. Patients will receive up to 4 intraperitoneal administrations of bevacizumab/placebo depending on clinical necessity of routine paracentesis for symptom relief. In case of unacceptable toxicity, treatment will be prematurely discontinued. A final follow-up regarding response and safety will performed for both arms at 4 weeks after EOT. Thereafter, all patients will be followed up for progression-free and puncture-free survival at 2-months intervals for a total of 12 months.
Randomization procedure	Permuted block randomization will be applied to guarantee balanced group numbers.
Statistical considerations	
Sample size calculation	The primary variable is efficacy as indicated by the paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death, whichever occurs first). Under the assumption of an expected median ParFS in the control group of 14 days [3] and a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions: • type I error: 5% (one-sided) • power: 80% • observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year (Parsons et al., 2007) [3] • exponential shape of the Kaplan-Meier [4] curves In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized. As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis (or other symptomatic treatments for ascites with the exception of diuretics or death or end of the standard 4-week follow-up) within in the 12 week observation period. However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data.

Analysis plan	Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison. Secondary analyses: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend. Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test. In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test). Quality of life as assessed by the standardized questionnaires (FACIT-AI) will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a comparable manner. Other analyses to be performed descriptively: • Overall survival will be analyzed analogous to ParFS • The proportion of patients with changes in ECOG perform-

ance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

INFORMATION TO BE GIVEN ON SAE / PREGNANCY

In the case of a serious adverse event (SAE) or pregnancy the following person must be contacted within one working day by fax:

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Address:	**GSO mbH** **Harvestehuder Weg 21** **D-20148 Hamburg**
Phone:	**+49 40 44 19 54 60**
Fax:	**+49 40 44 19 54 78**

FLOW CHART: SCHEDULE OF ASSESSMENTS DURING THE STUDY

	Screening[1, 3]		Baseline[3,9]	Treatment Period[3]				Safety FU	Survival FU[4]
Treatment Number[2]				**1**	**Variable (maximum number: 4 applications)**			**EOT[19] + 4 weeks**	**Every 2 months**
Study Week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent[5]	X								
In- / Exclusion Criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment History	X								
Pregnancy Test (if applicable)[6]		X							
Frequency of paracenteses required[7]	X			X	X	X	X	X	
Volume of ascites drained[8]	X			X	X	X	X		
ECOG Performance Status[11]			X[9]	X	X	X	X	X	
Physical Examination[11]			X[9]	X	X	X	X	X	
Body weight[11]	X			X	X	X	X	X	
Quality of life assessment[11]		X		X	X	X	X	X	
Vital Signs[10, 11]			X[9]	X	X	X	X	X	
12-lead ECG			X[9]	As clinically indicated					
Investigational analysis of plasma[12]				X	X	X	X	X	
Investigational analysis of ascites[12]				X	When paracentesis is clinically indicated			X[20]	
Urinalysis [11, 13]		X		X	X	X	X	X	
Hematology [11, 14]		X		X	X	X	X	X	
Clinical Chemistry [11, 15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X[20]	
Paracentesis for symptom control	As indicated			X	As clinically indicated			X[20]	
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X[18]
Survival				Continuously				X	X

Notes

1. The screening visit S1 will take place within the screening period and not earlier than 7 days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.
3. All assessments have to be performed before administration of the study drug
4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.
5. Prior to the first study-specific measures.
6. Applicable for women of childbearing potential. Serum β-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.
7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.
8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.
9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy)
10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.
11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. Measurements will be performed at biweekly intervals from first paracentesis until EOT. A final measurement will be performed at safety follow-up.
12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for pharmacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Sera will also be collected at 14-day intervals from first paracentesis until EOT and a final sample will be collected at safety follow-up.
13. Urinalysis: Dipstick test for protein only. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine is required.
14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.
15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.
16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) from 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.
17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.
18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.
19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.
20. When Paracentesis is clinically indicated

GLOSSARY OF ABBREVIATIONS

β-HCG	Beta human chorionic gonadotropin
AE	Adverse event
AIO	Arbeitsgemeinschaft Internistische Onkologie
ALT (SGPT)	Alanine aminotransferase
AMG	Arzneimittelgesetz (German Drug Law)
ANC	Absolute neutrophil count
aPPT	Activated partial thromboplastin time
AST (SGOT)	Aspartate aminotransferase
BR	Best Response
CHF	Congestive heart failure
CHO	Chinese hamster ovary
CNS	Central nervous system
CR	Complete response
CRF	Case report form
CT	Computed tomography
CTCAE	Common terminology criteria for adverse events
DGHO	Deutsche Gesellschaft für Hämatologie/Onkologie
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
EDTA	
ELISA	Enzyme-linked Immuno-absorbant Assay
EOT	End of treatment
ESF	Eligibility screening form
FACIT-AI	Functional Assessment of Chronic Illness Therapy - Ascites Index
GCP-V	GCP-Verordnung
h	Hour
IC	Informed consent
ICH	International Conference on Harmonization
IMP	Investigational medicinal product
INR	International normalized ratio
ITT	Intent to treat
iv	intravenous
LD	Longest diameter
LDH	Lactate dehydrogenase
LKP	Leiter der klinischen Prüfung (Co-ordinating Investigator)
m2	Square meter (body surface area)
mg	Milligram
min	Minute
mL	Milliliter
MRI	Magnetic resonance imaging
NB	Nota bene (please note)
NCI	National Cancer Institute
NCT	National Center for Tumor Diseases
NSAIDS	Non-steroidal anti-inflammatory drugs
NYHA	New York Heart Association
PVS	Peritovenous shunting
PD	Progressive disease
PR	Partial response
RECIST	Response Evaluation Criteria in Solid Tumors
rHuMAb	Recombinant humanized monoclonal antibody

rpm	Rounds per minute
SAE	Serious adverse event
SD	Stable disease
SmPC	Summary of product characteristics
UICC	International Union Against Cancer
ULN	Upper limit of normal
VEGF	Vascular Endothelial Growth Factor
VPF	Vascular Permeability Factor
w/wo	With or without

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PART I – STUDY DESIGN AND CONDUCT

1 BACKGROUND AND RATIONALE

1.1 Background

1.1.1 Malignant ascites

Growth of tumors in serous cavities such as the peritoneum is often accompanied by the accumulation of a protein-rich exudates and formation of malignant effusions is a common problem for patients with advanced-stage cancer. Malignant ascites is defined as an abnormal accumulation of fluid in the peritoneal cavity as a consequence of cancer occurring in association with a wide variety of neoplasms such as colorectal, stomach, pancreatic, ovarian, breast, and lung cancer [5].

Accumulation of massive amounts of malignant ascites is a significant cause of morbidity and mortality in patients with intra-abdominal tumors [6]. Accordingly, mean survival is only 20 weeks after ascites has been discovered [7]. Local fluid accumulations provoke discomfort and distress in many patients in advanced stages of their disease. By increasing abdominal pressure malignant ascites causes severe symptoms such as abdominal pain, bowel obstruction, shortness of breath, loss of appetite, nausea, cachexia, anorexia, reduced mobility, and fatigue [6]. Palliation of the symptomatic patient is the foremost goal and elimination of fluid accumulation in a patient with these symptoms will certainly improve the patient's quality of life and may even prolong survival [8]. Unfortunately, treatment of malignant ascites is problematic and challenging. No single method has been developed that works satisfactorily for the majority of patients and, accordingly, effective management of malignant ascites has been a frustrating problem for many physicians and their patients [8].

Currently, treatment modalities commonly applied for patients with malignant ascites include diuresis, salt restriction, paracentesis, and peritoneo-venous shunts, however, evidence for each of these treatment options is weak, and there are no randomized controlled trials evaluating their safety and efficacy. In addition, results achieved by the application of these conventional methods are variable, only occasionally provide prolonged relief from symptoms, and do not improve survival [9]. Accordingly, in contrast to treatments for the underlying cancer, there is no generally accepted and evidence-based guideline for the management of malignant ascites [10].

Reduced sodium intake together with diuretics are often used to treat malignant ascites but there is no consensus regarding effectiveness [11]. As in the case of other treatment modali-

ties for malignant ascites, there are no randomized controlled trials assessing the efficacy of diuretic therapy in malignant ascites. Available data are controversial and there are no clear predictors to identify which patients would benefit from diuretics. The use of diuretics should, therefore, be evaluated individually [10] and some authors have even concluded that medical therapies such as diuretics, and sodium and fluid restriction are not effective in ascites caused by malignancies [8].

Paracentesis has been the most common treatment option offering the advantage of a quick, simple, and relatively low-risk procedure with immediate symptom relief. However, as the patient's disease progresses, the frequency of hospital visits for the procedure increase as well. Patients are left with the choice between frequent hospital visits or waiting as long as possible between procedures until the ascites symptoms are no longer tolerable [8]. In addition, repeated paracenteses subject the patient to risks such as bleeding, infection, visceral perforation, and hypotension associated with invasive fluid depletion, and renal impairment [5]. Finally, the procedure itself is painful, inconvenient, and, most importantly, only temporarily effective [8].

To avoid repeated paracenteses, a peritoneo-venous shunting (PVS) may theoretically be considered. However, PVS requires hospitalization and surgery and is associated with significant risks both in shunt placement as well as in the ordinary function of the shunt [12]. Accordingly, major complications such as pulmonary edema, pulmonary emboli, clinically relevant disseminated intravascular coagulation, and infection, have to be expected in a significant number of patients undergoing PVS [8, 10]. Moreover, survival and quality are not improved in patients who have received PVS in comparison with patients treated with serial paracentesis [12]. Therefore, it is agreed that shunt insertion is contraindicated in patients with gastrointestinal cancer and malignant ascites due to relatively poor prognosis and limited survival [10, 13, 14].

Recently, the tri-specific antibody catumaxomab has been introduced to the treatment of malignant ascites [15, 16]. This CD3- and EpCAM-specific antibody is thought to stimulate the T cellular immune system as well as to induce MHC-unrestricted cytotoxicity and phagocytosis of tumor cells [17, 18]. Cohorts of patients suffering of ovarian cancer and gastric cancer have experienced symptom relieve after catumaxomab application and, more recently, a phase II/III trial has assessed catumaxomab in the treatment of malignant ascites, with significant results regarding to puncture-free survival and quality of life [19]. In conclusion, although these data need to be confirmed in daily clinical practice, catumaxomab might represent a new approach for the therapy of malignant effusions. However, the need for

placement of an intraperitoneal catheter for several days and the prolonged hospital admission required for the treatment severely limit its potential use in patients with end-stage cancer who require palliative treatment [16]. In addition, EpCAM is known to be expressed on a variety of healthy tissues including hepatocytes [20]. Accordingly, significant grade III-IV hepatic toxicity has been observed in patients treated with intravenous antibody, sometimes even associated with an impaired hepatic function [21]. Similar side effects have been described when catumaxomab was applied intraperitoneally [16]. Therefore, catumaxomab seems to be better suited for patients with a relatively good performance state, able to tolerate both the continuous presence of an intraperitoneal catheter in an inpatient setting as well as hepatic toxicity [21]. Most patients with malignant ascites, however, are unlikely to be candidates for this particular mode of treatment.

In conclusion, effective palliation of malignant ascites remains a difficult management issue. As patients are expected to survive only for a very limited period of time, a desirable treatment should (1) effectively alleviate associated symptoms, (2) be minimally invasive, (3) allow for rapid discharge from the hospital, (4) be relatively simple with low associated risk of complications, and (5) be of tolerable cost to the patient and his/her family [22].

1.1.2 Vascular Endothelial Growth Factor

A better understanding of the molecular mechanisms that regulate the formation of malignant effusions may offer ways to design novel and more effective modes of therapy for this severe cancer-related clinical problem. The etiology of malignant pleural effusions and ascites had traditionally been attributed to lymphatic obstruction caused by tumor spread into draining lymph vessels [23, 24, 25, 26]. It had also been suggested that tumor-induced angiogenesis might contribute to the development of ascites [27, 28]. In 1983, however, Senger et al. suggested an alternative possibility [29]. They isolated vascular permeability factor (VPF) from ascites of tumor-bearing animals and hypothesized that this factor secreted by tumor cells in a paracrine fashion was responsible for the cancer-related fluid accumulations [30]. A few years later, vascular endothelial growth factor (VEGF) was discovered as a potent stimulator of angiogenesis and was subsequently recognized to be identical to VPF [31, 32].

VEGF is a highly conserved 34-42 kD glycoprotein secreted by a large variety of human tumors [30, 33, 34]. In addition, peritoneal mesothelial cells [35], monocytes/macrophages infiltrating malignant effusions [36], and even tumor-infiltrating T cells [37] are capable of producing VEGF.

By interacting with two high affinity tyrosine kinase receptors (Flt-1 and KDR/Flk-1), which are selectively expressed in vascular endothelium [38], VEGF acts on endothelium both normal and newly induced by tumor angiogenesis [39].

Angiogenesis, the development of new blood vessels from pre-existing vasculature, is an essential component of solid tumor growth and metastasis [40, 41, 42, 43]. It is now generally accepted that solid tumor growth must be accompanied by angiogenesis to provide the vascular support necessary for the expanding tumor mass. However, not only does neovascularization permit further tumor growth of the primary tumor, but it also provides a pathway for migrating tumor cells to gain access to the systemic circulation and to establish distant metastases. Tumors express a variety of angiogenic factors in order to promote their own vascularization by activating the host endothelium. One angiogenic factor that is thought to play a decisive role in the vascularization of neoplastic tissue is VEGF which is a potent and specific mitogen for endothelial cells [44] and stimulates the full cascade of events required for angiogenesis *in vitro* and *in vivo* [45]. However, in addition to its ability to promote angiogenesis, VEGF is also capable of markedly augmenting the permeability of pre-existing microvasculature [29, 39, 46].

1.1.3 VEGF and malignant ascites

VEGF is over-expressed in a variety of tumors causing malignant ascites [47, 48, 49] and intratumoral VEGF expression correlates with an increased metastatic potential [49, 50] and poorer survival rates, among others, in gastrointestinal tumors, ovarian, breast, and lung cancer [48, 49, 51, 52, 53, 54, 55, 56]. Accordingly, serum concentrations of soluble VEGF have often been shown to be increased in patients with various solid tumors [47, 51, 57, 58, 59, 60, 61, 62] when compared to normal controls. Serum levels of VEGF correlate positively with the stage of the disease [58, 60, 61, 63], and elevated concentrations of VEGF in the peripheral blood of cancer patients might also be associated with a poorer overall and progression-free survival [58, 59, 61].

Initial studies had already indicated that the accumulation of malignant ascites results in large parts from an increased permeability of peritoneal lining vessels [64, 65]. However, until the identification of VEGF in malignant ascites, the molecular basis of peritoneal vascular hyperpermeability had not been deciphered. It was later shown that that malignant effusions derived from tumor-bearing mice and guinea pigs contain high concentrations of soluble VEGF [36, 66, 67, 68, 69] and that in mice injected with tumor cells increases in microvascular permeability of pre-existing small vessels located in tissues lining the peritoneal cavity as

well as the total volume of the peritoneal fluid correlated closely with the appearance of VEGF within the ascites [36, 67, 70]. Furthermore, it was shown that VEGF protein accumulated in the leaky blood vessels that line the peritoneal cavities of mice bearing ascites tumors [38, 39, 70] and that in low nanomolar or picomolar concentrations VEGF increased the permeability of venules and small veins for plasma proteins with a potency 10.000 times higher than histamine [29]. Finally, the expression level of VEGF by cancer cells has been shown to directly correlate with the tumor cell-induced production of ascites in the animal model [71, 72, 73]. Accordingly, transfection of renal cancer cells with VEGF cDNA or viral vectors encoding for VEGF increased the capacity of these cells to induce hyperpermeability of peritoneal blood vessels and ascites following implantation into mice [74, 75, 76, 77]. Even direct transfection of mouse peritoneum with VEGF was sufficient to cause an accumulation of ascites [75]. In contrast, transfection of tumor cell lines with VEGF antisense oligonucleotides resulted in a reduced formation of malignant effusions in the mouse [72, 76, 78]. Altogether, these collected findings allowed for the firm conclusion that local VEGF secretion is responsible in large parts for initiating and maintaining the ascites pattern of tumor growth.

In numerous human studies, markedly increased concentrations of VEGF have been found in malignant pleural effusions and ascites derived from patients with a large variety of solid tumors, such as ovarian cancer, gastric cancer, colorectal cancer, pancreatic cancer, breast cancer, and lung cancer. Generally, much lower concentrations were detected in non-malignant effusions caused by congestive heart failure, liver cirrhosis, or infections [34, 35, 38, 57, 62, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95]. In a very recent study by Atanackovic et al. concentrations of 21 different cytokines/chemokines were simultaneously analyzed in malignant and non-malignant ascites and it was clearly shown that VEGF is the one cytokine most strongly over-expressed in ascites related to cancer [96]. VEGF concentrations within the effusions are always higher than in the corresponding sera from the respective patients [47, 57, 92, 97] and the volume of malignant ascites correlates with VEGF concentrations within the effusion [98] and with the intensity of VEGF expression in abdominal tumors removed from the same patient [99], indicating a significant local release within the peritoneal or pleural cavity. Furthermore, concentrations of VEGF within human malignant effusions correlate with their capability to induce vascular leakage in an experimental model, an effect that can be blocked by treatment with an antibody directed against VEGF receptor Flk-1 [82, 83]. Most importantly, concentrations of VEGF in malignant ascites have recently been shown to correlate with chemosensitivity and represent an independent predictor of progression-free and overall survival of cancer patients [94, 97, 98].

1.1.4 Inhibition of VEGF activity as a potential therapy for malignant effusions

If VEGF is responsible for fluid accumulation in the environment of solid tumors then anti-VEGF therapies should be able to directly influence the development of malignant effusions in addition to possessing an immediate anti-tumor effect. Importantly, it has repeatedly been shown in *in vitro* experiments that the capacity of VEGF, present in the supernatant of tumor cell lines or in malignant ascites, to induce vascular hyperpermeability can indeed completely be neutralized using an antibody directed against VEGF [29, 30, 38, 67, 100]. Furthermore, it was already shown in an initial animal study by Senger et al. that an anti-VEGF antibody is able to block the increased peritoneal influx associated with the intra-abdominal presence of VEGF-secreting tumor cells *in vivo*. Since then, a number of studies have clearly demonstrated that the intraperitoneal application of anti-VEGF antibodies is safe and leads to impressive and often complete remissions of the local fluid accumulations in mice following inoculation with different carcinoma or sarcoma cell lines [67, 68, 73, 101, 102]. Consistent with these findings, the vascular permeability of microvessels lining the peritoneal cavity of tumor-bearing mice decreased significantly in the anti-VEGF antibody-treated animals compared with controls [67]. Antibody treatment to a lesser degree also inhibited tumor growth [67, 68, 101, 102] and prolonged the survival of mice inoculated with tumor cells [67]. A comparable preclinical efficacy was seen with tyrosine kinase inhibitors targeting VEGF receptors [103, 104] or with a soluble VEGF decoy receptor inhibiting VEGF [75, 105], and after intraperitoneal infusion of a VEGF antisense oligonucleotide [106], but not with conventional chemotherapy applied intraperitoneally alone [73]. Interestingly, the delay in tumor growth induced by the anti-VEGF antibody was mainly attributed to the blockage of ascites development and vascular permeability and not to the inhibition of VEGF-induced angiogenesis [67]. In this context, it has been hypothesized that increased vascular permeability within the peritoneum leading to the development of ascites might indeed result in increased shedding of mesothelial cells into the abdominal cavity facilitating indirectly peritoneal tumor dissemination [107].

Despite the very strong preclinical evidence for an obligatory role of VEGF in the formation of malignant ascites and for a possible therapeutic efficacy of anti-VEGF therapies in the setting of malignant effusions, there are currently no reports from clinical studies addressing this point in cancer patients. However, recently a number of articles reporting on small series of patients with malignant effusions treated off-label with bevacizumab have presented impressing results. It was first reported by Pichelmayer et al. that Bevacizumab might be active in malignant ascites [108]. Following their observation of a marked responses to treatment

with Bevacizumab in a patient with benign pleural effusion [109], they decided to apply a single dose of Bevacizumab intravenously at 15 mg/kg to two patients with malignant ascites due to colorectal cancer and adenocarcinoma of unknown origin, respectively. They found that both patients, in whom paracentesis was previously required at least every second week, treatment with bevacizumab was safe and highly successful. They observed significant reductions in ascites volume resulting in a discontinuation of repeat paracentesis. Moreover, both patients had a marked decrease in their VEGF plasma levels after treatment. [109]. In agreement with these early observations, Numnum et al. reported the intravenous application of bevacizumab (15 mg/kg every 3 weeks) to 4 heavily pretreated patients with end-stage ovarian cancer with the intent to palliate symptomatic ascites. In all 4 patients repeatedly applied paracenteses could be discontinued because of dramatically reduced levels of ascites after initiation of therapy with bevacizumab [110].

In a very recent publication, Hamilton et al. reported on the treatment of an 88-year-old patient receiving home hospice care with refractory ovarian cancer, a very poor functional status, and severe symptomatic ascites. They performed paracentesis and treated the patient with two subsequent doses (5 mg/kg) of intraperitoneal bevacizumab with dramatic improvement in her ascites and the quality of her final weeks of life [2]. The largest series of patients treated with intraperitoneal Bevacizumab has recently been presented by El-Shami et al. who evaluated the safety and efficacy if intraperitoneal administration of bevacizumab (5 mg/kg every 4 weeks) to a total of 9 patients with refractory ascites due to colorectal, breast, uterine, or ovarian cancer. Impressingly, malignant ascites resolved after a single intraperitoneal dose in every single patient without reaccumulation or repeat paracentesis over a median observation period of more than two months. Moreover, no grade 2-5 adverse events were observed [1].

1.1.5 Study Drug Bevacizumab

Bevacizumab (Avastin®) is a recombinant humanized monoclonal antibody (rHuMAb) to VEGF composed of human IgG1 framework regions and antigen-binding complementarity-determining regions from a murine antibody (A.4.6.1) that blocks the binding of human VEGF to its receptors [111] (further details can be found in the Investigator's Brochure). By neutralizing the biologic activity of VEGF produced by solid tumors, Bevacizumab has the capability to reduce the vascularization of tumors, thereby inhibiting the growth of the malignancy. In numerous mouse models, bevacizumab has clearly demonstrated an inhibition of human tumor growth. The administration of bevacizumab in hetero-transplant models of

colon carcinoma has shown to result in a reduction in microvessel formation and number of metastases.

A number of properties make some agents more favorable than others for intraperitoneal therapy. One important characteristic might be that the drug has activity in the malignancy to be treated. The efficacy of bevacizumab when used in combination with chemotherapy has been demonstrated in several prospective, randomized phase III studies [112, 113, 114, 115]. For example, in a phase III trial in patients with metastatic colon cancer, bevacizumab in combination with standard chemotherapy was found to increase overall and progression-free survival and response rates when compared with chemotherapy plus placebo [113]. Because of this and the results from other trials [116], bevacizumab has become widely used for the treatment of colorectal cancer and non-small-cell lung cancer and is being studied as part of the treatment regimen in a wide range of malignancies [117, 118, 119, 120]. In Germany, bevacizumab has been approved for first-line treatment of metastatic colorectal carcinoma (in combination with 5-FU/folic acid or 5-FU/folic acid/irinotecan [FOLFIRI]) in January 2005. Since January 2008, bevacizumab has been approved for the treatment of mCRC in combination with any fluoropyrimidin-based chemotherapy.

Bevacizumab is generally well tolerated and has an acceptable toxicity profile consisting primarily of hypertension and proteinuria. Other rare but important adverse effects, however, include delayed wound healing, arterial thrombosis, and bleeding [121]. Another potentially serious adverse effect of bevacizumab is gastrointestinal (GI) perforation and, although comparably infrequent, this potentially life-threatening complication has generated significant clinical interest. Overall, GI perforation was found to be an uncommon but well-documented side-effect of treatment in the phase III trials of bevacizumab, as well as in subsequent surveillance trials, with a reported incidence of 1% to 2% [113, 114, 117]. Accordingly, in an observational study by Hedrick et al. 1968 patients with unresectable colorectal cancer received bevacizumab and first-line chemotherapy and GI perforation was observed in 1.7% of patients [122]. Recently, a retrospective analysis was published examining adverse events in 1442 cancer patients who had received bevacizumab at M.D. Anderson Cancer Center over a 2-year period. Bowel perforation or fistula occurred in 1.7% of patients with a variety of malignancies including gastrointestinal cancers. In patients with colorectal cancer, for example, such adverse events were only observed in 6 of 478 cases (1.3%) Median time to perforation after the initiation of bevacizumab treatment was 71 days. Only five of all 1442 patients ultimately underwent surgical exploration and overall 30-day mortality rate was only 12.5% in these patients [123].

Though strong evidence identifying specific risk factors is lacking, investigators have urged caution when treating patients with known bowel implants or large tumor burden, prior radiation, and recent surgery or bowel obstruction [124]. Accordingly, in their phase II study with colorectal cancer patients Hurwitz et al. [113] identified colon surgery within 2 months as a risk factor, they also found a history of peptic ulcer disease and a partial or complete response to therapy as potential risk factors for perforation. Sugrue et al. [125] analyzed the same registry as Hedrick et al. and found no statistically significant associations between specific patient characteristics and an increased risk of GI perforations were identified, however, 67% of the patients with GI perforation showed at least one of the following findings: tumor at the site of perforation, obstruction, intra-abdominal abscess, intraabdominal carcinomatosis, acute diverticulitis, or prior abdominal or pelvic radiation therapy. Importantly, these potential risk factors were similar to those observed in the study by Badgwell et al. [123] as was the median time until first event of approximately two months. Finally, in a case report series of patients treated with bevacizumab for colorectal, lung, renal cell, and unknown primary cancer, ischemic bowel complications were more frequent in patients with a history of pelvic irradiation [126]. Although this series consisted of only 33 patients, bowel complications occurred in those three patients who had received infradiaphragmatic irradiation but in none of the 30 remaining patients who had not received this mode of treatment.

Further details on non-clinical and clinical data for bevacizumab are provided in the Investigator's Brochure.

1.2 Rationale

1.2.1 Rationale for the Study, Relevance of the Study and Study Design

Malignant ascites represents a severe clinical problem for physicians and patients being confronted with this common symptom of advanced-stage gastrointestinal cancer. Unfortunately, there is no standardized and evidence-based treatment for malignant ascites and therapies which are currently being used are, if anything, only temporarily effective. Newer modes of therapy, such as the application of the tri-functional antibody catumaxomab, are associated with significant side effects and are limited to patients in stages of good overall performance. Therefore, there is still an urgent need for more effective, longer-lasting, and less toxic modes of treatment for peritoneal effusions caused by gastrointestinal cancers.

Preclinical data strongly suggest that bevacizumab might be a very effective agent for the treatment of malignant ascites, which is in large parts caused by the hyperpermeability-promoting factor VEGF. Emerging clinical results from cancer patients with malignant ascites

treated with bevacizumab add further support to this idea. Bevacizumab has been tested in a variety of large clinical trials, has a good toxicity profile, and is effective in a number of human cancers underlying malignant ascites. While there are, so far, no reports on GI perforation resulting from intraperitoneal application of bevacizumab, there might still be a significant risk of such adverse reactions. However, we believe that palliative intraperitoneal treatment with bevacizumab is still indicated in these patients with advanced-stage gastrointestinal cancer patients who are capable of providing informed consent and who often severely suffer from symptoms associated with malignant ascites.

1.2.2 Rationale for the route of application and dosage selection

In the present study, Bevacizumab will be administered as an intraperitoneal infusion. The route of administration was chosen based on four considerations: (1) Intraperitoneal administration does not mean additional stress for the patients since routine paracentesis requiring the placement of an intraperitoneal catheter is one inclusion criteria of this study, (2) intraperitoneal application should build up the highest concentration of the study drug within the body compartment where malignant ascites is promoted by VEGF secretion, (3) the intraperitoneal route of administration was successfully used in most preclinical animal models of malignant ascites, and (4) within the study reporting the largest series of patients treated for malignant ascites Bevacizumab was administered intraperitoneally [1].

Bevacizumab will be administered intraperitoneally at an absolute standardized dosage of 400 mg. This dosage was chosen because it is comparable to the approved standard dosage for intravenous administration which was also used in both studies reporting the successful and safe intraperitoneal administration of Bevacizumab to patients with malignant ascites [1, 2]. Finally, a standardized dosage seems more practical in the particular patient population treated in this study.

2 OBJECTIVES OF THE STUDY

2.1 Primary Objectives

The **first** primary endpoint will consist of paracentesis-free survival (ParFS) which will be calculated as the time period between the initial puncture after randomization to the first subsequent paracentesis or other symptomatic treatments for ascites with the exception of diuretics or until death (whichever occurs first).

2.2 Secondary Objectives

Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study.

The **second** endpoint (Best response; BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or, from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU.

The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase

Further evaluation of the efficacy, feasibility, and general safety of an intraperitoneal application of Bevacizumab in patients with malignant ascites.

Other measures of efficacy will be:

- Volume of ascites drained by routine paracentesis (ascites volume minus lavage volumes, if applicable)
- Total volume of ascites present in the patient as indicated by body weight at each study visit during the treatment period
- Quality of life as assessed by standardized questionnaires (FACIT-AI) filled out by the patient and one questionnaire by the palliative group of the DGHO, which needs to be completed by the medical staff
- Secondary Analysis: Number of patients with CR/PR

Feasibility and Safety:

- Proportions of patients with adverse events (NCI CTC v3.0) grades 3, 4, or 5
- Proportions of patients with adverse events of special interest (any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events)
- All adverse events
- Changes in laboratory values and vital signs
- Changes in ECOG performance status

Pharmacokinetics of Bevacizumab and VEGF concentrations:

Serum and ascites VEGF and Bevacizumab concentrations will repeatedly be analyzed throughout the study as possible indicators for baseline responsiveness to Bevacizumab and as a parameter for biological response to the study treatment.

Evaluation of Bevacizumab concentrations and of VEGF concentrations in malignant ascites at the time of each routine paracentesis with study medication during the 8-week treatment period and, if possible, at safety follow-up.

Evaluation of serum Bevacizumab and VEGF concentrations at the screening visit, at each routine paracentesis with study medication during the 8-week treatment period, as well as at the time of routine safety follow-up. Measurements will be performed at bi-weekly intervals from first paracentesis until EOT and a final sample will be collected at the standard follow-up 4 weeks after EOT.

3 STUDY DESIGN

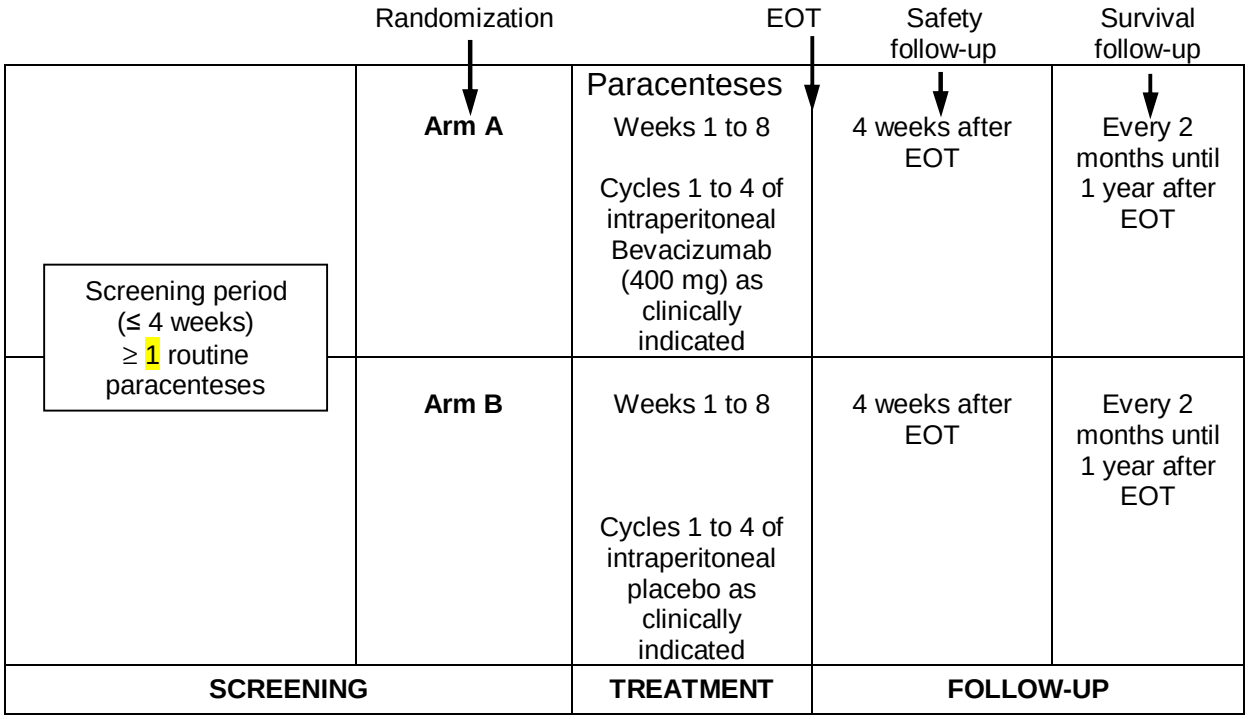
3.1 Overview of Study Design and Dosing Regimen

Double-blind, placebo-controlled, randomized phase II study investigating the efficacy of Bevacizumab for symptom control in patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers after conventional therapy. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the tri-functional antibody catumaxomab or intraperitoneal chemotherapy. At the end of a screening phase of up to 4 weeks during which at least 1 routine paracentesis for symptom control of malignant ascites must have taken place, the screening visit S1 will take place. The screening visit must not take place earlier than 7 days before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. Eligible patients will be randomized into arm A (Bevacizumab) or arm B (placebo) of the study. The treatment period starts with the application of the first paracentesis for study purposes and must not take place later than 4 weeks after the preceding paracentesis in screening phase. Patients will receive up to 4 intraperitoneal administrations of Bevacizumab (400 mg absolute dose) or a placebo depending on clinical necessity of paracentesis for symptom relief. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days. In case of unacceptable toxicity, treatment will be prematurely discontinued.

Patients will generally be followed regarding response and safety at 4 weeks (“safety follow-up”) following EOT. Puncture-free survival follow-up will take place every two months for one year after EOT.

A brief overview on the study design is given in Figure 1.

Figure 1. Study Design



EOT= End of treatment

3.2 Number of Patients/Assignment to Treatment

A total of 72 patients will be enrolled into the study (48 into treatment arm A, 26 into control arm B). At the baseline visit, each patient will receive an unique patient number that will be given to the investigator by FAX at the time of individual patient enrolment. The number assigned of each patient has to be documented by using a Patient Identification Log and on each patient's Case Report Form.

The rational for the 2:1 allocation is that the study may gain more information about patient responses to the new intervention, such as toxicity and side effects. Additionally, if the intervention turns out to be beneficial, more study subjects would benefit than under an equal allocation design. Moreover, from a psychological point of view, the higher chance to receive the intervention rather than placebo may render the trial participation more acceptable to the eligible patients. [155, 158]

3.3 Centers

An approximate number of 20 centers will participate in the study. Each center is expected to recruit at least 4 patients until the planned total number of 72 patients is reached. A list of all participating investigational sites including information regarding names of the principal investigators and contact details (address, phone, fax email) will be handled separately.

3.4 Study Duration

The study started in June 2010 with respect to first patient in (FPI) and a recruitment period of approximately three and a half year is planned.. The follow-up periods will be approximately 12 months in total. The total study duration is approximately 4 years and the last patient will probably complete the study (last patient out; LPO) in December 2014.

Submission to EC/CA: September 2009

First Patient in (FPI): June 2010

Recruitment Phase: 3.5 years

FU phase: 1 year

Last Patient out (LPO): December 2014

4 STUDY POPULATION

4.1 Target Population

Patients with symptomatic malignant ascites due to advanced-stage gastrointestinal cancers will be eligible for the study. Routine paracentesis for symptom control (and not only for diagnostic purposes) must have taken place at least once within the 4 weeks prior to treatment start. Under no circumstances are patients, who were once enrolled in this study, permitted to be re-enrolled into the same study.

4.2 Inclusion Criteria

To be eligible for this trial, patients must fulfill the following criteria:

1. Age ≥ 18 years
2. Written informed consent has been obtained prior to inclusion into the study
3. Patient is capable and willing to comply with the study
4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular, or colorectal carcinoma
5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites > 30 g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT , MRT or ultrasound
6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
7. Ascites clinically judged as not responsive to diuretics
8. At the time of inclusion paracentesis required at least once within past 4 weeks. The first application of study medication must not take place later than 4 weeks after the preceding paracentesis in screening phase.
9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period. This also excludes the application of the trifunctional antibody catumaxomab or intraperitoneal chemotherapy.
10. ECOG performance score 0-3
11. Life expectancy > 12 weeks

12. Laboratory parameters:

    Hematology

    – Neutrophils > 1,500/µl

    – Platelets > 100,000/µl

    – Hemoglobin ≥ 9 g/dl or 5.59 mmol/l

    Hemastasiology

    – INR ≤ 1.5 x ULN and aPTT ≤ 1.5 x ULN within past 7 d

    Clinical chemistry

    – Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN

    – Serum bilirubin < 3.0 x ULN

    – Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7 x ULN)

    Urinalysis:

    – Patients with < 2+ proteinuria on dipstick urinalysis.

    – Patients with ≥ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.

4.3 Exclusion Criteria

Patients with any of the following will not be eligible for the study:

1. Concomitant malignancies other than gastrointestinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
2. Bacterial peritonitis as indicated by laboratory results (neutrophil count > 250 / µl ascites) or clinical suspicion
3. Hemorrhagic ascites (ascites hematocrit > 2%)
4. ~~Transudative ascites (total protein in ascites < 30 g/l)~~
5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (– 4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed.
6. Therapy naïve patients

7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
8. Patients with extensive metastases of the liver making up > 70% of the total liver mass
9. Child C cirrhosis of the liver
10. Occlusion or thrombosis of the portal vein.
11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptoe, cardiovascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III/IV, symptomatic coronary heart disease, peripheral arterial disease stage ≥ II.
13. History of fistula formation involving an internal organ (e.g. tracheo-oesophagal, bronchopleural, biliary, vagina and bladder)
14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab for primary malignancy is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture.
17. Radiotherapy for purposes other than local control of symptoms.
18. Evidence of bleeding diathesis or coagulopathy.
19. Hematopoietic disease
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration.
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (≤ 6 months before treatment start).
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.

25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have previously participated in this study.
28. Women, lactating, pregnant or of childbearing potential and fertile men not using a highly effective contraceptive method[2]. [Women of childbearing potential must have a negative pregnancy test (serum β-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
32. Patients who possibly are dependent on the sponsor or investigator.

4.4 Concomitant Medication and Treatment

The initiation or continuation of any non-protocol-specific anti-tumor therapy is forbidden from inclusion into the study until EOT. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs will not be allowed from start of the screening phase until safety follow-up. This also excludes treatment with the tri-functional antibody catumaxomab or the intraperitoneal application of chemotherapy. Application of such treatments from start of the screening phase until safety follow-up will lead to the immediate discontinuation of the study for the given patient.

[2] Methods allowed for birth control (with a failure rate of ≤ 1% per year) are implants, injectables, combined oral contraceptives, intrauterine devices (only hormone spirals), sexual abstinence or vasectomized partner for up to 6 months after end of treatment.

All concomitant medication(s) must be reported in the Case Report Form (CRF). Any diagnostic, therapeutic, or surgical procedure performed during the study period should be recorded including the dates, description of the procedure(s) and any clinical findings. Patients should receive full supportive care including transfusion of blood and products, antibiotics, etc. where applicable. The treatment details should be recorded in the CRF.

With the only exception of full-dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.

Prophylactic low-dose aspirin is a recommended standard of care in patients at high-risk of an arterial thrombo-embolic event [127] and is supported by an extensive body of literature [128]. Safety data were pooled from three Genentech-sponsored trials in metastatic colorectal cancer (N=1203) in which patients were randomized to fluorouracil-based chemotherapy plus bevacizumab or placebo. In a retrospective exploratory analysis of patients in the bevacizumab arms, the incidence of grade 3-4 hemorrhagic events was 3.4% among those who used low-dose aspirin (≤ 325 mg daily) at enrolment or on study before a hemorrhagic event and 4.4% in those patients who did not use low-dose aspirin [129]. As low-dose aspirin does not appear to increase the risk of grade 3-4 hemorrhagic events when used with bevacizumab plus chemotherapy, the use of prophylactic low-dose aspirin in patients who are at high risk of an arterial thrombo-embolic event is not prohibited in this protocol.

The use of low-dose oral coumarin-derived anticoagulants, heparin, or low molecular weight heparins is permitted before and during study, as is low-dose aspirin (≦325 mg/day) and clopidogrel (≤ 75 mg/day).

Note: In patients who experience thrombo-embolic events during study treatment full dose anticoagulant are allowed and information on anticoagulant treatment (including doses) will be collected and recorded in the CRF.

INR will be assessed at baseline for all patients. In patients treated with oral coumarin-derived anticoagulants INR will be checked at least before start of application of Bevacizumab or routine paracentesis, respectively.

In patients treated with full-dose oral anticoagulants due to thrombo-embolic event during study treatment, INR must be checked at least every second day the first week of treatment, at least 2 times/week for the following treatment weeks until a stable therapeutic level of INR

has been achieved and at least once every 3rd week when the weekly dose has been established and INR is stable with this dose (see also section 7.3.2).

The following is recommended regarding the use of concomitant medications:

Oral contraceptives: No dose modifications are required for patients on oral contraceptives.

5 SCHEDULE OF ASSESSMENT AND PROCEDURE

5.1 Screening Examination and Eligibility Screening Form

At the end of a screening phase the screening visit S1 will take place. The screening visit must not take place earlier than 7 days respectively 3 days (see Flow Chart) before application of the first paracentesis for study purposes. On the screening visit, inclusion and exclusion criteria will be checked and all screening measurements will be performed. After detailed oral and written information about the study, all patients willing to participate in this study have to provide a written Informed Consent (IC) before any study-specific assessment is performed.

An eligibility screening form (ESF) documenting the patient's fulfillment of the entry criteria for all patients considered for the study and subsequently included or excluded, is to be completed by the investigator and forwarded to the GSO mbH, Harvestehuder Wege 21, D-20148 Hamburg. All patients undergoing screening activities (documented by completion of an ESF for each patient) must be listed in the Patient Screening Log.

Patients who are considered for study entry, but who fail to meet the eligibility requirements, should also have an ESF completed with the reason for lack of eligibility given, since this provides information on the selected trial population. These patients will not be entered on the clinical trials database. The ESFs for patients who fail to meet the eligibility requirements should be kept in the study files at the sites. The same applies to patients who fulfill the entry criteria at the screening visit, but no longer at the baseline visit. For patients who did not sign the IC in the first, the ESF have not to be filled in. The patients will only be present in the pre-screening log.

A CRF should be filled out only for patients fulfilling the entry criteria both at screening and at baseline visits.

5.2 Study Assessments

5.2.1 Clinical Assessments

5.2.1.1 Assessment of Severity of Ascites

Baseline severity of malignant ascites will be assessed by calculating the mean time frame (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before start of the treatment phase

(screening phase). In addition, mean volumes of ascites (minus the volume of lavage fluid, if applicable) as well as body weight will be calculated for paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within 4 weeks of screening.

Between first paracentesis performed within the treatment period and until safety follow-up, the frequency of paracenteses clinically required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses with (arm A) or without (arm B) application of Bevacizumab. The Best response (BR) will be calculated as the longest period of time (in days) from one paracentesis until next paracentesis within the treatment period, or (if longer) from the last paracentesis performed within the treatment period until first subsequent symptomatic treatment for ascites with the exception of diuretics (before end of the standard 4 week follow-up), or (if longer) from last paracentesis performed within the treatment period until standard 4 week follow-up or until death within treatment period and 4 week standard FU. Baseline severity of malignant ascites will be assessed by calculating the period (in days) between paracenteses that have been performed for symptom relief (and not only for diagnostic purposes) within the past 4 weeks before inclusion into the study. The best response value will be compared between groups and to the mean time frame between two paracenteses required for symptom relief (and not only for diagnostic purposes) during the screening phase.

Between first paracentesis within the treatment period and until EOT, volume of ascites (minus the volume of lavage fluid, if applicable) drained during routine paracentesis for symptom relief with (arm B) or without (arm B) subsequent application of Bevacizumab will be recorded. Mean volumes and total volumes of ascites removed will be compared between groups and to the respective baseline values (mean and total volumes of paracentesis performed for symptom relief and not only diagnostic purposes during screening period) within the same group.

5.2.1.2 ECOG Performance Status

To be eligible for study entry, patients must have an ECOG performance score between 0 and 3 (see also section 4.2, Item 10). The patients' ECOG performance status (see section 18.3) will be assessed at the screening visit, before every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In addition, assessment of ECOG performance status will be performed every 2 weeks from first paracentesis until EOT and at safety follow-up.

5.2.1.3 Assessment of Quality of Life

In order to assess quality of life, utilities will be generated using a standardized and validated instrument of quality of life questionnaire. Quality of life will be evaluated using the FACIT-AI questionnaire, which needs to be filled out by the patient. Another questionnaire developed by the DGHO palliative care group has to be completed by the medical staff. Quality of life (see section 18.4) will be assessed at the screening visit, at every paracentesis during the treatment phase, and at the safety follow-up visit that will take place 4 weeks after EOT. In addition, assessment of QoL performance status will be performed at biweekly intervals from first paracentesis until EOT and at safety follow-up. FACIT-AI will be applied at the same time point during the study as the other quality of life questionnaire. Analyses will reveal the typical severity of each symptom during the different study phases to further assess burden of disease and treatment benefit. Additionally, a total ascites score derive from all symptoms will be calculated for each point in regard to the FACIT-AI.

5.2.2 Safety Assessments

5.2.2.1 Assessment of Toxicity

Throughout the treatment period and the 4-week safety follow-up period, patients will be assessed for toxicities attributable to therapy. Common terminology criteria for adverse events (CTCAE v3.0; see Investigator's File) will be used for grading. If necessary, the patient may be withdrawn from the study treatment. For details, see Section 7.3.

- **Medical history** including cancer and treatment history will be reviewed and recorded at the screening visit.
- **Concomitant medications** will be documented throughout treatment phase and the 4-week safety follow-up period. During the survival follow-up period, only anti-tumor drugs will be documented.
- A **physical examination** will be performed at the screening visit, before start each paracentesis, biweekly and at the safety follow-up visit 4 weeks after the last treatment cycle (safety follow-up).
- **ECG** will be measured at screening and if clinically indicated.
- **Vital signs** (blood pressure, heart rate, body temperature and body weight), will be measured at the screening visit, before start of each paracentesis and 4 weeks after the last treatment cycle (safety follow-up). Body height will be measured at screening only. In addition, measurements will be performed at biweekly intervals form first paracentesis until EOT.

- **Adverse events** (see also Section 7.1): All patients will be closely monitored for adverse events (incl. survival) from Day 1 of the first treatment cycle through Week 4 after the last treatment cycle. Thereafter, patients will be followed up for progression and survival only. Adverse events will be graded according to the Common Terminology Criteria for Adverse Events (CTCAE v3.0).

5.2.3 Laboratory Assessments

5.2.3.1 Routine Laboratory Assessments

Blood samples will be taken for hematological and serum chemistry monitoring at each scheduled visit. The local laboratory will perform the analyses and provide reference ranges. All Assessments must be performed at the screening visit. Thereafter, assessments (with the exception of urinalysis and pregnancy test) will be repeated before each treatment cycle and 4 weeks after EOT (8 weeks after first paracentesis within the treatment period).

- **Hematology** - Hemoglobin, platelets, leukocytes, neutrophils,
- **Hemostasiology** - INR and aPTT.
- **Clinical Chemistry** - Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance, total protein.
- **Urinalysis -** Dipstick test will only be performed for protein (see Flow Chart).. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine is required. Measurements will be performed at screening, prior to each paracentesis within the treatment period and at biweekly intervals form first paracentesis until EOT.
- **Pregnancy test -** A serum β-HCG pregnancy test will be performed at the screening visit, if childbearing potential cannot be ruled out. Additional pregnancy test will be done as clinically indicated.

5.2.3.2 Routine analysis of malignant ascites

Samples of malignant ascites will be taken for hematological analysis and routine chemistry at screening and at each visit scheduled for paracentesis during the treatment period. Analyses will be performed in the local laboratory.

- **Hematology (2-5 ml EDTA-anticoagulated ascites)** – Hemoglobin, hematocrit, absolute numbers of total leukocytes and neutrophils.
- **Chemistry (5 ml heparinized ascites)** – Concentration of total protein and of albumin.

5.2.3.3 Pharmacokinetics of Bevacizumab and investigational analyses

10 ml of blood and 10 ml of ascites fluid will be collected starting at the time of first application of the study medication in heparinized tubes for the generation of the respective blood or ascites plasma samples. In addition, 10 ml of serum will be obtained at the same time points for the analysis of pharmacokinetics of Bevacizumab. Samples for investigational analyses and for the measurement of pharmacokinetics will be obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Heparinized peripheral blood and sera will be collected at 14-day intervals from first paracentesis until EOT and a final sample will be collected at safety follow-up. Serum concentrations of Bevacizumab will be analyzed by Xendo laboratory.

In addition to its role as a vascular permeability and as an angiogenic factor, VEGF also exerts a number of suppressive effects on the T cell-mediated immune system. First, VEGF functions as a chemo-attractant for CD34+ progenitor cells mobilizing these T cell-inhibiting cells into the tumor tissue where they inhibit the function of tumor-infiltrating T cells [130]. In addition, VEGF has repeatedly shown to reduce the phagocytic capacity and to inhibit the functional maturation of antigen-presenting dendritic cells (DC) *in vitro* and *in vivo* [131, 132, 133, 134, 135] and increased levels of VEGF are associated with reduced numbers of DC within the peripheral blood [136] and the tumor tissue [132]. In gastric cancer, for example, multivariate analysis showed that infiltration by DC was an independent prognostic indicator and there was an inverse correlation between the intratumoral density of DC and the expression of VEGF [132]. Accordingly, treatment with anti-VEGF antibody significantly improved the number and function of lymph node and spleen DC in tumor-bearing mice. Moreover, the efficacy of tumor vaccination with peptide-pulsed DC was dramatically enhanced by combining it with anti-VEGF antibody treatment [134, 137].

On the other hand, VEGF may also directly suppress immune effector cells by reducing cytokine-induced leukocyte-endothelial interactions *in vivo* [138] and by decreasing transendothelial migration of leukocytes [139]. Moreover, VEGF has recently been shown to exert an immediate effect on T cell immunity by inhibiting the thymic development of T cells [140] and enhancing Th2-type immunity [136, 141]. Accordingly, cytokines like IL-1β [107], IL-6 [142], and TGF-β [143, 144, 145] as well as a number of Th2-type cytokines (IL-4, IL-5, IL13) [145] might promote the production of intraperitoneal VEGF. In contrast, Th1-type cytokines, such as IFN-γ, inhibit VEGF production [145].

Finally findings for our group and others have recently indicated that the typical immune environment present within malignant ascites, which is marked by a dramatically elevated concentration of VEGF, might also contribute to the accumulation of immunosuppressive T regulatory cells (Tregs) within the effusion [96, 146]. These combined results suggest that expression of VEGF might be associated with tumor progression and poor prognosis not only because VEGF stimulates angiogenesis and vascular permeability, but also because it allows tumors to escape from attack by the immune system in patients with cancer.

In order to assess the effect of Bevacizumab-induced effects on angiogenic/vascular permeability factors as well as the acquired immune system 10 ml of serum and 10 ml of ascites fluid will be obtained from each patient included at each visit performed for routine paracentesis with study medication. Sera will also be collected at bi-weekly intervals from first paracentesis until EOT. A final serum (and ascites sample, if possible) will be obtained at safety follow-up.

As soon as possible after removal, heparinized blood and ascites samples, and sera will be centrifuged for 10 min at 1000g and supernatants will be divided into 3 x 1 ml volumes per sample type (3 ml total volume for each sample type, plasma and ascites fluid) and will immediately be frozen at a minimum of -20°C. After completion of the study by a given patient, the collective Plasma, ascites and serum samples of the respective patients will be picked up by TNT express who will also provide complete packaging and dry ice for the transport to the central investigational laboratory in Hamburg. For each participating center, four pick-up dates will be determined by the sponsor of the study.

All plasma and ascites samples in regard to the investigational analyses will be transported to the Laboratory for Tumor Immunology (Hamburg), Dr. Djordje Atanackovic. The pharmacokinetic samples will be sent to the Xendo Laboratory.

5.2.3.3.1 Central laboratory for pharmacokinetics

Xendo Laboratory

LE I D E N - N E T H E R L A N D S

Bio Science Park, Archimedesweg 17, 2333 CM Leiden

P.O. Box 255, 2300 AG Leiden, The Netherlands

Tel +31 (0)71 524 40 00 Fax +31 (0)71 524 40 01

E-Mail office.pharmaservices@xendo.com

5.2.3.3.2 Central laboratory for investigational analyses

Dr. Djordje Atanackovic
University Medical Center Hamburg-Eppendorf
Center of Oncology
Laboratory for Tumor Immunology
Building N27, 4th floor, Room 04.083
Phone: 0049-40-7410-55032
Mobile: 0049-177-7329398
Fax: 0049-40-7410-55735
E-Mail: D.Atanackovic@uke.uni-hamburg.de

Samples can only be sent from Monday to Wednesday of each week. The central laboratory needs to be informed by email at least one week before the samples are sent. In addition, the courier needs to be informed of the pick-up date at least 24 hours before the samples are expected to be sent:

TNT Express Hotline (01805-633725)

To be reached on working days until 4.00 p.m.

In a first step, serum and ascites samples derived from 5 representative patients will be analyzed by antibody arrays for immunomodulatory cytokines and chemokines as well as a broad variety of angiogenic factors. Results derived from these analyses will be confirmed by analyzing serum and ascites samples of the whole patient collective for the respective cytokines/chemokines/angiogenic factors by ELISA. Sera of 50 anonymized blood donors will serve as controls.

In addition, 20 ml of fresh heparinized blood will be collected at each appointment for collection of serum for investigational analyses and the complete volume of malignant ascites removed at a given paracentesis performed during the treatment period and at safety follow-up will be collected from each patient treated in centers being in close proximity to the laboratory performing the investigational analysis (Laboratory for Tumor Immunology, Center of Oncology, II. Medical Clinic, University Medical Center Hamburg-Eppendorf). From the ascites material, tumor cells as well as endothelial cells and T cells will be separated in order to analyze by real-time PCR-based arrays which cells produce the angiogenic and/or immune factors creating the typical immune environment within malignant effusions. The same patient material as well as the peripheral blood will also be analyzed by flow cytometry for the presence of Tregs and other immunomodulatory cell types

5.2.4 Additional Assessments

Additional assessments will be required in the case of hypertension, proteinuria, thrombosis and hemorrhagic events as specified below. These additional assessments will be recorded on specific CRF forms in addition to the completion of the adverse events form.

- **Hypertension:** In the case of grade 3/4 hypertension, additional blood pressure measurements should be performed on a weekly basis (for the duration of trial therapy) until resolution of the event. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Proteinuria:** A 24-h urine collection is required in case of a protein > 1+ dipstick result before the subsequent treatment cycle. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Thrombosis:** In case of grade 3/4 thrombosis a blood sample for the following laboratory values should be taken prior to initiation of treatment for the event: INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.
- **Hemorrhagic events:** In case of a hemorrhagic event of grade 2 or higher, a blood sample for the following laboratory values should be taken prior to treatment for the event: Platelet count, INR, aPTT. Further study treatment should be in line with the recommendations as defined in Section 7.3.2.

6 INVESTIGATIONAL PRODUCT

6.1 Investigational Medicinal Product (IMP)

According to § 3 (3) GCP-V an investigational medicinal product is a pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial, including products already with a marketing authorization but used or assembled (formulated or packaged) in a way different from the authorized form, or when used for an unauthorized indication, or when used to gain further information about the authorized form.

The IMP in this study is Bevacizumab and its corresponding placebo.

6.2 Background Medication

Background medication for treatment of the primary malignancy or other medical problems of the patient will be applied at the discretion of the investigator. Background medication will not be supplied or reimbursed by Roche. The investigator needs to observe the summary of product characteristics of the different component of the background medication with special attention to contraindications.

6.3 Dose and Schedule of Test Drug Bevacizumab and comparator drug

Patients will receive up to 4 intraperitoneal administrations of the study drugs depending on clinical necessity of paracentesis for symptom relief. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treatment period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.

Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control.

Following the application of an 18-22 G intraperitoneal catheter, the largest possible volume of malignant ascites will be drained. Thereafter, Bevacizumab or the comparator drug will be applied through the same catheter at a total volume of 100 ml. Bevacizumab will be applied at an absolute standardized dosage of 400 mg. The initial dose of the study drug will be delivered over 60±15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills) the following infusions may be delivered over 30±10

minutes. Following the complete application of the study drug the intraperitoneal distribution will be optimized by varying the patient's body position (10 min on the back, 10 minutes on right side, 10 min on left side).

6.4 Preparation and Administration of Bevacizumab

6.4.1 Drug Name, Formulation and Storage

Drug name:

INN: Bevacizumab

Trade name: Avastin®

Manufacturer: Roche Registration Ltd.

Formulation:

Bevacizumab is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The IMP has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

Drug name:

INN: Placebo

Trade name: NA

Manufacturer: Roche Registration Ltd.

Formulation:

Placebo is supplied as a clear to slightly opalescent, colorless to pale brown, sterile liquid for intravenous infusion in single-use vials that are preservative-free. The formulation contains sodium phosphate, trehalose, polysorbate 20, and Sterile Water for Injection (SWFI), USP.

Storage:

The Placebo has to be stored at a temperature between 2°C and 8°C. The adherence to the required storage condition has to be documented in a temperature-log. The vials have to be kept in the outer carton in order to protect them from light.

6.4.2 Packaging and Labeling

Bevacizumab (400 mg, 25 mg/mL) and placebo will be supplied in 20 mL glass vials with a fill of 16 mL. The investigational medicinal product will be labeled according to § 5 GCP-V and internal requirements for blinding purposes.

6.4.3 Preparation of Study Drug

Bevacizumab infusions will be prepared according to the SmPC.

6.4.4 Route of Administration

Bevacizumab will be administered as an intraperitoneal infusion. See also 6.3.

6.4.5 Blinding and Randomization

Randomization will be performed stratified by center using computer-generated lists consisting of permuted blocks of randomly varying size in order to ensure equal group sizes within strata. The randomization lists will be generated by WiSP GmbH and transferred to the facility responsible for blinding, labeling and packaging of the study drugs.

6.4.6 Compliance

A pre-printed drug dispensing log is provided in the Investigator Site File and must be kept current and must identify the patient, and the amount of medication dispensed to each patient at each visit with the corresponding dates.

All medication supplies (empty containers, as well as partly used and unused medication) must be available for inspection at every monitoring visit. All unused medication, partly-used and empty packages must be returned by the investigator to Roche at the end of the study.

7 SAFETY ISSUES

The Investigator's Brochure will be used as reference document for Bevacizumab and will be provided to the investigators in the Investigator's File.

7.1 Adverse Events and Laboratory Abnormalities

It is the responsibility of the investigator(s) to report all adverse events in the case report form. Any serious adverse event (SAE) must be reported to GSO within one working day. GSO will forward the SAEs to the sponsor and Roche within one working day.

7.1.1 Clinical Adverse Events

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. Pre-existing conditions that worsen during a study are to be reported as Adverse Events. They can become Serious Adverse Events if they fulfill one of the seriousness criteria described in section 18.2.

All clinical adverse events (AEs) encountered during the clinical study (treatment period and the 4-week safety follow-up) will be reported on the AE page of the CRF.

Intensity of adverse events will be graded using the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE), version 3.0 (see Investigator's File) and reported in detail as indicated on the CRF. If an adverse event occurs which is not contained in the CTCAE v3.0, the four-point scale below will be used.

Mild:	Discomfort noticed but no disruption of normal daily activity.
Moderate:	Discomfort sufficient to reduce or affect daily activity.
Severe:	Inability to work or perform normal daily activity
Life-threatening:	Represents an immediate threat to life

Relationship of the adverse event to the treatment should also be assessed. Description of scales can be found in section 18.1.

Progression or deterioration of the malignancy under study (including new metastatic lesions and death due to disease progression) will be part of the efficacy assessment and should NOT be reported as AE or SAE.

Signs and symptoms of the malignancy under study should only be reported if:

1. Newly emergent (i.e. not present at baseline) and the association with the underlying malignancy and old/new metastatic lesions is unclear and/or
2. The investigator attributed deterioration of malignancy-associated signs and symptoms directly to the study drug.

Should there be any uncertainty regarding the attribution of the malignancy under study to an AE, it should be reported as an AE or a SAE accordingly.

7.1.2 Laboratory Test Abnormalities

Laboratory test results will be recorded on the laboratory results pages of the Case Report Form, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable. Laboratory-test-value abnormalities as such should not be reported on the AE page of the CRF as adverse events, unless they are treatment-emergent and they satisfy one or more of the following conditions for clinical significance:

1. Accompanied by clinical symptoms.
2. Leading to a change in study medication (e.g. dose modification, interruption or permanent discontinuation).
3. Requiring a change in concomitant therapy (e.g. addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).

Please note: any laboratory result abnormality fulfilling the criteria for a serious adverse event (SAE) should be reported as such, in addition to being recorded as an adverse event in the CRF.

7.1.3 Adverse Events of Special Interest

Adverse events of special interest are any grade of gastrointestinal perforation, gastrointestinal fistulas or other internal fistulas, wound-healing disturbances, hemorrhagic events and arterial thrombo-embolic events. Adverse events of special interest are to be processed like serious adverse events.

7.2 Handling of Safety Parameters

7.2.1 Serious Adverse Events or Adverse Events of Special Interest (Immediately Reportable to GSO)

Any clinical adverse event or abnormal laboratory test value that is serious or of special interest occurring during the course of the study, irrespective of the treatment received by the patient, must be reported to the GSO within one working day of knowledge (expedited reporting). For each patient, all serious adverse events should be reported until 8 weeks since last Bevacizumab infusion. SAEs considered to have a causal relationship to the Investigational Product should be reported regardless of time elapsed since last Bevacizumab dose.

The definition and reporting requirements according to German Drug Law, GCP-V and ICH Guideline for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2 will be adhered (for details refer to section 18.2).

7.2.2 Treatment and Follow-up of Adverse Events

Adverse events, especially those for which the relationship to test drug is not "unrelated", should be followed up until they have returned to baseline status or stabilized. If a clear explanation is established it should be recorded on the CRF.

7.2.3 Follow-up of Abnormal Laboratory Test Values

In the event of unexplained abnormal laboratory test values, the tests should be repeated immediately and followed up until they have returned to the normal range and/or an adequate explanation of the abnormality is found. If a clear explanation is established it should be recorded on the CRF.

7.2.4 Pregnancy

A female patient must be instructed to immediately inform the investigator if she becomes pregnant during the study. The study treatment must immediately be stopped and the patient must be withdrawn from the study. Pregnancies occurring up to 90 days after the completion of the last treatment cycle must also be reported to the investigator. The investigator must report all pregnancies within one working day to the sponsor and the CRO. The investigator should counsel the patient, discuss the risks of continuing the pregnancy, and possible effects on the fetus. Monitoring of the patient should continue until conclusion of the pregnancy.

Pregnancy occurring in the partner of a patient participating in the study should also be reported to the investigator, the sponsor and the CRO. The partner should be counseled and followed as described above.

7.3 Dose Modifications for Toxicity

The observed toxicity will be graded according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events. Dose modifications will be implemented according to the observed toxicity grade as specified below.

7.3.1 General Notes Regarding Dose Modifications

For adverse events which are considered by the Investigator unlikely to develop into serious events and which do not result in a delay or interruption of therapy, treatment will be continued at the same dose without reduction or interruption. If the toxicity is attributable to a certain drug, dose modifications should only be made for this drug. In case of several toxicities occurring simultaneously, the highest dose reduction should be applied.

7.3.2 Dose Modifications for Bevacizumab

No dose reduction of Bevacizumab is foreseen for an individual patient. The dose of 400 mg Bevacizumab was proven to be a safe treatment for intravenous treatment. In addition, all studies applying Bevacizumab as an intraperitoneal infusion have used this dosage.

The initial study drug dose will be delivered over 60 ± 15 minutes. If the first infusion is tolerated without infusion-associated adverse events (fever and/or chills), the second infusion may be delivered over 30 ± 10 minutes.

Bevacizumab-specific toxicities:

Any patient who develops any one of the following toxicities should not further receive Bevacizumab. (for details see below):

- Gastrointestinal perforation
- Fistula Formation involving internal organ
- Arterial thrombo-embolic events
- Symptomatic grade 4 thrombosis
- Grade 3/4 hemorrhagic events
- Grade 4 hypertension (hypertensive crisis)
- Grade 4 proteinuria (nephrotic syndrome)

If the Bevacizumab treatment has to be discontinued permanently, the patient must be withdrawn from the study treatment and followed up for PD and survival only.

Gastrointestinal perforation

Bevacizumab should be permanently discontinued in patients who develop gastrointestinal perforation.

Fistula formation

Bevacizumab should be permanently discontinued in patients who develop fistula involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder).

Thrombosis/Embolism

For patients who develop grade 3 or 4 thrombosis/embolism the following action is recommended:

- Arterial thrombo-embolic events: Bevacizumab should be permanently discontinued.
- Grade 3 or 4 venous thrombosis: Bevacizumab should be permanently discontinued.

Hemorrhage

Patients who develop grade 3 or 4 hemorrhage should permanently discontinue Bevacizumab treatment.

Hypertension

Patients should be monitored for the development or worsening of hypertension via frequent blood pressure measurement. Blood pressure measurements should be taken after the patient has been in a resting position for ≥ 5 minutes. Repeat measurements of blood pressure for verification should be undertaken if the initial reading is ≥ 140 mmHg systolic and/or ≥ 90 mmHg diastolic blood pressure.

- Grade 1 hypertension: Asymptomatic, transient (< 24 h) increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Intervention not indicated.
- Grade 2 hypertension: Recurrent or persistent (> 24 h) or symptomatic increase by > 20 mmHg (diastolic) or to > 150/100 mmHg if previously within normal limits. Monotherapy of anti-hypertensive may be indicated. Once controlled to < 150/100 mmHg, patients may continue Bevacizumab therapy.
- Grade 3 hypertension: Requiring more than one anti-hypertensive or more intensive therapy than previously. Bevacizumab should be withheld for persistent or symptomatic hypertension and should be permanently discontinued if hypertension is not controlled.

- Grade 4 hypertension: Life threatening consequence (e.g. hypertensive crisis). Occurrence of grade 4 hypertension should lead to permanent discontinuation of Bevacizumab. All doses of anti-hypertensive medicines should be recorded at all visits.

Proteinuria

All patients will have a dipstick urinalysis performed within 48 h prior to each Bevacizumab dose. All proteinuria toxicity, as determined by 24 h urine collection, will be graded according to CTCAE v3.0 classification. Adjustment of Bevacizumab administration for proteinuria of ≥ 2 g/24 h will occur according to the following guidelines, listed below.

First occurrence of proteinuria:

- < 2+ (dipstick): Administer Bevacizumab as scheduled; NO additional evaluation is required.
- ≥ 2+ (dipstick): Administer Bevacizumab as scheduled. Collect 24-hour urine to determine the total protein within 3 days prior to the next scheduled dose:
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24 h total protein.
    - Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
    - Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g/24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.

Second and subsequent occurrence of proteinuria:

- < 3+ proteinuria (dipstick): administer Bevacizumab as planned. No additional evaluation is required.
- ≥3+ proteinuria (dipstick): administer Bevacizumab as planned and collect 24-h urine for determination of total protein within 3 days before the next scheduled Bevacizumab administration.
  - ⇒ 24-h proteinuria ≤ 2 g: Administer Bevacizumab as scheduled.
  - ⇒ 24-h proteinuria > 2 g: Bevacizumab treatment should be withheld pending next 24-h total protein.

- Repeat 24-h urine protein ≤ 2 g: Administer Bevacizumab as scheduled. 24-h protein should be further monitored prior to each administration of Bevacizumab until it has decreased to ≤ 1 g/24 h.
- Repeat 24-h urine protein > 2 g: Bevacizumab dose should be withheld until 24-h protein has decreased to ≤ 2 g. 24-h protein should be further monitored prior to each administration of bevacizumab until it has decreased to ≤ 1 g/24 h.

Nephrotic syndrome (grade 4, CTCAE v3.0): Discontinue Bevacizumab treatment.

7.3.3 Dose Modifications for Background Medication

Local standard practice and the recommendations provided in the respective SmPCs will drive dose reduction or interruption of chemotherapeutic compounds of the background medication.

7.4 Criteria for Discontinuation or Termination of the Study

7.4.1 Criteria for Discontinuation of the Treatment or Premature Withdrawal of the Patient

Treatment in both arms of the study will discontinue according to the protocol if any of the following apply:

- if any exclusion criteria develop
- Any patient who develops any one of the following toxicities:
  - Gastrointestinal perforation
  - Fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
  - Arterial thrombo-embolic events
  - Symptomatic grade 4 thrombosis
  - Grade 3/4 hemorrhagic events
  - Grade 4 hypertension (hypertensive crisis)
  - Grade 4 proteinuria (nephrotic syndrome)
- at the patient's request
- at the investigator's discretion
- Physician's judgment following an adverse event
- Termination by the Sponsor, or a regulatory authority

- Any other reason for withdrawal that the study physician or patient indicates is in the overall best interest of the patient

All patients who prematurely discontinue the treatment period will be followed up for safety and survival (exception: patient withdraws consent for further participation or patient is lost to follow-up).

7.5 Treatment after Discontinuation or Termination of the Study

After discontinuation or termination of the study, patients will be treated at the discretion of the investigator and according to medical routine. Further treatment modalities include continuing diuresis, salt restriction and repeated paracentesis. This would also include the possibility of applying the tri-functional antibody catumaxomab or intraperitoneal chemotherapy.

7.6 Warnings and Precautions

In some clinical trials anaphylactic and anaphylactoid-type reactions were reported more frequently in patients receiving Avastin in combination with chemotherapy than with chemotherapy alone. The incidence of these reactions in some clinical trials of Avastin is common (up to 5% in bevacizumab treated patients).
Patients may be at risk of developing infusion / hypersensitivity reaction. Close observation of the patient during and following the administration of bevacizumab is recommended as expected for any infusion of a therapeutic humanized monoclonal antibody. If a reaction occurs, the infusion should be discontinued and appropriate medical therapies should be administered. A systematic premedication is not warranted.

8 BIOSTATISTICAL ASPECTS

8.1 Trial design and hypotheses

The trial is designed as a two-arm parallel group phase II study with a 2:1 randomization. Its primary objective is to obtain evidence, that bevacizumab treatment is effective in the symptom control of patients with malignant ascites. The primary endpoint is defined as paracentesis-free survival (ParFS), i.e. the time period between the initial puncture after randomization to the first subsequent paracentesis or death, whichever occurs first). Thus, the following hypotheses will be tested:

H_0: ParFS (bevacizumab) ≤ ParFS (placebo)

H_1: ParFS (bevacizumab) > ParFS (placebo)

ParFS = paracentesis-free survival

As the bevacizumab treatment may eventually show a somewhat delayed but protracted efficacy, a second primary endpoint is defined as “best response” (BR): the longest period (in days) from one paracentesis to next paracentesis or death or end of the standard 4-week follow-up, whichever occurs first) within in the 12 week observation period.

H_0: BR (bevacizumab) ≤ BR (placebo)

H_1: BR (bevacizumab) > BR (placebo)

However, the sample size calculation is based solely on the ParFS, since the assumptions can be derived from published data. According to these hypotheses, the tests concerning the primary endpoints will be performed one-sided. In accordance with the phase II character of the trial, no type I error adjustment for multiplicity is performed.

8.2 Sample Size Calculation

Based on the results from Parsons et al. [3] the median ParFS in the untreated control group is expected to be around 14 days. In order to detect a prolongation of ParFS by 100% to a median of 28 days by bevacizumab (hazard ratio: 0.5) a total number of 60 evaluable patients is required (40 in the experimental group, 20 in the standard, according to the 2:1 randomization). This calculation is based on the following additional assumptions:

- type I error: 5% (one-sided)
- power: 80%
- observation of all patients until the occurrence of the ParFS event; this assumption will be fulfilled due to the extended follow-up period of up to one year [3]
- exponential shape of the Kaplan-Meier [4] curves

In order to allow for non-evaluable cases or drop-outs, a total of 72 patients will be randomized.

The sample size calculation concerning the analysis of ParFS is based on methods described by Lachin and Foulkes [147]. Since a group sequential design allowing for interim analyses and early discontinuation will be adopted for this trial (see section 8.5), the above fixed sample size calculations serve only as an orientation for the maximum of patient numbers needed. The expected sample size and/or follow-up duration for reaching a conclusion may be considerably less than the number given above. This depends on the number and time points of interim looks as well as the actual difference in efficacy and the actual rates of recruitment and treatment failure. In the case of only one “look” (i.e. no interim analysis before completion of recruitment) the sample size coincides with that of the fixed-sample approach given above.

8.3 Evaluation categories of the patients

8.3.1 Intent-to-Treat Population

Intent-to-treat population (ITT) for ParFS is defined to include all randomized patients with at least one day of follow-up after the initial paracentesis. With respect to BR, this population consists of all patients with at least one documented paracentesis after the initial one or reaching the observation point at 4 weeks after EOT without any second paracentesis.

8.3.2 Per Protocol Population

The per protocol population (PP) is defined to include the intent-to-treat population excluding those patients with major protocol violations. As major protocol violations are considered those that may have an influence to the primary variables (e.g. not obtaining at least one application of study medication according to the protocol and randomization; receiving unauthorized antineoplastic or ascites treatment before observation of a second paracentesis or EOT + 4 weeks). Criteria that are assumed to have such an influence will be defined in a review meeting before data base lock.

8.3.3 Safety Population

According to the definition of the safety population all patients who received at least one dose of the trial medication and a safety follow-up, whether prematurely withdrawn or not, will be included in the safety analysis.

8.4 Methods of Statistical Analysis

The primary endpoints of the trial will be analyzed confirmatively (within the phase II framework) considering a global level for each hypothesis of $p < 0.05$ as significant.

All other parameters will be evaluated in an explorative or descriptive manner, providing means, medians, ranges, standard deviations and/or confidence intervals. If additional p values are calculated (e.g. in subgroup analyses or for secondary endpoints), they will be presented explicitly without referring to hypotheses or a significance level. Usually, no error adjustment for multiple testing will be performed. Thus the p values will reflect the comparison-wise error and not the experiment-wise error. All p values will be two-sided if not stated otherwise. The statistical methods described in this section are suited for the data and distributions usually expected in this type of trials. The suitability will be checked after data entry. If necessary, the statistical method will be modified accordingly and sensitivity analyses performed.

Demographic and prognostic baseline data will be checked for homogeneity between treatment groups. In case of relevant imbalances of other important prognostic factors the statistical method will be adjusted in order to achieve best possible comparability of the groups, and the results will be critically reviewed in comparison to the unadjusted ones.

Primary endpoints: ParFS over one year according to Kaplan-Meier [4], with median ParFS (with confidence intervals) of both arms, the difference of the medians, the hazard ratio with confidence interval and a logrank test comparing both arms. If the Peto logrank test [148, 149] is not appropriate because of violation of the proportional hazard assumption [150], Gehan's generalization of the Wilcoxon rank sum test for censored data [151] will be applied, preferably in its modification by Peto [148] and Prentice [152]. If necessary or prospectively defined at randomization, prognostic strata will be taken into account [149, 153].

Best response will be analyzed providing mean, standard deviation, median, quartiles and range for each arm, using the Wilcoxon rank sum test for statistical comparison.

Secondary endpoints: Time to first subsequent paracentesis as well as best response will be compared to pre-baseline values (mean time interval between paracenteses performed for symptom control within the 4 weeks prior to inclusion into the study) in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt).

Additional response criteria are defined and analyzed as follows: Complete response (CR) will be reached if no additional paracentesis needs to be performed for symptom relief during the 12-week observation period after the first administration of the study medication. Partial response (PR) will be reached if less than 3 additional paracenteses are performed for symptom relief during the 12-week observation period after the first administration of the study medication. The CR/PR/NR (no response) distributions will be compared using an exact version of the Cochran-Armitage test for trend.

Mean values of volumes of ascites removed at paracenteses between first paracentesis within the treatment period and EOT will be calculated and will be compared to volumes of the two most recent paracenteses before inclusion into the study, applying the same statistical test.

Quality of life as assessed by the standardized questionnaires and analyzed according to the recommendations of the respective developer, will be compared to pre-baseline and baseline values in both groups using the Wilcoxon signed rank test for within-group comparisons (Pratt). Body weight assessed throughout the study will be analyzed in a similar way.

In addition, both groups will be compared regarding mean values of volumes and total volume of ascites removed at paracenteses during the treatment period (Wilcoxon rank sum test).

Other analyses to be performed descriptively:

- Overall survival will be analyzed analogous to ParFS
- The proportion of patients with changes in ECOG performance status will be displayed by frequency tables
- Essential laboratory values and/or vital signs will be compared to baseline and displayed by shift tables.
- For the ECOG performance status, the frequency of worsening, unchanged and improved status will be displayed by frequency tables in each scheduled visit.

The methods mentioned above are likewise suitable for the univariate evaluation of prognostic factors. Multivariate analyses may be performed by appropriate regression models (proportional hazard regression model [154], logistic regression).

Further details on the analysis will be given in a separate Statistical Analysis Plan that has to be finalized prior to data base closure. Likewise, the exclusion of patients from the analysis and the definition of a per-protocol population in addition to the ITT population have to be decided upon at this time point.

8.5 Interim and Final Analysis

In case of longitudinal studies in severe chronic diseases, the study design should allow for interim analyses and, consequently, early stopping of the trial for ethical reasons [155]. A group sequential design will be adopted, using the α error spending function methodology by Lan and DeMets [156], implementing a use function according to the O'Brien-Fleming [157] boundary guideline. The design chosen will allow drawing conclusions from interim analyses in the following respect:

- acceptance of superiority of the bevacizumab arm (rejecting H0)

The additional option of accepting the control arm as non-inferior, when an interim result strongly suggests that the anticipated large difference of HR=0.5 will not be detected, is discarded, since a smaller difference might be discussed as relevant, too, especially if supported by secondary findings. Moreover, there is a less stringent need to stop the trial from an ethical point-of-view, if the data tend to similar results in both arms.

In order to keep an overall type I error of 5%, stopping boundaries will be calculated at the respective time points of interim evaluation, using the EaSt software (Cytel Software Corp., Cambridge, USA). This allows for arbitrary interim analyses, irrespective of time schedules and recruitment number. Moreover, the expected sample size and/or study duration for reaching a conclusion may be considerably smaller than in a fixed sample design (cf. 8.2), especially if the therapeutic difference is even larger than expected. The extent of "saving" patients mainly depends on the actual difference in efficacy as well as the actual rates of recruitment and failures. However, subsequent interim analyses will not be performed, unless an increment of at least 10 further evaluable patients are included in the database.

The final biometrical evaluation of the study and the compilation of the statistical report as part of the integrated clinical/biometrical report will be performed after completion and/or correction of all case report forms.

9 DATA QUALITY ASSURANCE

Data will be entered into a database by GSO Hamburg. The data will be filed in digital format and two people will enter the data independently. Data will be checked for accuracy using range, validity and consistency checks, as well as by cross-checking. Implausible or missing data may be corrected or completed after discussing with the investigator. The notes of amendment shall be filed together with the case report form (CRF). The validated data will be stored in a database and this process shall also be documented.

This database conforms to the requirements of ICH-GCP regarding the following:

- Validation of the system and data
- Presentation of SOPs
- Access and back-up systems
- Traceability and documentation of data amendments (audit trail)

Only authorised persons may access the database. Unauthorised access will be prevented via a security system.

10 STUDY COMMITTEES – DATA SAFETY MONITORING BOARD

A Protocol Committee consisting of experienced oncologists will be emplaced and will ensure the development of a clinically appropriate protocol. The Protocol Committee will also organize collaboration with reviewing statisticians and will make suggestions regarding centers to participate in the study.

The Data Safety Monitoring Board (DSMB) will be an independent board consisting of a group of 3 physicians with experience in oncology. A physician is not allowed to participate in this clinical trial while serving on the DSMB. The DSMB will be supported by an independent statistician, if necessary. The DSMB will decide on the feasibility as soon as the first 10 patients will have received the first administration of the study drug and again as soon as a total of 20 patients have received their first intraperitoneal infusion. In addition, the DSMB will evaluate the feasibility as soon as the first 10 and 20 patients, respectively, will have completed the study (EOT). Safety variables (laboratory data, adverse events and serious adverse events) collected in the study will be reviewed every month during the first year. Later the DSMB can extend the regular intervals to e.g. 3 months. The collection and summary of these data will be prepared by the independent statistician. The members of the board will be primarily responsible for the clinical interpretation of the safety results. The board members can recommend a premature discontinuation of the trial or any other changes in the study conduct at any time, if required.

PART II - ETHICS AND GENERAL STUDY ADMINISTRATION

11 ETHICAL ASPECTS

11.1 Declaration of Helsinki/Good Clinical Practice

The Declaration of Helsinki is the accepted basis for clinical study ethics, and must be fully followed and respected by all engaged in research on human beings. Any exceptions must be justified and stated in the protocol. The latest version of the Declaration of Helsinki is available under http://www.wma.net/e/policy/b3.htm.

Additionally it is the responsibility of all engaged in research on human beings to ensure that the study is performed in accordance with the international Good Clinical Practice standards and according to all local laws and regulations concerning clinical studies.

11.2 Patient Information and Informed Consent

It is the responsibility of the investigator to obtain written informed consent from each patient participating in this study, after adequate explanation of aim, importance, anticipated benefits, and potential hazards and consequences of the study according to § 40 Abs. 2 and § 40 Abs. 2a AMG. Written informed consent must be obtained before any study specific procedures are performed. It must be also explained to the patient that they are completely free to refuse to enter the study or to withdraw from it at any time for any reason without incurring any penalty or withholding of treatment on the part of the investigator.

By signing the consent form, the subject/patient agrees with the "unwiderrufliche datenschutzrechtliche Einwilligung" according to § 40 Abs. 2a AMG. The subject/patient also agrees to allow the monitor/auditor/health authorities to verify the collected patient data against the subject's/patient's original medical records for the purpose of source data verification.

The informed consent form personally signed and dated by the patient must be kept on file by the investigator(s), and documented in the CRF and the subject's medical records. The investigator confirms obtaining the written informed consent to the sponsor.

If new safety information results in significant changes in the risk/benefit assessment, the consent form should be reviewed and updated if necessary. All patients (including those already being treated) should be informed of the new information and must give their written informed consent to continue in the study.

If the family doctors are informed of their patients' participation in the clinical study, this should be mentioned in the consent form.

11.3 Independent Ethics Committees and Regulatory Authorities

11.3.1 Approval of the Study by the Federal Regulatory Authority and Independent Ethics Committees

According to §§ 40-42 of the German drug law (AMG) it is the responsibility of the sponsor to obtain and maintain independent approval from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees to conduct the study.

The insurance coverage (study subject insurance) led down in § 40 AMG is in force. For each patient, the sponsor has provided insurance with HDI-Gerling Industrie Versicherung AG, Märkische Straße 23-33, 44141 Dortmund, contract number 48 158388 03055 390.

The sponsor names the "Leiter der klinischen Prüfung" (LKP) who has to be a physician with at least 2 years experience in the conduct of clinical trials of drugs according to § 4 (25) and § 40 (1) No. 5 AMG..

11.3.2 Notification of the Study

According to § 67 German drug law (AMG) the sponsor is responsible to notify competent regional authority about the study and all principal investigators of the participating investigational sites. If no other agreements are made, the sponsor will take over responsibility for investigator's obligation to report (§ 12 (3) GCP-V).

11.3.3 Report and Documentation Obligation

The sponsor is responsible to comply with the report and documentation obligation according to § 13 GCP-V.

The investigator is responsible to comply with the report and documentation obligation according to § 12 GCP-V.

12 CONDITIONS FOR MODIFYING THE PROTOCOL

Protocol modifications to ongoing studies must be made via amendment. The sponsor is responsible to obtain independent approval for the amendment from the federal regulatory authority (BfArM/PEI) and a positive opinion from the competent ethics committees if required according to § 10 GCP-V. According to § 67 AMG competent regional authorities and the federal regulatory authority must be notified about the amendment, if they concern items according to § 12 Abs. 1 GCP-V.

13 DISCONTINUATION OR EARLY TERMINATION OF THE STUDY

13.1 Discontinuation of the Treatment or Premature Withdrawal of the Patient

All study specific withdrawal criteria are described in Section 7.4.1.

In addition, all patients have the right to withdraw from the study at any time for any reason. The investigator also has the right to withdraw patients from the study in the event of inter-current illness, adverse events and treatment failure after a prescribed procedure, protocol violations, cure, administrative reasons or other reasons.

The study has to be terminated when the patient starts a new tumor therapy.

An excessive rate of withdrawals can render the study uninterpretable; therefore, unnecessary withdrawal of patients should be avoided.

Should a patient decide to withdraw, all efforts will be made to complete and report the observations as thoroughly as possible.

The investigator should contact the patient to determine as completely as possible the reason for the withdrawal. A complete final evaluation at the time of the patient's withdrawal should be made with an explanation of why the patient is withdrawing from the study. If the reason for removal of a patient from the study is an adverse event or an abnormal laboratory test result, the principal specific event or test will be recorded on the Case Report Form.

13.2 Discontinuation or Early Termination of the Study

Both the sponsor and the investigator reserve the right to terminate the study at any time. Should this be necessary, both parties will arrange the procedures on an individual study basis after review and consultation. In terminating the study, the sponsor and the investigator will assure that adequate consideration is given to the protection of the patient's interests.

Following criteria could lead to a discontinuation or early termination of the study:

- Safety reason regarding patients' safety
- Negative benefit/risk assessment due to new information

In case of premature termination of the study all collected data have to be analyzed and a report has to be written. The sponsor has to inform the federal regulatory authority and the ethics committees within 15 days, giving detailed reason for the premature termination.

14 STUDY DOCUMENTATION, CRFS AND RECORD-KEEPING

14.1 Investigator's Files / Retention of Documents

The Investigator must maintain adequate and accurate records to enable the conduct of the study to be fully documented and the study data to be subsequently verified. These documents should be classified into two different separate categories Investigator's Study File, and subject/patient data.

The Investigator's Study File will contain all essential documents as the protocol/amendments, Case Report and Query Forms, patient information and informed consent form, Ethics Committee and federal regulatory authority approval, notification of the federal regulatory authority and competent regional authorities, drug records, staff curriculum vitae and authorization forms and other appropriate documents/correspondence etc.

Patient data include patient hospital/clinic records (medical reports, OP reports appointment book, medical records, pathology and laboratory reports, ECG, EEG, X-ray, etc.) and signed informed consent forms and patient screening and eligibility screening forms.

The investigator must keep these two categories of documents on file for at least 15 years (or more as legally required) after completion or discontinuation of the study. The documents must be archived in a secure place and treated as confidential material.

Should the investigator wish to assign the study records to another party or move them to another location, the sponsor must be notified in advance.

If the investigator can not guarantee this archiving requirement at the investigational site for any or all of the documents, special arrangements must be made between the investigator and the sponsor to store these in a sealed container(s) outside of the site so that they can be returned sealed to the Investigator in case of a regulatory audit. Where source documents are required for the continued care of the patient, appropriate copies should be made for storing outside of the site.

The sponsor must archive the protocol, documentation, approvals and all other essential documents related to the study, including certificates that satisfactory audit and inspection procedures have been carried out, as long as the test product(s) remains on the market.

All documents must be archived in a secure place and treated as confidential material.

14.2 Source Documents and Background Data

The investigator shall supply the sponsor on request with any required background data from the study documentation or clinic records. This is particularly important when Case Report Forms are illegible or when errors in data transcription are suspected. In case of special problems and/or governmental queries or requests for audit inspections, it is also necessary to have access to the complete study records, provided that patient confidentiality is protected. According to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy.

14.3 Audits and Inspections

This study may be audited by the sponsor, any person authorized by the sponsor or the competent health authority in order to determine the authenticity of the recorded data and compliance with the study protocol.

The investigator should understand that source documents for this trial should be made available to appropriately qualified personnel from sponsor/monitors/auditor/health authority inspectors after appropriate notification needed for source data verification and proper review of the study progress. The verification of the Case Report Form data must be by direct inspection of source documents. The investigator agrees to comply with the sponsor and regulatory authority requirements regarding the auditing of the study.

All material used in clinical studies are subjected to quality control.

14.4 Case Report Forms

For each patient enrolled, a Case Report Form must be completed and signed by the principal investigator or authorized delegate from the study staff. This also applies to records for those patients who fail to complete the study (even during a screening period if a Case Report Form was initiated). If a patient withdraws from the study, the reason must be noted on the Case Report Form. If a patient is withdrawn from the study because of a treatment-limiting adverse event, thorough efforts should be made to clearly document the outcome.

All forms should be typed or filled out using indelible ink, and must be legible. Errors should be crossed out but not obliterated, the correction inserted, and the change initialed and dated by the investigator or his/her authorized delegate. The investigator should ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the CRFs and in all required reports.

15 MONITORING THE STUDY

The monitor has the responsibility to familiarize the investigator(s) and the entire center staff involved in the study with all study procedures including the administration of study drug.

The GSOmbH must provide a trained monitor to assist the investigator(s) in conducting the clinical study. The monitor must visit the clinical study center on a regular basis and at least before the first patient has been enrolled, once during the course of the study, and at study completion. The monitor has the responsibility of reviewing the ongoing study with the investigator(s) to verify adherence to the protocol and to deal with any problems that arise. At all times the GSOmbH must maintain the confidentiality of the study documents. It is the responsibility of the study monitor to verify the study documents against the patient's original medical records.

The investigator (or his/her deputy) agrees to cooperate with the monitor to ensure that any problems detected in the course of these monitoring visits are resolved.

16 CONFIDENTIALITY OF TRIAL DOCUMENTS AND PATIENT RECORDS

The investigator and the sponsor (or designee) must assure that according to the standards of the data protection law, all data obtained in the course of a clinical study must be treated with discretion in order to guarantee the rights of the patient's privacy. CRFs or other documents should be submitted to the sponsor in a pseudonymous manner. The investigator should keep a patient identification log showing codes and names. The investigator should maintain documents not for submission to Sponsor, e.g., patients' written consent forms, in strict confidence.

17 PUBLICATION OF DATA

This study will be entered into the clinical trial protocol registry and clinical results database.

The sponsor is responsible for the timely reporting of study data. An integrated clinical study report (CSR) has to be completed one year after end of the study (whether completed or prematurely terminated). The report has to be approved by the responsible specialist chosen by the sponsor, the project manager of the CRO, the statistician and the principal investigator/LKP (for multicenter studies) by provision of their signatures.

The results of this study may be published or presented at scientific meetings as soon as after completion of the study. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor prior to submission.

In a multicenter study, it must be ensured that the data from one center are not published before the publication of the whole study. Roche reserves the right to review the manuscript(s) before their submission for publication or presentation. This is not intended to restrict or hinder publication or presentation, but is to allow the sponsor to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the investigator(s).

In accord with standard editorial and ethical practice, Roche will generally support publication of multicenter trials only in their entirety and not as individual center data. In this case, a coordinating investigator will be designated by mutual agreement.

Any formal publication of the study in which input of Roche personnel exceeded that of conventional monitoring will be considered as a joint publication by the investigator and the appropriate Roche personnel. Authorship will be determined by mutual agreement.

18 APPENDICES

Appendix 1: Adverse Events Categories for Determining Relationship to Test Drug

Appendix 2: Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

Appendix 3: ECOG Performance Status

Appendix 4: Quality of Life Questionnaires

18.1 Appendix 1 – Adverse Events Categories for Determining Relationship to Test Drug

(a) Probable (must have first three)

This category applies to those adverse events that are considered, with a high degree of certainty, to be related to the test drug. An adverse event may be considered probable, if:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It cannot be reasonably explained by the known characteristics of the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It disappears or decreases on cessation or reduction in dose. (There are important exceptions when an adverse event does not disappear upon discontinuation of the drug, yet drug-relatedness clearly exists: e. g. (1) bone marrow depression, (2) tardive dyskinesias).
4. It follows a known pattern of response to the suspected drug.
5. It reappears upon rechallenge.

(b) Possible (must have first two)

This category applies to those adverse events in which the connection with the test drug administration appears unlikely but cannot be ruled out with certainty. An adverse event may be considered possible if, or when:

1. It follows a reasonable temporal sequence from administration of the drug.
2. It may have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.
3. It follows a known pattern of response to the suspected drug.

(c) Remote (must have first two)

In general, this category is applicable to an adverse event that meets the following criteria:

1. It does <u>not</u> follow a reasonable temporal sequence from administration of the drug.
2. It may readily have been produced by the subject's clinical state, environmental or toxic factors, or other modes of therapy administered to the subject.

3. It does not follow a known pattern of response to the suspected drug.
4. It does not reappear or worsen when the drug is readministered.

(d) Unrelated

This category is applicable to those adverse events which are judged to be clearly and incontrovertibly due only to extraneous causes (disease, environment, etc.) and do not meet the criteria for drug relationship listed under remote, possible, or probable.

	Probable	**Possible**	**Remote**	**Unrelated**
Clearly due to extraneous causes	–	–	–	+
Reasonable temporal association with drug administration	+	+	–	–
May be produced by subject clinical state, etc.	–	+	+	+
Known response pattern to suspected drug	+	+	–	–
Disappears or decreases on cessation or reduction in dose	+	–	–	–
Reappears on rechallenge	+	–	–	–

18.2 Appendix 2 – Definitions according to AMG and GCP-V, ICH Guidelines for Clinical Safety Data Management, Definitions and Standards for Expedited Reporting, Topic E2

An adverse event is any untoward medical occurrence in a patient or clinical investigation subject, administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

Adverse reactions are all untoward and unintended responses to an investigational medicinal product related to any dose administered.

A serious adverse event or serious adverse reaction is any experience that suggests a significant hazard, contraindication, side effect or precaution. It is any Adverse Event that at any dose fulfils at least one of the following criteria:

- is fatal (results in death) *(NOTE: death is an outcome, not an event)*
- is life-threatening *(NOTE: the term "life-threatening" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not refer to an event which could hypothetically have caused a death had it been more severe.)*
- required in-patient hospitalization or prolongation of existing hospitalization
- results in persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- is medically significant or requires intervention to prevent one or other of the outcomes listed above

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the outcomes listed in the definitions above. These situations should also usually be considered serious.

Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

An unexpected Adverse Event is one, the nature or severity of which is not consistent with the applicable product information.

Causality is initially assessed by the investigator. With respect to report and documentation obligation (regulatory authorities, ethics committees and other investigators) for Serious Adverse Events, causality can be one of 2 possibilities:

- No (unrelated; equals not drug related).
- Yes (remotely, possibly, probably or definitely drug related).

All adverse events not assessed as definitive "not drug related" by either the investigator or Roche will be considered as adverse drug reaction.

A suspected unexpected serious adverse reaction (SUSAR) is a serious adverse reaction, the nature, or severity of which is not consistent with the applicable product information.

It is important that the severity of an adverse event is not confounded with the seriousness of the event. For example, vomiting which persists for many hours may be severe, but is not necessarily a serious adverse event. On the other hand, stroke which results in only a limited degree of disability may be considered a mild stroke, but would be a serious adverse event.

A serious adverse event occurring during the study or which comes to the attention of the investigator within 8 weeks after stopping the treatment or during the protocol-defined follow-up period, if this is longer, whether considered treatment-related or not, must be reported. In addition, a serious adverse event that occurs after this time, if considered related to test "drug", should be reported.

Such preliminary reports will be followed by detailed descriptions later that will include copies of hospital case reports, autopsy reports and other documents.

For serious adverse events, the following must be assessed and recorded on the adverse events page of the Case Report Form: intensity, relationship to test substance, action taken, and outcome to date.

Document and report obligation have to be adhered according to the national and international laws and regulations.

Contact details and Fax No. for SAE and pregnancy reporting refer to page 17.

18.3 Appendix 3 – ECOG Performance Status

GRADE	SCALE
0	Fully active, able to carry out all pre-disease performance without restriction. (Karnofsky 100%)
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light housework, office work. (Karnofsky 80-90%)
2	Ambulatory and capable of all self-care, but unable to carry out work activities. Up and about more than 50% of waking hours (Karnofsky 60-70%)
3	Capable of only limited self-care, confined to bed or chair more than 50% of waking hours. (Karnofsky 40-50%)
4	Completely disabled. Cannot carry out any self-care. Totally confined to bed or chair. (Karnofsky 10-30%)
5	Dead (Karnofsky 0%)

18.4 Appendix 4 – Quality of Life Questionnaires

FACIT ASCITES INDEX (Patientenfragebogen)

Nachfolgend finden Sie eine Liste von Aussagen, die von anderen Personen mit Ihrer Krankheit für wichtig befunden wurden. **Bitte geben Sie jeweils an, wie sehr jede der folgenden Aussagen im Laufe der letzten 7 Tage auf Sie zugetroffen hat, indem Sie die entsprechende Zahl ankreuzen.**

		Überhaupt nicht	Ein wenig	Mäßig	Ziemlich	Sehr
C6	Ich habe einen guten Appetit	0	1	2	3	4
GF5	Ich schlafe gut	0	1	2	3	4
BMT5	Ich bin in der Lage, mich alleine fortzubewegen	0	1	2	3	4
B1	Ich leide unter Atemnot	0	1	2	3	4
GP2	Mir ist übel	0	1	2	3	4
O2	Ich habe mich übergeben	0	1	2	3	4
ACT11	Ich habe Schmerzen in der Magengegend	0	1	2	3	4
O1	Ich habe Schwellungen im Magenbereich	0	1	2	3	4
GP1	Mir fehlt es an Energie	0	1	2	3	4
ACT10	Wenn ich esse, fühle ich mich rasch satt	0	1	2	3	4
BL2	Ich muss häufiger Wasserlassen als üblich	0	1	2	3	4
Cx6	Ich leide an Verstopfung	0	1	2	3	4
AI1	Ich bin bekümmert	0	1	2	3	4

Patientenlebensqualitätsfragebogen

(modifiziert nach der Palliativgruppe der DGHO)

Der Fragebogen ist durch das medizinische Personal zu ergänzen

Betreuungsverlauf

Datum des ersten Besuchs ☐☐☐☐☐☐

Ort des ersten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Datum des letzten Besuchs ☐☐☐☐☐☐

Ort des letzten Besuchs
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Krankenhaus
- ● sonstiges

Begründung des Abschlusses der Home Care-Versorgung ★
- ● Tod des Patienten
- ● Änderung des Wohnorts des Patienten
- ● unerwartete Verbesserung des Gesundheitszustands des Pat.
- ● Fortsetzung der tumorspezifischen Therapie
- ● Krankenhauseinweisung
- ● Wechsel des HC-Arztes
- ● sonstiges:_____

Sterbedatum ☐☐☐☐☐☐

Sterbeort
- ● (eigene) Wohnung
- ● Senioren-/Pflegeheim
- ● stationäres Hospiz
- ● Kurzzeitpflege
- ● Palliativstation
- ● andere Krkhs.-Station
- ● sonstiges

Ausprägung von Symptomen zum Zeitpunkt der Aufnahme und nach erfolgter Einstellung

alles Zutreffende nach Schweregrad ausfüllen (Schweregrad: 0-ohne, 1-gering, 2-mittel, 3-stark) ■ vor Intervention ☐ nach Intervention

■	Schmerzen ☐	■	Dysphagie ☐	■	(Lymph-)Ödem ☐	■	Krampfanfälle ☐
■	Schwäche ☐	■	Obstipation ☐	■	Juckreiz ☐	■	motor. Unruhe ☐
■	Appetitlosigkeit ☐	■	Diarrhoe ☐	■	Dekubitus ☐	■	Verwirrtheit ☐
■	Übelkeit ☐	■	Ascites ☐	■	exulc. Wunde ☐	■	Schlafstörung ☐
■	Erbrechen ☐	■	Dyspnoe ☐	■	Harnverhalt ☐	■	Angst ☐
■	(Tumor-)Blutung ☐	■	Husten ☐	■	Lähmungen ☐	■	Depression ☐

Therapie bei Aufnahme und im Verlauf / zum Ende

Schmerztherapie WHO
- ● keine ○
- ● nur bei Bedarf ○
- ● WHO Stufe I ○
- ● WHO Stufe II ○
- ● WHO Stufe III ○

Koanalgetika / Begleitmedikation ★
- ● Nichtopioide Analget. ○
- ● Antiemetika ○
- ● Antidepressiva ○
- ● Antikonvulsiva ○
- ● Kortikosteroide ○
- ● Laxanzien ○
- ● Neuroleptika ○
- ● Sedativa / Hypnotika ○

Opioidtherapie ★

Substanz(en) ★
- ● Morphin ○
- ● Hydromorphon ○
- ● Fentanyl ○
- ● Oxycodon ○
- ● Buprenorphin ○
- ● Levomethadon ○
- ● Piritramid ○

Applikationsform(en) ★
- ● oral / PEG / rektal ○
- ● transdermal ○
- ● s.c. Injektion ○
- ● s.c. Dauerinfusion ○
- ● i.v. Dauerinfusion ○
- ● peridural ○

sonstige Palliativmaßnahmen ★
- ● PEG / transnasale Sonde ○
- ● zentraler venöser Zugang ○
- ● enterale Ernährung ○
- ● parenterale Ernährung ○
- ● i.v. Flüssigkeitssubstitution ○
- ● s.c. Flüssigkeitssubstitution ○
- ● Ascitespunktion(en) ○
- ● Pleurapunktion(en) ○
- ● palliative Chirurgie ○
- ● palliative Radiatio ○
- ● palliative Chemotherapie ○
- ● palliative endoskop. Eingriffe ○
- ● nichtspezialisierter Pflegedienst ○
- ● Palliativpflegedienst ○

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AIO-SUP-0108

Double-blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

EudraCT-No: 2009-014725-16

Administrative Change to Protocol Version 4.0 dated 18.04.2012

Reason for Administrative Change:

Change of Sponsor's address

The protocol of the study is to be changed as follows:

Section affected: **Page 1**

Previous text:
AIO-Studien-gGmbH
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New text:
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Section affected: **Page 2**

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Signed Agreement to the Administrative Change

I hereby approve the Administrative Change to the protocol.

Dr. Aysun Karatas
Sponsor

17.10.12
Date

16.1.2 Sample CRF

AIO-Trial-SUP-0108

Double blind, placebo-controlled, randomized phase II-study investigating the efficacy of Bevacizumab for symptom control in patients with malignant ascites due to advanced-stage gastrointestinal cancers

Case Report Form

Patient randomisation number: |_|_|_|_|

Date of birth: |_|_| |_|_| 1 9 |_|_|
Day Month Year

Centre: _____

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FLOW CHART: Scheduled assessments during the Study

Notes:

1. The screening visit S1 will take place within the screening period and not earlier than 7days before inclusion of the patient into the study and application of the first paracentesis for study purposes. No treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.

2. The treatment period starts with the first paracentesis applied after the screening visit S1 but not later than 7 days after that visit.

3. All assessments have to be performed before administration of the study drug.

4. The first visit of the survival follow-up period will take place two months after the last infusion of the study drug. The last visit will take place as soon as the patient has completed 1 year after EOT.

5. Prior to the first study-specific measures.

6. Applicable for women of childbearing potential. Serum ß-HCG test within 7 days before the first dose of study drug. To be repeated by urine test, if date of first result exceeds the 7-day window.

7. Baseline frequency of paracenteses clinically required will be assessed by calculating the mean time frame (in days) between paracenteses which have been performed for symptom relief (and not only for diagnosis purposes) within the past 4 weeks before inclusion into the study (screening period). Thereafter, the frequency of paracenteses required will be individually calculated for both groups as number of days between each pair of two subsequent paracenteses from start of the treatment period with the first infusion of the study drug until safety follow-up.

8. Baseline volumes of ascites will be assessed by calculating the mean and total volumes of ascites for paracenteses that have been performed for symptom relief within the past 4 weeks prior to inclusion into the study. Thereafter, volumes of ascites removed will be monitored during the treatment period.

9. Baseline measurements not more than 3 days before Day 1 of the first treatment cycle (start of therapy).

10. Vital signs: Blood pressure, heart rate, body temperature. Body height will be measured at screening only.

11. Measurements will be performed at the screening visit and on each visit for routine paracentesis. Measurements will be performed at biweekly intervals form first pracentesis until EOT. A final measurement will be performed at safety follow-up.

12. 10 ml of heparinized blood (plasma) and 10 ml of ascites fluid for investigational analyses and for phamacokinetics of Bevacizumab (10 ml Serum) will be obtained before each routine paracentesis with study medication performed during the treatment period and, if possible in the case of malignant ascites, at safety follow-up. Sera will also be collected at 14-day intervals from first paracentesis until EOT and a final sample will be collected at safety follow-up.

13. Urinalysis: Dipstick test for protein only. In case of protein > 1+ with dipstick: Quantitative determination in 24 h urine is required.

14. Hematology: Leukocytes, platelets, hemoglobin, neutrophils.

15. Clinical Chemistry: Alkaline phosphatase, AST, ALT, LDH, total bilirubin, creatinine, creatinine clearance (Cockroft-Gault formula), total protein.

16. Analysis of differential cell count (hemoglobin, hematocrit, total leukocytes, neutrophils) form 2-5 ml EDTA-anticoagulated ascites and chemistry (total protein, albumin) from 5 ml heparinized ascites.

17. Study drugs (Bevacizumab, arm A; placebo, arm B) will be administered to patients after placement of an intraperitoneal catheter and removal of the maximum volume of ascites for symptom control. Patients will receive a maximum number of four cycles of intraperitoneal Bevacizumab/placebo. Maximum duration of the treament period is 8 weeks. Minimum interval between applications of the study drug during the treatment period will be 14 days.

18. After the last cycle of treatment period only anti-tumor drugs administered should be documented.

19. EOT is set at 8 weeks after first application of the study drug for both arms of the study.

20. When paracentesis is clinically indicated.

	Screening [1,3]		Baseline [3,9]	Treatment Period [3]				Safety FU	Survival FU [4]
Treatment Number [2]				**1**	**Variable (maximum number: 4 applications)**			**EOT [19] + 4 weeks**	**Every 2 months**
Study week	-4 to 0	-7 d. to 0	-3 d. to 0	1	total duration: 8 weeks				
Informed Consent [5]	X								
In- / Exclusion criteria	X								
Demographics	X								
Medical History	X								
Cancer and Treatment history	X								
Pregnancy Test (if applicable) [6]		X							
Frequency of paracenteses required [7]	X			X	X	X	X	X	
Volume of ascites drained [8]	X			X	X	X	X		
ECOG Performance Status [11]			X [9]	X	X	X	X	X	
Physical Examination [11]			X [9]	X	X	X	X	X	
Body weight [11]	X			X	X	X	X	X	
Quality of life assessment [11]		X		X	X	X	X	X	
Vital Signs [10,11]			X [9]	X	X	X	X	X	
12-lead ECG			X [9]	As clinically indicated					
Investigational analysis of plasma [12]				X	X	X	X	X[20]	
Investigational analysis of ascites [12]				X	When paracentesis is clinically indicated			X	
Urinalysis [11,13]		X		X	X	X	X	X	
Hematology [11,14]		X		X	X	X	X	X	
Clinical Chemistry [11,15]		X		X	X	X	X	X	
aPTT, INR [11]		X		X	X	X	X	X	
Routine analysis of ascites [16]	X			X	As clinically indicated			X[20]	
Paracentesis for symptom control	As indicated			X	As clinically indicated			X[20]	
Study drug infusion [17]				X	Depending on paracentesis frequency				
Adverse Events				Continuously				X	
Concomitant Diseases	X			Continuously				X	
Concomitant Treatment	X			Continuously				X	X [18]
Survival				Continuously				X	X

AIO-SUP-0108

General instructions for the Investigator

- **Please use a ballpoint , fine-tip pen (permanent ink) in black or blue, to make sure that all copies are clear and legible. The use of a pencil is not allowed.**
- **Please fill in boxes with only one letter or digit.**
- **All fields must be completed. Was a specific test not performed please note this as following**
  - **ND (=not done)**
  - **NA (=not available)**
  - **UK (=unknown).**
- **The field are of a maximum length. Please fill in the field completely. If the entry is a number consisting of only one digit, please fill in all prior boxes with zeros.**
  Example: 3 digit field, Entry of number 5

  Right: 0 0 5 Wrong: 5
- **Note all dates in the following manner:**
  Example for a date with 2 digit date (05.10.09)

  0 5 | 1 0 | 0 9
  Day Month Year

  Please note that day and month data, if they were one digit data, filled in with a prior zero, at any one time.

  If a date can not accurate defined, please fill in the unknown day (and possibly the unknown month) with „-“.

  Example:

  - - | 1 0 | 0 9
  Day Month Year
- **Necessary corrective actions must be made by the investigator or a authorised person.**
  The corrections may basically performed according to GCP/ICH-Guidelines. That means:
  - **The corrective entries shall be crossed out in the way that the entries still are readable.**
  - **The valid version shall be written readable above or beside the wrong entry.**
  - **The correction (or other entries) must be signed with date, initials and possibly with the reason for the correction by the person who makes the modifications.**

  Example:

  0 0 5 1 with date, initials and possibly reason Ø 0 5
- **Please writte all entries in block letters.**

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Randomisation No. ____

Screening
(within 4 weeks before 1st application of study drug)

1

Written Informed Consent

Important: Signed and dated informed consent has to be obtained before the start of first protocol specific procedures.

Date of written informed consent ____ ____ ____ (Day Month Year)

Demographics

Date of birth ____ ____ 1 9 ____ (Day Month Year)

Gender ☐ male ☐ female

Cancer History

Cancer type:
- ☐ esophageal carcinoma
- ☐ gastric carcinoma
- ☐ pancreatic carcinoma
- ☐ cholangiocellular carcinoma
- ☐ hepatocellular carcinoma
- ☐ colorectal carcinoma

Histology:
- ☐ adenocarcinoma
- ☐ squamous cell carcinoma
- ☐ other
- ☐ unknown

TNM Staging at Diagnosis: T ☐ N ☐ M ☐

Stage at screening (if applicable): ☐ I ☐ II ☐ III ☐ IV

Date of first diagnosis ____ ____ (Month Year)

Date of metastatic or locally advanced disease (if applicable): ____ ____ (Month Year)

Previous anti-cancer treatment

Did the patient receive any anti-tumor therapy prior to screening?

☐ No ☐ Yes, please specify

Please only document previous anti-cancer treatments. Please document any ongoing anti-cancer treatment on concomitant medication page.

Regime	Start date	Stop date	Reason 1= adjuvant 2 = neo-adjuvant 3= palliative-1st line 4= palliative - 2nd line 5= palliative - > 3rd line
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	
	Day Month Year	Day Month Year	

Relevant Medical History / Concomitant Diseases other than gastrointestinal cancer

Does the patient have any relevant medical history / concomitant illness other than gastrointestional cancer?

☐ No ☐ Yes, please specify

Description	Start date	Stop date	Ongoing
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐
	Month Year	Month Year	☐

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure within screening phase?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“

AIO-SUP-0108 Randomisation No.	Screening (within 7 days before 1st application of study drug)	4

Pregnancy test

Serum beta-HCG Date of test Day Month Year

☐ positive ☐ negative ☐ NA (male, woman without childbearing potential)

Urine beta-HCG Date of test Day Month Year

☐ positive ☐ negative ☐ NA (male, woman without childbearing potential), or in case Serum beta-HCG was performed within 7 days prior to administration of study drug.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason: ☐ Patient's wish ☐ Organisatory reason ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Hematology

Sampling date Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl		☐	
Platelets		/nl		☐	
White Blood count		/nl		☐	
Neutrophils		/nl		☐	

* Evaluation: **0 = normal**
1 = abnormal, not clinically not relevant
2 = abnormal and clinically relevant → Please be aware to follow inclusion/exclusion criteria!!

Clinical chemistry

Sampling date ___ ___ ___ (Day Month Year)

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
Albumin		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation: **0 = normal**
1 = abnormal, not clinically not relevant
2 = abnormal and clinically relevant → Please be aware to follow inclusion/exclusion criteria!!

Urinalysis

Date of analysis ___ ___ ___ (Day Month Year)

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse events - additional assessments“.

1st and 2nd Paracentesis performed for symptom relief

According to study protocol at least 2 routine paracenteses for symptom control of malignant ascites must have taken place at the end of the screening phase of up to 4 weeks.

Please document details for each paracentesis.

1st paracenteses performed for symptom relief

Date ___ ___ ___ (Day Month Year)

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) ____ ml

Body weight *(before paracentesis)* ___,___ kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

2nd paracenteses performed for symptom relief

Date ___ ___ ___ (Day Month Year)

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) ____ ml

Body weight *(before paracentesis)* ___,___ kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

3rd and 4th Paracentesis performed for symptom relief

According to study protocol at least 2 routine paracenteses for symptom control of malignant ascites must have taken place at the end of the screening phase of up to 4 weeks.

Please document details for each paracentesis.

In case that a 3rd or 4th paracentesis for symptom relief was not performed please tick „not applicable“.

3rd paracenteses performed for symptom relief

☐ NA Date |_|_| Day |_|_| Month |_|_|_|_| Year

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) |_|_|_|_| ml

Body weight *(before paracentesis)* |_|_|_|,|_| kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

4th paracenteses performed for symptom relief

☐ NA Date |_|_| Day |_|_| Month |_|_|_|_| Year

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) |_|_|_|_| ml

Body weight *(before paracentesis)* |_|_|_|,|_| kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

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Randomisation No. ⌊_____⌋

Screening
(within 4 weeks before 1st application of study drug)
Additional Paracentesis Log

7.⌊_⌋

Additional paracentesis performed for symptom relief

☐ NA Date ⌊_____⌋ Day Month Year

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) ⌊_____⌋ ml

Body weight *(before paracentesis)* ⌊_____⌋,⌊_⌋ kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

Additional paracentesis performed for symptom relief

☐ NA Date ⌊_____⌋ Day Month Year

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) ⌊_____⌋ ml

Body weight *(before paracentesis)* ⌊_____⌋,⌊_⌋ kg

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites): ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites) ☐ Not done

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐ ↓ EXCLUSION!

Physical Examination

Date: Day Month Year

Were physical examinations performed? ☐ No ☐ Yes, please specify

Physical examination	Normal / abnormal	if abnormal, clinically relevant?
General appearance	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Cardiovascular	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Respiratory	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Liver, Hepatic	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Other: _____	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Other: _____	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes
Other: _____	☐ normal ☐ abnormal, specify _____	☐ No ☐ Yes

Inclusion Criteria

1. Age > 18 years
2. Written informed consent has been obtained prior to inclusion into the study
3. Patient is capable and willing to comply with the study
4. Histologically confirmed esophageal, gastric, pancreatic, cholangiocellular, hepatocellular or colorectal carcinoma
5. Cytologically confirmed ascites OR diagnosis of an exsudate (total protein in ascites >30g/l) clinically suggestive for malignant ascites OR morphological diagnosis of peritoneal carcinosis by CT, MRT or ultrasound
6. Ascites clinically judged as not responsive to conventional systemic therapies for primary malignancy
7. Ascites clinically judged as not responsive to diuretics
8. At the time of inclusion paracentesis required at least twice within past 4 weeks.
9. Before inclusion of the patient into the study, a 4-week screening period will allow for a stringent evaluation of the patient regarding fulfillment of inclusion and exclusion criteria. Importantly, no treatments for malignant ascites other than paracentesis and diuretics are allowed during the 4-week screening period.
10. ECOG peformance score 0-3
11. Life expectancy > 12 weeks
12. Laboratory parameters:
    Hematology
    - Neutrophils > 1,500/µl
    - Platelets > 100,000/µl
    - Hemoglobin $\geq$ 9 g/dl or 5.59 mmol/l
    Hemastasiology
    - INR $\leq$ 1.5 x ULN and aPTT $\leq$ 1.5 x ULN within past 7 d
    Clinical chemistry
    - Creatinine clearance > 30 ml/min, serum creatinine < 2.5 x ULN
    - Serum Bilirubin < 3.0 x ULN
    - Alkaline phosphatase and transaminases < 3.0 x ULN (in case of liver metastases < 7x ULN)
    Urinalysis
    - Patients with < 2+ proteinuria on dipstick urinalysis.
    - Patients with $\geq$ 2+ proteinuria on dipstick urinalysis, who demonstrate < 2.0 g of protein/24 h on 24-h urine collection.

Exclusion Criteria

Patients with any of the following will not be eligible for participation:

1. Concomitant malignancies other than gastrointesinal cancers (Patients with curatively treated basal and squamous cell carcinoma of the skin and / or in-situ carcinoma of the cervix are eligible).
2. Bacterial peritonitis as idicated by laboratory results (neutrophil count > 250/µl ascites) or clinical suspicion
3. Hemorrhagic ascites (ascites hemtocrit > 2%)
4. Transudative ascites (total protein in ascites < 30 g/l)
5. Parallel treatment with anti-tumor agents other than the study medication from inclusion into the study until safety follow-up. Chemotherapy may be continued if started before screening phase (-4 weeks before inclusion). Parallel Treatment with Bevacizumab i.v. is not allowed.
6. Therapie naive patients
7. Parallel treatment of ascites with measures other than paracentesis, diuretics, and the study drugs from 4 weeks before inclusion into the study until safety follow-up.
8. Patients with extensive metastases of the liver making up > 70% of the total liver mass
9. Child C cirrhosis of the liver
10. Occlusion or thrombosis of the portal vein.
11. Evidence of current and symptomatic central nervous system (CNS) metastases or spinal cord compression.
12. Clinically significant cardiovascular diseases, e.g., uncontrolled hypertension, uncontrolled arrhythmia, hemoptoe, cardiocascular accident within the last 6 months before treatment start, unstable angina, congestive heart failure (CHF) NYHA grade III /IV, symptomatic coronary heart disease, peripheral arterial disease stage > II.
13. History of fistula formation involving an internal organ (e.g. trachea-oesophageal, bronchopleural, biliary, vagina and bladder)
14. Major surgical procedure, open biopsy, or significant traumatic injury within 28 days prior to study treatment start, or anticipation of the need for major surgical procedure during the course of the study.
15. Concomitant treatment with intravenous Bevacizumab for primary malignancy from inclusion into study until safety follow-up. Prior treatment with Bevacizumab is not exclusionary.
16. Serious non-healing wound, ulcer or bone fracture
17. Radiotherapy for purposes other than local control of symptoms
18. Evidence of bleeding diathesis or coagulopathy
19. Hematopoietic diseases
20. Known intra-abdominal inflammatory process or serious gastrointestinal ulceration
21. History of chronic intestinal diseases associated with severe diarrhea.
22. Thrombo-embolic events or severe hemorrhage (< 6 months before treatment start)
23. Known hypersensitivity to the test drug Bevacizumab
24. Evidence of any other disease, metabolic dysfunction, physical examination finding, or laboratory finding giving reasonable suspicion of a disease or condition that contraindicates the use of an investigational drug or puts the patient at high risk for treatment-related complications.
25. With the only exception of full dose (INR > 1.5) oral coumarin-derived anticoagulants, the use of full dose anticoagulants is allowed as long as the INR or a PTT is within therapeutic limits (according to the medical standard in the institution) and the patient has been on a stable dose for at least two weeks at the time of randomisation.
26. Patients who participated within the last 30 days prior to enrolment in a clinical trial and received a non approved investigational drug (e.g. follow up within the trial is not exclusionary).
27. Patients who have participated in this study before.
28. Women, lactating, pregnant or of childbearing potentioal and fertile men not using a highly effective contraceptive method . [Women of childbearing potenial must have a negative pregnancy test (serum ß-HCG) within 7 days before the first dose of study drug].
29. Patients who are committed to an institution by virtue of an order issued either by the judicial or the administrative authorities (according to § 40 (1) 4 AMG).
30. Patients who are underage or patients who are incapable to understand the aim, importance and consequences of the study and to give legal informed consent (according to § 40 (4) and § 41 (2) and (3) AMG).
31. Patients with a history of a psychological illness or condition such as to interfere with the patient's ability to understand the requirements of the study.
32. Patients who possibly are dependent on the sponsor or investigator.

AIO-SUP-0108

Randomisation No. |_|_|_|_|

Baseline
(within 3 days before 1st application of study drug)

9

Vital Signs

Date: |_|_| Day |_|_| Month |_|_|_|_| Year

Blood pressure |_|_|_| / |_|_|_| mmHg (syst. / diast.)

Heart rate |_|_|_| beats/min

Body temperature |_|_|,|_| °C

Body height |_|_|_| cm

12-lead ECG

Date of record: |_|_| Day |_|_| Month |_|_|_|_| Year

- ☐ normal
- ☐ abnormal, clinically not relevant
- ☐ abnormal, clinically relevant, please specify: _____

Eligibility and Randomisation

Does the patient fullfill all inclusion / exclusion criteria?

- ☐ No, please describe violation:

  _____

  _____

  _____

- ☐ Yes, please specify date of randomisation

  Date: |_|_| Day |_|_| Month |_|_|_|_| Year

Comments

none ☐

_____

_____

_____

_____

Herewith I confirm completeness and correctness of documentation on CRF page 1 to 9.

_____
Stamp / Signature Investigator

|_|_| Day |_|_| Month |_|_|_|_| Year
Date

All assessments have to be performed before administration of study drug!

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: Day Month Year

Was there any clinical significant worsening from baseline?

☐ No ☐ Yes, please document on CRF-form „Adverse Events“.

Vital Signs

Date: Day Month Year

Blood pressure ____ / ____ mmHg (syst. / diast.)

Heart rate ____ beats/min

Body temperature ____,__ °C

Body weight ____,__ kg

Hematology

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl			
Platelets		/nl			
White Blood count		/nl			
Neutrophils		/nl			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Clinical chemistry

Sampling date Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis Day Month Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse Events - additional assessments“.

Paracentesis

Date of first paracentesis performed in treatment period for symptom relief: Day Month Year

Please complete CRF-form „Paracentesis Log“.

AIO-SUP-0108 Randomisation No.	Week 0 (1st paracentesis in treatment period)	12

Adverse Events

Did the patient experience any adverse event since 1st application of study drug?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since start of study treatment?

☐ No ☐ Yes, please specify on CRF-form „Concomitant medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason:
- ☐ Patient‘s wish
- ☐ Organisatory reason
- ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a plasma sample for investigational analysis collected?
- ☐ No, please give reason _____
- ☐ Yes, Date ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Was a serum sample for pharmacokinetics collected?
- ☐ NA
- ☐ No, please give reason _____
- ☐ Yes, Date ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Comments

none ☐

_____

_____

_____

All assessments have to be performed before administration of study drug!

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: Day Month Year

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Events“.

Vital Signs

Date: Day Month Year

Blood pressure ____ / ____ mmHg (syst. / diast.)

Heart rate ____ beats/min

Body temperature ____,__ °C

Body weight ____,__ kg

Hematology

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl		☐	
Platelets		/nl		☐	
White Blood count		/nl		☐	
Neutrophils		/nl		☐	

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Clinical chemistry

Sampling date Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation: **0 = normal**
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis Day Month Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse events - additional assessments“.

Paracentesis

Were there any paracenteses performed for symptom relief since last vist?

☐ No

☐ Yes, please complete CRF-form „Paracentesis Log“.

Adverse Events

Did the patient experience any adverse event since last visit?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since last visit?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason:
- ☐ Patient‘s wish
- ☐ Organisatory reason
- ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a <u>plasma</u> sample for investigational analysis collected?
- ☐ No, please give reason _____
- ☐ Yes, Date ___ ___ ___ (Day Month Year)

Was a <u>serum</u> sample for pharmacokinetics collected?
- ☐ NA
- ☐ No, please give reason _____
- ☐ Yes, Date ___ ___ ___ (Day Month Year)

All assessments have to be performed before administration of study drug!

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: Day Month Year

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Event“.

Vital Signs

Date: Day Month Year

Blood pressure ____ / ____ mmHg (syst. / diast.)

Heart rate ____ beats/min

Body temperature ____ , ____ °C

Body weight ____ , ____ kg

Hematology

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl		☐	
Platelets		/nl		☐	
White Blood count		/nl		☐	
Neutrophils		/nl		☐	

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Clinical chemistry

Sampling date Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation: **0 = normal**
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis Day Month Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse events - additional assessments“.

Paracentesis

Were there any paracenteses performed for symptom relief since last visit?

☐ No

☐ Yes, please complete CRF-form „Paracentesis Log“.

Adverse Events

Did the patient experience any adverse event since last visit?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since last visit?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason: ☐ Patient‘s wish ☐ Organisatory reason ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a plasma sample for investigational analysis collected? ☐ No, please give reason

_____

☐ Yes, Date |__|__|__|__|__|__|__|__| Day Month Year

Was a serum sample for pharmacokinetics collected? ☐ NA

☐ No, please give reason

_____

☐ Yes, Date |__|__|__|__|__|__|__|__| Day Month Year

All assessments have to be performed before administration of study drug!

ECOG Performance status

Date: ____ (Day) ____ (Month) ____ (Year)

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: ____ (Day) ____ (Month) ____ (Year)

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Event“.

Vital Signs

Date: ____ (Day) ____ (Month) ____ (Year)

Blood pressure ____ (syst.) / ____ (diast.) mmHg

Heart rate ____ beats/min

Body temperature ____,____ °C

Body weight ____,____ kg

Hematology

Sampling date: ____ (Day) ____ (Month) ____ (Year)

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl		☐	
Platelets		/nl		☐	
White Blood count		/nl		☐	
Neutrophils		/nl		☐	

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Clinical chemistry

Sampling date ____ Day ____ Month ____ Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl		☐	
Total bilirubin		mg/dl		☐	
Total protein		g/dl		☐	
ASAT / GOT		U/l		☐	
ALAT / GPT		U/l		☐	
LDH		U/l		☐	
Alkaline phosphatase		U/l		☐	
Creatinine Clearance		ml/min		☐	
INR				☐	
aPTT		sec		☐	

* Evaluation: **0 = normal**
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis ____ Day ____ Month ____ Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse events - additional assessments“.

Paracentesis

Were there any paracenteses performed for symptom relief since last visit?

☐ No

☐ Yes, please complete CRF-form „Paracentesis Log“.

Adverse Events

Did the patient experience any adverse event since last visit?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since last visit?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason:
- ☐ Patient‘s wish
- ☐ Organisatory reason
- ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a <u>plasma</u> sample for investigational analysis collected?
- ☐ No, please give reason _____
- ☐ Yes, Date |__|__| Day |__|__| Month |__|__|__|__| Year

Was a <u>serum</u> sample for pharmacokinetics collected?
- ☐ NA
- ☐ No, please give reason _____
- ☐ Yes, Date |__|__| Day |__|__| Month |__|__|__|__| Year

All assessments have to be performed before administration of study drug!

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: Day Month Year

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Event“.

Vital Signs

Date: Day Month Year

Blood pressure ___ (syst.) / ___ (diast.) mmHg

Heart rate ___ beats/min

Body temperature ___,___ °C

Body weight ___,___ kg

Hematology

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl			
Platelets		/nl			
White Blood count		/nl			
Neutrophils		/nl			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Clinical chemistry

Sampling date ____ Day ____ Month ____ Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl		☐	
Total bilirubin		mg/dl		☐	
Total protein		g/dl		☐	
ASAT / GOT		U/l		☐	
ALAT / GPT		U/l		☐	
LDH		U/l		☐	
Alkaline phosphatase		U/l		☐	
Creatinine Clearance		ml/min		☐	
INR				☐	
aPTT		sec		☐	

* Evaluation: **0 = normal**
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis ____ Day ____ Month ____ Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on CRF-form „Adverse events - additional assessments“.

Paracentesis

Were there any paracenteses performed for symptom relief since last visit?

☐ No

☐ Yes, please complete CRF-form „Paracentesis Log“.

Adverse Events

Did the patient experience any adverse event since last visit?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since last visit?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason:
- ☐ Patient‘s wish
- ☐ Organisatory reason
- ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a <u>plasma</u> sample for investigational analysis collected?
- ☐ No, please give reason _____
- ☐ Yes, Date ___ ___ ___ (Day Month Year)

Was a <u>serum</u> sample for pharmacokinetics collected?
- ☐ NA
- ☐ No, please give reason _____
- ☐ Yes, Date ___ ___ ___ (Day Month Year)

Course of Treatment

☐ Treatment phase completed regularly 8 weeks after 1st application of study drug: Day Month Year

☐ Treatment phase prematurely discontinued*: Day Month Year

* All patients who prematurely discontinue the treatment period will be irrespectively followed up for safety and survival (exceptions: patient withdrawal of consent for further study participation, lost to follow-up, death).

Reason:

☐ Any exclusion criteria developed, specify: _____

☐ Withdrawal of consent

☐ Lost to follow-up

☐ Death, date of death: Day Month Year → Please fill in death report form!

☐ Toxicity:

- ☐ Gastrointestinal perforation
- ☐ Fistula formation involving an internal organ (e.g. tracheo-oesophageal, bronchopleural, biliary, vagina and bladder)
- ☐ Arterial thrombo-embolic events
- ☐ Symptomatic grade 4 thrombosis
- ☐ Grade 3/4 hemorrhagic event
- ☐ Grade 4 hypertension (hypertensive crisis)
- ☐ Grade 4 proteinuria (nephrotic sydrome)
- ☐ Other grade 3/4 toxicities: _____

☐ At the patients request

☐ At the investigator‘s request, specify: _____

☐ Physician‘s judgment following an adverse event

☐ Termination by sponsor or a regulary authority

☐ Initiation of non-protocol-specific anti-tumor therapy

☐ Treatment of ascites with measures other than paracentesis, diuretics and study drugs

☐ Other, specify: _____

AIO-SUP-0108

Randomisation No. ____

Safety Follow-Up

(12 weeks after 1st application of study drug)

26

Survival status

☐ Ongoing, patient alive on ____ Day ____ Month ____ Year

☐ Patient lost to follow-up — Date of last contact: ____ Day ____ Month ____ Year

☐ Patient died on ____ Day ____ Month ____ Year → Please fill in death report form!

ECOG Performance status

Date: ____ Day ____ Month ____ Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: ____ Day ____ Month ____ Year

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Event“.

Vital Signs

Date: ____ Day ____ Month ____ Year

Blood pressure ____ / ____ mmHg (syst. / diast.) — Heart rate ____ beats/min

Body temperature ____ , ____ °C — Body weight ____ , ____ kg

Hematology

Sampling date: ____ Day ____ Month ____ Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl		☐	
Platelets		/nl		☐	
White Blood count		/nl		☐	
Neutrophils		/nl		☐	

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

AIO-SUP-0108

Randomisation No.

Safety Follow-Up

(12 weeks after 1st application of study drug)

27

Clinical chemistry

Sampling date Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis

Date of analysis Day Month Year

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

Paracentesis

Were there any paracenteses performed for symptom relief since last visit?

☐ No

☐ Yes, please complete CRF-form „Paracentesis Log“.

AIO-SUP-0108

Randomisation No. |_|_|_|_|

Safety Follow-Up

(12 weeks after 1st application of study drug)

28

Adverse Events

Did the patient experience any adverse event since last visit?

☐ No ☐ Yes, please specify on CRF-form „Adverse Event“.

Concomitant Medication, Diagnostic, Therapeutic or Surgical Procedure

Did the patient receive any concomitant medication, diagnostic, therapeutic or surgical procedure since last visit?

☐ No ☐ Yes, please specify on CRF-form „Concomitant Medication“.

Quality of life assessments

Did the patient complete the FACIT-AI quality of life questionnaire?

☐ Yes ☐ No, please specify reason:
- ☐ Patient‘s wish
- ☐ Organisatory reason
- ☐ Other

Did the medical staff/investigator complete the DGHO palliative care goup questionnaire?

☐ Yes ☐ No, please specify reason: _____

Investigational analysis

Was a <u>plasma</u> sample for investigational analysis collected?
- ☐ No, please give reason _____
- ☐ Yes, Date |_|_| |_|_| |_|_|_|_| (Day Month Year)

Was a <u>serum</u> sample for pharmacokinetics collected?
- ☐ NA
- ☐ No, please give reason _____
- ☐ Yes, Date |_|_| |_|_| |_|_|_|_| (Day Month Year)

Comments

none ☐

_____

_____

Herewith I confirm completeness and correctness of documentation on CRF pages 10 to 28, Paracenteses Log PL1 to 4, Adverse Events and Concomitant Medication.

_____
Stamp / Signature Investigator

|_|_| |_|_| |_|_|_|_|
Day Month Year
Date

AIO-SUP-0108

Randomisation No. ____

Follow-Up

(every 2 months)

____ (provide No. of FU visit)

Survival status

☐ Ongoing, patient alive on ____ Day ____ Month ____ Year

☐ Patient lost to follow-up, date of last contact: ____ Day ____ Month ____ Year

☐ Patient died on ____ Day ____ Month ____ Year → Please fill in death report form!

Anti-tumor therapy

Did the patient receive any further anti-tumor therapy? ☐ No ☐ Yes, specify

Drug generic name (if constitutional drug use trade name)	Date started If continuing after safety follow-up, please leave date empty and tick: ongoing		Date stopped If continuing after follow-up, please leave date empty and tick: ongoing	
	☐	Day Month Year	Day Month Year	☐
	☐	Day Month Year	Day Month Year	☐
	☐	Day Month Year	Day Month Year	☐
	☐	Day Month Year	Day Month Year	☐

Paracentesis for symptom relief

Was there any paracentesis performed for symptom relief since last visit?

☐ Not Applicable (in case a 2nd paracentesis was already clinically required and documented after application of 1st paracentesis for study purposes (week 0))

☐ No

☐ Yes, specify date of 1st paracentesis during Follow-up ____ Day ____ Month ____ Year

Herewith I confirm completeness and correctness of documentation on CRF page Follow-Up.

_____

Stamp / Signature Investigator

____ Day ____ Month ____ Year
Date

AIO-SUP-0108 Randomisation No.	Death Report	DR1

Date of death: Day Month Year

Was an autopsy performed? ☐ No ☐ Yes ☐ Unknown

Reason of death

☐ Progressive disease

☐ Toxicity of study medication, please specify: _____

☐ Other (serious) adverse event,please specify: _____

☐ Other reasons, please specify: _____

☐ Unknown

Comments

none ☐

_____

_____

_____

_____

_____

Herewith I confirm completeness and correctness of documentation on CRF page „Death Report“.

_____
Stamp / Signature Investigator

Day Month Year
Date

AIO-SUP-0108 Randomisation No.	**Paracentesis Log** **(between 1st paracentesis for study purposes and Safety-Follow-Up)**	Paracentesis No.	PL1

Date of paracentesis performed for symptom relief (and not only for diagnostic purposes) ___ ___ Day ___ ___ Month ___ ___ Year

Volume of ascites (i.e. Volume minus volume of lavage fluid, if applicable) ___ ___ ___ ___ ml

Routine analysis of malignant ascites

Hematology (from 2-5 ml EDTA-anticoagulated ascites):

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Hemoglobin		g/dl		
Hematocrit		%		
Total Leukocytes		/nl		
Neutrophils		/nl		

Chemistry (from 5ml heparinised ascites)

	Value	Unit	Unit, if other	converted value (for Data Managment only)
Total protein		g/dl		
Albumin		g/dl		

Investigational analysis of plasma / serum

☐ not applicable*

*** In case that date of paracentesis coincides with date of routine examination, already documented on CRF pages week 0, 2, 4, 6 or 8 or in case of paracenteses performed at Safety-Follow-up.**

Was a plasma sample for investigational analysis collected?

☐ No, please give reason

_____

☐ Yes, Date ___ ___ Day ___ ___ Month ___ ___ Year

Was a serum sample for pharmacokinetics collected?

☐ NA

☐ No, please give reason

_____

☐ Yes, Date ___ ___ Day ___ ___ Month ___ ___ Year

Investigational analysis of ascites

Was an ascites sample for investigational analysis collected?

☐ No, please give reason

_____

☐ Yes, Date ___ ___ Day ___ ___ Month ___ ___ Year

AIO-SUP-0108 Randomisation No.	**Paracentesis Log** **(between 1st paracentesis for study purposes and Safety-Follow-Up)**	Paracentesis No.	PL2

☐ **Not Applicable, in case that date of paracentesis coincides with date of routine examinations, already documented on CRF pages week 0, 2, 4, 6, 8 or at Safety-Follow-Up.**

ECOG Performance status

Date: Day Month Year

ECOG 0	ECOG 1	ECOG 2	ECOG 3	ECOG 4
☐	☐	☐	☐	☐

Physical Examination

Date: Day Month Year

Was there any clinical significant worsening since last visit?

☐ No ☐ Yes, please document on CRF-form „Adverse Event“.

Vital Signs

Date: Day Month Year

Blood pressure ___ (syst.) / ___ (diast.) mmHg

Heart rate ___ beats/min

Body temperature ___,___ °C

Body weight *(before paracentesis)* ___,___ kg

Hematology

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Managment only)
Hemoglobin		g/dl			
Platelets		/nl			
White Blood count		/nl			
Neutrophils		/nl			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

AIO-SUP-0108 Randomisation No.	**Paracentesis Log** **(between 1st paracentesis for study purposes and Safety-Follow-Up)**	Paracentesis No.	PL3

☐ **Not Applicable, in case that date of paracentesis coincides with date of routine examinations, already documented on CRF pages week 0, 2, 4, 6, 8 or at Safety-Follow-Up.**

Clinical chemistry

Sampling date ___ ___ ___ (Day Month Year)

	Value	Unit	Unit, if other	Evaluation*	converted value (for Data Management only)
Creatinine		mg/dl			
Total bilirubin		mg/dl			
Total protein		g/dl			
ASAT / GOT		U/l			
ALAT / GPT		U/l			
LDH		U/l			
Alkaline phosphatase		U/l			
Creatinine Clearance		ml/min			
INR					
aPTT		sec			

* Evaluation:
0 = normal
1 = abnormal, not clinically significant
2 = abnormal and clinically significant. In case of treatment-emergent abnormality, please complete adverse event form.

Urinalysis within 48 h prior study treatment

Date of analysis ___ ___ ___ (Day Month Year)

	Negative	Traces	Positive			
			+	++	+++	≥++++
Protein	☐	☐	☐	☐	☐	☐

Please check maximum value only.

If Protein > 1+ (dipstick) please refer to section 7.3.2 and 5.2.4 of study protocol and document protein in 24h urine on Adverse Event additional pages.

AIO-SUP-0108 Randomisation No.	**Paracentesis Log** **(between 1st paracentesis for study purposes and Safety-Follow-Up)**	Paracentesis No.	PL4

Bevacizumab / Placebo application

Was Bevacizumab/Placebo administered after paracentesis?

- ☐ No, give reason
  - ☐ time span between paracenteses < 14 days
  - ☐ temporary treatment discontinuation, please specify reason _____
  - ☐ permanent treatment discontinuation, please specify reason _____
- ☐ Yes, specify dose to be administered
  - ☐ Full dose of 400 mg administered
  - ☐ Dose reduction, please specify

    Actual dose: ___ mg

    Reason: _____

Comments

none ☐

_____

_____

_____

_____

Herewith I confirm completeness and correctness of documentation on CRF page PL1-PL4.

_____

Stamp / Signature Investigator

Day Month Year

Date

Concomitant Medication

Please describe
- anti-tumor therapy
- symptomatic treatment for ascites (other than paracentesis and study drug)
Please document diuretics also. (In case of catumaxomab or intraperitoneal application of chemo therapy, study treatment is to be discontinued immediately)
- other concomitant medications
- diagnostic, therapeutic or surgical procedure
- transfusions

Indication

1=Adverse event
2=Prophylaxis
3=Concomitant disease
4=Pre-Medication for chemotherapy
5=Symptomatic treatment for ascites
6=Anti-tumor drugs
7=Transfusion of blood and products
8=Diagnostic, therapeutic or surgical procedure
9=Other

AIO-SUP-0108

Randomisation No. ____

Concomitant Medication

(between start of treatment and Safety Follow-up)

CM ___/___

Did the patient receive any concomitant medication, concomitant treatment, anti-tumor drugs and/or concomitant procedures? ☐ **No** ☐ **Yes, please specify**

Drug / concomitant treatment / concomitant procedure generic name (if constitutional drug use trade name)	Date Started	Date stopped If continuing at safety follow-up, please leave date empty and tick: ongoing		Indication If AE or other, specify
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	
	Day Month Year	Day Month Year	☐	

Adverse Events

Please refer to recommendations defined in section 7.1.1, 7.1.2 and 7.1.3 of the study protocol.

Adverse events

- all new adverse events
- all adverse events which change in seriousness
- all medical conditions present at start of treatment which had worsened
- all treatment-emergent laboratory - test value abnormalities with clinical significance

CTC-Grade

Please insert the worst grade according to NCI - Common Toxicity Criteria Version 3.0.

Causality

1=probable
2=possible
3=remote
4=unrelated

Outcome

1=recovered/resolved
2=recovering/resolving
3=not recovered/not resolved
4=recovered/resolved with resident effects
5=fatal
6=unknown

SAE

1=yes, please fill in SAE Report form
2=no

Therapy necessary

1=yes, please complete also concomitant medication pages
2=no

Most relevant study drug action

1=none
2=drug withdrawn
3=dose reduced
4=not applicable

Report on Adverse Events

Adverse events are to be documented from the first application of study drug of the first treatment week through Safety Follow-Up (EOT + 4 weeks)

none ☐

Adverse Events CTC Term	Start Date	Stop Date	CTC-Grade	Causality	Out-come	SAE	Therapy necessary?	Most relevant study drug action
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						
	Day Month Year	Day Month Year						

In case of grade 3/4 hypertension, proteinuria or grade 3 thrombosis or grade 2 or higher hemorrhagic event, please complete also CRF page „Adverse Events - Additional Assessments".

AIO-SUP-0108 Randomisation No.	Adverse Events - Additional Assessments	AEa1

Hypertension

Did the patient experience any grade 3/4 hypertension? ☐ No ☐ Yes, specify on Adverse Event page

Start of grade 3/4 hypertension ___ Day ___ Month ___ Year

Please document blood pressure at start of event and thereafter on a weekly basis until resolution of event or Safety-FU visit, whatever occurs first:

Date	Blood pressure (mmHg)	Date	Blood pressure (mmHg)
Day Month Year	syst. / diast.	Day Month Year	syst. / diast.
Day Month Year	syst. / diast.	Day Month Year	syst. / diast.
Day Month Year	syst. / diast.	Day Month Year	syst. / diast.
Day Month Year	syst. / diast.	Day Month Year	syst. / diast.
Day Month Year	syst. / diast.	Day Month Year	syst. / diast.

Proteinuria

Did patient experience proteinuria (protein > 1+ dipstick)? ☐ No ☐ Yes, specify on Adverse Event page

Please provide the routine 24h-urine results according to section 5.2.4 and 7.3.2 of study protocol.

Date	24-Urine (g/24h)	Date	24-Urine (g/24h)
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	
Day Month Year		Day Month Year	

AIO-SUP-0108 Randomisation No.	Adverse Events - Additional Assessments	AEa2

Grade 3/4 Thrombosis

Did the patient experience any grade 3/4 thrombosis? ☐ No ☐ Yes, specify on Adverse Event page

Was there any treatment of this event started? ☐ No ☐ Yes, specify on Concomitant medication page

Please provide INR, aPPT prior to initiation of therapy.

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	Converted value (for Data Managment only)
INR					
aPTT		sec			

* Evaluation: **0 = normal**
1 = abnormal, clinically significant
2 = abnormal and clinically significant.

Grade 2 or higher hemorrhagic event

Did the patient experience any grade 2 or higher hemorrhagic event? ☐ No ☐ Yes, specify on Adverse Event page

Was there any treatment of this event started? ☐ No ☐ Yes, specify on Concomitant medication page

Please provide platelet count, INR, aPTT prior to initiation of therapy for the event.

Sampling date: Day Month Year

	Value	Unit	Unit, if other	Evaluation*	Converted value (for Data Managment only)
Platelet count		/nl			
INR					
aPTT		sec			

* Evaluation: **0 = normal**
1 = abnormal, clinically significant
2 = abnormal and clinically significant.

AIO-SUP-0108 Randomisation No.	FACIT QoL	Visit: ☐ Screening ☐ Week 06 ☐ Week 00 ☐ Week 08 ☐ Week 02 ☐ Safety FU ☐ Week 04 ☐ Paracentesis

FACIT ASCITES INDEX (Patientenfragebogen)

Tag Monat Jahr
Heutiges Datum

Nachfolgend finden Sie eine Liste von Aussagen, die von anderen Personen mit Ihrer Krankheit für wichtig befunden wurden. **Bitte geben Sie jeweils an, wie sehr jede der Aussagen im Laufe der letzten 7 Tage auf Sie zugetroffen hat, indem Sie die entprechende Zahl ankreuzen.**

		Überhaupt nicht	Ein wenig	Mäßig	Ziemlich	Sehr
C6	Ich habe einen guten Appetit	0	1	2	3	4
GF5	Ich schlafe gut	0	1	2	3	4
BMT5	Ich bin in der Lage, mich alleine fortzubewegen	0	1	2	3	4
B1	Ich leide unter Atemnot	0	1	2	3	4
GP2	Mir ist übel	0	1	2	3	4
O2	Ich habe mich übergeben	0	1	2	3	4
ACT11	Ich habe Schmerzen in der Magengegend	0	1	2	3	4
O1	Ich habe Schwellungen im Magenbereich	0	1	2	3	4
GP1	Mir fehlt es an Energie	0	1	2	3	4
ACT10	Wenn ich esse, fühle ich mich rasch satt	0	1	2	3	4
BL2	Ich muss häufiger Wasserlassen	0	1	2	3	4
Cx6	Ich leide an Verstopfung	0	1	2	3	4
AI1	Ich bin bekümmert	0	1	2	3	4

AIO-SUP-0108

Randomisation No.

DGHO QoL

Visit: ☐ Screening ☐ Week 06 ☐ Week 00 ☐ Week 08 ☐ Week 02 ☐ Safety FU ☐ Week 04 ☐ Paracentesis

Patientenlebensqualitätsfragebogen
(modifiziert nach der Palliativgruppe der DGHO)

Der Fragebogen ist durch das medizinische Personal zu ergänzen.

Tag Monat Jahr
Heutiges Datum

Ausprägung von Symptomen zum Zeitpunkt der Aufnahme und nach Erfolgter Einstellung

alles Zutreffende nach Schweregrad ausfüllen (schweregrad 0=ohne, 1=gering, 2=mittel, 3=stark) vor Intervention ☐ nach Intervention ☐

Schmerzen ☐	Dysphagie ☐	(Lymph-)Ödem ☐	Krampfanfälle ☐
Schwäche ☐	Obstipation ☐	Juckreiz ☐	motor. Unruhe ☐
Appetitlosigkeit ☐	Diarrhoe ☐	Dekubilus ☐	Verwirrtheit ☐
Übelkeit ☐	Ascites ☐	exulc. Wunde ☐	Schlafstörungen ☐
Erbrechen ☐	Dyspnoe ☐	Harnverhalt ☐	Angst ☐
(Tumor-)Blutung ☐	Husten ☐	Lähmungen ☐	Depression ☐

Therapie bei Aufnahme und im Verlauf / zum Ende

Schmerztherapie WHO

- keine ○
- nur bei Bedarf ○
- WHO Stufe I ○
- WHO Stufe II ○
- WHO Stufe III ○

Koanalgetika / Begleitmedikation

- Nichtopioide Analget. ○
- Antiemetika ○
- Antidepressiva ○
- Antikonvulsiva ○
- Kortikosteroide ○
- Laxanzien ○
- Neuroleptika ○
- Sedativa / Hypnotika ○

Opioidtherapie

Substanz(en)

- Morphin ○
- Hydromorphon ○
- Fentanyl ○
- Oxycodon ○
- Buprenorphin ○
- Levomethadon ○
- Piritramid ○

Applikationsform(en)*

- oral/PEG/rektal ○
- transdermal ○
- s.c. Injektion ○
- s.c. Dauerinfusion ○
- i.v. Dauerinfusion ○
- peridural ○

sonstige Palliativmaßnahmen

- PEG / transnasale Sonde ○
- zentraler venöser Zugang ○
- enterale Ernährung ○
- parenterale Ernährung ○
- i.v. Flüssigkeitssubstitution ○
- s.c. Flüssigkeitssubstitution ○
- Ascitespunktion(en) ○
- Pleurapunktion(en) ○
- palliative Chirurgie ○
- palliative Radiatio ○
- palliative Chemotherapie ○
- palliative endoskop. Eingriffe ○
- nichtspezialisierter Pflegedienst ○
- Palliativpflegedienst ○

16.1.3 List of ethics committees - Representative written information for patient and sample consent forms

16.1.3 List of ethics committees

10 sites were already deregistered during the study (crossed)

Site	Address	Ethics Committee	Local Authority
01	Universitätsklinikum Halle-Wittenberg Klinik für Innere Medizin IV Onkologie / Hämatologie Ernst-Grube-Straße 40 06120 Halle (Saale)	Ethik-Kommission der Medizinischen Fakultät der Martin-Luther Universität Halle-Wittenberg Magdeburger Straße 16 06112 Halle (Saale)	Landesverwaltungsamt Sachsen-Anhalt Referat: Arzneimittel- und Apothekenwesen Ernst-Kamieth-Str. 2 06112 Halle/Saale
02	Universitätsklinikum Hamburg-Eppendorf Onkologisches Zentrum Martinistraße 52 20246 Hamburg	Ethik-Kommission der Ärztekammer Hamburg Humboldtstraße 67a 22083 Hamburg	Behörde für Gesundheit und Verbraucherschutz der Freien und Hansestadt Hamburg Fachabteilung V 14 Patientenschutz und Sicherheit in der Medizin Billstraße 80 20539 Hamburg
03	Charité Campus Virchow-Klinikum Hämatologie und Onkologie Augustenburger Platz 1 13353 Berlin	Landesamt für Gesundheit und Soziales Ethik-Kommission des Landes Berlin Fehrbelliner Platz 1 10707 Berlin	Landesamt für Gesundheit und Soziales Berlin Referat I B Krankenhausaufsicht, Arzneimittelwesen, Apothekenwesen Turmstr. 21, Haus A 10559 Berlin
~~04~~	~~Klinikum Ludwigsburg Klinik für Innere Medizin, Gastroenterologie, Hämatologie und Diabetologie Posilipostraße 4 71640 Ludwigsburg~~	~~Ethik-Kommission bei der Landesärztekammer Baden-Württemberg Jahnstraße 40 70565 Stuttgart~~	~~Regierungspräsidium Stuttgart Ruppmannstraße 21 70565 Stuttgart~~
05	Onkologische Schwerpunktpraxis 311XX Hildesheim	Ethik-Kommission bei der Ärztekammer Niedersachsen Berliner Allee 20 30175 Hannover	Staatliches Gewerbeaufsichtsamt Hannover Inspektorat Hannover Am Listholze 74 30177 Hannover
~~06~~	~~Universitätsklinikum der Johannes-Gutenberg-Universität Mainz I. Medizinische Klinik und Poliklinik Langenbeckstraße 1 55131 Mainz~~	~~Ethik-Kommission der Landesärztekammer Rheinland-Pfalz Deutschhausplatz 3 55116 Mainz~~	~~Landesamt für Soziales, Jugend und Versorgung Koblenz Referat 55: Arzneimittel, Tierarzneimittel Baedeckerstraße 2-10 56073 Koblenz~~
07	Klinikum der Johann-Wolfgang-Goethe Universität Frankfurt (Main) Theodor-Stern-Kai 7 60590 Frankfurt	Ethik-Kommission des Fachbereichs Medizin der Johann Wolfgang Goethe Universität Frankfurt Theodor-Stern-Kai 7 60590 Frankfurt	Regierungspräsidium Darmstadt Dezernat II 23.2 Pharmazie Luisenplatz 2 64283 Darmstadt
08	Kliniken Maria Hilf GmbH, Krankenhaus St. Franziskus Medizinische Klinik I Viersener Str. 450 41063 Mönchengladbach	Ethik-Kommission der Ärztekammer Nordrhein Tersteegenstraße 9 40474 Düsseldorf	Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf

~~09~~	~~Klinikum Fulda gAG Tumorklinik, Medizinische Onkologie/Hämatologie Pacelliallee 4 36043 Fulda~~	~~Ethik-Kommission der Landesärztekammer Hessen Im Vogelgesang 3 60488 Frankfurt~~	~~Regierungspräsidium Darmstadt Dezernate II 23.1: Pharmazie I und II 23.2: Pharmazie II Luisenplatz 2 64278 Darmstadt~~
10	Onkologische Schwerpunktpraxis 202XX Hamburg	Ethik-Kommission der Ärztekammer Hamburg Humboldtstraße 67a 22083 Hamburg	Behörde für Gesundheit und Verbraucherschutz der Freien und Hansestadt Hamburg Fachabteilung V 14 Patientenschutz und Sicherheit in der Medizin Billstraße 80 20539 Hamburg
~~11~~	~~Prosper-Hospital Medizinische Klinik I Mühlenstraße 27 45659 Recklinghausen~~	~~Ethik-Kommission der Ärztekammer Westfalen-Lippe und der Medizinischen Fakultät der Westfälischen Wilhelms-Universität Münster Gartenstr. 210 - 214 48147 Münster~~	~~Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf~~
12	Onkologische Schwerpunktpraxis 732XX Wendlingen	Ethik-Kommission bei der Landesärztekammer Baden-Württemberg Jahnstraße 40 70565 Stuttgart	Regierungspräsidium Stuttgart Ruppmannstraße 21 70565 Stuttgart
~~13~~	~~Ernst von Bergmann Klinikum Zentrum für Hämatologie, Onkologie und Strahlenheilkunde Charlottenstraße 72 14467 Potsdam~~	~~Ethikkommission der Landesärztekammer Brandenburg Dreifertstraße 12 03044 Cottbus~~	~~Landesamt für Umwelt, Gesundheit und Verbraucherschutz Apotheken, Arzneimittel, Medizinprodukte Seeburger Chaussee 2 14476 Potsdam~~
14	Lahn-Dill-Kliniken GmbH Klinik für Hämatologie/Onkologie und Palliativmedizin Forsthausstraße 1 35578 Wetzlar	Ethik-Kommission der Landesärztekammer Hessen Im Vogelgesang 3 60488 Frankfurt	Regierungspräsidium Darmstadt Dezernat II 23.2 Pharmazie Luisenplatz 2 64283 Darmstadt
~~15~~	~~Klinikum Region Hannover GmbH Krankenhaus Siloah Medizinische Klinik III für Hämatologie und Onkologie Roesebeckstraße 15 30449 Hannover~~	~~Ethik-Kommission bei der Ärztekammer Niedersachsen Berliner Allee 20 30175 Hannover~~	~~Staatliches Gewerbeaufsichtsamt Hannover Inspektorat Hannover Am Listholze 74 30177 Hannover~~
~~16~~	~~Onkologische Schwerpunktpraxis 141XX Berlin~~	~~An das Landesamt für Gesundheit und Soziales Ethik-Kommission des Landes Berlin Fehrbelliner Platz 1 10707 Berlin~~	~~Landesamt für Gesundheit und Soziales Berlin Referat I B Krankenhausaufsicht, Arzneimittelwesen, Apothekenwesen Turmstr. 21, Haus A 10559 Berlin~~
~~17~~	~~Onkologische Schwerpunktpraxis 220XX Hamburg~~	~~Ethik-Kommission der Ärztekammer Hamburg Humboldtstraße 67a 22083 Hamburg~~	~~Behörde für Gesundheit und Verbraucherschutz der Freien und Hansestadt Hamburg Fachabteilung V 14 Patientenschutz und Sicherheit in der Medizin~~

			~~Billstraße 80~~ ~~20539 Hamburg~~
~~18~~	~~Onkologische Schwerpunktpraxis~~ ~~224XX Hamburg~~	~~Ethik-Kommission der~~ ~~Ärztekammer Hamburg~~ ~~Humboldtstraße 67a~~ ~~22083 Hamburg~~	~~Behörde für Gesundheit und~~ ~~Verbraucherschutz der~~ ~~Freien und Hansestadt~~ ~~Hamburg~~ ~~Fachabteilung V 14~~ ~~Patientenschutz und~~ ~~Sicherheit in der Medizin~~ ~~Billstraße 80~~ ~~20539 Hamburg~~
19	Klinikum Leverkusen gGmbH Medizinische Klinik III Am Gesundheitspark 11 51375 Leverkusen	Ethik-Kommission der Ärztekammer Nordrhein Tersteegenstraße 9 40474 Düsseldorf	Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf
~~20~~	~~Onkologische Schwerpunktpraxis~~ ~~018XX Neustadt (Sachsen)~~	~~thikkommission der~~ ~~Sächsischen~~ ~~Landesärztkammer~~ ~~Schützenhöhe 16~~ ~~01099 Dresden~~	~~Landesdirektion Leipzig~~ ~~Referat 24:~~ ~~Veterinärwesen und~~ ~~Lebensmittelüberwachung,~~ ~~Pharmazie~~ ~~Braustraße 2~~ ~~04107 Leipzig~~
22	Städtisches Klinikum Magdeburg Klinik für Allgemein- und Viszeralchirurgie Birkenallee 34 39130 Magdeburg	Ethik-Kommission des Landes Sachsen-Anhalt - Geschäftsstelle - Kühnauer Straße 70 06846 Dessau-Roßlau	Landesverwaltungsamt Sachsen-Anhalt Referat: Arzneimittel- und Apothekenwesen Ernst-Kamieth-Str. 2 06112 Halle/Saale
23	Vivantes Klinikum Spandau Klinik für Innere Medizin Neue Bergstraße 6 13585 Berlin	An das Landesamt für Gesundheit und Soziales Ethik-Kommission des Landes Berlin Fehrbelliner Platz 1 10707 Berlin	Landesamt für Gesundheit und Soziales Berlin Referat I B Krankenhausaufsicht, Arzneimittelwesen, Apothekenwesen Turmstr. 21, Haus A 10559 Berlin
24	Evang. Huyssens-Stiftung Klinik für internistische Onkologie und Hämatologie Henricistraße 92 45136 Essen	Ethik-Kommission der Ärztekammer Nordrhein Tersteegenstraße 9 40474 Düsseldorf	Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf
25	Universitätsklinikum Essen Innere Klinik (Tumorforschung) Hufelandstr. 55 45147 Essen	Ethik-Kommission Medizinischen Fakultät der Universität Duisburg-Essen Universitätsklinikum Essen Robert-Koch-Straße 9-11 45147 Essen	Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf
26	Klinikum Deggendorf Medizinische Klinik II Perlasberger Str. 41 94469 Deggendorf	Ethik-Kommission der Bayerischen Landesärztekammer Mühlbauerstraße 16 81677 München	Regierung von Oberbayern Zentrale Arzneimittelüberwachung Bayern -ZAB Sachgebiet 53.2: Pharmazie Maximilianstrasse 39 80538 München
27	Vivantes Klinikum am Urban Onkologisches Zentrum Mitte	Landesamt für Gesundheit und Soziales	Landesamt für Gesundheit und Soziales Berlin Referat I

	Dieffenbachstr. 1 10967 Berlin	Ethik-Kommission des Landes Berlin Fehrbelliner Platz 1 10707 Berlin	B Krankenhausaufsicht, Arzneimittelwesen, Apothekenwesen Turmstr. 21, Haus A 10559 Berlin
28	Vivantes Klinikum Neukölln Onkologisches Zentrum Vivantes Süd Rudower Straße 48 12351 Berlin	Landesamt für Gesundheit und Soziales Ethik-Kommission des Landes Berlin Fehrbelliner Platz 1 10707 Berlin	Landesamt für Gesundheit und Soziales Berlin Referat I B Krankenhausaufsicht, Arzneimittelwesen, Apothekenwesen Turmstr. 21, Haus A 10559 Berlin
29	Friedrich-Ebert-Krankenhaus GmbH Klinik für Hämatologie/Onkologie/Nephrologie Friesenstr. 11 24534 Neumünster	Ethik-Kommission bei der Ärztekammer Schleswig-Holstein Bismarckallee 8-12 23795 Bad Segeberg	Landesamt für soziale Dienste des Landes Schleswig-Holstein Dezernat 31: Arzneimittelüberwachung Adolf-Westphal-Straße 4 24143 Kiel
31	Onkologische Schwerpunktpraxis Städteregion Aachen	Ethik-Kommission der Ärztekammer Nordrhein Tersteegenstraße 9 40474 Düsseldorf	Stadt Düsseldorf - Gesundheitsamt - Zentrales Inspektorat für klinische Prüfstellen in NRW Kölner Straße 180 40200 Düsseldorf

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Site No.1

Patienteninformation/-einwilligungserklärung

Studiennummer: AIO-SUP-0108

EudraCT-Nummer: 2009-014725-16

Titel der Studie: Eine randomisierte, doppelblinde, Placebo kontrollierte Phase II-Studie zur Untersuchung der Wirksamkeit von Bevacizumab zur Linderung vom Symptomen bei Patienten mit malignem Aszites bei fortgeschrittenen Krebserkrankungen gastrointestinalen Ursprungs

Patientennummer: _____

Sehr geehrte Patientin, sehr geehrter Patient,

wir möchten Sie fragen, ob Sie bereit sind, an der nachfolgend beschriebenen klinischen Prüfung (Studie) teilzunehmen.

Klinische Prüfungen sind notwendig, um Erkenntnisse über die Wirksamkeit und Verträglichkeit von Arzneimitteln zu gewinnen oder zu erweitern. Deshalb schreibt der Gesetzgeber im Arzneimittelgesetz vor, dass Arzneimittel klinisch geprüft werden müssen. Die klinische Prüfung, die wir Ihnen hier vorstellen, wurde – wie es das Gesetz verlangt – von der zuständigen Ethikkommission zustimmend bewertet und von der zuständigen Behörde genehmigt. Diese klinische Prüfung wird an mehreren Orten durchgeführt; insgesamt sollen 72 Personen daran teilnehmen. Die Studie wird veranlasst, organisiert und finanziert durch die AIO-Studien-gGmbH, Kuno-Fischer-Straße 8, 14057 Berlin, den Sponsor dieser Studie.

Kosten für Maßnahmen, die der Routine-Behandlung entsprechen, sind nicht Bestandteil der Unterstützung durch diese Firma. Für den Prüfarzt erwächst aus der Unterstützung kein Interessenskonflikt.

Ihre Teilnahme an dieser klinischen Prüfung ist freiwillig. Sie werden in diese Prüfung also nur dann einbezogen, wenn Sie dazu schriftlich Ihre Einwilligung erklären. Sofern Sie nicht an der klinischen Prüfung teilnehmen oder später aus ihr ausscheiden möchten, erwachsen Ihnen daraus keine Nachteile.

Sie wurden bereits auf die geplante Studie angesprochen. Der nachfolgende Text soll Ihnen die Ziele und den Ablauf erläutern. Anschließend wird ein Prüfarzt das Aufklärungsgespräch mit Ihnen führen. Bitte zögern Sie nicht, alle Punkte anzusprechen, die Ihnen unklar sind. Sie werden danach ausreichend Bedenkzeit erhalten, um über Ihre Teilnahme zu entscheiden.

Insgesamt nehmen deutschlandweit circa 72 Patienten in circa 20 Kliniken an dieser klinischen Prüfung teil. Die klinische Prüfung wird seit dem Jahr 2010 durchgeführt.

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1. Warum wird diese Prüfung durchgeführt?

Sie leiden an einer Flüssigkeitsansammlung im Bauchraum (Aszites). Diese wird durch Ihre Krebserkrankung verursacht, Das Abpunktieren der Flüssigkeit stellt eine übliche Therapieform dar, die damit verbundene Linderung der Beschwerden ist jedoch in der Regel nicht von Dauer. Ihr behandelnder Arzt hat Ihnen die freiwillige Teilnahme an einer klinischen Prüfung vorgeschlagen, in der ein Teil der Patienten das Medikament Bevacizumab erhält. Sie werden daher im Rahmen dieser Studie nach der ohnehin stattfindenden Abpunktion der Flüssigkeit im Bauchraum durch die bereits liegende Kanüle das Medikament Bevacizumab (Handelsname Avastin®) oder Placebo /(Scheinmedikament) erhalten. Die intravenöse Gabe von Bevacizumab ist bereits seit längerem zur Behandlung von verschiedenen Krebsarten, einschließlich des metastasierenden Dick- oder Enddarmkrebses, zugelassen. Neuere Erkenntnisse legen außerdem nahe, dass eine Injektion von Bevacizumab in den Bauchraum auch die tumorbedingte Ansammlung von Flüssigkeit in dieser Körperhöhle verzögern oder unterbinden kann.

Das Ziel dieser Studie ist, herauszufinden, ob die Gabe von Bevacizumab im Vergleich zu Placebo (Scheinmedikament) zusätzlich zu der üblichen Abpunktion der tumorbedingten Flüssigkeitsansammlung im Bauchraum das Wiederauftreten derselben verzögern oder sogar unterbinden kann. Außerdem werden im Rahmen dieser klinischen Prüfung Daten zur Wirksamkeit, Durchführbarkeit und Sicherheit einer Applikation von Bevacizumab in den Bauchraum erhoben.

Informationen zur Prüfsubstanz Bevacizumab

Der vaskuläre endotheliale Wachstumsfaktor (VEGF) ist ein Eiweiß, das von Tumorzellen freigesetzt wird. Es bewirkt, dass die Blutgefäße im Bauchraum durchlässiger werden und sich zunehmend Flüssigkeit in dieser Körperhöhle ansammelt. Außerdem lässt VEGF Blutgefäße in Richtung des Tumors wachsen, die diesen dann mit Nährstoffen versorgen. Bevacizumab ist ein Antikörper, der nach dem Schloss-Schlüssel-Prinzip VEGF blockiert, so dass die tumorbedingte Ansammlung von Flüssigkeit in der Bauchhöhle möglicherweise eingeschränkt werden kann. Möglicherweise hat Bevacizumab, in die Bauchhöhle injiziert, auch einen hemmenden Effekt auf das Tumorwachstum, indem es dessen Versorgung mit Nährstoffen blockiert. Die Firmen Roche Pharma AG (Roche) und Genentech Inc. haben dieses Medikament zusammen entwickelt.

2. Erhalte ich das Prüfpräparat auf jeden Fall?

Sie haben eine Chance von 2:1, Bevacizumab (Prüfpräparat) oder das Scheinmedikament zu erhalten. Die Zuordnung zu der jeweiligen Behandlungsgruppe erfolgt rein zufällig (so wie das Werfen einer Münze) und kann weder von Ihnen noch von Ihrem Arzt beeinflusst werden. Keiner, der direkt an der Durchführung der klinischen Studie beteiligt ist (einschließlich Ihres Arztes) wird wissen, ob Sie Bevacizumab oder Placebo (Scheinmedikament) erhalten. Diese Medikamente werden „verblindet“ verabreicht. Dieses „Verblindungsverfahren“ ist notwendig, um sicherzustellen, dass die Ergebnisse der Studie aussagekräftig genug sind und auch, um beurteilen zu können, ob Bevacizumab zweifelsfrei die beste Therapiemöglichkeit für Patienten mit tumorbedingter Flüssigkeitsansammlung in der Bauchhöhle darstellt. Die Zuordnung zur jeweiligen Behandlungsgruppe wird nur dann „entblindet“, wenn schwere Nebenwirkungen auftreten, um so eine Schädigung der Gesundheit aller an der Studie teilnehmenden Patienten zu verhindern.

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Behandlung/Zuordnung der Patienten zu den einzelnen Behandlungsgruppen

Es wird mittels Losverfahren entschieden werden, welche Patienten im Rahmen der Studie das Placebo (Scheinmedikament) und welche Patienten zusätzlich das Medikament Bevacizumab erhalten.

Arm A (Zwei Drittel aller Patienten): Sie erhalten Bevacizumab

Arm B (Ein Drittel aller Patienten): Sie erhalten das Placebo (Scheinmedikament)

3. Beschreibung des Studienablaufes sowie aller invasiven Vorgänge

Die geplante Behandlungsdauer für Sie als Patient/Patientin im Rahmen der klinischen Prüfung beträgt maximal acht Wochen. Während dieser Zeit wird ein Drittel der Patienten mit der üblichen Abpunktion der tumorbedingten Flüssigkeitsansammlung im Bauchraum behandelt werden. Durch die bereits liegende Kanüle wird nach der Abpunktion das Placebo (Scheinmedikament) über eine Infusion verabreicht. Bei zwei Dritteln der Patienten wird nach Abpunktieren der Flüssigkeit das Medikament Bevacizumab durch die bereits liegende Kanüle in den Bauchraum über eine Infusion verabreicht.

Insgesamt werden die Patienten eine maximale Anzahl von vier Gaben von Bevacizumab/Placebo (Scheinmedikament) erhalten. Nach der 8-wöchigen Behandlungsphase folgt dann eine vierwöchige Nachbeobachtungsphase, in der eventuell später auftretende Nebenwirkungen erfasst werden sollen. Darüber hinaus möchten wir Ihren Krankheitsverlauf in Abständen von acht Wochen solange weiter beobachten, bis Sie eine Gesamtdauer in der Studie von einem Jahr (gerechnet von der ersten Behandlung im Rahmen der Studie an) erreicht haben.

Für den Behandlungserfolg und die Erreichung des Studienzieles ist es sehr wichtig, dass Sie alle Besuchstermine wie geplant einhalten.

a) Vor Behandlungsbeginn

Wenn Sie sich mit der Teilnahme an dieser Studie einverstanden erklären, wird Ihr Arzt Sie zunächst bitten, diese Information und Einwilligungserklärung zu unterzeichnen. Dann wird er Sie in der maximal vierwöchigen Prüfungsvorphase der klinischen Prüfung gründlich medizinisch untersuchen, um herauszufinden, ob Sie für die Studienteilnahme geeignet sind.

Ihr Arzt befragt Sie zu Ihrer medizinischen Vorgeschichte. Von speziellem Interesse sind dabei Herz- sowie andere schwere Erkrankungen, größere Gewichtsveränderungen in den letzten sechs Monaten, Episoden von Eiweiß oder Blut im Urin sowie Ihre Tumorvorgeschichte und deren Behandlung.

Auch die Medikamente, die Sie momentan einnehmen, die Behandlungen/Operationen, denen Sie sich in der jüngsten Vergangenheit unterzogen haben, sowie Ihr generelles Wohlbefinden sind von Interesse und werden abgefragt.

Darüber hinaus erfolgen frühestens drei Tage vor Behandlungsbeginn eine allgemeine körperliche und neurologische Untersuchung inklusive EKG und eine Bestimmung Ihrer Vitalfunktionen (Körpergröße, Körpergewicht, Blutdruck, Puls und Temperatur).

Um den Verlauf Ihrer Erkrankung genau beurteilen zu können, wird routinemäßig außerdem erfasst werden, wie oft bei Ihnen bereits Flüssigkeit aus dem Bauchraum abpunktiert werden musste und wie viel Flüssigkeit dabei jeweils abgelassen wurde.

Wenn Sie eine Frau im gebärfähigen Alter sind, wird innerhalb von einer Woche vor Behandlungsbeginn ein Schwangerschaftstest durchgeführt.

Frühestens 7 Tage vor Behandlungsbeginn wird eine Urinprobe benötigt. Zudem wird Ihnen Blut abgenommen (35 ml, entspricht etwa 7 Teelöffeln), um verschiedene Routinelaborwerte zu bestimmen. Des Weiteren sollen in einem Teil der Blutprobe bestimmte Faktoren

analysiert werden, die möglicherweise eine Rolle bei der Entstehung der tumorbedingten Flüssigkeitsansammlung im Bauchraum bzw. einem Fortschreiten der Krebserkrankung selbst spielen. Zusätzlich möchten wir einige Patienten zur Blutspiegelmessung von Bevacizumab (Pharmakokinetik) einladen. Dazu ist eine gesonderte Aufklärung vorgesehen.

b) Während der Behandlung

Am Tag der ersten Behandlung werden Sie vor der ersten Gabe nochmals kurz untersucht, um sicherzustellen, dass Sie auch weiterhin für die Studienteilnahme geeignet sind. Sie werden gefragt, ob seit der letzten Untersuchung irgendwelche gesundheitlichen Beeinträchtigungen aufgetreten sind oder ob Sie in der Zwischenzeit neue Medikamente eingenommen haben bzw. einnehmen.

Sofern auch zu diesem Zeitpunkt nichts gegen Ihre Teilnahme an der klinischen Prüfung spricht, beginnt im Anschluss daran die Behandlung mit Abpunktieren der tumorbedingten Flüssigkeitsansammlung aus dem Bauchraum. 10 ml der entnommenen Flüssigkeit werden für die oben genannten wissenschaftlichen Untersuchungen verwendet werden, der Rest wird verworfen. Anschließend wird Bevacizumab oder das Placebo (Scheinmedikament) durch die bereits liegende Kanüle in den Bauchraum verabreicht werden. Für diese Gabe sind keinerlei zusätzliche Eingriffe notwendig. Die Verabreichung von Bevacizumab/Placebo (Scheinmedikament) erfordert einzig das Legen einer Kanüle in den Bauchraum und dieses muss ohnehin zum Abpunktieren der tumorbedingten Flüssigkeitsansammlung erfolgen. Bevacizumab oder das Placebo (Scheinmedikament) wird mittels Tropfinfusion (100 ml) in den Bauchraum verabreicht werden. Die erste Infusion dauert 60 Minuten. Die Dauer der nächsten Infusionen kann auf 30 Minuten reduziert werden, wenn während der vorangegangenen Infusion keine Nebenwirkungen aufgetreten sind.

Wie oft Sie innerhalb der achtwöchigen Behandlungsphase mit einer Punktion und anschließender Tropfinfusion mit Bevacizumab oder Placebo (Scheinmedikament) behandelt werden, hängt davon ab, wie schnell es zu einem Wiederauftreten der Flüssigkeitsansammlung kommt und wie rasch wieder eine Linderung der damit verbundenen Beschwerden notwendig wird. Maximal erhalten Sie jedoch 4 Behandlungen in einem Abstand von mindestens 14 Tagen.

Bei jeder Visite misst Ihr Prüfarzt vor den Medikationsgaben Ihre Vitalfunktionen (Blutdruck, Puls, Temperatur und Körpergewicht), führt eine körperliche Untersuchung durch und prüft, ob Eiweiß in Ihrem Urin nachweisbar ist. Wenn Sie Eiweiß im Urin haben, sind weitere Urinproben über einen Zeitraum von 24 Stunden notwendig. Des Weiteren wird bei jeder Visite Blut abgenommen (35 ml, entspricht etwa 7 Teelöffeln), um verschiedene Routinelaborwerte und die oben genannten Faktoren, die möglicherweise zur Entstehung des Bauchraumergusses und dem Fortschreiten der Krebserkrankung beitragen, zu bestimmen. Nach Abschluss dieser Untersuchungen entscheidet Ihr Prüfarzt, ob es in Ihrem besten Interesse liegt, die Behandlung wie zuvor fortzusetzen. Falls die Nebenwirkungen der Behandlung zu stark sein sollten, hat er die Behandlung zu verschieben oder die Behandlung komplett abzubrechen. Bitte teilen Sie Ihrem Prüfarzt auch sämtliche Veränderungen Ihres Gesundheitszustandes seit dem letzten Besuch mit.

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c) Ende der Behandlung und Weiterbeobachtung

Vier Wochen nachdem die 8-wöchige Behandlungsphase abgeschlossen ist, möchten wir Sie bitten, sich für eine abschließende Untersuchung bei ihrem behandelnden Arzt vorzustellen. Ihr Prüfarzt wird Sie dann nochmals körperlich untersuchen, Ihre Vitalfunktionen bestimmen und Ihren Allgemeinzustand einschätzen. Auch ist nochmals eine Blutentnahme von circa 35 ml (entspricht etwa 7 Teelöffeln) für die oben genannten Routineuntersuchungen und wissenschaftlichen Analysen notwendig. Bitte teilen Sie dem Prüfarzt auch bei diesem Besuch mögliche Veränderungen Ihres gesundheitlichen Zustandes mit.

Im Anschluss daran möchten wir Sie bitten, sich alle 2 Monate bei Ihrem Prüfarzt vorzustellen, damit er die Ausbreitung Ihrer Erkrankung (wie während der klinischen Prüfung) weiter beurteilen kann. Dies soll solange erfolgen, bis Sie insgesamt ein Jahr ab erstem Behandlungszyklus abgeschlossen haben.

Verantwortlichkeiten des Patienten

Von großer Wichtigkeit ist es, dass Sie zu allen vereinbarten Terminen erscheinen und sich genau an die Anweisungen Ihres Prüfarztes halten. Nennen Sie Ihrem Prüfarzt alle Begleiterkrankungen sowie alle Medikamente, die Sie zeitgleich mit der Studie einnehmen oder in den letzten Monaten eingenommen haben. Sollten Sie diese Medikamente benötigen, halten Sie bitte vorher mit Ihrem Prüfarzt Rücksprache. Bitte nennen Sie Ihrem Arzt auch immer Änderungen von Medikationsdosierungen. Sie müssen mit Ihrem Prüfarzt auch die mögliche Gabe neuer Medikamente wegen anderer Beschwerden besprechen. Dies schließt auch nicht verschreibungspflichtige (im Handel frei käufliche) und alternative Medikamente ein. Während Ihrer Teilnahme an dieser klinischen Prüfung dürfen Sie an keiner anderen klinischen Prüfung teilnehmen.

Alle gesundheitlichen Beschwerden und gesundheitliche Verbesserungen, die während Ihrer Teilnahme an dieser klinischen Prüfung neu auftreten, müssen Sie umgehend Ihrem Prüfarzt mitteilen. Dies gilt auch für Beschwerden, für die Sie keinen Zusammenhang mit Ihrer Teilnahme an dieser Prüfung vermuten. Bei erheblichen gesundheitlichen Beschwerden (z. B. Bluthusten o. ä.) müssen Sie sich sofort in medizinische (Notfall-) Behandlung begeben sowie sofort Kontakt mit Ihrem Prüfarzt aufnehmen!

Eine evtl. eintretende Schwangerschaft, bei Ihnen oder Ihrem Partner, während Ihrer Teilnahme an dieser Prüfung, muss umgehend gemeldet werden. In einem solchen Fall wird Sie Ihr Prüfarzt über die notwendigen Schritte ausführlich beraten. Die Beratung umfasst Informationen zu den Risiken der Fortführung der Schwangerschaft und mögliche Effekte auf die Entwicklung des Fötus.

Ein Abbruch der Studienbehandlung könnte möglicherweise die Wirkung auf den tumorbedingten Erguss beenden. Brechen Sie Ihre Studienbehandlung daher nicht ab, ohne vorher darüber mit Ihrem Prüfarzt zu sprechen.

Bitte kommen Sie im Falle eines vorzeitigen Studienabbruchs zur Abschlussuntersuchung.

4. Messung der Lebensqualität

Die Messung der Lebensqualität ist ein wichtiges Ziel in dieser Studie. Die Ergebnisse werden helfen herauszufinden, ob die Therapie mit Bevacizumab mögliche körperliche Symptome, die Ihre Lebensqualität beeinträchtigen, zu lindern vermag. Dazu werden 2 Lebensqualitätsfragebögen verwendet.

1. Durch Sie selber auszufüllen: FACIT-Aszites-Index (Functional Assessment of Chronic Illness therapy), bestehend aus 13 standardisierten symptomorientierten krankheitsbezogenen Fragen

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2. Befragung durch das medizinische Personal: Modifizierter Lebensqualitätsfragebogen der Palliativgruppe der Deutschen Gesellschaft für Hämatologie und Onkologie, insgesamt 25 symtomorientierte krankheitsbezogene Fragen

Der Zeitaufwand für Sie beträgt schätzungsweise 15 Minuten. Der Lebensqualitätsfragebogen soll von Ihnen vor Beginn der Studie, bei den 2-wöchigen Besuchen, sowie bei der Abschlussuntersuchung ausgefüllt werden. Die Auswertung erfolgt selbstverständlich pseudonymisiert. Diese Ergebnisse sind wichtig für zukünftige Therapieentscheidungen. Zudem ermöglichen Sie Ihren Ärzten und Studienauswertern, Ihre Lebens- und Behandlungssituation besser zu verstehen.

5. Welchen persönlichen Nutzen habe ich von der Teilnahme an der Studie?

Wir hoffen, dass Bevacizumab die Behandlung von tumorbedingten Ergüssen der Bauchhöhle verbessert und dabei hilft, die damit verbundenen Beschwerden wirksamer zu bekämpfen. Die Wirkung von in die Bauchhöhle applizierten Bevacizumab wurde bisher aber noch nicht eingehend untersucht.

Die Teilnahme an dieser Studie kann für Sie nützlich sein oder auch nicht. Wir hoffen, dass diese Studie Ihnen und anderen hilft, aber wir können nicht sagen, ob Sie einen direkten Nutzen haben werden. Ihre Teilnahme kann für andere hilfreich sein, indem durch ihre Behandlung wichtige Erkenntnisse über die Behandlung eines tumorbedingten Ergusses mittels Bevacizumab gewonnen werden, auch dann, wenn Sie nicht persönlich von der Behandlung profitieren. Durch die Teilnahme an dieser klinischen Prüfung werden Sie engmaschiger überwacht und so wird eine Verschlechterung Ihrer Erkrankung frühzeitig erkannt.

6. Mögliche Belastungen und Risiken

Um unnötige Risiken zu vermeiden, klärt Ihr Arzt ab, ob Sie für eine Studienteilnahme geeignet sind. Bitte informieren Sie Ihren Arzt zu Ihrer eigenen Sicherheit umfassend über bisher bestehende Erkrankungen und darüber, welche Medikamente Sie eingenommen haben bzw. noch einnehmen.

Die nachfolgend beschriebenen Nebenwirkungen sind bei Gabe von Bevacizumab in die Vene aufgetreten. Über die Nebenwirkungen von Bevacizumab bei Verabreichung in die Bauchhöhle liegen bisher nur wenige Erkenntnisse vor. Es ist aber davon auszugehen, dass die Nebenwirkungen bei intravenöser Gabe ausgeprägter sind als bei Gabe des Medikamentes in den Bauchraum.

Nebenwirkungen von Bevacizumab

Grundsätzlich kann Bevacizumab, wie alle Arzneimittel, Nebenwirkungen haben, die jedoch nicht bei jedem Patienten auftreten müssen. Diese Nebenwirkungen können unterschiedlich schwer sein. Sie können lebensbedrohlich sein und unter Umständen auch zum Tod führen. Informieren Sie bitte Ihren Arzt, wenn eine der aufgeführten Nebenwirkungen Sie erheblich beeinträchtigt oder Sie Nebenwirkungen bemerken, die nicht in dieser Patienteninformation angegeben sind. Ihr Arzt kann die Behandlung vorübergehend unterbrechen oder endgültig beenden, falls die Reaktion auf das Medikament schwerwiegend ist. Sie werden hinsichtlich der möglichen Nebenwirkungen des Medikaments sorgfältig überwacht. Sie werden außerdem über neue Erkenntnisse informiert, die Ihre Bereitschaft, die Studie fortzusetzen, beeinflussen könnten. Sie werden dann gebeten, auf einem Formblatt mit Ihrer Unterschrift zu bestätigen, dass mit Ihnen über diese neuen Informationen gesprochen wurde.

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Durch die intraperitoneale (in den Bauchraum) Gabe der Prüfmedikation kann es gelegentlich zu örtlichen Gewebeschäden (Spritzenabszesse, Nekrosen, Nervenreizungen) oder Entzündungen kommen. Wir bitten Sie daher, Ihren behandelnden Arzt über eventuelle Entzündungen oder Schwellungen an der Einstichstelle zu informieren. In der bisher durchgeführten Studie wurde ein Fall einer Transportstörung des Darms beschrieben. Außerdem traten milde Bauchschmerzen sowie leichte Übelkeit auf.

Das Gesamtsicherheitsprofil von Bevacizumab basiert auf den Daten von Patienten mit verschiedenen Krebserkrankungen, die im Rahmen klinischer Studien Bevacizumab entweder als alleinige Behandlung oder in Kombination mit einer Chemotherapie erhielten.

Die schwerwiegendsten unerwünschten Ereignisse waren:

- Lochbildung im Magen-Darm-Bereich
- Blutungen, einschließlich Lungeneinblutung/Bluthusten, die bei Patienten mit nicht kleinzelligem Lungenkrebs häufiger auftreten
- Verschluss von Schlagadern durch ein Blutgerinnsel

Die am häufigsten beobachteten unerwünschten Ereignisse in den klinischen Studien waren bei Patienten unter Bevacizumab-Behandlung mit oder ohne Chemotherapie: Bluthochdruck, Kraftlosigkeit, Durchfall, Übelkeit und Bauchschmerzen.

Im Folgenden finden Sie die Nebenwirkungen, die in Kombination mit Bevacizumab berichtet wurden, nach Häufigkeiten sortiert.

Berichtete schwere Nebenwirkungen (Grad 3 - 5), die schwerwiegend sein können

Dieser Abschnitt ist so angeordnet, dass die schwersten Nebenwirkungen, die am häufigsten auftraten, zuerst behandelt werden. Die weniger schweren und weniger häufigen Nebenwirkungen werden im Anschluss daran dargestellt. Um zu beschreiben, wie häufig Nebenwirkungen auftreten und wie schwer die Symptome sein können, wurden Standard-Definitionen verwendet. Es wird immer der schlimmste mögliche Fall dargestellt.

Nebenwirkungen, die als „sehr häufig“ beschrieben werden, treten bei mehr als 1 von 10 Patienten auf, die Bevacizumab in Kombination mit anderen Arzneimitteln erhalten, während Nebenwirkungen, die als „häufig“ bezeichnet werden, bei mehr als 1 von 100 Patienten auftreten.

Schwerwiegende Nebenwirkungen sind solche, die entweder lebensbedrohlich sind oder, wenn sie nicht entsprechend behandelt werden, lebensbedrohlich sein können, sowie Nebenwirkungen, die einen Krankenhausaufenthalt erforderlich machen oder den Tod zur Folge haben. Schwere Nebenwirkungen müssen nicht unbedingt schwerwiegend oder lebensbedrohlich sein, aber sie bedürfen normalerweise einer Behandlung.

<u>Sehr häufig (bei mehr als 1 von 10 Patienten):</u>

- Bluthochdruck
- Blutgerinnsel in den Bein- und Beckenvenen bis hin zur Lungenembolie
- Periphere sensorische Neuropathie (z.B. Taubheitsgefühl oder Gefühllosigkeit in den Fingern oder Zehen oder Probleme bei feinmotorischen Tätigkeiten mit den Fingern, wie z.B. dem Zuknöpfen eines Hemdes)
- Leukopenie (Verminderung der weißen Blutkörperchen im Blut)
- Durchfall
- Müdigkeit, Kraftlosigkeit

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Häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten), geordnet nach Organsystemen:

Organsystemübergreifend:

- Blutungen, meist mit dem Tumor zusammenhängend, auch mit tödlichem Verlauf
- Wundheilungsstörungen

Herzerkrankungen:

- Herzschwäche, Herzrasen

Erkrankungen des Blutes und des Lymphsystems:

- Verringerung des Hämoglobingehaltes (roter Blutfarbstoff) im Blut, verringerte Anzahl von Blutplättchen im Blut

Allgemeine Erkrankungen und Beschwerden am Verabreichungsort:

- Schmerzen, Kraftlosigkeit, Muskelschwäche

Erkrankungen des Magen-Darm-Trakts:

- Bauchschmerzen, Darmverschluss, Erkrankung des Magen-Darm-Trakts, Lochbildung und Fistelbildung (abnorme Verbindung zwischen Organen) im Magen-Darm-Trakt jeweils auch mit tödlichem Verlauf

Erkrankungen der Nieren und Harnwege:

- Eiweiß im Urin

Infektionen und parasitäre Erkrankungen:

- Blutvergiftung, Eiteransammlung, Harnwegsinfektion, Infektion

Laboruntersuchungen:

- Verlängerte Blutgerinnungszeit

Stoffwechsel- und Ernährungsstörungen:

- Erniedrigter Kaliumgehalt (ein Mineralstoff) im Blut
- Mangel an Körperflüssigkeit (Dehydratation)

Erkrankungen des Nervensystems:

- Durchblutungsstörung des Gehirns, kurzdauernde Bewusstlosigkeit, Benommenheit mit starker Schläfrigkeit

Erkrankungen der Atemwege, des Brustraums und des Mittelfellraums:

- Atemnot, erniedrigte Sauerstoffversorgung des Körpergewebes
- Bluthusten, auch mit tödlichem Verlauf
- Blutungen in der Lunge, auch mit tödlichem Verlauf

Gefäßerkrankungen:

- Blutgerinnsel in den Schlagadern, die zu einem Schlaganfall oder einem Herzinfarkt führen können und tödlich verlaufen können

Sonstige Erkrankungen:

- In einigen klinischen Studien wurden anaphylaktische und anaphylaktoide Reationen (allergische Allgemeinreaktion) häufiger bei Patienten berichtet, die Bevacizumab in Kombination mit Chemotherapie erhielten als bei alleiniger Chemotherapie (bis zu 5%).

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Gelegentlich (bei mehr als 1 von 1000 und weniger als 1 von 100 Patienten):

Organsystemübergreifend:

- Fisteln (abnorme Verbindungen zwischen Organen) außerhalb des Magen-Darm-Trakts, auch mit tödlichem Verlauf

Wenn eine der oben genannten Nebenwirkungen bei Ihnen auftritt, sollten Sie unverzüglich ärztliche Hilfe aufsuchen und den Arzt über ihre Studienteilnahme informieren. Ferner muss umgehend der in der Studie für Sie zuständige Prüfarzt konsultiert und informiert werden.

Berichtete, nicht schwere Nebenwirkungen

Sehr häufig (bei mehr als 1 von 10 Patienten):

Allgemeine Erkrankungen und Beschwerden:

- Bluthochdruck
- Appetitlosigkeit
- Schleimhautblutungen, meist Nasenbluten
- Eiweiß im Urin

Erkrankungen des Magen-Darm-Trakts:

- Blutungen im Enddarm, Mundschleimhautentzündung, Verstopfung

Allgemeine Erkrankungen und Beschwerden am Verabreichungsort:

- Kraftlosigkeit, Schmerzen, Fieber

Häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten):

- Augenerkrankungen
- Verändertes Geschmacksempfinden
- Nasenbluten
- Nasenschleimhautentzündung
- Atemnot
- Schuppende Hautentzündung, Hautverfärbung, trockene Haut

Weitere, weniger häufige Nebenwirkungen:

- Zahnfleisch- und Vaginalblutungen

Weitere, seltene Nebenwirkungen (bei mehr als 1 von 10.000 und weniger als 1 von 1.000 Patienten):

- Epileptische Anfälle, Kopfschmerzen, Verwirrtheit oder Änderungen des Sehvermögens (RPLS; reversibles posteriores Leukoenzephalopathie-Syndrom)

Weitere, sehr seltene Nebenwirkungen (bei weniger als 1 von 10.000 Patienten):

- Loch in der Nasenscheidewand (= Trennwand zwischen den beiden Nasenlöchern)
- Krankhafte Hirnveränderung infolge Bluthochdrucks (hypertensive Enzephalopathie), auch tödlich verlaufend

Weitere Nebenwirkungen bei Studien zum Lungenkrebs:

- Wirbelkörperbruch

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Weitere Nebenwirkungen bei Studien zum metastasierten Nierenkrebs

Schwerwiegend und häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten)

- Bluthusten

Nicht schwerwiegend und sehr häufig (bei mehr als 1 von 10 Patienten)

- Blut im Urin

Nicht schwerwiegend und häufig (bei mehr als 1 von 100 und weniger als 1 von 10 Patienten

- Bluthusten

Wenn eine der oben genannten Nebenwirkungen bei Ihnen auftritt, sollten Sie sobald wie möglich ärztliche Hilfe aufsuchen.

Veränderungen von Laborwerten

Unter Bevacizumab Therapie kann es zu einer Veränderung der folgenden Laborwerte kommen:

1. Verringerte Anzahl weißer Blutkörperchen, insbesondere der Neutrophilen im Blut (eine bestimmte Art weißer Blutkörperchen)
2. erniedrigter Hämoglobinwert im Blut
3. erniedrigter Natrium-, Kalium- und Phosphorgehalt im Blut (Mineralstoffe)
4. erhöhte alkalische Phosphatase im Blut (ein Enzym)
5. erhöhter Blutzucker
6. Eiweiß im Urin

Hinweis:
Eiweiß im Urin tritt unter der Anwendung von Bevacizumab sehr häufig auf. Der Schweregrad reicht von vorübergehender, leichter Ausprägung bis hin zu einer schwerwiegenden Ausprägung (nephrotisches Syndrom). In der Mehrzahl besteht eine leichte Symptomatik.

Belastung im täglichen Leben durch die Studienteilnahme

Mit Ausnahme der zusätzlichen Gabe von Bevacizumab oder Placebo (Scheinmedikament), was jeweils einen etwa 2-3 Stunden längeren Aufenthalt in der Klinik bedeutet, unterscheidet sich der zeitliche Aufwand und damit die Belastung im täglichen Leben nicht wesentlich davon, als wenn lediglich die übliche Abpunktierung des Bauchraumergusses vorgenommen wird.

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7. Wer darf an dieser klinischen Studie nicht teilnehmen?

An dieser klinischen Prüfung dürfen Sie nicht teilnehmen, wenn Sie gleichzeitig an anderen klinischen Prüfungen oder anderen klinischen Forschungsprojekten teilnehmen oder vor kurzem (innerhalb von 4 Wochen) teilgenommen haben.

Schwangere Frauen dürfen an dieser klinischen Prüfung **nicht teilnehmen**.

Zu Beginn der klinischen Prüfung müssen sich deshalb alle Frauen einem Schwangerschaftstest unterziehen. Davon ausgenommen sind Frauen nach den Wechseljahren oder solche, die operativ sterilisiert wurden. Durch einen Schwangerschaftstest kann jedoch eine Schwangerschaft erst einige Tage nach der Empfängnis verlässlich nachgewiesen werden.

Im Falle Ihrer Teilnahme an dieser klinischen Prüfung müssen Sie zuverlässige Maßnahmen zur Schwangerschaftsverhütung bis mindestens sechs Monate nach der letzten Behandlung anwenden. Diese sind die Hormonspirale, die Depot-Spritze (Drei-Monats-Spritze), ein hormonabgebendes Implantat (Verhütungsstäbchen, welches in den Oberarm implantiert wird), die Transdermale Kontrazeption (Verhütungspflaster), Abstinenz oder die Sterilisation des Partners. Da es bei Ihrem Krankheitsbild eventuell zu Erbrechen und Durchfällen kommen kann, ist eine Empfängnisverhütung durch die Pille allein nicht ausreichend. Der männliche Partner sollte ein Kondom benutzen.

Der Grund dafür ist, dass aus Tierversuchen Belege für ein erhöhtes Risiko einer Schädigung des ungeborenen Lebens vorliegen.

Sollten Sie während der klinischen Prüfung schwanger werden oder den Verdacht haben, dass Sie schwanger geworden sind, müssen Sie umgehend den Prüfarzt informieren.

Auch **stillende Frauen** dürfen an dieser klinischen Prüfung **nicht teilnehmen**, da Bevacizumab mit der Muttermilch in den Körper des Kindes gelangen und zu seiner Schädigung führen könnten. Während einer Behandlung und auch noch mindestens sechs Monate nach der letzten Dosis von Bevacizumab dürfen Sie nicht stillen, da Bevacizumab das Wachstum und die Entwicklung Ihres Kindes beeinträchtigen kann.

Männer, die an dieser klinischen Prüfung teilnehmen möchten, müssen durch Abstinenz oder eine sichere Verhütung mit einer der oben geschilderten Methoden für Ihre Partnerin (Hormonspirale, Depot-Spritze, hormonabgebendes Implantat, orale Kontrazeption plus Kondom) sicherstellen, während der Studienteilnahme sowie über einen Zeitraum von einem halben Jahr über das Ende der Behandlung hinaus keine Kinder zu zeugen, da die Chemotherapie zu Missbildungen bei Ungeborenen führen kann. Sollte Ihre Partnerin trotz der zwingend erforderlichen Verhütung während Ihrer Teilnahme an der Studie oder innerhalb von 6 Monaten nach Behandlungsende schwanger werden, ist Ihr Prüfarzt zu informieren.

8. Informationen zur Basistherapie

Über die Nebenwirkungen und Risiken der Basistherapie mittels Abpunktion des tumorbedingten Ergusses wird Ihr behandelnder Arzt Sie informieren. Bitte beachten Sie, dass die Nebenwirkungen und Risiken dieser Basistherapie auch ohne Studienteilnahme auf Sie zukommen würden.

9. Belastung durch Studienprozedere (Blutentnahmen)

Im Verlauf der klinischen Prüfung werden Ihnen einige Blutproben entnommen. Mit Ausnahme der 20 ml Blut (etwa 4 Teelöffel), die speziell für die Bestimmung der prognostischen Faktoren (s. Seite 4) entnommen werden, entsprechen alle anderen Blutentnahmen der klinischen Routine. Die Entnahme einer Blutprobe ist grundsätzlich nur mit einem geringen Risiko verbunden. An der Einstichstelle kann es zu leichten Schmerzen kommen oder es kann ein blauer Fleck (Bluterguss) entstehen, der evtl. einige Tage sichtbar ist. In äußerst seltenen Fällen kann auch die Bildung eines Blutgerinnsels (Thrombose), eine örtliche begrenzte Entzündung, eine Verletzung von Nerven oder eine Infektion an der Einstichstelle auftreten.

10. Andere therapeutische Möglichkeiten und Behandlungen sowie deren potentielle Vorteile und Risiken

Eine alternative kürzlich zugelassene Behandlungsmöglichkeit ist die Therapie mit dem Antikörper Catumaxumab, der ebenfalls in den Bauchraum gespritzt werden muss. Bei dieser Therapie ist allerdings eine 4malige Applikation innerhalb von 10 Tagen über einen dauerhaft liegenden Verweilkatheder notwendig. Die Therapieeinleitung dazu sollte unbedingt stationär erfolgen. Ihr Arzt hat Sie über die möglichen Vor-und Nachteile dieser Therapie ausführlich aufgeklärt. Eine Vorbehandlung oder auch Nachbehandlung mit Catumaxumab ist kein Ausschlusskriterium für die Studie.

Die Teilnahme an dieser Studie ist völlig freiwillig. Wenn Sie sich gegen eine Teilnahme entscheiden oder wenn Sie sich zu irgendeinem Zeitpunkt entscheiden, Ihre Einwilligung zurückzunehmen, werden Ihnen alternative Möglichkeiten, wie z. B. eine alleinige Abpunktion des Ergusses angeboten. Sie sollten die alternativen Behandlungen mit Ihrem Arzt besprechen bevor Sie entscheiden, diese Studie nicht fortzusetzen.

11. Freiwilligkeit der Teilnahme an der Studie

Die Teilnahme an dieser Studie ist freiwillig. Voraussetzung für die Studienteilnahme ist Ihre schriftliche Einwilligung. Mit Ihrer Unterschrift bestätigen Sie, dass Sie von Ihrem behandelnden Arzt über Wesen, Bedeutung, Risiken und Tragweite der klinischen Prüfung (der auch dieses Informationsblatt dient) aufgeklärt worden sind und dass Sie an dieser Studie teilnehmen wollen.

Sie werden auch während der Studie rechtzeitig über sich neu ergebende Besonderheiten oder Risiken der Studie, die für Ihre Teilnahme (oder den Behandlungsabbruch) relevant sein könnten, informiert werden.

Die zu Studienbeginn erteilte Einwilligung zur Teilnahme an der Prüfung kann jederzeit und ohne Angabe von Gründen widerrufen werden, ohne dass Ihnen daraus Nachteile entstehen oder das Vertrauensverhältnis zum behandelnden Arzt in irgendeiner Weise leidet.

12. Kosten für den Patienten

Durch die Teilnahme an dieser klinischen Studie entstehen für Sie keine zusätzlichen Kosten. Sie erhalten keine Aufwandsentschädigung. *Fahrtkosten werden in der Regel nicht übernommen. Bitte wenden Sie sich an Ihren Prüfarzt, um zu überprüfen, ob im Einzelfall eine Ausnahme möglich ist.*

13. Bin ich während der klinischen Prüfung versichert?

Bei der klinischen Prüfung eines Arzneimittels sind alle Studienteilnehmer gemäß dem Arzneimittelgesetz versichert. Der Umfang des Versicherungsschutzes ergibt sich aus den Versicherungsunterlagen, die Sie ausgehändigt bekommen.

Wenn Sie vermuten, dass durch die Teilnahme an der klinischen Prüfung Ihre Gesundheit geschädigt oder bestehende Leiden verstärkt wurden, müssen Sie dies unverzüglich dem Versicherer

Name und Anschrift der Versicherung:
HDI-Gerling Industrie Versicherung
Märkische Str. 23-33
44141 Dortmund
Telefon: 0231 / 5481-492
Fax: 0231 / 5481-302
Versicherungsscheinnummer: 48 158388 03055 390

direkt anzeigen, gegebenenfalls mit Unterstützung durch Ihren Prüfarzt, um Ihren Versicherungsschutz nicht zu gefährden. Sofern Ihr Prüfarzt Sie dabei unterstützt, erhalten Sie eine Kopie der Meldung. Sofern Sie Ihre Anzeige direkt an den Versicherer richten, informieren Sie bitte zusätzlich Ihren Prüfarzt.

Bei der Aufklärung der Ursache oder des Umfangs eines Schadens müssen Sie mitwirken und alles unternehmen, um den Schaden abzuwenden und zu mindern.

Während der Dauer der klinischen Prüfung dürfen Sie sich einer anderen medizinischen Behandlung – außer in Notfällen – nur nach vorheriger Rücksprache mit dem Prüfarzt unterziehen. Von einer erfolgten Notfallbehandlung müssen Sie den Prüfarzt unverzüglich unterrichten.

Sie erhalten ein Exemplar der Versicherungsbestätigung einschließlich der Versicherungsbedingungen. *Wir weisen Sie insbesondere auf Punkt 1.4 (zu den Ausschlüssen), Punkt 3.1 (zum Umfang der Leistungen) und Punkt 4.3 sowie Punkt 4.4. (zu Ihren Obliegenheiten) hin.")*

Wir weisen Sie ferner darauf hin, dass Sie auf dem Weg von und zur Prüfstelle nicht unfallversichert sind.

14. Was geschieht mit meinen Daten

Während der klinischen Prüfung werden medizinische Befunde und persönliche Informationen von Ihnen erhoben und in der Prüfstelle in Ihrer persönlichen Akte niedergeschrieben oder elektronisch gespeichert. Die für die klinische Prüfung wichtigen Daten werden zusätzlich in pseudonymisierter Form gespeichert, ausgewertet und gegebenenfalls weitergegeben.

Pseudonymisiert bedeutet, dass keine Angaben von Namen oder Initialen verwendet werden, sondern nur ein Nummern- und/oder Buchstabencode, evtl. mit Angabe des Geburtsjahres. Nur im Zusammenhang mit der Meldung schwerwiegender unerwünschter Ereignisse erfolgt zusätzlich die Übermittlung des Geburtsdatums.

Die Daten sind gegen unbefugten Zugriff gesichert. Eine Entschlüsselung erfolgt nur unter den vom Gesetz vorgeschriebenen Voraussetzungen.

Das Arzneimittelgesetz enthält nähere Vorgaben für den erforderlichen Umfang der Einwilligung in die Datenerhebung und -verwendung. **Einzelheiten, insbesondere zur Möglichkeit eines**

Widerrufs, entnehmen Sie bitte der Einwilligungserklärung, die im Anschluss an diese Patienteninformation abgedruckt ist.

15. Wer entscheidet, ob ich aus der klinischen Studie ausscheide?

Es kann Gründe geben, dass die Studienbehandlung bei Ihnen vorzeitig abgebrochen werden muss. Zum Beispiel, wenn Ihr Arzt den Eindruck hat, dass eine weitere Teilnahme an der klinischen Prüfung nicht in Ihrem gesundheitlichen Interesse ist, oder bei Ihnen eine schwere Krankheit auftritt. Dasselbe gilt, wenn bei Ihnen zu starke Nebenwirkungen auftreten.

Auf jeden Fall wird bei einem Progress, d.h. einem Fortschreiten der Entwicklung des tumorbedingten Bauchraumergusses, die Behandlung im Rahmen der klinischen Prüfung abgebrochen. Unabhängig vom Grund des Abbruches führt der Arzt dann eine Abschlussuntersuchung durch und bespricht mit Ihnen die weitere Therapie.

Gegebenenfalls kann auch der Sponsor der Studie, die Studien-gGmbH der Arbeitsgemeinschaft Internistische Onkologie, die Entscheidung treffen, die gesamte klinische Prüfung abzubrechen oder lediglich Ihre Teilnahme zu beenden. Neben den oben genannten Sicherheitsgründen, wären weitere Gründe um Ihre Teilnahme vorzeitig zu beendigen zum Beispiel, wenn Sie sich nicht an die vereinbarten Besuchstermine hielten oder auch wenn Sie keine weitere Tumorbehandlung benötigen würden.

Selbstverständlich haben auch Sie das Recht, die Behandlung im Rahmen dieser klinische Prüfung jederzeit auf eigenen Wunsch abzubrechen (siehe auch Seite 12; *Freiwilligkeit der Teilnahme*). Bitte kommen Sie im Falle eines vorzeitigen Abbruchs der klinischen Prüfung zur Abschlussuntersuchung.

Der Prüfarzt wird mit Ihnen besprechen, wie und wo Ihre weitere Behandlung stattfindet.

16. Was geschieht mit meinen Blutproben und Aszitesproben?

Die Blutproben und Aszitesproben werden ausschließlich für diese klinische Prüfung verwendet. Etwaiges Restmaterial wird bei Abschluss der Prüfung vernichtet.

Zum einen werden anhand dieser Proben bei einem lokalen Labor, welches Ihr Prüfarzt routinemäßig nutzt, bestimmte Routinelaborwerte zur Beurteilung Ihres Gesundheitsstatus bestimmt. Zu diesem gehören bei der Untersuchung Ihres Blutes ein Blutbild, Bestimmung von Gerinnungsfaktoren, verschiedener Enzyme Enzyme, Elektrolyte und Spurenelemente. In der Aszitesflüssigkeit wird routinemäßig der Blut- und Proteingehalt ermittelt.

Außerdem sollen mit Hilfe dieser Proben bestimmte Faktoren analysiert werden, die möglicherweise eine Rolle bei der Entstehung der tumorbedingten Flüssigkeitsansammlung im Bauchraum bzw. einem Fortschreiten der Krebserkrankung selbst spielen. In diesem Zusammenhang werden Botenstoffe untersucht, die in der Steuerung der Gefäßneubildung und der Gefäßdurchlässigkeit von Bedeutung sind.

Diese Untersuchungen der Proben finden im Labor für Tumorbiologie von Herrn Dr. Djordje Atanackovic in der Universitätsklinik Eppendorf in Hamburg statt. Ihre Proben und erhobenen Daten werden in pseudonymisierte Form an das Labor weitergegeben.

17. An wen wende ich mich bei weiteren Fragen?

Bei Unklarheiten, Notfällen, unerwarteten oder unerwünschten Ereignissen, die während der Studie oder nach deren Abschluss auftreten oder für weitere Fragen, die Ihre Rechte als Patient dieser klinischen Studie betreffen, können Sie sich jederzeit an die untenstehende Kontaktperson wenden:

Prüfarzt:

Name des Arztes: _____

Anschrift: Musterklinik, Hämatologie und Onkologie

Musterstraße 1, 12345 Musterstadt

Telefon-Nr.: _____

Eine unabhängige Kontaktstelle zur Information über klinische Prüfungen ist beim PEI (Paul-Ehrlich-Institut) eingerichtet

Paul Ehrlich Institut
Paul-Ehrlich-Straße 51-59
63225 Langen
Tel: 06103 – 771 810
Fax: 06103 – 771 275
E-Mail: klinpruefung@pei.de

Sponsor:
Studien-gGmbH der Arbeitsgemeinschaft Internistische Onkologie (AIO)
in der Deutschen Krebsgesellschaft e.V.
Kuno-Fischer-Straße 8
14057 Berlin

Beauftragte Unternehmen
Auftragsforschungsinstitut (Contract Research Organisation,CRO)
GSO Gesellschaft für Studienmanagement und Onkologie mbH
Harvestehuder Weg 21
20148 Hamburg

Leiter der klinischen Prüfung (LKP)
Dr. Karin Jordan
Universitätsklinikum Halle
Onkologie/Hämatologie
Department für Innere Medizin IV
Ernst-Grube-Str. 40
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Emil-Barell-Str. 1
79639 Grenzach-Wyhlen

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Patienteneinwilligungserklärung

Studiennummer: AIO-SUP-0108

EudraCT-Nummer: 2009-014725-16

Titel der Studie: Eine randomisierte, doppelblinde, Placebo kontrollierte Phase II-Studie zur Untersuchung der Wirksamkeit von Bevacizumab zur Linderung vom Symptomen bei Patienten mit malignem Aszites bei fortgeschrittenen Krebserkrankungen gastrointestinalen Ursprungs

Ich, ... geb. am .....

wurde von ..... (Name des Prüfarztes)

über Wesen, Zielsetzung und Ablauf, Bedeutung und Tragweite der klinischen Prüfung des Präparates Bevacizumab (Avastin®) aufgeklärt und eingehend über Wirksamkeit, Nebenwirkungen sowie Risiken dieses Präparates informiert; eine Kopie dieser schriftlichen Patienteninformation und Einwilligungserklärung wurde mir ausgehändigt; ich hatte ausreichend Zeit, diese zu lesen und mich zu entscheiden. Den Inhalt habe ich verstanden. Der Prüfarzt steht mir darüber hinaus jederzeit für ein Beratungsgespräch über weitere Einzelheiten der klinischen Prüfung zur Verfügung.

Ich erkläre mich mit der Behandlung einverstanden. Mir ist bekannt, dass ich berechtigt bin, die Teilnahme an der klinischen Prüfung jederzeit ohne Angabe von Gründen zu beenden ohne dass mir daraus Nachteile entstehen.

Ich wurde darüber informiert, dass gemäß Arzneimittelgesetz eine Probandenversicherung besteht.

Ort	Datum	Unterschrift

(Muss von der Patientin/vom Patienten eigenhändig ausgefüllt werden)

Vom Prüfarzt auszufüllen:

Kommentar oder Notizen zum Aufklärungsgespräch:

Ich habe heute Herrn/Frau ..... aufgrund der mir vorliegenden Unterlagen (Patienteninformation/-einwilligungserklärung, Prüfplan, Prüfvereinbarung) über die klinische Prüfung im obigen Sinne aufgeklärt. **Die Patienteninformation wurde ihm/ihr ausgehändigt.**

Ort	Datum	Unterschrift und Stempel

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Datenschutzrechtliche Einwilligung

Mir ist bekannt, dass bei dieser klinischen Prüfung personenbezogene Daten, insbesondere medizinische Befunde über mich erhoben, gespeichert und ausgewertet werden sollen. Die Verwendung der Angaben über meine Gesundheit erfolgt nach gesetzlichen Bestimmungen und setzt vor der Teilnahme an der klinischen Prüfung folgende freiwillig abgegebene Einwilligungserklärung voraus, das heißt ohne die nachfolgende Einwilligung kann ich nicht an der klinischen Prüfung teilnehmen.

1. Ich erkläre mich damit einverstanden, dass im Rahmen dieser klinischen Prüfung personenbezogene Daten, insbesondere Angaben über meine Gesundheit, über mich erhoben und in Papierform sowie auf elektronischen Datenträgern bei/in GSO, Hamburg. Soweit erforderlich, dürfen die erhobenen Daten pseudonymisiert (verschlüsselt) weitergegeben werden:

   a) an die AIO-Studien-gGmbH, den Sponsor oder eine von diesem beauftragte Stelle zum Zwecke der wissenschaftlichen Auswertung,

   b) im Falle eines Antrags auf Zulassung: an den Antragsteller und die für die Zulassung zuständige Behörde, das Paul-Ehrlich-Institut,

   c) im Falle unerwünschter Ereignisse: an AIO-Studien-gGmbH, den Sponsor, an die jeweils zuständige Ethik-Kommission und die zuständige Bundesoberbehörde, das Paul-Ehrlich-Institut, sowie von dieser an die Europäische Datenbank und

   d) die Roche Pharma AG, Emil-Barell-Str. 1, 79639 Grenzach-Wyhlen

2. Außerdem erkläre ich mich damit einverstanden, dass autorisierte und zur Verschwiegenheit verpflichtete Beauftragte des Sponsors sowie die zuständigen Überwachungsbehörden in meine beim Prüfarzt vorhandenen personenbezogenen Daten, insbesondere meine Gesundheitsdaten, Einsicht nehmen, soweit dies für die Überprüfung der ordnungsgemäßen Durchführung der Studie notwendig ist. Für diese Maßnahme entbinde ich den Prüfarzt von der ärztlichen Schweigepflicht.

3. Die Einwilligung zur Erhebung und Verarbeitung meiner personenbezogenen Daten, insbesondere der Angaben über meine Gesundheit, ist unwiderruflich. Ich bin bereits darüber aufgeklärt worden, dass ich jederzeit die Teilnahme an der klinischen Prüfung beenden kann. Im Fall eines solchen Widerrufs meiner Einwilligung, an der Studie teilzunehmen, erkläre ich mich damit einverstanden, dass die bis zu diesem Zeitpunkt gespeicherten Daten weiterhin verwendet werden dürfen, soweit dies erforderlich ist, um

   a) Wirkungen des zu prüfenden Arzneimittels festzustellen,

   b) sicherzustellen, dass meine schutzwürdigen Interessen nicht beeinträchtigt werden,

   c) der Pflicht zur Vorlage vollständiger Zulassungsunterlagen zu genügen.

4. Ich erkläre mich damit einverstanden, dass meine Daten nach Beendigung oder Abbruch der Prüfung mindestens zehn Jahre aufbewahrt werden, wie es die Vorschriften über die klinische Prüfung von Arzneimitteln bestimmen. Danach werden meine personenbezogenen Daten gelöscht, soweit nicht gesetzliche, satzungsmäßige oder vertragliche Aufbewahrungsfristen entgegenstehen.

5. Ich bin über folgende gesetzliche Regelung informiert: Falls ich meine Einwilligung, an der Studie teilzunehmen, widerrufe, müssen alle Stellen, die meine personenbezogenen Daten, insbesondere Gesundheitsdaten, gespeichert haben, unverzüglich prüfen, inwieweit die gespeicherten Daten für die in Nr. 3 a) bis c) genannten Zwecke noch erforderlich sind. Nicht mehr benötigte Daten sind unverzüglich zu löschen.

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Musterklinik, Hämatologie und Onkologie
Musterstraße 1, 12345 Musterstadt
Site No.1

6. Ich bin damit einverstanden, dass mein Hausarzt über meine Teilnahme an dieser Studie informiert wird und Informationen über meinen Gesundheitszustand an den Prüfarzt weiterleiten darf

   .....

   Name

   über meine Teilnahme an der klinischen Prüfung informiert wird (falls nicht gewünscht, bitte streichen).

Ort	Datum	Unterschrift

(Muss von der Patientin/vom Patienten eigenhändig ausgefüllt werden)

Patienteninformation/-einwilligungserklärung

zur begleitenden Pharmakokinetikuntersuchung der Studie SUP-0108

Studiennummer: **AIO-SUP- 0108**

EudraCT-Nummer: **2009-014725-16**

Titel der Studie: **Eine randomisierte, doppelblinde, Placebo kontrollierte Phase II-Studie zur Untersuchung der Wirksamkeit von Bevacizumab zur Linderung vom Symptomen bei Patienten mit malignem Aszites bei fortgeschrittenen Krebserkrankungen gastrointestinalen Ursprungs**

Patientennummer: _____

Sehr geehrte Patientin,
sehr geehrter Patient,

Sie wurden gefragt, ob Sie an einer begleitenden Untersuchung zur klinischen Prüfung mit Bevacizumab, der so genannten Pharmakokinetik, teilnehmen wollen. Bevor Sie sich entscheiden, sollten Sie diese Patienteninformation ausführlich lesen und alle Fragen mit Ihrem Arzt besprechen.

Untersuchung der Pharmakokinetik

Bei der Pharmakokinetik handelt es sich um einen zusätzliche wissenschaftliche Untersuchung an Patienten, die an der Hauptprüfung teilnehmen. Ihre Teilnahme an dieser Zusatzuntersuchung ist freiwillig und unabhängig von Ihrer sonstigen Teilnahme an der klinischen Prüfung. Es entstehen Ihnen keine Nachteile, wenn Sie nicht daran teilnehmen wollen.

Auch wenn Sie dieser zusätzlichen Untersuchung nicht zustimmen möchten, können Sie dennoch an der klinischen Prüfung teilnehmen!

Bei der Pharmakokinetik handelt es sich um eine Messung der Konzentration des Medikamentes Bevacizumab in Ihrem Blut. Man erhält dadurch Information über die Verstoffwechselung des Medikamentes im Körper.
Für diese Untersuchung werden Ihnen insgesamt maximal 60 ml Blut entnommen. Es werden mehrere Blutentnahmen à jeweils 10 ml Blut (entspricht ca. 2 Teelöffel) zu folgenden Zeitpunkten während der klinischen Prüfung durchgeführt:

- Vor jeder Abpunktion der Bauchraumflüssigkeit mit anschließender Gabe des Studienmedikamentes

- Sollte während des 8-wöchigen Studienzeitraumes nur eine Abpunktion der Bauchraumflüssigkeit notwendig sein, erfolgt die Blutabnahme alle 14 Tage nach der letzten Abpunktion zu Ihren regulär geplanten Vorstellungsterminen.
- 4 Wochen nach Ende des Behandlungszeitraumes erfolgt die letzte Blutentnahme

Zusätzliche Termine außer den regulär vereinbarten Terminen, um die o.g. Blutentnahmen durchzuführen, sind nicht vorgesehen.
Die Proben werden im Forschungslabor der Firma Xendo in den Niederlanden untersucht.

Die Ergebnisse dieser Untersuchung werden weder Ihnen noch Ihrem Prüfarzt zugänglich gemacht noch haben sie einen Einfluss auf Ihre Behandlung. Es entstehen Ihnen keine zusätzlichen Kosten durch diese Untersuchung. Für den Fall, dass Ergebnisse dieses Tests dazu benutzt werden, Patente oder Lizenzen anzumelden, ist nicht vorgesehen, dass Sie eine finanzielle Entschädigung erhalten.

Die Ergebnisse dieser Untersuchung können dazu beitragen, die Erkrankung und ihr Ansprechen auf eine bestimmte Behandlung besser zu verstehen. Persönlich können Sie keinen Nutzen aus dieser zusätzlichen Untersuchung ziehen, aber möglicherweise können zukünftig Patienten von den Erkenntnissen profitieren. Die mit dieser Zusatzuntersuchung verbundnen Risiken sind dieselben, die mit einer normalen Blutentnahme verbunden sind. Die Entnahme einer Blutprobe ist grundsätzlich nur mit einem geringen Risiko verbunden. An der Einstichstelle kann es zu leichten Schmerzen kommen oder es kann ein blauer Fleck (Bluterguss) entstehen, der evtl. einige Tage sichtbar ist. In äußerst seltenen Fällen kann auch die Bildung eines Blutgerinnsels (Thrombose), eine örtliche begrenzte Entzündung, eine Verletzung von Nerven oder eine Infektion an der Einstichstelle auftreten.

Datenschutzrechtliche Informationen

Im Rahmen dieser Untersuchungen ist auch geplant, Ihre krankheitsbezogenen Daten zu erheben. Ihre personenbezogenen Daten werden jederzeit vertraulich behandelt.

Die persönlichen Informationen, Informationen in Zusammenhang mit dem von Ihnen zur Verfügung gestellten Blutes und im Rahmen der Verlaufsbeobachtung Ihrer Erkrankung erhobene klinische Informationen (Alter, Geschlecht, Krankheitsstadium, Therapieansprechen, Nebenwirkungen der Therapie, weitere Therapien, Überlebensdaten) werden entsprechend der geltenden gesetzlichen Vorschriften pseudonymisiert und gespeichert.

Das heißt, dass Ihr Name und anderer Identifikationsmerkmale durch ein Kennzeichen ersetzt werden, mit dem Ziel, die Bestimmung des Ihrer Person auszuschließen oder wesentlich zu erschweren. Die Datenerhebung erfolgt ausschließlich zum Zweck des o.g. Forschungsvorhabens. Die Daten werden vom lokalen Prüfarzt verschlüsselt und in einer zentralen Datenbank gespeichert. Bei der Verarbeitung der Daten werden die Bestimmungen des Datenschutzgesetzes eingehalten.

Es wird nur Personen, die an den wissenschaftlichen Untersuchungen bzw. ihrer Auswertung beteiligt sind, Zugang zu Ihren Daten gewährt. Alle erhobenen Daten werden nur unter vollständiger Anwendung der geltenden gesetzlichen Bestimmungen zum Datenschutz behandelt. Im Falle von Veröffentlichungen bleibt die Vertraulichkeit Ihrer persönlichen Daten gewährleistet.

Aufgrund gesetzlicher Regelungen haben bestimmte Personen (autorisierte Dritte) ein Recht auf Einsichtnahme in Ihre personenbezogenen Daten. Dazu zählen Monitore, Auditoren, sonstige Beauftragte des Auftraggebers, Mitarbeiter der zuständigen Überwachungsbehörde oder der zuständigen Bundesoberbehörde. Die Einsichtnahme erfolgt nur im Rahmen der

gesetzlich geregelten Aufgaben der Einsichtnehmenden, nämlich zum Zweck der Überprüfung der Daten. Diese Personen sind zur Verschwiegenheit verpflichtet.

Die personenbezogenen Daten werden nach Ende des Forschungsvorhabens, spätestens jedoch nach 20 Jahren, gelöscht, soweit gesetzliche Vorgaben nicht längere Archivierungspflichten vorsehen.

Ihre Zustimmung zur Archivierung und Untersuchung Ihres Blutes ist vollkommen freiwillig und kann von Ihnen jederzeit widerrufen werden. In diesem Fall erfolgen mit Hilfe der Fallnummer die Löschung aller Ihrer gespeicherten Daten. Ihnen entstehen daraus keinerlei Nachteile hinsichtlich Ihrer Behandlung.

Das Forschungsvorhaben wurde durch die zuständige Ethikkommission ethisch geprüft und positiv bewertet. Wir möchten Sie darauf hinweisen, dass die Probandenversicherung der Hauptstudie ebenfalls die Risiken der hier beschriebenen begleitenden Untersuchungen abdeckt.

Ihre Zustimmung zur Untersuchung Ihres Blutes ist freiwillig und völlig unabhängig von Ihrer Zustimmung zur Teilnahme an der vorliegenden Therapiestudie. Sie können jederzeit ohne Angabe von Gründen die Teilnahme an der Studie beenden, ohne dass Ihnen dadurch Nachteile im Hinblick auf die Behandlung oder Ihr Verhältnis zu Ihrem behandelnden Arzt entstehen. Nach Beendigung Ihrer Teilnahme werden keine weiteren Daten von Ihnen erhoben. Ihre bisherigen Daten werden pseudonymisiert (d. h. Sie können nicht mehr anhand der Daten identifiziert werden).

Sollten weitere Fragen bezüglich der Studie haben, wenden Sie sich bitte an Ihren Prüfarzt:

Ich erkläre hiermit meine Bereitschaft zur Teilnahme an der zusätzlichen Untersuchung der Pharmakokinetik. Gleichzeitig erkläre ich meine Einwilligung zum Datenschutz	☐ Ja ☐ Nein

Eine Kopie dieser Patienteninformation und Einwilligungserklärung habe ich erhalten.

Ort	Datum	Patient	Unterschrift

(Muss von der Patientin/vom Patienten eigenhändig ausgefüllt werden)

Ort	Datum	Aufklärender Arzt	Unterschrift

16.1.4 List of sites

16.1.4 List of sites

Site	Address
01	Universitätsklinikum Halle-Wittenberg Klinik für Innere Medizin IV Onkologie / Hämatologie Ernst-Grube-Straße 40 06120 Halle (Saale)
02	Universitätsklinikum Hamburg-Eppendorf Onkologisches Zentrum Martinistraße 52 20246 Hamburg
03	Charité Campus Virchow-Klinikum Hämatologie und Onkologie Augustenburger Platz 1 13353 Berlin
~~04~~	~~Klinikum Ludwigsburg~~ ~~Klinik für Innere Medizin, Gastroenterologie, Hämatologie und Diabetologie~~ ~~Posilipostraße 4~~ ~~71640 Ludwigsburg~~
05	Onkologische Schwerpunktpraxis 311XX Hildesheim
~~06~~	~~Universitätsklinikum der Johannes-Gutenberg-Universität Mainz~~ ~~I. Medizinische Klinik und Poliklinik~~ ~~Langenbeckstraße 1~~ ~~55131 Mainz~~
07	Klinikum der Johann-Wolfgang-Goethe Universität Frankfurt (Main) Theodor-Stern-Kai 7 60590 Frankfurt
08	Kliniken Maria Hilf GmbH, Krankenhaus St. Franziskus Medizinische Klinik I Viersener Str. 450 41063 Mönchengladbach
~~09~~	~~Klinikum Fulda gAG~~ ~~Tumorklinik, Medizinische Onkologie/Hämatologie~~ ~~Pacelliallee 4~~ ~~36043 Fulda~~
10	Onkologische Schwerpunktpraxis 202XX Hamburg
~~11~~	~~Prosper-Hospital~~ ~~Medizinische Klinik I~~ ~~Mühlenstraße 27~~ ~~45659 Recklinghausen~~
12	Onkologische Schwerpunktpraxis 732XX Wendlingen
~~13~~	~~Ernst von Bergmann Klinikum~~ ~~Zentrum für Hämatologie, Onkologie und Strahlenheilkunde~~ ~~Charlottenstraße 72~~ ~~14467 Potsdam~~
14	Lahn-Dill-Kliniken GmbH Klinik für Hämatologie/Onkologie und Palliativmedizin Forsthausstraße 1 35578 Wetzlar

Site	Address
~~15~~	~~Klinikum Region Hannover GmbH~~ ~~Krankenhaus Siloah~~ ~~Medizinische Klinik III für Hämatologie und Onkologie~~ ~~Roesebeckstraße 15~~ ~~30449 Hannover~~
~~16~~	~~Onkologische Schwerpunktpraxis~~ ~~141XX Berlin~~
~~17~~	~~Onkologische Schwerpunktpraxis~~ ~~220XX Hamburg~~
~~18~~	~~Onkologische Schwerpunktpraxis~~ ~~224XX Hamburg~~
19	Klinikum Leverkusen gGmbH Medizinische Klinik III Am Gesundheitspark 11 51375 Leverkusen
~~20~~	~~Onkologische Schwerpunktpraxis~~ ~~018XX Neustadt (Sachsen)~~
22	Städtisches Klinikum Magdeburg Klinik für Allgemein- und Viszeralchirurgie Birkenallee 34 39130 Magdeburg
23	Vivantes Klinikum Spandau Klinik für Innere Medizin Neue Bergstraße 6 13585 Berlin
24	Evang. Huyssens-Stiftung Klinik für internistische Onkologie und Hämatologie Henricistraße 92 45136 Essen
25	Universitätsklinikum Essen Innere Klinik (Tumorforschung) Hufelandstr. 55 45147 Essen
26	Klinikum Deggendorf Medizinische Klinik II Perlasberger Str. 41 94469 Deggendorf
27	Vivantes Klinikum am Urban Onkologisches Zentrum Mitte Dieffenbachstr. 1 10967 Berlin
28	Vivantes Klinikum Neukölln Onkologisches Zentrum Vivantes Süd Rudower Straße 48 12351 Berlin
29	Friedrich-Ebert-Krankenhaus GmbH Klinik für Hämatologie/Onkologie/Nephrologie Friesenstr. 11 24534 Neumünster
31	Onkologische Schwerpunktpraxis Städteregion Aachen

10 sites were already deregistered during the study (crossed)

16.1.5 Signature LKP

Please refer to page 2 of the Clinical Study Report.

16.1.6 List of patients receiving test drug(s)/investigational product(s) from specific batches where more than one batch was used

NA

16.1.7 Randomisation scheme and codes

Please refer to Appendix 16.2.4

16.1.8 Audit certificates

NA

16.1.9 Documentation of statistical methods

Please refer to section 9.7.1

16.1.10 Documentation of inter-laboratory standardisation methods and quality assurance procedures if used

NA

16.1.11 Publications based on this study

2014 ESMO (Abstract)

Intraperitoneal Bevacizumab (Bev) for control of malignant ascites due to advanced-stage gastrointestinal cancers (GI): A multicenter double-blind, placebo (Pl)-controlled, randomized phase II study. AIO SUP-0108

Karin Jordan, Djordje Atanackovic, Christina Gog, Brigitta Killing, Michael Stahl, Werner Freier, Dirk Arnold, Jörn Rüssel, Axel Hinke, Siegfried Wagner, Ernst Späth-Schwalbe, Uwe Pelzer, Dirk Megdenberg, Susanna Hegewisch-Becker

Background: Malignant Ascites is debilitating for pts. with advanced cancer which negatively impacts the quality of life (QoL). Causative treatment approaches are still limited. It has previously been shown that tumor cell production and/or increases of Vascular Endothelial Growth Factor (VEGF) might be a major cause of the formation of malignant ascites. Intraperitoneal Bev could therefore be an option for symptom control of refractory malignant ascites.

Methods: Pts. with advanced GI cancer and malignant ascites who had received paracentesis at least once within the past 4 weeks were randomly assigned in a 2:1 ratio to intraperitoneal Bev (400mg absolute in 100 ml NaCl 0.9%) or Pl after paracentesis. During the 8-week treatment period, a minimum interval of 14d was kept between the applications of the study drug. First primary endpoint was the paracentesis-free survival (ParFS). Second primary endpoint was best response (BR) defined as the longest paracentesis-free period within the 12-week observation period. Further endpoints were overall survival, QoL, serum and ascites VEGF. ParFS and BR were compared using the logrank test and the Wilcoxon-Mann-Whitney test, respectively.

Results: 53 Patients (median age 63y) were randomized (37 Bev/16 Pl arm) whereas 49 pts. received at least one application of the study drug and qualified for the intention to treat analysis. The median ParFS was 14d (CI:11-17d) in the Bev arm and 10.5d (CI: 7-21d) in the Pl arm (hazard ratio 0.74, CI:0.40-1.37; p= 0.16). The BR was 19d for the Bev arm (range 6-66d) and 17.5d for the Pl arm (range 4-42d) with a p value of 0.85. Median OS was 64d (CI: 45-103d) for the Bev arm and only 31.5d (CI: 20-117d) for the Pl arm (p=0.31). The proportion of pts. with at least one CTC grade 3-5 event occurred was similar with 20/33 (61%) in the Bev arm and 11/16 (69%) in the Pl arm. Results on serum and ascites VEGF will be presented at the meeting.

Conclusions: In this unfavorable group of terminally ill pts. intraperitoneal Bev was well tolerated but did not result in a significantly better symptom control of malignant ascites compared to the control.

16.1.12 Important publications referenced in the report

NA

16.2 Patient data listings

16.2.1 Discontinued patients

16.2.1 Patient data listings: Discontinued patients

Patient no.	Randomisation group	Reason: death	Reason: other
0101	Bevacizumab	yes	
0102	Placebo	yes	
0103	Bevacizumab	yes	
0104	Bevacizumab	yes	
0105	Placebo	yes	
0106	Bevacizumab	yes	
0201	Bevacizumab	yes	
0203	Placebo	yes	
0207	Bevacizumab	yes	
0302	Placebo		non-protocol ascites treatment
0502	Bevacizumab	yes	
0503	Placebo	yes	
0702	Placebo	yes	
0703	Bevacizumab		patient's request
0705	Bevacizumab	yes	
0706	Bevacizumab		withdrawal of consent
0707	Bevacizumab	yes	
1006	Bevacizumab		investigator's decision (clinical progression)
1201	Bevacizumab	yes	
1402	Bevacizumab	yes	
1404	Bevacizumab		patient's request
1405	Bevacizumab	yes	
1406	Placebo	yes	
1407	Bevacizumab		pain in both legs due to open wounds
2301	Bevacizumab	yes	
2302	Placebo	yes	
2401	Placebo		investigator's decision (worsening of general condition)
2402	Bevacizumab		bacterial peritonitis
2403	Bevacizumab		hospitalisation due to bacterial peritonitis
2601	Bevacizumab		investigator's decision (worsening of general condition)
2701	Placebo	yes	

16.2.2 Protocol deviations

16.2.2 Protocol deviations

Patient No.	All inclusion criteria fulfilled yes/no	If no, no. of violated inclusion criteria	Any exclusion criteria violated yes/no	If yes, no. of violated exclusion criteria
0101	yes		no	
0102	yes		no	
0103	no	12[1]	no	
0104	yes		no	
0105	yes		no	
0106	yes		no	
0201	yes		yes	2, 3[3]
0202	yes		yes	2, 3, 4[4]
0203	yes		yes	2, 3, 4[4]
0204	no	12[5]	yes	2,3[3]
0205	no	12[6]	yes	3[2]
0206	yes		yes	2, 3, 4[4]
0207	yes		yes	2, 3, 4[4]
0301	no	12[5]	yes	2, 3[3]
0302	yes		no	
0501	yes		no	
0502	yes		yes	22[7]
0503	yes		no	
0701	yes		no	
0702	yes		no	
0703	yes		no	
0704	yes		yes	3[2]
0705	yes		yes	3[2], 22[7]
0706	yes		yes	2[8], 3[2]
0707	no	12[10]	yes	3, 4[9]
1001	yes		yes	10, 22[11]
1002	yes		no	
1003	yes		no	
1004	yes		no	
1005	yes		no	
1006	yes		no	
1201	no	12[12]	yes	2, 3[3]
1401	yes		no	
1402	yes		yes	22[7]
1403	yes		no	
1404	yes		no	
1405	no	5[13]	no	
1406	yes		no	
1407	yes		no	
1901	yes		no	
1902	yes		no	
2301	yes		yes	3[2]
2302	no	4[14], 12[12]	no	
2303	yes		no	
2401	no	12[5]	yes	2, 3, 4[4]
2402	no	12[5]	no	
2403	yes		no	

2404	yes		no	
2501	yes		no	
2601	no	12[5]	no	
2602	no	4[14], 8[15]	no	
2603	yes		no	
2701	yes		no	

[1] Alkaline Phosphatase and ASAT is elevated
[2] Hematocrit in ascites not assessable
[3] Hematocrit and neutrophil count in ascites not assessable
[4] Hematocrit, neutrophil count and protein in ascites not assessable
[5] Urinalysis not done
[6] Alkaline phosphatase not assessable
[7] Patient included with history of thrombosis < 6 months prior to treatment start
[8] Neutrophils in ascites > 250/µl
[9] Hematocrit and protein in ascites not assessable
[10] Alkaline Phosphatase is elevated
[11] Patient included with history of portal vein thrombosis and pulmonary embolism
[12] Creatinine clearance < 30ml/min
[13] Malignity of ascites not cytologically confirmed
[14] Histology of carcinoma not confirmed
[15] Only one paracentesis within 4 week screening phase (before amendment 02)

16.2.3 Patients excluded from the efficacy analysis

16.2.3 Patients excluded from the efficacy analysis

Four patients did not receive any paracentesis with study drug administration after randomisation, and, thus, were excluded from all quantitative analyses.

Exclusions due to lack of receiving any study drug administration

Pat. no.	Reason for early drop-out before any study treatment
# 0206	The patient was randomized on 16.01.12. On 19.01.12 an SAE occurred (severe hyponatremia), and the patient died because of the underlying tumor disease on 24.01.12.
# 0301	The patient was randomized on 14.01.11. On the same day, a severe pulmonary infection was diagnosed, precluding any study treatment. The patient died on 07.02.11 due to recurrent aspiration pneumonia.
# 1403	The patient was randomized on 22.06.11. As the general condition of the patient deteriorated following randomization, the patient was withdrawn prior to the first administration of study drug.
# 2603	The patient was randomized on 16.02.11. Start of therapy was planned for 18.02.11, but the patient did not come back to the site for treatment.

16.2.4 Demographic data

16.2.4 Patient data listings: Demographic data

Pat. no.	Randomisation group	Age	Gender
0101	Bevacizumab	58	male
0102	Placebo	66	male
0103	Bevacizumab	35	male
0104	Bevacizumab	50	female
0105	Placebo	66	male
0106	Bevacizumab	61	male
0201	Bevacizumab	64	female
0202	Bevacizumab	63	female
0203	Placebo	47	male
0204	Bevacizumab	62	female
0205	Bevacizumab	50	male
0207	Bevacizumab	67	female
0302	Placebo	50	female
0501	Bevacizumab	67	male
0502	Bevacizumab	70	male
0503	Placebo	53	male
0701	Bevacizumab	72	male
0702	Placebo	65	male
0703	Bevacizumab	69	female
0704	Placebo	72	male
0705	Bevacizumab	47	male
0706	Bevacizumab	67	female
0707	Bevacizumab	52	female
1001	Placebo	46	male
1002	Bevacizumab	72	male
1003	Bevacizumab	62	female
1004	Placebo	65	male
1005	Bevacizumab	61	male
1006	Bevacizumab	52	male
1201	Bevacizumab	71	female
1401	Placebo	67	female
1402	Bevacizumab	60	male
1404	Bevacizumab	81	female
1405	Bevacizumab	63	male
1406	Placebo	73	male
1407	Bevacizumab	74	male
1901	Bevacizumab	76	female
1902	Placebo	74	female
2301	Bevacizumab	44	female
2302	Placebo	75	male
2303	Bevacizumab	69	male
2401	Placebo	62	male
2402	Bevacizumab	52	female
2403	Bevacizumab	49	male
2404	Bevacizumab	49	male
2501	Bevacizumab	66	female
2601	Bevacizumab	51	male
2602	Placebo	61	female
2701	Placebo	73	male

16.2.5 Compliance and/or drug concentration data

NA

16.2.6 Individual efficacy response data

16.2.6 Patient data listings: Individual efficacy response data

Patient no.	Randomisation group	Paracentesis-free survival [days]	Event occurred	Best response [days]
0101	Bevacizumab	14	yes	16
0102	Placebo	6	yes	5
0103	Bevacizumab	5	yes	11
0104	Bevacizumab	28	yes	21
0105	Placebo	4	yes	4
0106	Bevacizumab	10	yes	9
0201	Bevacizumab	6	yes	10
0202	Bevacizumab	20	yes	35
0203	Placebo	8	yes	4
0204	Bevacizumab	7	yes	18
0205	Bevacizumab	16	yes	29
0207	Bevacizumab	19	yes	12
0302	Placebo	8	yes	42
0501	Bevacizumab	14	yes	15
0502	Bevacizumab	11	yes	19
0503	Placebo	7	yes	16
0701	Bevacizumab	15	yes	39
0702	Placebo	20	yes	19
0703	Bevacizumab	20	yes	14
0704	Placebo	21	yes	28
0705	Bevacizumab	10	yes	11
0706	Bevacizumab	15	yes	14
0707	Bevacizumab	13	yes	9
1001	Placebo	15	yes	21
1002	Bevacizumab	17	yes	29
1003	Bevacizumab	35	yes	44
1004	Placebo	22	yes	39
1005	Bevacizumab	40	yes	35
1006	Bevacizumab	13	yes	10
1201	Bevacizumab	4	yes	6
1401	Placebo	22	yes	40
1402	Bevacizumab	21	yes	20
1404	Bevacizumab	11	yes	61
1405	Bevacizumab	11	yes	7
1406	Placebo	6	yes	5
1407	Bevacizumab	24	yes	15
1901	Bevacizumab	14	yes	55
1902	Placebo	6	yes	21
2301	Bevacizumab	24	yes	23
2302	Placebo	9	yes	8
2303	Bevacizumab	8	yes	21
2401	Placebo	17	yes	9
2402	Bevacizumab	7	yes	28
2403	Bevacizumab	4	yes	6
2404	Bevacizumab	14	yes	28
2501	Bevacizumab	10	yes	21
2601	Bevacizumab	10	yes	66
2602	Placebo	14	yes	30
2701	Placebo	12	yes	8

16.2.7 Adverse events listings (each patient)

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
0101	Bevacizumab	01.07.2011	anemia	anemia	0	3	2
0101	Bevacizumab	17.06.2011	anemia	anemia	0	3	2
0101	Bevacizumab	10.07.2011	fatigue	fatigue	0	4	3
0101	Bevacizumab	10.07.2011	sepsis	Infection – Other (Specify, Blood)	1	4	2
0102	Placebo	18.11.2011	agitation	mood alteration: agitation	0	4	1
0103	Bevacizumab	25.01.2012	pneumothorax	pneumothorax	0	4	1
0103	Bevacizumab	19.01.2012	increase of CRP	Metabolic/Laboratory: Other - Increase of CRP	0	4	3
0103	Bevacizumab	18.01.2012	Keratitis on both sides	Keratitis	0	4	3
0103	Bevacizumab	ND.01.2012	AZ-worsening	ascites	0	4	3
0103	Bevacizumab	ND.01.2012	loss of weight	weight loss	0	4	3
0104	Bevacizumab	18.04.2012	hypocalcaemia	Calcium, serum-low	1	4	3
0105	Placebo	30.11.2012	Leukocytosis in Ascites	Metabolic/Laboratory – Other - Leukocytosis in Ascites	0	4	6
0105	Placebo	08.12.2012	Paraparesis due to osseus metastasis of BWK2	neuropathy: motor	1	4	6
0106	Bevacizumab	13.05.2013	anemia	anemia	0	4	1
0106	Bevacizumab	07.05.2013	Staph. Capilis in Ascites	Infection – Other - specify- staph. Capilis in Ascities	0	4	1
0106	Bevacizumab	ND.05.2013	Fatigue	fatigue	0	4	4
0106	Bevacizumab	NK.05.2013	tumor progression	n.a.	1	4	5
0201	Bevacizumab	19.07.2010	decubitus	Skin breakdown/ decubitus ulcer	0	4	3

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
0201	Bevacizumab	20.07.2010	(feaculent) vomiting	Gastrointestinal – Other - Specify - feaculent vomiting	0	4	1
0201	Bevacizumab	20.07.2010	duodenal compression	Gastrointestinal other: duodenal obstruction	1	4	3
0201	Bevacizumab	02.07.2010	diarrhea	diarrhea	0	4	1
0201	Bevacizumab	16.07.2010	infection	infection	0	4	1
0201	Bevacizumab	04.07.2010	painful micturation	Pain - RENAL/GENITOURINARY – Bladder	0	4	1
0201	Bevacizumab	11.07.2010	dyspnea	dyspnae	0	4	1
0201	Bevacizumab	21.07.2010	dyspnea	dyspnae	0	4	1
0201	Bevacizumab	16.07.2010	acute renal failure	renal failure	1	4	1
0202	Bevacizumab		no adverse events	n.a.			
0203	Placebo	04.10.2011	edema legs	edema: limb	0	4	1
0203	Placebo	04.10.2011	acute renal failure	renal failure	1	4	1
0204	Bevacizumab	04.01.2012	decubitus	Skin breakdown/ decubitus ulcer	0	4	1
0204	Bevacizumab	04.01.2012	obstipation	constipation	0	4	1
0204	Bevacizumab	18.12.2011	hyperglycemia	Glucose, serum-high (hyperglycemia)	1	4	1
0204	Bevacizumab	29.11.2011	dyspnea	dyspnae	0	4	1
0205	Bevacizumab	15.12.2011	anemia	anemia	0	4	1
0205	Bevacizumab	15.12.2011	fatigue	fatigue	0	4	3
0205	Bevacizumab	15.12.2011	weight loss	weight loss	0	4	3
0205	Bevacizumab	01.12.2011	nausea	nausea	0	4	1
0205	Bevacizumab	29.12.2011	nausea	nausea	0	4	3
0205	Bevacizumab	01.12.2011	vomiting	vomiting	0	4	1
0205	Bevacizumab	29.12.2011	vomiting	vomiting	0	4	3

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
0205	Bevacizumab	07.01.2012	constipation	constipation	0	4	3
0205	Bevacizumab	01.12.2011	diarrhea	diarrhae	0	4	1
0205	Bevacizumab	29.12.2011	heartburn	Heartburn/dyspepsia	0	4	1
0205	Bevacizumab	28.12.2011	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	1
0206	n/a	11.01.2012	hyponatremia	Sodium, serum-low (hyponatremia)	1	4	5
0207	Bevacizumab	18.12.2012	nausea	nausea	0	4	6
0207	Bevacizumab	18.12.2012	vomiting	vomiting	0	4	6
0301	n/a	14.01.2011	pulmonary infection	PULMONARY/UPPER RESPIRATORY – Lung (pneumonia)	1	NA	3
0302	Placebo	04.10.2011	subileus	ileus	1	4	1
0501	Bevacizumab	09.08.2011	loss of appetite (anorexia)	anorexia	0	4	3
0501	Bevacizumab	14.06.2011	dyspnea	dyspnae	0	4	3
0502	Bevacizumab	11.04.2012	bad general condition	constitutional symptoms - other - bad general condition	1	4	3
0502	Bevacizumab	10.04.2012	vomiting	vomiting	0	4	1
0502	Bevacizumab	10.04.2012	infection food	Infection – Other (Specify - food poisoning)	0	4	1
0503	Placebo	14.12.2012	anemia	anemia	0	4	1
0503	Placebo	02.01.2013	bad general condition	constitutional symptoms - other - bad general condition	1	4	
0701	Bevacizumab	19.11.2010	weakness	Muscle weakness, - Whole body/generalized	0	4	3
0701	Bevacizumab	19.11.2010	insomnia	insomnia	0	4	3
0701	Bevacizumab	19.11.2010	itching	Pruritis / itching	0	4	3

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
0701	Bevacizumab	19.11.2010	nausea	nausea	0	4	3
0701	Bevacizumab	19.11.2010	vomiting	vomiting	0	4	3
0701	Bevacizumab	20.12.2010	obstipation	constipation	0	4	6
0701	Bevacizumab	19.11.2010	diarrhea	diarrhea	0	4	1
0701	Bevacizumab	19.11.2010	anorexia	anorexia	0	4	3
0701	Bevacizumab	03.12.2010	ascites	ascites	0	4	6
0701	Bevacizumab	19.11.2010	depression	Mood alteration -depression	0	4	6
0701	Bevacizumab	20.12.2010	anxiety	Mood alteration - anxiety	0	4	6
0701	Bevacizumab	19.11.2010	dyspnea	dyspnae	0	4	3
0701	Bevacizumab	03.12.2010	pain	pain - unspecified	0	4	3
0702	Placebo	02.03.2011	edema limb	edema: limb	0	4	1
0702	Placebo	14.03.2011	increased tumor pain	pain: GENERAL Tumor pain	0	4	3
0702	Placebo	24.02.2011	pleural effusion	pleural effusion	1	4	3
0703	Bevacizumab		no adverse events	n.a.			
0704	Placebo		no adverse events	n.a.			
0705	Bevacizumab	07.02.2012	dyspnea	dyspnae	0	4	5
0705	Bevacizumab	07.02.2012	pulmonary embolism	Thrombosis/thrombus/embolism	1	4	5
0706	Bevacizumab		no adverse events	n.a.			
0707	Bevacizumab	28.02.2012	nausea	nausea	0	4	3
0707	Bevacizumab	19.04.2012	vomiting	vomiting	0	4	1
0707	Bevacizumab	04.04.2012	edema limb	edema: limb	0	4	3
0707	Bevacizumab	28.03.2012	urinary retention	urinary retention	0	4	3
0707	Bevacizumab	19.04.2012	worsening of general constitution	constitutional symptoms - other - bad general condition	1	4	3
0707	Bevacizumab	04.04.2012	pain back	Pain: MUSCULOSKELETAL – Back	0	4	1
1001	Placebo	29.09.2010	coagulopathy	coagulation / other	0	2	NK
1001	Placebo	01.09.2010	nausea	nausea	0	4	1

16.2.7 Adverse events listings (each patient)

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1001	Placebo	21.09.2010	anorexia	anorexia	0	4	3
1001	Placebo	21.09.2010	abdominal fullness	gastrointestinal other: fullness	0	4	3
1001	Placebo	05.09.2010	hematoma in the left flank region	hematoma	0	4	1
1001	Placebo	02.09.2010	pain post ascites puncture	pain - other - post ascites puncture	0	4	1
1001	Placebo	15.09.2010	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	NK
1001	Placebo	29.09.2010	dyspnea	dyspnae	0	NK	3
1001	Placebo	22.10.2010	tumor related pulmonary embolism	Thrombosis/thrombus/embolism	1	4	3
1002	Bevacizumab	08.11.2010	leucocytosis	Metabolic/Laboratory – Other (Specify - Leukocytosis	0	4	1
1002	Bevacizumab	29.10.2010	leukocytopenia	Metabolic/Laboratory – Other (Specify, Leukocytopenia	0	4	1
1002	Bevacizumab	29.09.2010	fatigue	fatigue	0	4	3
1002	Bevacizumab	27.10.2010	nausea	nausea	0	4	3
1002	Bevacizumab	27.10.2010	vomiting (intermittent)	vomiting	0	4	3
1002	Bevacizumab	20.10.2010	diarrhea	diarrhea	0	4	1
1002	Bevacizumab	16.10.2010	anorexia	anorexia	0	4	3
1002	Bevacizumab	09.12.2010	dehydration	dehydration	0	4	6
1002	Bevacizumab	02.12.2010	dysphagia	dysphagia	0	4	6
1002	Bevacizumab	21.10.2010	polyneuropathy	neuropathy:sensory	0	4	3
1002	Bevacizumab	14.09.2010	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	1
1002	Bevacizumab	29.09.2010	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	3

16.2.7 Adverse events listings (each patient)

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1002	Bevacizumab	14.09.2010	ardent feeling in the abdominal region	gastrointestinal other: ardent feeling	0	1	1
1003	Bevacizumab	02.02.2011	nausea	nausea	0	4	3
1003	Bevacizumab	02.02.2011	vomiting	vomiting	0	4	1
1003	Bevacizumab	NK.03.2011	constipation	constipation	0	4	6
1003	Bevacizumab	NK.04.2011	anorexia	anorexia	0	4	3
1003	Bevacizumab	NK.03.2011	heartburn	Heartburn/dyspepsia	0	4	6
1003	Bevacizumab	01.02.2011	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	3
1004	Placebo	01.05.2011	worsening of general constitution	constitutional symptoms - other - bad general condition	0	4	3
1004	Placebo	NK.06.2011	freezing	Rigors/ chills	0	4	3
1004	Placebo	07.05.2011	anorexia	anorexia	0	4	3
1005	Bevacizumab	02.11.2011	fatigue	fatigue	0	1	1
1005	Bevacizumab	30.10.2011	Acroedema	Edema:limb	0	4	3
1005	Bevacizumab	07.11.2011	Insomnia	Insomnia	0	4	1
1005	Bevacizumab	09.11.2011	Anemia	anemia	0	4	1
1005	Bevacizumab	16.11.2011	Edema on the inside of both ankles	edema: limb	0	4	3
1005	Bevacizumab	18.11.2011	thoracic rash	Rash/desquamation	0	2	3
1005	Bevacizumab	18.11.2011	forearm rash	Rash/desquamation	0	2	3
1005	Bevacizumab	19.01.2011	constipation	constipation	0	4	1
1005	Bevacizumab	13.11.2011	fatigue	fatigue	0	3	1
1005	Bevacizumab	13.12.2011	chill	Rigors/ chills	0	4	1
1005	Bevacizumab	21.11.2011	alopecia	Hair loss/alopecia	0	4	3
1005	Bevacizumab	20.12.2011	pleural effusion	Pleural effusion	0	4	3

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
1005	Bevacizumab	13.12.2011	infection upper respiratory	Infection – Other (Specify, Upper aerodigestive NOS	0	4	6
1006	Bevacizumab	19.11.2012	constipation	constipation	0	4	1
1006	Bevacizumab	11.11.2012	nausea	nausea	0	4	3
1006	Bevacizumab	12.11.2012	vomiting	vomiting	0	4	1
1006	Bevacizumab	27.11.2012	Rib pain	Pain – Other (Specify, Rib	0	4	6
1006	Bevacizumab	07.12.2012	nausea	nausea	0	4	1
1006	Bevacizumab	07.12.2012.	vomiting	vomiting	0	4	1
1006	Bevacizumab	05.12.2012	back pain	Pain – Other (Specify - back	0	4	6
1006	Bevacizumab	08.12.2012	fatigue	fatigue	0	4	6
1006	Bevacizumab	09.11.2012	urinary retention	Urinary retention	0	4	6
1006	Bevacizumab	01.12.2012	anorexia	anorexia	0	4	3
1006	Bevacizumab	08.12.2012	anorexia	anorexia	0	4	3
1006	Bevacizumab	07.12.2012	decrease of prothrombosine time	coagulation / other: decrease of prothrombin time	0	4	1
1006	Bevacizumab	11.12.2012	vomiting	vomiting	0	4	6
1006	Bevacizumab	11.12.2012	anorexia	anorexia	0	4	6
1006	Bevacizumab	07.12.2012	worsening of general condition	constitutional symptoms - other - bad general condition	0	4	3
1006	Bevacizumab	13.12.2012	anemia	anemia	0		1
1006	Bevacizumab	10.12.2012	hematoma lower abdomen	hematoma	0	4	3
1201	Bevacizumab	10.02.2011	cerebrovascular ischemia	CNS cerebrovascular ischemia	1	2	5
1201	Bevacizumab	08.02.2011	nausea	nausea	0	4	1

16.2.7 Adverse events listings (each patient)

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1201	Bevacizumab	08.02.2011	vomiting	vomiting	0	4	1
1401	Placebo	22.09.2010	polyneuropathy	neuropathy:sensory	0	3	3
1401	Placebo	25.08.2010	dyspnea	dyspnea	0	4	2
1401	Placebo	30.07.2010	nausea	nausea	0	4	1
1401	Placebo	11.08.2010	obstipation	constipation	0	4	1
1401	Placebo	22.09.2010	obstipation	constipation	0	4	3
1402	Bevacizumab	18.11.2010	fatigue	fatigue	0	2	1
1403	n/a	24.06.2011	(general) weakness	Muscle weakness, - Whole body/generalized	1	4	5
1404	Bevacizumab	16.09.2011	fatigue	fatigue	0	1	3
1405	Bevacizumab	14.08.2012	vomiting	vomiting	1	4	1
1405	Bevacizumab	14.08.2012	hepato-renal syndrome	Syndromes – Other (Specify, Hepato-renal syndrome	0	4	3
1405	Bevacizumab	07.08.2012	worsening of general constitution	constitutional symptoms - other - bad general condition	0	4	2
1405	Bevacizumab	14.08.2012	hyponatremia	Sodium, serum-low (hyponatremia)	0	4	2
1406	Placebo		no adverse events	n.a.			
1407	Bevacizumab	24.09.2013	pain	pain - unspecified	1	2	2
1407	Bevacizumab	18.10.2013	abscess perineal	Infection – Other (Specify, GASTROINTESTINAL Anal/perianal	1	4	1
1407	Bevacizumab	02.08.2013	massive ascites	ascites	1	4	1
1901	Bevacizumab	17.03.2011	weakness, fatigue	fatigue	0	4	3
1901	Bevacizumab	17.03.2011	lost of appetite	anorexia	0	4	1
1901	Bevacizumab	20.04.2011	cough	cough	0	4	1
1901	Bevacizumab	09.05.2011	nausea	nausea	0	4	1

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1901	Bevacizumab	14.06.2011	ascites	ascites	0	4	1
1901	Bevacizumab	14.06.2011	edema lower leg both sides	edema: limb	0	4	3
1902	Placebo	16.05.2013	fatigue	fatigue	0	4	3
1902	Placebo	02.05.2013	nausea	nausea	0	4	1
1902	Placebo	02.05.2013	insomnia	insomnia	0	4	1
1902	Placebo	09.05.2013	agitation	mood alteration: agitation	0	4	1
1902	Placebo	09.05.2013	dyspepsia	Heartburn/dyspepsia	0	4	1
1902	Placebo	27.05.2013	insomnia	Insomnia	0	4	1
1902	Placebo	29.05.2013	nausea	nausea	0	4	1
1902	Placebo	08.06.2013	insomnia	insomnia	0	4	1
1902	Placebo	08.06.2013	nausea	nausea	0	4	1
1902	Placebo	20.06.2013	nausea	nausea	0	4	1
1902	Placebo	20.06.2013	Insomnia	Insomnia	0	4	1
1902	Placebo	27.06.2013	Insomnia	Insomnia	0	4	1
1902	Placebo	06.06.2013	Acute epigastric pain	Pain- GASTROINTESTINAL - Stomach	1	2	1
1902	Placebo	11.06.2013	agitation	mood alteration: agitation	0	4	1
1902	Placebo	07.06.2013	Eye irritation	Ocular/Visual – Other (Specify, Eye irritation	0	4	1
1902	Placebo	06.06.2013	pain	pain - unspecified	0	4	1
1902	Placebo	27.06.2013	pain	pain - unspecified	0	4	3
1902	Placebo	13.07.2013	nausea	nausea	0	4	1
1902	Placebo	25.07.2013	nausea	nausea	0	4	1
1902	Placebo	25.07.2013	insomnia	Insomnia	0	4	1
1902	Placebo	03.08.2013	agitation	mood alteration: agitation	0	4	3
1902	Placebo	01.08.2013	bile duct stenosis	Stricture/stenosis (including anastomotic) Bile duct	1	4	3

16.2.7 Adverse events listings (each patient)

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2301	Bevacizumab	19.06.2010	worsening general condition	constitutional symptoms - other - bad general condition	0	4	3
2301	Bevacizumab	13.06.2010	(worsening of) emesis	vomiting	1	4	5
2301	Bevacizumab	13.06.2010	mucositis	Mucositis/stomatitis	0	4	2
2301	Bevacizumab	13.06.2010	obstipation	constipation	0	4	2
2301	Bevacizumab	24.06.2010	uneasiness	mood alteration: depression	0	4	3
2301	Bevacizumab	23.06.2010	dyspnea	dyspnea	0	4	3
2302	Placebo	30.06.2010	thrombocytopenia	platelets	0	4	1
2302	Placebo	21.06.2010	anemia	anemia	0	4	2
2302	Placebo	06.07.2010	anasarca	dermal change lymphedema	0	4	3
2302	Placebo	10.06.2010	hyperkalemia	Calcium, serum-high (hypercalcemia)	0	4	1
2302	Placebo	06.07.2010	hyponatremia	Sodium, serum-low (hyponatremia)	0	4	3
2302	Placebo	09.07.2010	uneasiness	mood alteration: depression	0	4	3
2302	Placebo	08.07.2010	dyspnea	dyspnea	0	4	3
2302	Placebo	03.07.2010	diarrhea	diarrhea	0	4	3
2302	Placebo	30.06.2010	soor oralmucosa	mucositis / oral	0	4	2
2302	Placebo	08.07.2010	clostridia infection	infection - unspecified	0	4	3
2303	Bevacizumab	20.04.2011	leucocytopenia	leucocytes	0	4	1
2303	Bevacizumab	10.05.2011	leukocytopenia	leucocytes	0	4	3
2303	Bevacizumab	17.05.2011	leukocytopenia	leucocytes	0	4	2
2303	Bevacizumab	10.05.2011	thrombocytopenia	platelets	0	4	1
2303	Bevacizumab	10.05.2011	nose bleed	Hemorrhage, pulmonary/upper respiratory - Nose	0	4	2
2303	Bevacizumab	20.04.2011	lower leg edema	Edema limb	0	4	2
2401	Placebo	20.08.2010	anemia	anemia	0	4	1

16.2.7 Adverse events listings (each patient)

Patient No.	IMP	AE start	Description of the AE	CTCAE Term (Version 3.0)	Serious yes (1), no (0)	Causality probable (1), possible (2), remote (3), unrelated (4)	Outcome recovered/resresolved (1), recovering/resolving (2), not recovered/not resolved (3), recovered/resolved with resident effects (4), fatal (5), unknown (6)
2401	Placebo	18.08.2010	fatigue	fatigue	0	4	3
2401	Placebo	21.08.2010	insomnia	insomnia	0	4	1
2401	Placebo	26.08.2010	insufficiency of liver	liver dysfunction	1	4	5
2401	Placebo	18.08.2010	focal seizures	seizure	1	4	1
2401	Placebo	22.08.2010	restlessness	Neurology – Other (Specify, restlessness)	0	4	1
2401	Placebo	21.08.2010	amentia	Confusion	0	4	1
2401	Placebo	17.08.2010	pain	pain - unspecified	0	4	1
2402	Bevacizumab	03.06.2012	Pain due to peritonitis	pain – peritoneum	1	4	1
2403	Bevacizumab	04.07.2012	insomnia	insomnia	1	4	1
2403	Bevacizumab	11.07.2012	bacterial peritonitis	infection other- peritoneum	0	3	2
2403	Bevacizumab	03.07.2012	pain	pain - unspecified	0	4	3
2404	Bevacizumab			n.a.			
2501	Bevacizumab	29.06.2012	thrombocytopenia	platelets	0	4	1
2501	Bevacizumab	24.08.2012	fatigue	fatigue	0	4	3
2501	Bevacizumab	17.08.2012	weight loss	weight loss	0	4	3
2501	Bevacizumab	05.07.2012	edema	edema - unspecified	0	4	1
2501	Bevacizumab	22.06.2012	port thrombosis	Thrombosis/thrombus/embolism	0	4	1
2501	Bevacizumab	24.08.2012	abdominal pain	Pain - GASTROINTESTINAL – Abdomen NOS	0	4	3
2501	Bevacizumab	NK.08.2012	thorax pain	Pain - Chest/thorax NOS	0	4	1
2601	Bevacizumab	06.08.2010	worsening general condition	constitutional symptoms - other - bad general condition	0	3	1
2601	Bevacizumab	03.08.2010	fatigue	fatigue	0	3	1
2601	Bevacizumab	01.08.2010	diarrhea	diarrhea	0	3	1
2601	Bevacizumab	03.08.2010	anorexia	anorexia	0	3	1
2601	Bevacizumab	03.08.2010	leg pain on both side	Pain / extremity limb	0	3	1
2601	Bevacizumab	05.08.2010	dyspnea	dyspnea	0	4	1

16.2.7 Adverse events listings (each patient)

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2601	Bevacizumab	29.08.2010	dyspnea	dyspnea	0	4	1
2602	Placebo	04.10.2010	fatigue	fatigue	0	3	1
2602	Placebo	04.10.2010	nausea	nausea	0	3	1
2602	Placebo	04.10.2010	anorexia	anorexia	0	3	1
2602	Placebo	04.10.2010	bacterial peritonitis	infection other- peritoneum	1	4	1
2603			no adverse events	n.a.			
2701	Placebo	29.07.2013	disease progression	n.a.	0	4	5
2701	Placebo	22.07.2013	proteinuria	proteinuria	0	4	1

16.2.8 Listing of individual laboratory measurements by patient, if required by regulatory authorities

NA