Simultaneous Study of Gemcitabine-Docetaxel Combination adjuvant treatment, as well as Biological Targeted Treatment

SUCCESS B-Trial

EudraCT No. 2007-001094-29

SUCCESS B-Trial is an open-label, multicenter, randomized controlled, Phase III study comparing the disease free survival after randomisation in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy and biological targeted treatment after chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy and biological targeted treatment after chemotherapy

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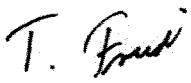
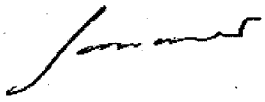


PD Dr. Thomas Friedl		15.11.2019
Trial Statistician	Signature	Date

Sponsor's responsible medical officer

I certify that this report accurately describes the conduct and the results of this clinical study.



Prof. Dr. med. Harald Sommer		15.11.2019
Medical Director	Signature	Date

Investigator signature

I have read this report and confirm that to the best of my knowledge it accurately describes the conduct and results of the study

Prof. Dr. med. Brigitte Rack		15.11.2019
Investigator	Signature	Date

Affiliation

1. Study information

Study title:

Simultaneous Study of Gemcitabine-Docetaxel Combination adjuvant treatment, as well Biological Targeted Treatment

Test drug/investigational product:

Epirubicin-Fluorouracil-Cyclophosphamide(FEC)+ Docetaxel/ Gemcitabine

Tamoxifen, Letrozole, Goserelin, Trastuzumab

Indication studied: Adjuvant cancer therapy in early primary breast cancer patients where chemotherapy was indicated.

Study design:

This is an open-label, multicenter, randomized controlled, Phase III study comparing the disease free survival after randomization in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine- Docetaxel(DG) chemotherapy. All patients were required to have HER2-positive disease and therefore both groups received biological anti-HER2 treatment with Trastuzumab according to the general therapy guidelines.

Postmenopausal patients with positive hormone receptor status of the primary tumor received Letrozole treatment for 5 years, after the end of chemotherapy. Premenopausal patients received Tamoxifen treatment. In addition to Tamoxifen, all patients with positive hormone receptor status of the primary tumor and under the age of 40 or restart of menstrual bleeding within 6 months after the completion of cytostatic treatment or with premenopausal hormone levels as defined below received Goserelin 3.6 mg subcutaneously every 4 weeks over a period of 2 years following chemotherapy (1, 2).

Sponsor:

Prof. Dr. H. Sommer, Poliklinik und Klinik für Frauenheilkunde und Geburtshilfe Campus Maistraße, Maistraße 11, 80337 München, Germany.

Protocol identification:

EudraCT No.: 2007-001094-29

Development phase of study: Phase III.

Study initiation date: 10-June-2008 (first patient's first visit).

Early termination date: The study was terminated after 793 randomized. Last patient's last visit on regular study was on 08.12.2016.

Principal or Coordinating Investigator(s): The coordinating investigator ("Leiter der klinischen Prüfung" [LKP] according to German Drug Law) was: Prof. Dr. H. Sommer, Poliklinik und Klinik für Frauenheilkunde und Geburtshilfe Campus Maistraße, Maistraße 11, 80337 München, Germany.

Company/Sponsor signatory: Prof. Dr. H. Sommer, Poliklinik und Klinik für Frauenheilkunde und Geburtshilfe Campus Maistraße, München, Maistraße 11, 80337 Germany

GCP Compliance Statement: This study was conducted in compliance with Good Clinical Practice (GCP), including the archiving of essential documents.

Report date(s): 26.06.2019

2. Synopsis

This was an open-label, multicenter randomized controlled, Phase III study comparing the disease free survival after randomization in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy. Patients were required to have HER2-neu positive disease and histopathological proof of axillary lymph node metastases (pN1-3) or high risk node negative, defined as: pT ≥ 1c or histopathological grade ≥ 2, or age ≤ 35 or negative hormone receptor', if chemotherapy was indicated, but were not allowed to have evidence of distant disease. Patients had to be entered into the study no later than 6 weeks after complete resection of the primary tumor. No other antineoplastic treatment other than surgical treatment, the defined cytotoxic and endocrine treatment and radiotherapy was allowed prior to study entry and during the course of the study.

After surgery, leading to R0 resection of the invasive and intraductal components of the primary tumor, patients were randomized to one of the following treatments:

Randomization

A: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3 cycles of Docetaxel 75 mg/m² body surface area i.v. (D), and Gemcitabine 1000 mg/m² i.v. (30 min infusion) (G), administered on day 1, followed by Gemcitabine 1000 mg/m² i.v. (30 min infusion) on day 8, repeated on day 22

B: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3 cycles of Docetaxel 100 mg/m² body surface area i.v. (D), administered on day 1, repeated on day 22

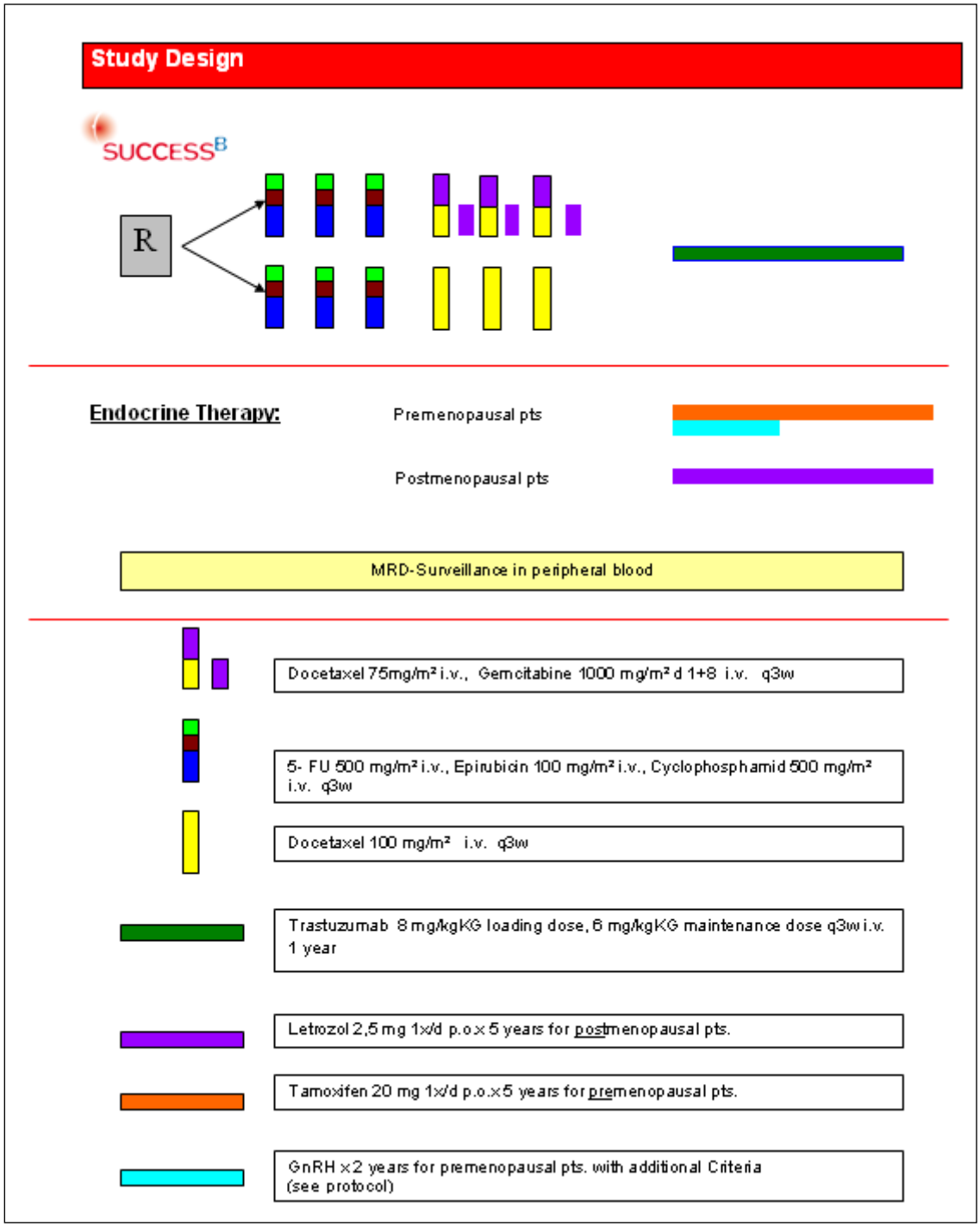
After the end of chemotherapy all patients received biological anti-HER2 treatment with Trastuzumab according to the general therapy guidelines.

Postmenopausal patients with positive hormone receptor status (≥ 10 % positively stained cells for estrogen and/or progesterone) of the primary tumor received Letrozole treatment 2,5 mg p.o. per day for 5 years, after the end of chemotherapy. Premenopausal patients received Tamoxifen treatment 20 mg p.o. qd. In addition to tamoxifen, all patients with positive hormone receptor status of the primary tumor and under the age of 40 or restart of menstrual bleeding within 6 months after the completion of cytostatic treatment or with premenopausal hormone levels as defined below received Goserelin 3.6 mg subcutaneously every 4 weeks over a period of 2 years following chemotherapy.(2;3) Premenopausal endocrine status was assumed, if the following serum levels were met: LH < 20 mIE/ml, FSH < 20 mIE/ml and E2 > 20 pg/ml. Endocrine therapy started after the end of chemotherapy.

All patients with breast conserving therapy or more than 3 axillary lymph node metastases or in the following cases after mastectomy:

- T3/T4-carcinoma
- T2-carcinoma > 3 cm
- multicentric tumor growth
- lymphangiosis carcinomatosa or vessel involvement
- involvement of the pectoralis fascia or a safety margin < 5 mm.(4) (5;6)

received adjuvant radiotherapy.


Study Design
SUCCESS B
R
Endocrine Therapy:
Premenopausal pts
Postmenopausal pts
MRD-Surveillance in peripheral blood
Docetaxel 75mg/m² i.v., Gemcitabine 1000 mg/m² d 1+8 i.v. q3w
5- FU 500 mg/m² i.v., Epirubicin 100 mg/m² i.v., Cyclophosphamid 500 mg/m² i.v. q3w
Docetaxel 100 mg/m² i.v. q3w
Trastuzumab 8 mg/kgKG loading dose, 6 mg/kgKG maintenance dose q3w i.v. 1 year
Letrozol 2,5 mg 1x/d p.o.x 5 years for postmenopausal pts.
Tamoxifen 20 mg 1x/d p.o.x5 years for premenopausal pts.
GnRH x2 years for premenopausal pts. with additional Criteria (see protocol)

3. Table of content

3.1 Tables

3.2 Figures

4 List of abbreviation and definition of term

AE	Adverse Event
AGO	Arbeitsgruppe gynäkologischer Onkologen" (Working Group for Gynecological Oncology)
AI	Aromatase Inhibitor
ALT (SGPT)	Alaninaminotransferase
AMG	Arzneimittelgesetz (German Drug Law)
ASCO	American Society of Clinical Oncology
AST (SGOT)	Aspartataminotransferase, (Glutamic Oxalacetic Transaminase)
AUC	Area under the (Concentration - Time) - Curve
BfArM	Bundesinstitut für Arzneimittel und Medizinprodukte (German competent authority)
BUN	Blood Urea Nitrogen
CHF	Congestive heart failure
CR	Complete Response
CRF	Case Report Form
CT	Computerized Tomograph
CTC	Circulating Tumor Cells
CTCAE	Common Toxicity Criteria for Adverse Events
DCR	Disease Control Rate
DNA	Desoxy Ribonucleic Acid
DFS	Disease Free Survival
ECG	Electrocardiogram
ECOG	Eastern Cooperative Oncology Group
eCRF	Electronic Case Report Form

EORTC	European Organization for Research and Treatment of Cancer
ER	Estrogen Receptor
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FDA	Food and Drug Administration (USA)
FISH	Fluorescent In Situ Hybridization
FSH	Follicle-Stimulating Hormone
GCP	Good Clinical Practice
GCP-V	Verordnung über die Anwendung der Guten Klinischen Praxis bei der Durchführung von klinischen Prüfungen mit Arzneimitteln zur Anwendung am Menschen (German regulation on GCP)
G-CSF	Granulocyte Colony-Stimulating Factor
GI	Gastrointestinal
GM-CSF	Granulocyte-Macrophage Colony
HER2	Human Epidermal Growth factor Receptor 2
HIV	Human Immunodeficiency Virus
ICH	International Conference on Harmonization
IF	Immunofluorescence
IHC	Immunohistochemistry
IMP	Investigational Medicinal Product
ISF	Investigator Site File
ITT	Intention to Treat
IU	International Units
IV	Intravenous
L	Liter
LD	Lesion Diameter
LH	Luteinizing Hormone

MBC	Metastatic Breast Cancer
mL	Milliliter
MRD	Minimal residual disease
MRI	Magnetic Resonance Imaging
NSABP	The National Surgical Adjuvant Breast Project
NCI	National Cancer Institute
NRS	Numeric Rating Scale
NSAI	Non-steroidal Aromatase Inhibitor
OD	Once Daily
ORR	Overall Response Rate
OS	Overall Survival
pCR	Pathological Complete Response
PCR	Polymerase Chain Reaction
PD	Progressive Disease
PET	Positron Emission Tomography
PFS	Progression Free Survival
PgR	Progesterone Receptor
PP	Per Protocol
PR	Partial Response
QOL	Quality of Life
REC	Response Related Committee
RNA	Ribonucleic Acid
SABCS	San Antonio Breast Cancer Symposium
SAE	Serious Adverse Event
SAR	Suspected Adverse Reaction

SD	Stable Disease
SGPT	Serum Glutamic-Pyruvic Transaminase
SPC	Summary of Product Characteristics
SRE	Skeletal Related Event
SUSAR	Suspected Unexpected Serious Adverse Reaction
TTP	Time To Progression
TtTF	Time to treatment failure
YOB	Year of Birth

5 Ethics

5.1 Independent ethics committee or institutional review board

The study protocol and the amendment as well as the informed consent document has been accepted by German ethical boards (lead ethical board: Ethikkommission der Medizinischen Fakultät der Ludwig-Maximilians-Universität München). The ethical review board agreed to monitor the conduct of the study and agreed to review it periodically. The study coordinators provided all study centers with information about any revisions to the informed consent document or amendments to the protocol. Furthermore the study coordinators were responsible for SAE transmission to the ethical review board and report the premature or regular study discontinuation.

The study sites themselves were responsible for informing the local ethical review board about the study to attain approval prior to patient's inclusion.

This study was conducted in accordance to the ethical principles stated in the most recent version of the Declaration of Helsinki and the applicable guidelines of the International Conference on Harmonization Good Clinical Practice Guideline 1998, whichever represents the greater protection of the individual.

5.2 Patient information and consent

The informed consent document was signed by the patient and the investigator before the patient entered the study. It explained in a patient orientated way, the risks and benefits of the study to the patient. The informed consent document contained, that the patient is aware of the risks and benefits of entering the study, and that the patient is free to withdraw from the study at any time.

The investigator was responsible for checking that informed consent was obtained from each patient and for obtaining the appropriate signatures and dates on the informed consent document prior to the performance of any protocol procedures and

prior to the administration of any study drug. Informed consent for the SUCCESS B chemotherapy trial had to be obtained prior to the performance of any protocol procedures and prior to the administration of any study drug. Samples of the written information handed out to each patient and the consent form for the core study and the pharmacokinetic/apoptosis substudies are included in Appendix.

6 Investigators and study administrative structure

Physicians with a specialty in medical oncology or gynecological oncology participated as investigators in this clinical trial.

The names, titles, institutions and professional addresses of the investigators are listed in the appendix.

If investigators were added after the trial had been approved by the Steering Committee, an ethical review committee or a regulatory agency, these updates were not considered as changes to the protocol, but rather to the Contracts for Protocol SUCCESS B Trial.

Study director (sponsor):

Prof. Dr. H. Sommer,
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Co-Study Directors:

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Advisory Board:

Chair: Prof. Dr. A. Schneeweiß
Members:
Prof. Dr. Beck, Rosenheim; Prof. Dr. Dall. Lüneburg;
Prof. Dr. Diedrich, Lübeck; Prof. Dr. Dietl, Würzburg; Prof. Dr. Emons, Göttingen;
Prof. Dr. Fiekau, Erlangen; Prof. Dr. Friese, München; Prof. Dr. Hilfrich, Hannover;
Prof. Dr. Hillemanns, Hannover; Prof. Dr. Jänicke, Hamburg; Prof. Dr. Jonat, Kiel;

Prof. Dr. Kimming, Essen; Prof. Dr. Pantel, Hamburg; Prof. Dr. Porzsolt, Ulm; Prof. Dr. Reimer, Rostock; Prof. Dr. Runnebaum, Jena; Prof. Dr. Schneider, Berlin; Prof. Dr. Sohn, Heidelberg; Prof. Dr. Tulusan, Bayreuth; Prof. Dr. Wallwiener, Tübingen; Prof. Dr. Wez, Mannheim; Prof. Dr. Wischnik, Augsburg

Steering Commitee:

Chair: Dr. med. D. Chatsiproios

Members:

Dr. Bauernfeind, Landshut; Dr. Beldermann, Stuttgart; Dr. de Waal, Dachau; Prof. Dr. Fasching, Erlangen; Prof Dr. Friedrich, Krefeld; Frau Haidinger, München; Prof. Dr. Janni, Ulm; Dr. Kleie-Tebbe, Berlin; Dr. Kuhn, Stuttgart; Dr. Kümmel, Essen; Prof. Dr. Maass, Kiel; PD Dr. Rody, Frankfurt am Main; Dr. med. Schilling, Berlin; Frau Schmitt Augsburg; Prof. Dr. Sehouli, Berlin; Prof. Dr. Solomayer, Tübingen; Dr. Steinfeld-Birg, Augsburg; Prof. Dr. Stickler, Freiburg; Prof. Dr. Sütterlin, Mannheim; Dipl. Ing. Zwingers, Augsburg

The study was conducted and monitored by the CRO:

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35394 Gießen, Germany

Statistics

Estimate GmbH
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86150 Augsburg, Germany

External participants

The study is supported by the pharmaceutical companies Sanofi-Aventis, AstraZeneca, Novartis, Lilly, Chugai and Veridex.
Docetaxel medication for node negative patients was provided by Sanofi-Aventis at its own cost. Gemcitabine medication for all patients in treatment arm A was provided by Lilly at its own cost. Veridex supported the translational research program of the study.

7 Introduction

Breast cancer is the most frequent malignoma of the female in all countries of the industrialized western hemisphere. Approximately 28 % of all malignancies in the countries of the European Community (EC) were attributed to neoplasms of the female breast. There were 73.557 deaths caused by breast cancer in the countries of the EC in 1990 (3). While the combination of locoregional and systemic treatment has early been shown to improve the prognosis of the disease considerably, (4) only 25 % of all patients will be cured after primary therapy.

Halsted's demand for radical mastectomy as treatment of choice for breast cancer dates back to the 1880's and was based on the understanding of breast cancer as a locoregional disease. This view was questioned by a series of studies between 1950 and 1970 (5). These studies established the advantages of limited local therapy and finally led the way to breast conserving surgery and systemic treatment. Breast conserving treatment, once controversially discussed, is now an established alternative to modified radical mastectomy for surgically manageable breast cancer. Several prospective, randomized controlled trials have uniformly reported similar rates of distant disease free survival and overall survival after long-term follow-up (6-11). While one of the major concerns in breast conserving treatment is the risk for local recurrence, the literature reports wide variation ranging from 3% to 25% (12, 13). This risk continuously increases with time at a yearly conditional event probability of approximately 1% (14). This is in contrast to chest wall recurrences, which predominantly take place within the first 3 years after primary treatment. However, local recurrences after breast conserving therapy stay without major impact on the overall survival of these patients.

Systemic treatment is widely accepted as adjuvant, integral part of primary therapy in patients with average to high risk for relapse according the St. Gallen risk criteria (15):

Table 1 St. Gallen Risk Criteria 2007 (16)

Risk category	
Low risk[a]	**Node negative** AND **all** of the following features: pT* ≤2 cm, AND Grade 1**, AND Absence of extensive peritumoral vascular invasion[b], AND ER and/or PgR*** expressed[c], AND HER2/*neu* gene neither overexpressed nor amplified[d], AND Age ≥5 years
Intermediate risk[e]	**Node negative** AND **at least one** of the following features: pT* >2 cm, OR Grade 2-3**, OR Presence of extensive peritumoral vascular invasion[b], OR ER and PgR absent[c], OR HER2/*neu* gene overexpressed or amplified[d], OR Age <35 years **Node positive (1-3 involved nodes)** AND ER and/or PgR expressed, AND HER2/*neu* gene neither overexpressed nor amplified[d]
High risk	**Node positive (1-3 involved nodes)** AND ER and PgR absent, OR HER2/*neu* gene overexpressed or amplified[d] **Node positive (4 or more involved nodes)**

According to this risk assessment, all breast cancer patients should receive adjuvant systemic treatment, except those, who meet all of the following favorable risk criteria (12):

- no axillary lymph node metastases
- age ≥ 35 years
- endocrine-responsive disease
- tumor ≤ 2 cm
- histopathological grading G1
- no HER2-neu overexpression
- no vascular space invasion.

In all other patients, systemic treatment, either primarily systemic or adjuvant should be considered.

Treatment options should be considered and decided upon in the context of case management meetings, which should include:

- Weekly multidisciplinary case management meetings dealing with
    - diagnosis (surgeons, radiologists and pathologists)

  - further case management (surgeons, radiologists, pathologists, oncologists)
    - advanced breast cancer management
- San Antonio Database (Adjuvant Online) estimate support for decision making
- National guideline conformity assurance
- Daily counselling clinics to support decision making for the relevant patients

Within this protocol, patients were treated according to the relevant national and international guidelines.

7.1 Chemotherapy in breast cancer

Present data indicate that only women with node-negative breast cancers < 2 cm in diameter and histopathological grade 1 (pT1, pN0, G1) have similar survival likelihood as age-matched women without breast cancer (17). Therefore, women in this subgroup should only receive chemotherapy in the setting of a controlled clinical trial with carefully informed consent.

The cyclophosphamide, methotrexate, and fluorouracil (CMF) regimen was among the first multiagent regimens to show disease-free survival and overall survival benefits in patients with a more advanced stage of disease (18, 19). CMF still represents the adequate standard of care for patients with low risk disease (15); for elderly patients and for those patients who have pre-existing cardiac dysfunction or hypertension, the nonanthracycline regimen CMF may be preferable (20). However, for patients with an increased risk for recurrence, several randomized studies and the 2000 Oxford overview confirms that anthracycline-based multiagent chemotherapy offers a significant survival benefit compared with CMF (21-25). The meta-analysis of the Early Breast Cancer Trialists' Collaborative Group 1998 estimates a recurrence free survival benefit of 3.2% and an overall survival benefit of 2.7 % for anthracycline-based multi agent chemotherapy compared to CMF (23).

Anthracycline-based chemotherapy therefore can be assumed as the minimum standard in treating breast cancer patients who need cytostatic treatment. Unfortunately, there is no national or international consensus on which regimen is

preferable. Currently, there are two regimen internationally accepted as standard for anthracycline based chemotherapy: the Canadian FEC120 ‘Levine’ regimen (22) and the French FEC100 ‘Bonneterre’ regimen.

The results published by Levine et al. showed clear superiority of FEC chemotherapy over CMF in terms of both disease-free and overall survival. The 5-year recurrence-free survival rates were 63 % in the patient group treated with FEC and 53 % in the patient group treated with CMF (p= .009). The corresponding 5-year actuarial survival rate were 70% and 77%, respectively (p= .03). However, it should be noted, that the rate of hospitalization for febrile neutropenia was significantly higher in the FEC group (8.5 %, compared to 1,1% in the CMF group). None of the febrile neutropenic episodes was fatal. There was no case of congestive heart failure noted in the FEC group. Five patients in the FEC group experienced acute leukemia. In general, patients who received FEC had more acute toxicity than CMF patients. 42 % of the FEC patients had grade 2 or more vomiting compared with only 18 % of CMF patients (p = .0001). Similar differences were seen for nausea and stomatitis. For FEC patients, the median nadir of the white blood cell count was 1.0*109/l, compared to 1.7*109/l in the CMF group (22).

The French Bonneterre FEC100 regimen (5-FU 500mg/m², Epirubicin 100mg/m², Cyclophosphamide 500mg/m², all i.v. q3s) is even more popular and has proofed to be significantly superior to the FEC50 regimen (26, 27). The 10-year DFS was 45.3% in FEC 50 and 50.7% in FEC 100, with a relative risk (RR) reduction of 24% (Wilcoxon, p=.03). The 10-year OS was 50% and 54.8%, respectively, with a RR reduction of 29% (Wilcoxon, p=.03). In the multivariate analysis including patients and tumor characteristics, FEC 100 remained significantly superior to FEC 50 for both DFS (p=.08) and OS (p=.04). In the FEC 50 arm, long-term side effects (not related to treatment) were: myocardial infarction, and acute lymphoblastic leukemia; in FEC 100: 3 congestive heart failures and 1 acute myeloblastic leukemia FAB 4 (probably related to chemotherapy). Overall, the 10-year DFS/EFS was 44.5% and 49.3%, respectively (Wilcoxon, p=.06). This regimen lacks a direct comparison to the classical CMF-regimen, but also meets the standard criteria of a multi-agent anthracycline regimen, containing at least 30mg/m² epirubicin per week. It appears most unlikely that the Bonneterre FEC100 regimen is inferior to the FEC120 regimen,

despite the fact that there is no data on a direct comparison available. Therefore, the Bonneterre FEC100 regimen is widely accepted as alternative anthracycline standard.

Further dose escalations of anthracyclines and of cyclophosphamide beyond the already intense dose have so far not proven superior in large randomized controlled trials (28-30).

Epirubicin had been chosen as anthracycline component of the standard and experimental therapy regimen for the following reasons:

- Epirubicin (4'-epidoxorubicin) is an antineoplastic agent derived from doxorubicin. The compounds differ in the configuration of the hydroxyl group at the 4' position. Epirubicin, like doxorubicin, exerts its antitumor effects by interference with the synthesis and function of DNA and is most active during the S phase of the cell cycle. The overall activity of epirubicin appears to be comparable with that of doxorubicin,(31) while toxicity is more frequent and more pronounced in patients receiving doxorubicin instead of epirubicin (32).
- Epirubicin has been successfully used in the Bonneterre regime(22)
- Epirubicin is used more widely in countries of the European Union, particularly in Germany. Epirubicin based regimens are the corner stone for recommendations of the Gravenbruch and St. Gallen consensus recommendations (16).
- To evaluate the potential benefit of using gemcitabine in addition to docetaxel, it seemed important and appropriate to use the same anthracycline in the experimental treatment arm as in the standard arm.

According to today’s standards, we also added a Taxane-based treatment period sequential to the Epirubicin-based treatment period. Since about 1995, there is increasing evidence that the taxanes are among the most promising new chemotherapy agents for the treatment of breast cancer (33). While the majority of data on the efficacy of these agents have been generated with the agent paclitaxel, there now was also sufficient data available on the efficacy of docetaxel containing

regimen as new possible standard of care, in order to justify the initiation of this phase III trial.

Subsequently, several phase III studies have been conducted, comparing therapeutic efficacy and toxicity of docetaxel with that of anthracyclines. In a recently published study of the 303 Study Group, patients were randomized to receive an intravenous infusion of docetaxel 100 mg/m² or doxorubicin 75 mg/m² every 3 weeks for a maximum of seven treatment cycles (34). 326 patients were followed for a median of 23 months. Overall, docetaxel produced a significantly higher rate of objective response than did doxorubicin (47.8% v 33.3%; P=.008). Docetaxel was also significantly more active than doxorubicin in patients with negative prognostic factors, such as visceral metastases (objective response, 46% v 29%) and resistance to prior chemotherapy (47% v 25%). Median time to progression was 26 weeks in the docetaxel group, compared to 21 weeks in the doxorubicin group (difference not significant). However, median overall survival was similar in the two groups (docetaxel, 15 months; doxorubicin, 14 months). Febrile neutropenia occurred more frequently in the doxorubicin group (12.3 %, compared to 5.7 % in the docetaxel group).

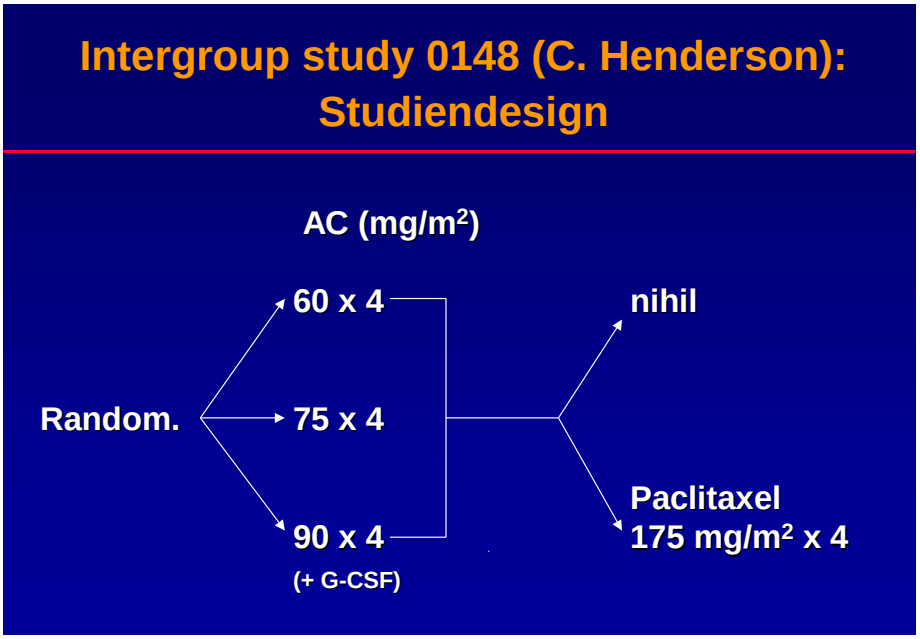
At the 1999 ASCO meeting, Nabholtz et al. presented a pivotal randomized phase III study of doxorubicin plus docetaxel (50/75) versus doxorubicin plus cyclophosphamide (60/600) as first-line chemotherapy for 429 patients with metastatic breast cancer, doxorubicin/docetaxel emerged as the more effective regimen. The response rate in patients with doxorubicin plus docetaxel was 60 % compared to 47 % in patients with doxorubicin plus cyclophosphamide (35, 36).

In another phase III study by this author, docetaxel was compared with mitomycin plus vinblastine (MV) in patients with metastatic breast cancer (MBC) progressing despite previous anthracycline-containing chemotherapy. 392 patients were randomized to receive either docetaxel 100 mg/m2 intravenously (i.v.) every 3 weeks or mitomycin 12 mg/m2 i.v. every 6 weeks plus vinblastine 6 mg/m2 i.v. every 3 weeks. Median time to progression and overall survival were significantly longer with docetaxel than MV (19 vs. 1 weeks, P=.001, and 1.4 vs. 8.7 months, P=.0097, respectively)(37).

At the ASCO 2001 meeting, Bonneterre et al. presented a randomized phase II study to evaluate the activity of ET (75/75 mg/m2) combination versus a standard anthracycline based regimen (FEC 75) in first line metastatic breast cancer patients. In this study, out of 105 evaluable patients, the response rate was 65% in the ET-arm and 37% in the FEC-arm. The authors concluded that the ET activity appeared considerably higher than the FEC activity.(38) In another phase III study at this meeting, Docetaxel, Doxorubicin and Cyclophosphamide (75/50/500 mg/m2) was compared to FAC (500/50/500 mg/m2) d1q3wk (maximum 8 cycles) as first line chemotherapy for metastatic breast cancer. As depicted in the following table, this study confirmed superiority of docetaxel-anthracycline based regimen compared to anthracycline containing multi-agent chemotherapy in terms of response rates (39).

Based on the 20- and 30-months results of the Cancer and Leukemia Group B (CALBG) 9344 trial, the addition of paclitaxel to the adjuvant treatment of node-positive breast cancer has been approved in the United States, but not in Europ (30). In this trial, patients were prospectively randomized to receive three different doses of adriamycin (60 mg/m², 75 mg/m², 90 mg/m² as part of four cycles AC chemotherapy), followed by a randomization between nihil (AC) or subsequent therapy with 175 mg/m² paclitaxel (AC→T).

Figure 1 CALGB 9344 prospectively randomized trial to compare 4 cycles of AC with 4 cycles of AC followed by Paclitaxel (30)



At the time of first presentation of these data, the recurrence free survival rate at 18 months follow-up was 86 % in the AC treatment arm and 90 % in the AC→T treatment arm (P=.0077). The overall survival rates were 95 % and 97 %, respectively (P=.039). No differences were seen between the patient groups with different doses of anthracycline. However, currently up-dated study results, presented by Henderson et al. at the 2000 San Antonio Breast Cancer Symposium and at the National Institute of Health CDC Meeting, did not maintain the same level of significance.

Table 2 Development of study results of the CALGB 9344 by extended follow-up

	ASCO 98	*SNDA 99*	*NIH CDC 2000*
Median Follow-up (months)	21	30	52
Number of recurrences	423	624	901
Number of deaths	200	342	589
Reduction of hazard ratio (recurrence)	22 %	22 %	13 %
Reduction of hazard ratio (death)	26 %	26 %	14 %

Further criticisms of the CALGB trial include that it is not clear to what extent the duration of treatment, which was longer in the AC→T treatment arm, may have influenced the study results. In 2003, the final analysis of the study was published, mostly confirming the initially indicated survival benefit (40). The hazard reductions from adding Paclitaxel to AC were 17% for recurrence (adjusted Wald chi(2) P =.0023; unadjusted Wilcoxon P =.0011) and 18% for death (adjusted P =.0064; unadjusted P =.0098). At 5 years, the disease-free survival (+/- SE) was 65% (+/- 1) and 70% (+/- 1), and overall survival was 77% (+/- 1) and 80% (+/- 1) after AC alone or AC plus Paclitaxel, respectively. The effects of adding paclitaxel were not significantly different in subsets defined by the protocol, but in an unplanned subset analysis, the hazard ratio of AC plus Paclitaxel versus AC alone was 0.72 (95% confidence interval, 0.59 to 0.86) for those with estrogen receptor-negative tumors and only 0.91 (95% confidence interval, 0.78 to 1.07) for patients with estrogen

receptor-positive tumors, almost all of whom received adjuvant Tamoxifen. The additional toxicity from adding four cycles of paclitaxel was generally modest. The authors concluded that the addition of four cycles of paclitaxel after the completion of a standard course of AC improves the disease-free and overall survival of patients with early breast cancer

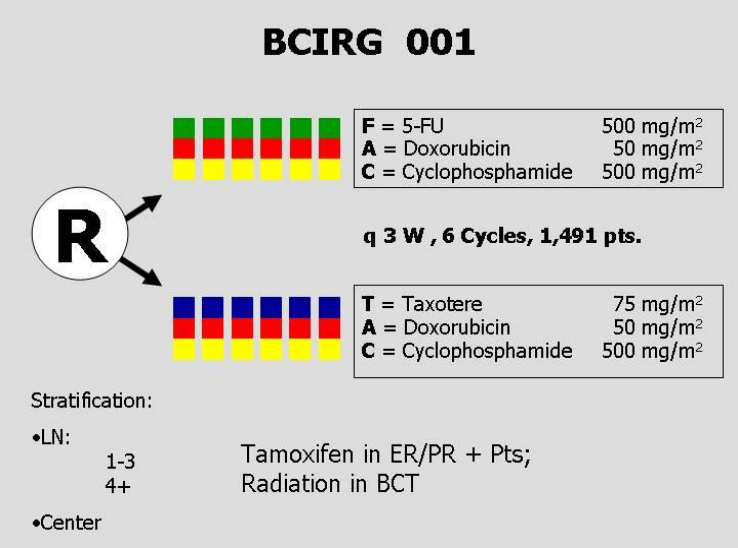
Preliminary results of the other major trial, including paclitaxel in the adjuvant treatment of breast cancer, the NSABP B-28 trial were presented at the ASCO 2003 meeting.(41)

In this study with similar design, paclitaxel 225 mg/m² q4w following four cycles of AC were applied instead of 175 mg/m². All patients over 50 years and those < 50 with ER-or PgR-positive tumors also received tamoxifen 20 mg p.o. daily for 5 years, starting with AC. Most frequently reported grade 3+ toxicity on AC (% of pts): day 1 granulocytopenia: 8%, febrile neutropenia: 7%, nausea: 6%, vomiting: 5%, infection: 3%, thromboembolic events: 2% and stomatitis: 2%. Most frequently reported grade 3+ toxicities on T (% of pts): neurotoxicity: 19%, arthralgia/myalgia: 11%, day 1 granulocytopenia: 4%, febrile neutropenia: 2%, infection: 2%, thromboembolic events: 2% and hypersensitivity reactions: 1%. While after a median follow-up of 64.6 months, disease free survival was significantly improved in patients receiving paclitaxel (p=0.008), overall survival did not differ statistically between the two treatment arms (p=0.46).

7.2 Docetaxel in the Adjuvant Treatment of Primary Breast Cancer

To date, there are results of two major randomized Phase III trials including docetaxel in the adjuvant treatment of breast cancer available. The first, more mature trial, the BCIRG 001 trial compared TAC (75/50/500 mg/m2 q3wk x 6) with FAC (500/50/500 mg/m2 q3wk x 6) in node positive breast cancer.

Figure 2 BCIRG 001 Study Design



At the ASCO Annual Meeting 2002, a planned interim analysis at 33 mos median follow-up (range 0-49 mos) was presented. (42)

Cox analysis for disease free survival showed a relative risk ratio for TAC/FAC of 0.64 (0.50, 0.81; p=0.0002) and for overall survival 0.71 (0.50,1.00; p=0.049). For DFS, there were 119 events on TAC and 170 on FAC; 82% of patients on TAC and 74% on FAC were alive and disease-free. However, in patients with more than 3 metastatic axillary lymph nodes, neither disease free, nor overall survival differed significantly between the two study arms. Febrile neutropenia (24% vs 2%) and grade 3/4 infection (2.8% vs 1.3%) were higher with TAC. No septic deaths occurred. Other grade 3/4 toxicities in > 5% of patients included nausea (9%), vomiting (7%), asthenia (5%) with FAC and asthenia (11%), stomatitis (7%) with TAC. Congestive heart failure incidence was 1.2% on TAC and 0.1% on FAC.

Table 3 2nd Interim Analysis of the BCIRG 001 Study(49)

Intent-to-Treat Efficacy Analyses Prospectively Powered (n=1,491)

DFS	Hazard Ratio TAC/FAC (95% CI)	P-value
Adjusted for N status (Primary endpoint)	0.72 (0.59-0.88)	0.0010
1-3 nodes (n=923)	0.61 (0.46-0.82)*	0.0009
4+ nodes (n=568)	0.82 (0.63-1.08)*	0.1629
Hormone Receptor Positive†	0.73 (0.57-0.94)	0.0132
Hormone Receptor Negative†	0.66 (0.47-0.93)	0.0163
Overall Survival		
Adjusted for N status	0.70 (0.53-0.91)	0.0080

**Ratio of Hazard Ratios: 1.34 (0.90-2.00), p= 0.1476, †: Centrally reviewed*

At the SABCS 2003, the second interim analysis was presented. At a median follow-up of 55 months and 399 DFS events, a statistical boundary of 0.001 for DFS adjusted for nodal (N) status was defined for this analysis.(43)

For DFS, there were 172 events on TAC and 227 on FAC: 80% and 75% of pts on TAC were alive and disease-free at 4 and 5 years respectively, vs.71% and 68% on FAC. For OS, there were 91 events on TAC and 130 on FAC: 89% and 87% of pts on TAC were alive at 4 and 5 years respectively, vs. 85% and 81% on FAC. HER2neu amplification was centrally reviewed. TAC/FAC/DFS hazard ratio was 0.61 (0.42-0.90; p=0.0118) in HER2+ pts, and 0.76 (0.58-0.99; p=0.0380) in HER2- pts. There were no changes in the toxicity profile since the first interim analysis.

Figure 3 Disease Free Survival of BCIRG 001 at 2nd Interim Analysis (43)

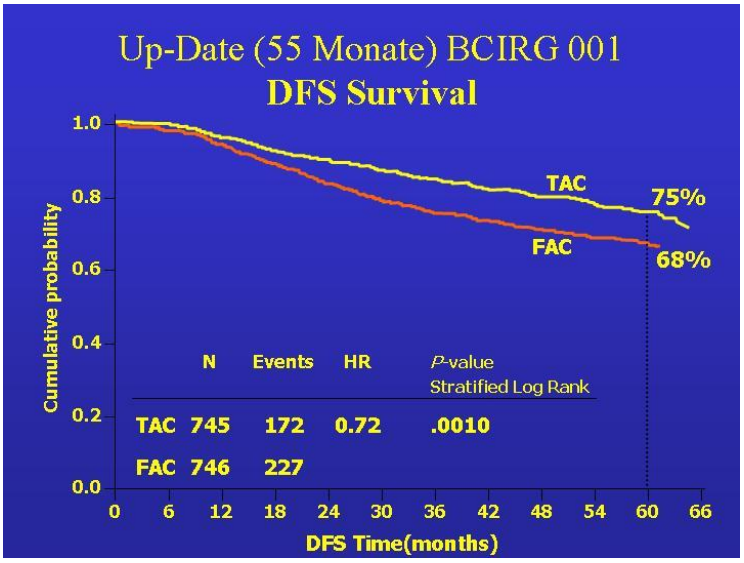
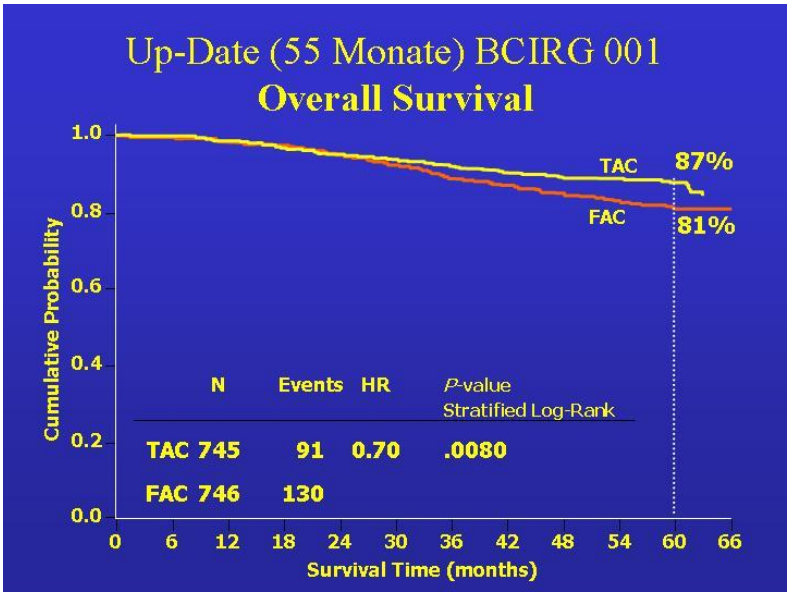
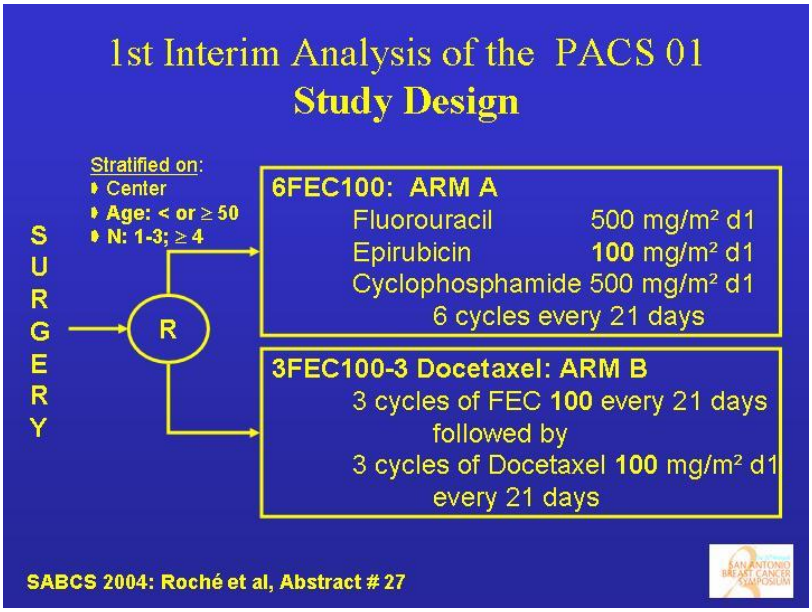


Figure 4 Overall Survival of BCIRG 001 at 2nd Interim Analysis(43)



The results of the first interim analysis of the other randomized Phase III trial, the French PACS 01 study were presented at the SABCS 2004. (44) Pts. had localized, resectable, non pre-treated, unilateral breast cancer. Main inclusion criteria were: age < 65 years, at least one positive node, no metastasis, normal cardiac, hepatic, haematological and renal functions. Arm A: 6 cycles of FEC100 (5FU/epirubicin/cyclophosphamide 500/100/500 mg/m^2 day 1, every 3 weeks); Arm B: docetaxel 100 mg/m^2 (day 1, every 3 weeks) replaced FEC100 for the last 3 cycles. First chemotherapy was to be started no more than 42 days after surgery. G-CSF was given in cases of febrile neutropenia or delay of neutrophil recovery by day 21. Radiotherapy was mandatory after conservative surgery and tamoxifen was given for 5 years if tumors were positive for at least one hormone receptor (HR). To ensure a minimal power of 90%, the analysis was to be carried out at a median follow-up of 60 months and, if at that time, 469 events or more have been observed.

Figure 5 PACS 001 Study Design



Treatment was completed for 95% and 93.4% of pts in arms A and B, respectively. Toxicity was reported at 2003 SABCS (abstract 144). More febrile neutropenia and nail disorders were observed in Arm B and a more decreased and subnormal LVEF at the end of chemotherapy Arm A. Five cases of leukaemia (3 Arm A; 2 Arm B) were observed. No toxic deaths have been reported. As of 30 April 2004, 465 pts have experienced at least one event: 93 locoregional relapses, 324 metastasis, 38 contralateral breast cancers, and 10 deaths as first event. A total number of 37 second cancers and 210 deaths were registered. The 5 year efficacy results revealed a significant benefit for patients receiving 3 cycles of FEC100 followed by 3 cycles of docetaxel in terms of disease free survival (p=0.014) and overall survival (p=0.017).

Figure 6 Disease Free Survival of PACS 01 at 5 Years of Follow-up(44)

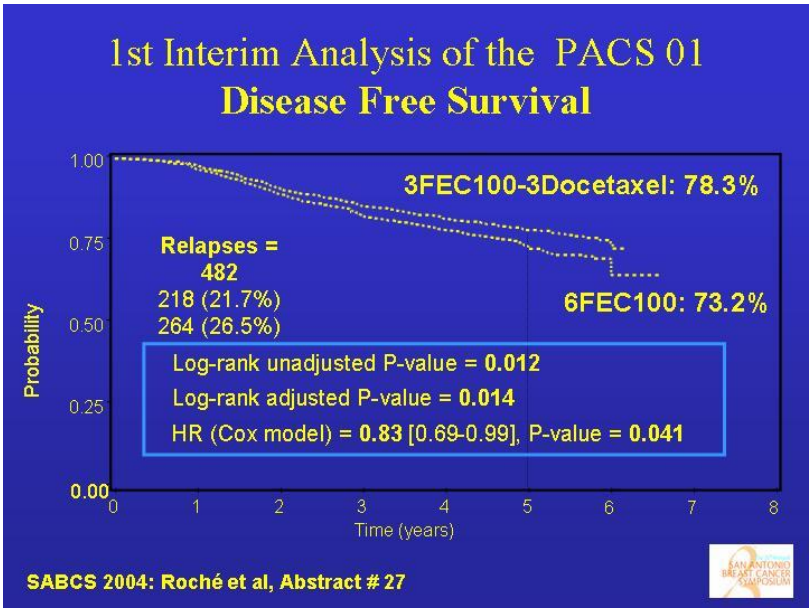
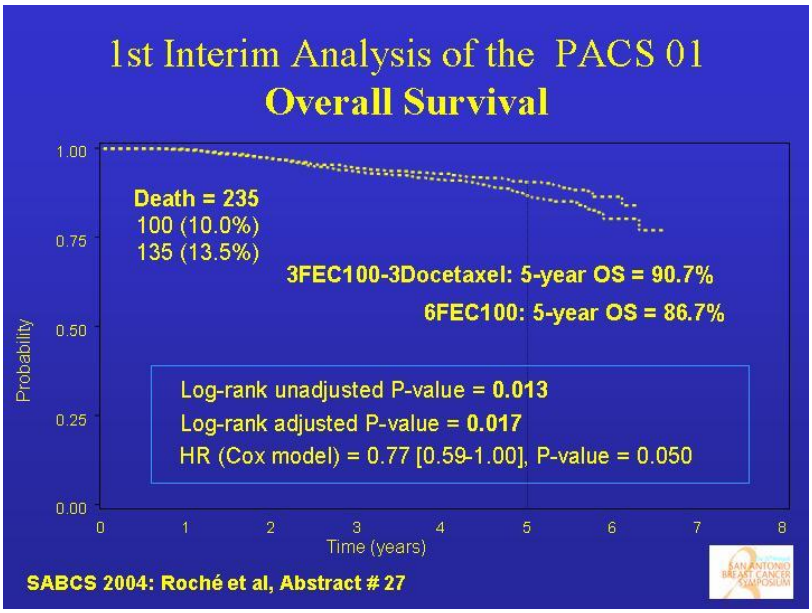


Figure 7 Overall Survival of PACS 01 at 5 Years of Follow-up(44)

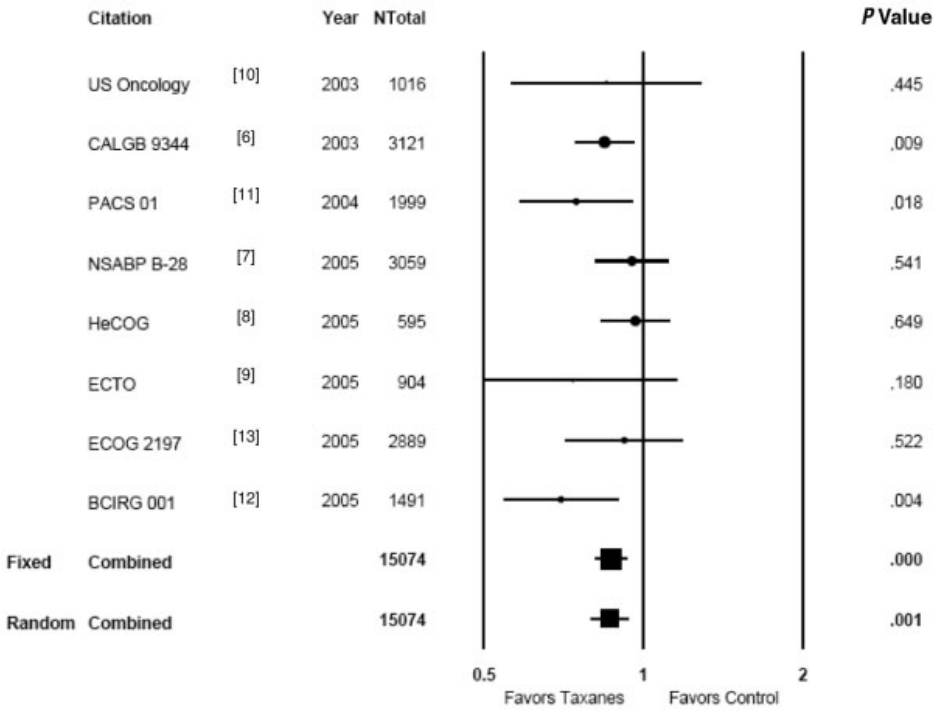


The results of this study provide sufficient evidence to choose the superior treatment arm of the PACS01 study as control arm of this protocol.

A recent pooled analysis of 9 available adjuvant Phase III taxane trials showed significant differences in favor of taxanes. Such differences were seen in DFS in the overall (RR: 0.86;95% CI, 0.81-0.90 [P_.00001]) and lymph node-positive population (RR: 0.84; 95%CI, 0.79-0.89 [P_.0001]), and in OS in the overall (RR: 0.87; 95% CI,

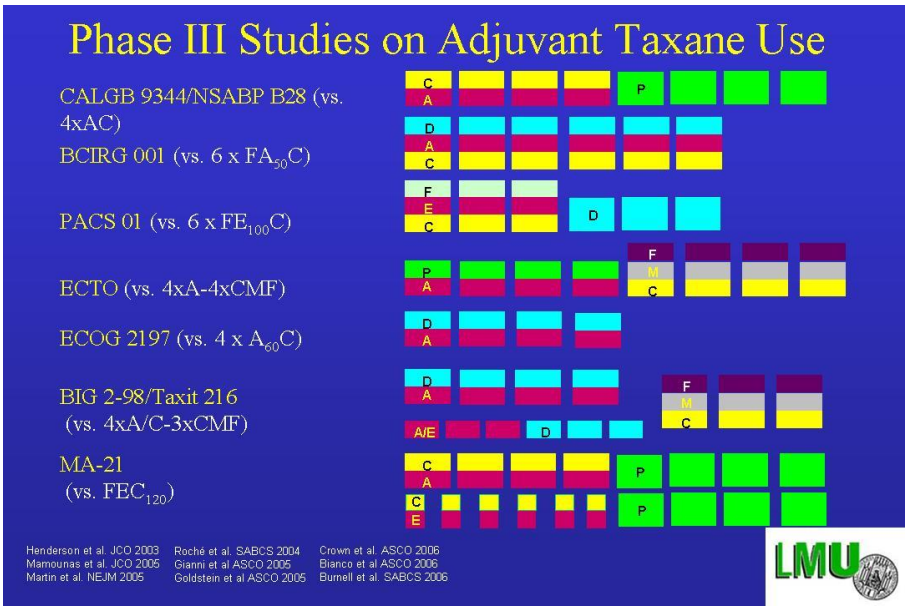
0.81-0.83 [P_.0001]) and lymph node-positive population (RR: 0.84; 95% CI, 0.77-0.92 [P_.0001]). The absolute benefits in DFS and OS in favor of taxanes ranged from 3.3% to 4.6% and from 2.0% to 2.8%, respectively. The authors concluded that considering all the available Phase III trials, taxane-based adjuvantchemotherapy for early breast cancer seems to add a significant benefit.(45)

Figure 8 Overall Survival in the Overall Population of pooled analysis of 9 available adjuvant Phase III taxane trials(46)



Right now, there are 8 treatment protocols evaluating the optimization of docetaxel in the adjuvant treatment of breast cancer with currently 14,104 patients to be enrolled.(46) However, no efficacy results have been demonstrated so far. Optimal use of anthracyclines and taxanes in early breast cancer remains a promising area of research, (47) as will be evaluated by this study.

Figure 9 Synopsis of Trials Evaluating the Role of Taxanes in the Adjuvant Treatment of Breast Cancer



7.3 Gemcitabine in the Treatment of Breast Cancer

Gemcitabine is an antimetabolite drug effective in breast cancer as a single agent and in combination with other chemotherapeutic agents. Its unique mechanisms of action, which involve masked DNA chain termination and several self-potentiating effects on DNA and RNA synthesis enzymes, result in broad and potent activity across many cancer types.(48) In a total of nine studies, gemcitabine monotherapy has reached response rates of up to 37% in the first-line setting, 26% in the second-line setting, and 18% or better in the third-line setting. Gemcitabine is an excellent choice for combination therapy by its unique mechanism of action and favorable toxicity profile, thus limiting the risk of pretreatment-related drug resistance and overlapping toxicity, and by its potential for synergistic interaction with some combination partners as indicated in preclinical studies.(49)

In breast cancer, as a single agent, gemcitabine yields response rates ranging from 14%-37% as first-line therapy for advanced breast cancer and 23%-42% as salvage therapy. However, these were small studies with large confidence intervals around all the indices of benefit including response rate, response duration, and time to disease progression. Gemcitabine is associated with higher response rates when used in

combination with other agents.(48) The combination of gemcitabine and anthracyclines-containing double- and triple-drug combinations used to treat patients with early-stage and advanced breast cancer were promising, with good tolerability and overall response rates ranging from 33%-89% in advanced disease. (50) Numerous phase II clinical studies have combined gemcitabine with other active agents such as taxanes, vinorelbine, vindesine, cisplatin, 5-fluorouracil, as well as anthracyclines across various regimens and conditions of pretreatment. Most of these two-drug combinations have consistently demonstrated higher efficacy than either single agent, particularly in pretreated patients. Even higher efficacy has been obtained with triple-drug regimens including gemcitabine, anthracyclines (epirubicin or doxorubicin), and paclitaxel; these regimens have yielded overall response rates of 58-92% as first-line treatment. (49)

The taxanes are recognized as some of the most active single agents in breast cancer and demonstrate remarkable activity with manageable toxicity in combination with gemcitabine. Gemcitabine, a novel S-phase specific cytidine nucleoside analogue of deoxycytidine, has broad antitumor activity with significant monotherapy activity in breast cancer, with response rates ranging from 22% to 42%, depending on the pretreatment characteristics of the patients. In general, gemcitabine`s favorable single-agent activity and novel mechanism of action, in addition to its largely nonoverlapping toxicities, have facilitated its further development in combination with a variety of chemotherapy agents, including the taxanes. Several phase I and II trials have reported impressive activity for the gemcitabine/taxane doublet with the suggestion of synergism between these 2 classes of agents. Given the remarkable and durable activity reported for this doublet, subsequent phase II trials have focused on optimizing doses and schedules.(51)

Initial studies evaluated a variety of gemcitabine/taxane administration schedules (Table nächste). (52-58)

Early trials focused on fractionated gemcitabine schedules (days 1, 8, and 15) with taxane administration as a single dose. Murad et al reported the results of 29 patients with mostly anthracycline refractory metastatic disease who were treated with 175

mg/m 2 paclitaxel on day 1 followed by gemcitabine on days 1, 8, and 15 at a 1000 mg/m2 every 28 days.(59)

Significant thrombocytopenia (18.5%) experienced by the first 5 patients resulted in elimination of the day 15 gemcitabine dose with the remaining patients treated on a 21-day schedule. Well tolerated, an overall response rate of 55% was noted, including 5 complete responses (CRs). Growth factor use in one third of the patients resulted in only a 9% rate of grade 3/4 neutropenia. The paclitaxel/gemcitabine combination was highly active with more than half of the patients achieving objective responses translating into a median survival of 12 months. However, the highest response rate and median survival was achieved for a gemcitabine/taxane doublet, utilizing the gemcitabine days 1, 8, and 15 schedule with docetaxel.(52) In this multicenter trial, 39 patients (33 had received prior anthracyclines) were treated with docetaxel 100 mg/m2 on day 1 and with gemcitabine 800 mg/m2 on day 1 and 8. Responses were dramatic with an overall response rate of approximately 79% with 2 CRs, 29 partial responses (PR), and 3 with stable disease (SD). Grade 3/4 neutropenia was universal, occurring in all enrolled patients, and with the stipulation that no growth factor use was permitted. Febrile neutropenia, however, was infrequent and evident in only 3 patients. Thrombocytopenia was also infrequent with only 1 patient with grade 3 thrombocytopenia and no grade 4 occurrences. The only remarkable grade 3/4 nonhematologic toxicity, occurring in 13 patients, was asthenia.

Vici et al evaluated higher doses of gemcitabine at 1500 mg/m2 with paclitaxel at 150 mg/m2 on days 1 and 15 at 28-day intervals. (53) All 20 patients evaluable for efficacy had received prior anthracyclines. Overall response rate was 45% with 10% CRs. As a result of mandated growth factor support on days 7-9 and 20-22, only 11% of the patients demonstrated grade 3/4 neutropenia. Median time to progression (TTP) was 8 months. At least 5 additional phase II trials have reported significant activity evident for the gemcitabine/taxane doublet, each exploring a gemcitabine schedule of days 1 and 8 in patients with previously treated MBC. Both Fountzilas et al and Schneeweiss et al used lower doses of gemcitabine at 1000 mg/m2 on days 1 and 8, with docetaxel 75 mg/m 2 on day 1 only. (54, 55) In the first trial, despite all patients being notably anthracycline resistant, median survival surpassed 1 year. Only 19 of 29 enrolled patients were evaluable for efficacy at the time Schneeweiss

reported results with a noted response rate of 47% with 5 CRs. In both trials, grade 3/4 neutropenia occurred in nearly 50% of the patients with nonhematologic toxicity consisting primarily of asthenia and fatigue. (58) Mucositis was the most frequent nonhematologic toxicity with this schedule.

A suggestion of synergism between gemcitabine and taxanes has been evident in at least 2 studies. A day-8 docetaxel schedule with gemcitabine 900 mg/m2 given on days 1 and 8 was administered to 52 patients with anthracycline-pretreated breast cancer, half of whom had also previously been treated with a prior taxane-based regimen. (56) Growth factor used on days 9-16 resulted in only 29% grade 3/4 neutropenia. The overall response rate was 54%, which is surprising, particularly in the light of the fact that just over 50% of enrolled patients had been exposed to prior taxane therapy. Eleven of 25 patients (44%) previously treated with taxane-based regimen demonstrated 1 CR and 10 PRs to the gemcitabine/taxane doublet. Furthermore, 4 of these 11 responders had progressed while being actively treated with the taxane and in 3 of these 4 responders, docetaxel was the front-line taxane administered. Grade 3/4 thrombocytopenia was evident in 21%, reflecting the extensively pretreated patient characteristics. Another study of Alexopoulos et al possessed an entirely unique design. Thirty-six patients with anthracycline-resistant MBC demonstrated either stable (n = 22) or progressive (n = 14) disease after 4-6 cycles of single-agent docetaxel. (57) They went on to continue treatment with docetaxel 100 mg/m2 every 21 days to which gemcitabine was added at 900 mg/m2 on days 1 and 8. There were 3 CRs and 23 PRs among these 36 patients, for an overall response rate of 72%. Of the 14 patients who demonstrated progression to single-agent docetaxel, 9 responded to the combination, as did 17 of the 22 patients with SD. The gemcitabine/docetaxel doublet provided additional benefit over single-agent docetaxel, although no toxicity data were available. The remarkable responses evident in the gemcitabine/docetaxel doublet, despite previous taxane exposure, imply that noteworthy synergism exists between these 2 agents. Collectively, these studies demonstrate that the gemcitabine/taxane doublet may serve as a potent regimen, particularly following anthracycline and/or taxane pretreatment. Irrespective of the schedule used, response rates for the gemcitabine/taxane doublet have ranged from 36% to 79%, depending on the pretreatment characteristics of the

patients. As a final point, although preclinical data are inconclusive with regards to the synergism evident between taxanes and gemcitabine, they clearly support the use of this combination, serving as an impetus to further evaluate this doublet in future phase III trials. (51)

Table 4 Phase II Studies of Gemcitabine and Taxanes in Patients with Pretreated Metastatic Breast Cancer

Study	Number of Patients	Gemcitabine Dosage	Taxane Regimen	ORR
Murad et al[22]	29	1000 mg/m^2 days 1 and 8 every 21 days*	Paclitaxel 175 mg/m^2 day 1 every 21 days	55%
Laufman et al[23]	39	800 mg/m^2 days 1, 8, and 15 every 28 days	Docetaxel 100 mg/m^2 day 1 every 28 days	79%
Vici et al[24]	27	1500 mg/m^2 days 1 and 15 every 28 days	Paclitaxel 150 mg/m^2 days 1 and 15 every 28 days	45%
Fountzilas et al[25]	40	1000 mg/m^2 days 1 and 8 every 21 days for 6 cycles	Docetaxel 75 mg/m^2 day 1 every 21 days for 6 cycles	36%
Lenz et al[26]	29	1000 mg/m^2 days 1 and 8 every 21 days	Docetaxel 75 mg/m^2 day 1 every 21 days	47%
Mavroudis et al[27]	52	900 mg/m^2 days 1 and 8 every 21 days	Docetaxel 100 mg/m^2 day 8 every 21 days	54%
Alexopoulos et al[28]	36	900 mg/m^2 days 1 and 8	Docetaxel 100 mg/m^2 day 8 every 21 days	72%
Brandi et al[29]	37	1000 mg/m^2 days 1 and 8 every 21 days	Docetaxel 80 mg/m^2 day 8 every 21 days	60%

*Reduced from 1000 mg/m^2 on days 1, 8, and 15 every 28 days after the first 5 patients experienced significant thrombocytopenia (18.5%).

As first-line treatment in patients with MBC, the gemcitabine/taxane doublet has generally reported high response rates. (60-62)With paclitaxel at 175 mg/m2 and gemcitabine 1200 mg/m 2 on a days 1 and 8, Genot et al reported outcomes for 40 patients.(67) Two CRs and 13 PRs for an overall response rate of 42% was noted with 76 events of grade 3/4 granulocytopenia. Median TTP was nearly 8 months. Employing the identical regimen, Delfino et al reported a 14% CR rate with an overall response rate of 55%.(60) Only 14% of patients demonstrated grade 3/4 leukopenia. Regardless of the pretreatment characteristics of patients, the paclitaxel/gemcitabine combination resulted in manageable myelosuppression. A first-line treatment of docetaxel 35 mg/m2 given on days 1, 8, and 15 in combination with gemcitabine is underway.(62) Preliminary results for 8 of a goal of 50 patients demonstrate an impressively high response rate of 75%, a figure reminiscent of the 79% response rate noted for standard day 1 full dose docetaxel in the trial by Laufman and

colleagues. Although less hematologic toxicity is expected than was noted in that trial, no toxicity data were available at the time of this preliminary report.

An alternative option, which is followed frequently in the palliative setting, is the gemcitabine-taxane combination in a biweekly schedule (**Fehler! Verweisquelle konnte nicht gefunden werden.**), (63-67) which is, however, not suitable for this study. Since in the SUCCESS B-Study, the question, whether the addition of gemcitabine to docetaxel leads to an optimized taxane efficacy in the adjuvant settings, needed to be addressed in a clear concept, differences in the scheduling between the control and the study arm were not be allowed.

Table 5 Phase II Studies of Gemcitabine and Taxanes in Patients Metastatic Breast Cancer as Biweekly Treatment

Study	Number of Patients	Gemcitabine Dosage	Taxane Regimen	ORR
Colomer et al[34]	43	2500 mg/m^2 day 1 every 14 days	Paclitaxel 150 mg/m^2 day 1 every 14 days	Overall, 69% HER2−, 85% HER2$^+$, 40%
Llombart et al[35]	43	2500 mg/m^2 day 1 every 14 days	Paclitaxel 150 mg/m^2 day 1 every 14 days	68%
Sanchez-Rovira et al[36]	44	2500 mg/m^2 days 1 and 15 every 28 days	Paclitaxel 135 mg/m^2 days 1and 15 every 28 days	45%
Kornek et al[37]	52	1500 mg/m^2 days 1 and 15 every 28 days	Docetaxel 50 mg/m^2 days 1 and 15 every 28 days	61%
Pelegri et al[38]	49	2500 mg/m^2 day 1 every 14 days	Docetaxel 65 mg/m^2 day 1 every 14 days	66%

Because of numerous phase II trials demonstrating efficacy of gemcitabine/taxane combinations as either a first- or second-line treatment of MBC, a phase III randomized trial was undertaken to evaluate the specific question of added benefit of gemcitabine to paclitaxel, an agent approved for monotherapy as first-line treatment of MBC.

An interim analysis of this large, global phase III study, presented at the ASCO 2003 Annual Meeting, demonstrated that first-line treatment with gemcitabine/paclitaxel is more efficacious than paclitaxel alone. (68) This study compared Gemcitabine and Paclitaxel with T in 529 pts with MBC previously treated with an anthracycline, but no prior chemotherapy for MBC. Objectives were overall survival (OS), progression-free survival (PFS), overall response rate (ORR), QoL, palliation of pain, toxicity, and time to progressive disease (TTP; primary interim analysis objective). Patients with

histologically confirmed, measurable metastatic breast cancer with prior adjuvant/neoadjuvant anthracyclines (or non-anthracyclines if clinically contraindicated) and Karnosfyk Prognostic Score >70 were randomized to Gemcitabine and Paclitaxel (G 1250 mg/m2 d1,8; T 175 mg/m2 d1) or Paclitaxel (175 mg/m2 d1) q21d until progressive disease. Between 8/99 and 4/02, 529 pts were randomized (267 Gemcitabine and Paclitaxel and 262 Paclitaxel) at 98 sites. Median age was 53 yrs. Arms were balanced; >70% had visceral metastases, 75% had >2 sites of metastatic disease, one-third had receptor-positive disease, and 96% had prior anthracyclines. Median cycles given were 6 for Gemcitabine and Paclitaxel and 5 for Paclitaxel. Median TTP was 5.4 mos (95% CI, 4.6-6.1 mos) for Gemcitabine and Paclitaxel and 3.5 mos (95% CI, 2.9-4.0 mos) for Paclitaxel (p=0.0013). The Hazard ratio was 0.734 (95% CI, 0.607-0.889; p=0.0015) with an increased probability of ~50% for Gemcitabine and Paclitaxel of being progression-free at 6 mos. PFS was significantly better with Gemcitabine and Paclitaxel (p=0.0021). ORR was 39.3% (95% CI, 33.5%-45.2%) for Gemcitabine and Paclitaxel and 25.6% (95% CI, 20.3%-30.9%) for Paclitaxel (p=0.0007). Gemcitabine and Paclitaxel had numerical improvement in analgesic level, pain relief, and global QoL (p=NS). CTC grade 4 hematologic toxicity was more pronounced with Gemcitabine and Paclitaxel vs Paclitaxel (17.2% vs 6.6% neutropenia, 1.1% vs 0.4% anemia, 0.4% vs 0% thrombocytopenia, 0.4% vs 0% F/N). Non-hematologic toxicity was manageable in both arms. There was 1 toxic death per arm. Conclusions: Gemcitabine and Paclitaxel demonstrated significant efficacy advantages over Paclitaxel in pts with MBC. Toxicity was manageable and expected.(68)

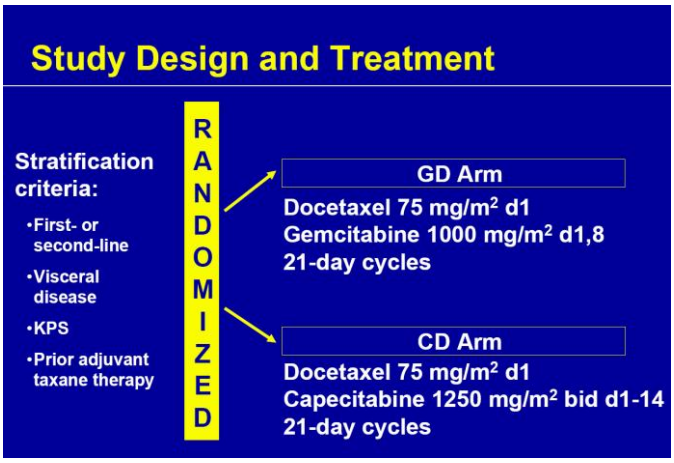
At the ASCO 2005 Annual Meeting in Orlando, USA, Chan et al. reported the results of a prsopectiven phase III study comparing Gemcitabine-Docetaxel with Capecitabine-Docetaxel as first- or second-line treatment in metastatic breast cancer patients pretreated with an anthracycline. Primary objective was progression-free survival (PFS); secondary objectives were overall response rate (ORR), time to treatment failure (TtTF), overall survival (OS), toxicity, and quality of life (QOL). 305 patients with histologically confirmed metastic breast cancer who relapsed after an anthracycline-based regimen either in neoadjuvant or first-line metastatic disease were randomized to Gemcitabine-Docetaxel (G 1000 mg/m2 d1,8; D 75 mg/m2 d1) or

Capecitabine-Docetaxel (C 2500 mg/m2 d1-14; D 75 mg/m2 d1) q21 days; neoadjuvant pretreatment with taxanes was allowed if completed >6 months before entry.

17% of the patients received prior taxane; 34% received prior chemotherapy for metastatic disease. At time of analysis, 287 pts were evaluable for safety and 229 for response. On Gemcitabine-Docetaxel arm, pts received 754 cycles; 642 cycles were given on Capecitabine-Docetaxel arm. ORR was 27% (95% CI, 18.4%-34.7%) for Gemcitabine-Docetaxel, and 31% (95% CI, 22.6%-39.5%) for Capecitabine-Docetaxel (p=.4537).

CTC grade 3/4 hematologic toxicity was similar in both arms, but febrile neutropenia was higher in Capecitabine-Docetaxel arm (12% vs 7%).

Figure 10 Study Design of a European phase III study GD vs. CD



Nonhematologic toxicity was low in both arms, but diarrhea (17% vs 7%), mucositis (16% vs 4%), and hand-foot syndrome (24% vs 0%) were more pronounced in Capecitabine-Docetaxel arm. More serious adverse events occurred in Capecitabine-Docetaxel arm (36% vs 28%), causing discontinuation in 27% (Capecitabine-Docetaxel arm) and 14% (Gemcitabine-Docetaxel arm) of pts. There were 2 toxic deaths, both in Capecitabine-Docetaxel arm.

The authors concluded that Gemcitabine-Docetaxel demonstrated similar efficacy to Capecitabine-Docetaxel in pts with MBC. Nonhematologic toxicity was higher in the Capecitabine-Docetaxel arm.

The available data on the combination of gemcitabine and taxanes justify the use of this combination as experimental treatment in this study for the following reasons:

- Gemcitabine is highly effective in breast cancer
- Gemcitabine and docetaxel might develop synergistic efficacy
- Toxicity profiles of the two substances combine favorably

7.4 Endocrine therapy in breast cancer

Updated results have demonstrated a remarkable risk/benefit ratio for endocrine therapeutic agents in general. It is therefore essential that patients with high risk for recurrence (i.e. all patients recruited into this study) should receive optimal endocrine therapy in case of positive hormone receptor status (≥ 10 % positively stained cells for estrogen and/or progesterone) of the primary tumor.

Tamoxifen, a first generation selective estrogen receptor modulator, has been studied most extensively. The cellular actions of tamoxifen are not completely understood, but it appears that the drug's antiproliferative effects are mediated primarily by inhibition of the activities of estrogen through binding to estrogen receptors. Disease-free and overall survival rates have been increased in postmenopausal women with ER-positive tumors when tamoxifen has been used as adjuvant therapy (irrespective of nodal status). In premenopausal women, adjuvant therapy with tamoxifen has been associated with prolongation of disease-free survival, while its impact on survival is strongly assumed.

(69)

However, it has also conclusively been shown that the benefit of tamoxifen does not accrue to those with estrogen receptor negative primary tumors. Other selective estrogen receptor modulators, such as raloxifen, have not been studied sufficiently to proof comparable efficacy as tamoxifen and therefore will not be used in this study.

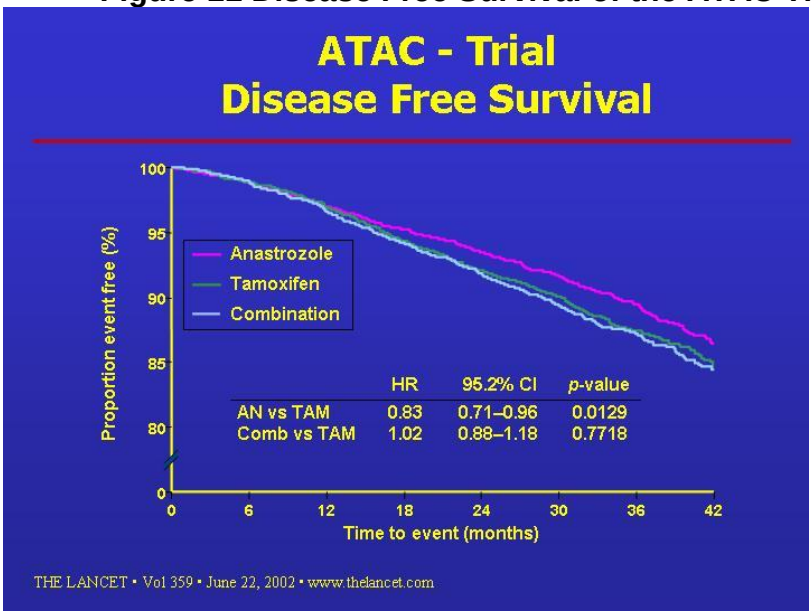
The development of third-generation aromatase inhibitors led to a significant advance in the treatment of metastatic breast cancer. (70)

Aromatase agents result in a total blockade of the peripheral aromatization of androgens in muscle, fat, skin and breast, required for estrogen synthesis. Therefore,

estrogen levels in postmenopausal women are suppressed to approximately 1-10% of pretreatment levels. Whereas in metastatic breast cancer, aromatase agents are established as either first- or second-line therapy for postmenopausal women, only preliminary data for the adjuvant setting are available. In the ATAC trial, comparing five years of tamoxifen versus five years of anastrozole versus five years of both agents in combination, 9366 postmenopausal with positive or unknown hormone receptor status were accrued. A recent update of the efficacy analysis after a median follow-up of 47 months confirmed anastrozole to be superior to tamoxifen in all major efficacy endpoints. DFS estimates were 86.9 % and 84.5 % for anastrozole and tamoxifen, respectively. In the subgroup of hormone receptor positive patients, the benefit for anastrozole was even more apparent (89.0 % vs. 86.1 %). The combination arm showed no significant difference to tamoxifen alone. (71)

In the final analysis, presented at the SABCS 2004 meeting, despite the robust benefit for disease free survival, no significant difference in overall survival for the anastrozole group could be demonstrated. After 68 months of follow-up the hazard ratio for overall survival was 0,97 (p=0.7).(72)

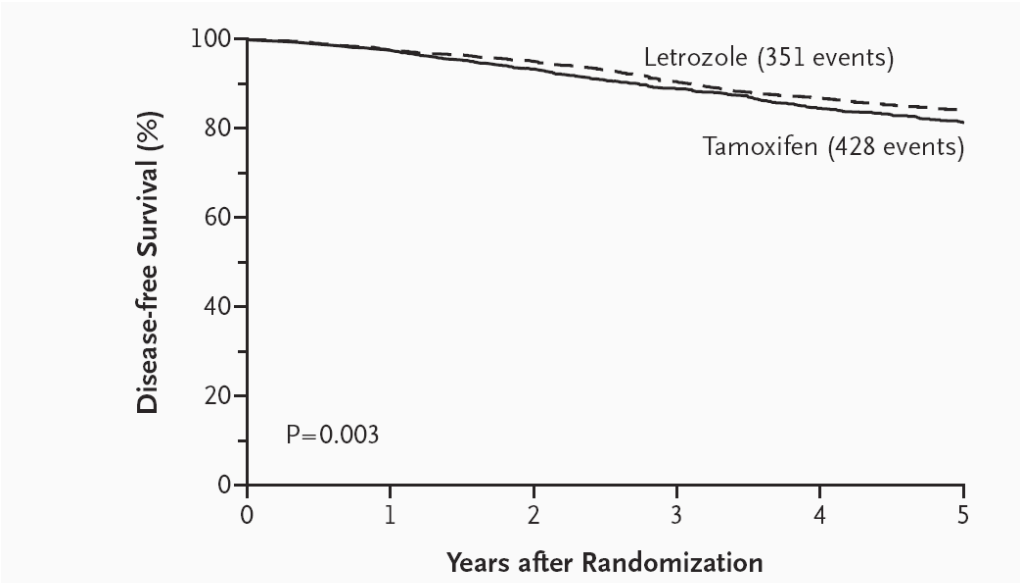
Figure 11 Disease Free Survival of the ATAC Trial (73)



The Breast International Group (BIG) 1–98 trial is another important study that should resolve the issue of when and how aromatase inhibitors should be used in relation to tamoxifen. Briefly, 5 years of letrozole or Tamoxifen monotherapy will be compared with each other, as well as with two sequential treatment regimens (letrozole 2 years

then tamoxifen 3 years or tamoxifen 2 years then letrozole 3 years).Results of the primary core analysis of BIG 1–98 were unavailable and, therefore, not included in the 2005 ASCO guidelines. The 2005 and 2007 St. Gallen international guidelines and the updated NCCN guidelines, however, have acknowledged the first results from the monotherapy groups in the BIG 1–98 trial, including the improvement in diseasefree survival and the significant improvement in distant disease- free survival observed for letrozole compared with tamoxifen.

Figure 12 Disease Free Survival of the BIG 1-98 Trial



Letrozole						
No. at risk	4003	3892	2964	1261	892	567
Disease-free survival (%)		97.7	95.1	90.5	86.8	84.0
Tamoxifen						
No. at risk	4007	3896	2926	1238	866	544
Disease-free survival (%)		97.6	93.4	89.0	84.6	81.4

For receptor-positive, premenopausal women ovarian ablation by surgical means (i.e. laparoscopic oophorectomy) or by pelvic irradiation as one-time treatment offers a reduction in the annual odds of death of 24 %, (74) which is comparable to the results of multi-agent chemotherapy. (75) However, there are concerns about its permanence leading to indefinite loss of childbearing, and the potential of long-lasting adverse effects on heart and bone. For this reason, goserelin has been tested extensively as a component of adjuvant therapy for breast cancer. (69, 70)

Goserelin is a gonadotropin-releasing hormone (GnRH) agonist. Continuous pituitary stimulation by GnRH, normally under pulsatile control, leads to an eventual downregulation of LH and FSH secretion with subsequent diminution of androgen levels. Even though there are no definite results available to answer the question, whether the addition of gosererelin to chemotherapy and tamoxifen provides adequate benefit, an update of the results by Davidson et al. suggests that the effects of adjuvant tamoxifen may be greater among women who have had cessation of ovarian function as a result of either chemotherapy alone or chemotherapy plus goserelin.(76) In this study, enrolling 1,537 patients, premenopausal patients were randomized to six cycles of FAC-chemotherapy, FAC + goserelin and FAC + goserelin and tamoxifen. The overall survival rate for the three groups was 77 %, 78 % and 80 %, respectively. In an update, presented at the 2003 ASCO Annual Meeting, exploratory retrospective subset analysis showed a trend towards a benefit with the addition of goserelin after FAC for women <40 yr, women with premenopausal estradiol level after FAC, or those not amenorrheic after FAC. (77)

In the Zoladex Early Breast Cancer Research Association Trial (ZEBRA) there was no significant difference in outcome after the first 2.5 years of follow-up among goserelin treated patients who continued to be amenorrheic and those whose menses returned. (78) This raises the possibility that a period of transient amenorrhea is sufficient to improve the survival of premenopausal women with hormone receptor positive tumors.(79)

Based on the evidence summarized above, in this study, postmenopausal patients with positive hormone receptor status of the primary tumor received letrozole 2,5 mg p.o. per day for 5 years after the end of chemotherapy. In addition to tamoxifen, all patients with positive hormone receptor status of the primary tumor and under the age of 40 or restart of menstrual bleeding within 6 months after the completion of cytostatic treatment or with premenopausal hormone levels as defined below received goserelin 3.6 mg subcutaneously every 4 weeks over a period of 2 years.(79, 80) Premenopausal endocrine status will be assumed, if the following serum levels are met: LH < 20 mIE/ml, FSH < 20 mIE/ml and E2 > 20 pg/ml. Endocrine therapy will start after the end of chemotherapy. Patients, who meet the

above named criteria for premenopausal status will stay on Tamoxifen treatment for a total of 5 years and will not switch to anastrozole.

7.5 Trastuzumab in the adjuvant treatment of breast cancer

Growth factors and their receptors are known to play critical roles in cell development, growth, and differentiation. (81) Many receptors possess intrinsic tyrosine kinase activity that is activated upon interaction of the receptor with its cognate ligand. The human epidermal growth factor receptor-2 (HER2) is coded by a proto-oncogene mapped to chromosome 17q21.(82) The HER2 gene has homology with the rodent gene neu, and is also referred to as HER2/neu or c-erbB-2. The HER2 gene encodes a 185 kD transmembrane glycoprotein (p185HER2) with tyrosine kinase activity. An overexpression of the HER2 receptor (about 10- to 100-fold compared to normal cells) is observed in a number of primary tumors, suggesting that the overexpression of this growth factor receptor may contribute to transformation and tumorigenesis. In most cases, HER2 protein overexpression is thought to result from gene amplification.(83-86) Approximately 25% to 30% of patients with breast and ovarian cancers overexpress HER2. (87) Similar correlations may exist for other epithelial tumors (lung adenocarcinoma, gastric cancers etc.).

Several lines of evidence support a direct role for p185HER2 expression in the pathogenesis and poor clinical course of human tumors. First, mutation of the rat neu proto-oncogene is associated with the induction of neuroblastomas. (88, 89) Second, when the gene is transfected into mouse fibroblast cells (NIH-3T3) it causes transformation, and the resulting cells are tumorigenic in the nude mouse. (90, 91) Studies using a non-mutated, human HER2 gene have demonstrated that NIH-3T3 cell transformation efficiency, as well as tumorgenicity in the nude mouse, are directly related to the level of HER2 gene expression. (92)

Additionally, studies utilizing the mutated rat neu gene to develop transgenic mice have revealed that animals expressing high levels of the mutated neu transgene (93) as well as normal neu (94) develop breast cancer. Finally, specific antibodies to the extracellular domain of the membrane-based protein encoded by the neu gene or the human HER2 gene inhibit the growth of tumors that overexpress the gene. (95-97) These data are consistent with a direct role for the HER2 proto-oncogene in both

malignant transformation and enhanced tumor genicity, and indicate a potential target for cancer therapy.

The role of HER2/neu overexpression as independent prognostic factor by means of time to disease relapse and overall survival in women with breast cancer was described for the first time by Slamon et al.in 1987. In the meantime, the prognostic importance of HER2/neu overexpression has been confirmed in several analyses.(98-100) However, the independence of this factor may be varying in different groups and stages of the disease, and currently remains a point of discussion. (101) The same holds for the predictive value of HER2 overexpression, i.e. its influence on the probability of therapy success. (101-110)

To improve the course of patients with HER2 overexpressing breast cancer by antagonizing the abnormal function of overexpressed HER2, murine monoclonal antibodies (muMAbs) were produced against the extracellular domain of the HER2 receptor to inhibit the proliferation of human tumor cells overexpressing p185HER2. The most encouraging results were obtained with muMAb 4D5, which produced significant antiproliferative effects in vitro against human breast cell lines that overexpress the HER2 receptor. (111) MuMAb 4D5 has no effect on cell lines that do not overexpress the receptor. (112)

Preclinical in vivo studies with muMAb 4D5 were conducted using both human breast and ovarian cancer heterotransplants from surgical excised human tumor specimens. The tumors were characterized to determine which had amplification/overexpression of the HER2 gene/protein. Results of these studies again established a clear antiproliferative effect against those human tumors characterized by overexpression of the HER2 receptor. No effect was seen on tumor xenografts that did not overexpress the receptor.

The clinical use of therapeutic murine monoclonal antibodies is usually limited because they are immunogenic and the development of neutralising human anti-murine antibodies often precludes repeated administration in patients. To avoid this probable limitation with muMAb 4D5 during clinical use, a humanised chimeric monoclonal antibody containing the hypervariable antigen-binding portions of muMAb 4D5 and a human immunoglobulin variable region framework was constructed.

(113)The resulting recombinant humanised anti-HER2 monoclonal antibody was trastuzumab (rhuMAbHER2), which is 95% human and 5% murine. (113)

Data from four open-label single agent clinical trials (335 patients) and one randomized, controlled clinical trial in combination with chemotherapy support the use of trastuzumab in patients with metastatic breast cancer who have tumors that overexpress HER2.

Trastuzumab was studied as a single agent in four multicenter, open-label, single-arm clinical trials. The largest study recruited 222 patients with HER2 overexpressing metastatic breast cancer who had relapsed following one or two prior chemotherapy regimens for metastatic disease. (114) Patients were treated with 2 mg/kg trastuzumab IV weekly. Patients had extensive prior therapy: 68% had prior adjuvant chemotherapy, 32% had one and 68% had two previous chemotherapy regimens for metastatic disease, and 26% had prior myeloablative treatment with hematopoietic rescue. The overall response rate (CR + PR) in the patients treated with trastuzumab, as determined by an independent Response Evaluation Committee (REC), was 16%, with a 4% complete response and a 12% partial response rate.

The median duration of response as determined by the REC was 9.1 months (range 1.6 to >26 months). Among all treated patients, the median time to disease progression was 3.1 months (range 0 to >28 months). The median survival for all enrolled patients was 13 months (range 0.5 to >30 months). At 5.8 months, 24% of treated patients were free of progression. Quality of life in terms of ‘global quality of live’ and ‘social function’, meassured by the EORTC QLQ-C30, was significantly improved during treatment; no change was seen in physical or role function and in fatigue. (115)

A further study evaluated the effect of trastuzumab as a single agent in 113 patients with HER2 overexpressing metastatic breast cancer with no prior chemotherapy for metastatic disease. Patients were treated with 2 mg/kg trastuzumab IV weekly or 4 mg/kg IV weekly until progression. The overall response rate (CR + PR) was 24% for patients treated with 2 mg/kg and 22% for patients treated with 4 mg/ kg. (116) The most comprehensive data originate from a prospective randomized trial performed on 469 patients. Patients were randomly assigned to receive standard chemotherapy (anthracycline and cylophosphamide or paclitaxel in case

anthracycline-pretreatment) or standard chemotherapy plus trastuzumab. (117) The addition of trastuzumab to chemotherapy was associated with a longer time to disease progression (p<0.001), a higher rate of objective response (p<0.001), a lower rate of death at one year (p=0.008) longer survival (p=0.046), and a 20 percent reduction in the risk of death. The most important adverse event in this study was cardiac dysfunction, which occurred in 27 percent of the group given anthracycline, cyclophosphamide and trastuzumab; 8 percent of the group given an anthracycline and cyclophosphamide alone and 13 percent of the group with paclitaxel and trastuzumab.

In the randomized trial on 469 patients, the addition of Herceptin to chemotherapy improved the RR in the FISH-positive subgroup (54.0% vs 30.8%; P<0.0001), but no such improvement was seen in the FISH-negative subgroup (38.0% vs 37.5%; P=NS). (118) The addition of Herceptin to chemotherapy in the FISH-positive group also provided a significant survival benefit (OR, 0.71; 95% CI, 0.54, 0.92; P=0.009) that was not detected in the FISH-negative group (OR, 1.11; 95% CI, 0.70, 1.80; P=NS). The group of HER2-positive patients in this study >60 years of age appeared to have a worse overall outcome compared with the group ≤60 years of age, possibly related to adverse baseline characteristics. (119) However, in the group >60 years of age, the survival benefit seen with the addition of Herceptin to chemotherapy was significant (odds ratio, 0.64; 95% CI, 0.41-0.99). These data suggest that older (age >60 years) patients with metastatic breast cancer should be considered for first-line use of Herceptin plus chemotherapy.

At the ASCO 2005 Annual Meeting in Orlando, USA, the first prospective data on the efficacy of trastuzumab in the adjuvant treatment of breast cancer were presented. In summary, the data strongly suggest a significant and clinically most relevant benefit in terms of disease free and overall survival, if patients with HER2-positive breast cancer are treated with trastuzumab for one year following chemotherapy.

In the combined analysis of the NSABP-B31/NCCTG-N9831 studies (Doxorubicin and Cyclophosphamide Followed by Paclitaxel with or without Trastuzumab as Adjuvant Therapy for Patients with HER-2 Positive Operable Breast Cancer by Romond EH et al.), more than 3.300 patients were randomized between a sequential AC-Paclitaxel chemotherapy, followed or not by one treatment of trastuzumab. In

summary, the results showed that for node positive HER-2 positive breast cancer, trastuzumab given concurrently with paclitaxel following AC chemotherapy, reduces the risk of a first breast cancer event at 3 years by 52%. The relative risk reduction benefit was present and of similar magnitude in all subsets of patients analyzed. The addition of trastuzumab reduced the probability of distant recurrence by 53% at 3 years, and the hazard of developing distant metastases appears, thus far, to decrease over time.

Figure 13 Disease free survival of NSABP B-31 and N9831 separately

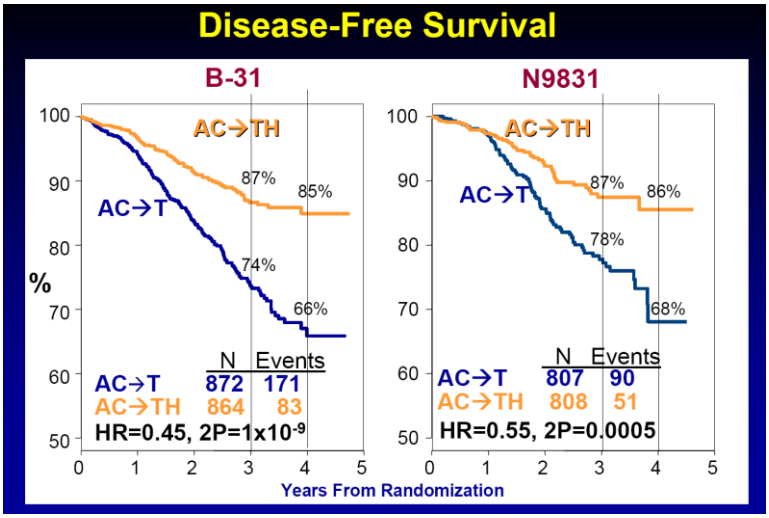
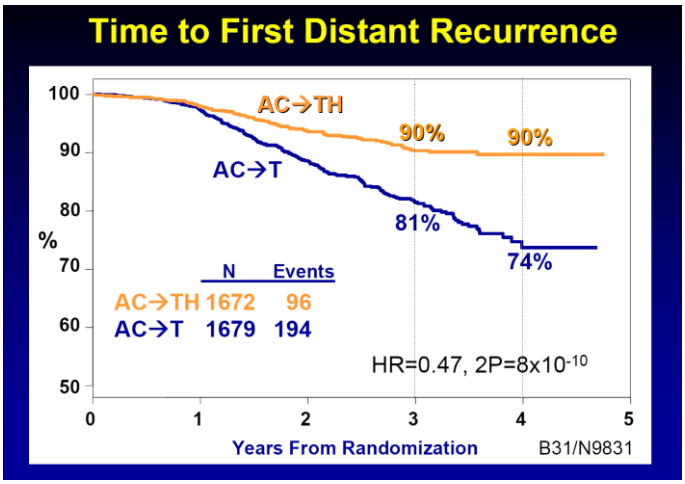


Figure 14 Kaplan Meier analysis of combined analysis for DDFS

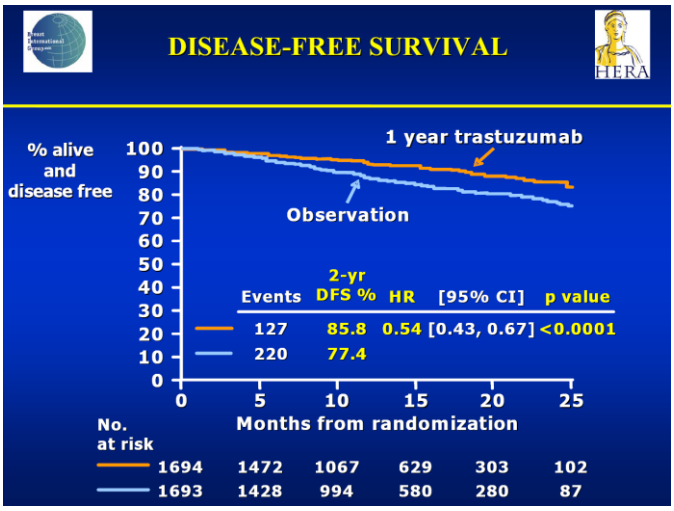


The results at a median follow-up of 2 years also showed a statistically significant survival advantage with a relative risk reduction of 33%. However, the combination of trastuzumab and chemotherapy has a significant risk of cardiac toxicity. The authors

conclude that careful monitoring of cardiac function is of vital importance if trastuzumab is to be used in the adjuvant setting.

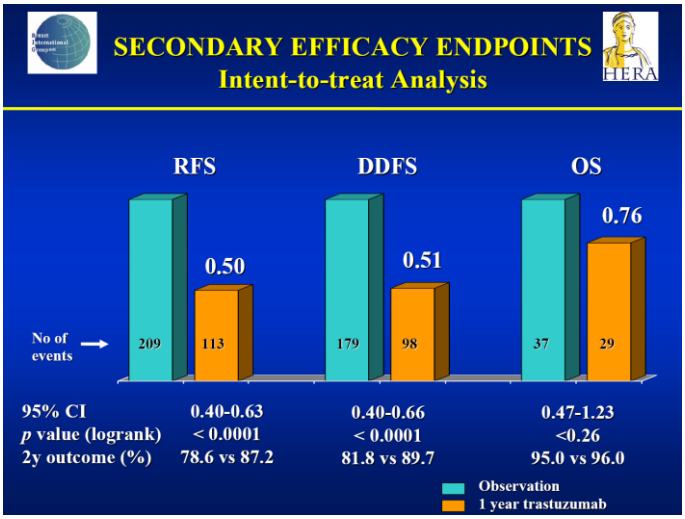
The results were in essence confirmed by the HERA-Study, which was presented by M. Piccart-Gebhardt. The HERA-Study was a randomized three-arm multi-centre comparison of 1 year trastuzumab, 2 years trastuzumab or no trastuzumab in women with HER-2 positive primary breast cancer who have completed adjuvant chemotherapy. At one year median follow-up, trastuzumab given every 3 weeks for one year following adjuvant chemotherapy, significantly prolonged disease free survival and recurrence free survival for women with HER-2 positive early breast cancer. Trastuzumab also significantly reduced the risk of distant metastases. These benefits were independent of patients' baseline characteristics (nodal status, hormone receptor status) and of type of adjuvant chemotherapy received.

Figure 15 Kaplan Meier analysis of the HERA Study for DFS



Overall survival was not significant between the study groups at the time of interimanalysis. Trastuzumab therapy was associated with a low incidence of severe symptomatic congestive heart failure.

Figure 16 Kaplan Meier analysis of the HERA Study for DDFS and OS



Although trastuzumab-based regimens have improved both systemic control and overall survival in patients with HER2 positive breast cancer, some patients continue to develop tumour progression in spite of these trastuzumab-containing regimens, as a result of de novo or acquired resistance to the drug.

Although the overall benefit versus risk ratio of trastuzumab is clearly a favorable one, further progress would be made with the use of anti-HER2 therapies, retaining the activity of trastuzumab but showing less cardiotoxicity.

HER2 overexpression/amplification appears to be an important risk factor for the development of brain metastases. Central nervous system (CNS) progression is emerging as a major clinical problem in HER2 positive patients. The increased incidence of brain metastases on trastuzumab therapy is generally not thought to be the result of decreased HER2 expression in the CNS, but rather, the inability of the drug to penetrate the blood-brain barrier. The increased incidence of brain metastasis is likely a reflection on both the inherent behaviour of HER2-positive tumours, as well as the prolonged survival in these patients, which has allowed more CNS metastases to become clinically evident before death.

7.6 Radiotherapy

Halsted's demand for radical mastectomy as treatment of choice for breast cancer dates back to the 1880's and was based on the understanding of breast cancer as a locoregional disease. This view was questioned by a series of studies between 1950 and 1970.(120) These studies established the advantages of limited local therapy and finally led the way to breast conserving surgery. (76) Adjuvant radiotherapy is undoubtedly an integral part of the concept of breast conserving surgery. Several randomized trials comparing conservative surgery alone with conservative surgery plus radiotherapy have demonstrated an average reduction in the risk of disease recurrence in the breast of 84% with the use of radiotherapy. This reduction in the recurrence rate translates into a significant survival benefit (EBCTCG ESTRO 2006) At present, a group of patients who do not require radiotherapy has not been reproducibly identified, and radiotherapy should remain a part of breast-conserving therapy for invasive carcinoma. (78, 79)

Until the late 1970's, post mastectomy radiotherapy was routinely administered. In the face of increased risk of local recurrence in patients who received no adjuvant radiotherapy, it was assumed that optimal tumor control could be achieved by routine postoperative radiotherapy. The expanding knowledge about early systemic dissemination of tumor cells and its predominant role in overall prognosis of the disease changed this policy in the early 1980's. (81) It has now been widely accepted for many years that postoperative radiotherapy of the chest wall after mastectomy should be restricted to cases with advanced stages of disease and/or with extensive lymphangiosis carcinomatosa and positive margins of resection. (82, 83) However, more recent studies have shown that the increased risk for local recurrence, which is associated with a more selective use of chest wall irradiation, might also lead to a reduced overall survival. (84)

Beginning in the 1970's, however, data were published showing that the increased incidence of local recurrence is not necessarily associated with a less favorable overall survival rate. (85, 86) In 1986, the results of two large, international, randomized, controlled trials (Stockholm Study (87) and Oslo Study (88)) provided a more definite and detailed understanding that, while post mastectomy irradiation might improve overall survival in advanced disease (> pT2 tumors), it did not do so in

early, locally limited breast cancer. (89) This was also confirmed in 1987 by Griem et al. in patients undergoing chemotherapy. In their study, 510 patients, with T1-T3 tumors and positive nodes or tumors larger than 5 cm and negative nodes, were treated with mastectomy and chemotherapy. Patients were then randomized to receive either no further treatment or adjuvant radiotherapy. The rate of local recurrence in patients with chemotherapy alone was 14 % compared to 5 % in those who received both chemotherapy and radiotherapy. However, no significant difference was seen in the overall survival rate. (90, 91)

Adjuvant radiotherapy of the chest wall following mastectomy reduces the risk of local recurrence in breast cancer of all stages. While this decrease may have no impact on the overall survival rate in early breast cancer(92), the update of the EBCTCG meta-analysis based on >7500 patients showed a significant improvement of overall survival at 15 years of absolute 6.2 % in patients with <3 involve lymph nodes, and a borderline significant (p=0.05) survival advantage of 5.3% in patients with 1-3 involves axillary lymph nodes EBCTCG ESTRO 2006). In accordance to the interdisciplinary German S3-guideline for diagnostics and therapy of breast cancer, adjuvant radiotherapy after mastectomy is recommended in the following situations:

in all cases if at least of the following factors is present:

- T3/T4
- R1-/R2-Resection
- pN+(> 3 involved axillary lymph nodes)
- in cases of 1-3 involved axillary lymph nodes if additional risk factors are present. (additional risk factors are: lymphangiosis carcinomatosis, vessel involvement, multicentric growth, involvement of the pectoralis fascia, safety margin<5m

8 Study objectives

8.1 Primary objective

The primary objective of this study was to compare the disease free survival after randomisation in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy, both arms followed by biological anti-HER2 treatment respectively.

8.2 Secondary objectives

The secondary objectives of this study were to compare the following items in the four regimen arms:

- Overall survival time after randomization
- Distant disease free survival
- Toxicity
- Changes in quality of life over time as defined by EORTC QLQ-C30 and QLQ-
- BR23 questionnaire
- Skeletal related events
- Incidence of secondary primaries
- Endpoints of adjunct translational research program

8.3 Additionaly scientific objectives

- The predictive and prognostic value of MRD surveillance as defined in the relevant section of this protocol
- The predictive and prognostic value of additional surveillance markers as defined in the relevant section of this protocol
- Evaluation of genomic alterations in respect to tumor biology, treatment efficacy and systemic toxicity of antitumor agents.

9 Investigational plan

9.1 Overall Study Design and Plan-Description

This was an open-label, multicenter, randomized controlled, Phase III study comparing the disease free survival after randomisation in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy, both arms followed by biological anti-HER2 treatment respectively. Patients were required to have histopathological proof of a HER2-neu positive tumor and: axillary lymph node metastases (pN1-3) or high risk node negative, defined as: 'pT ≥ 1c or histopathological grade ≥ 2, or age ≤ 35 or negative hormone receptor', if chemotherapy was indicated, but were not allowed to have evidence of distant disease. Patients had to be entered into the study no later than 6 weeks after complete resection of the primary tumor. No other antineoplastic treatment other than surgical treatment, the defined cytotoxic and endocrine treatment and radiotherapy wereallowed prior to study entry and during the course of the study.

After surgery, leading to R0 resection of the invasive and intraductal components of the primary tumor, patients were randomized to one of the following treatments:

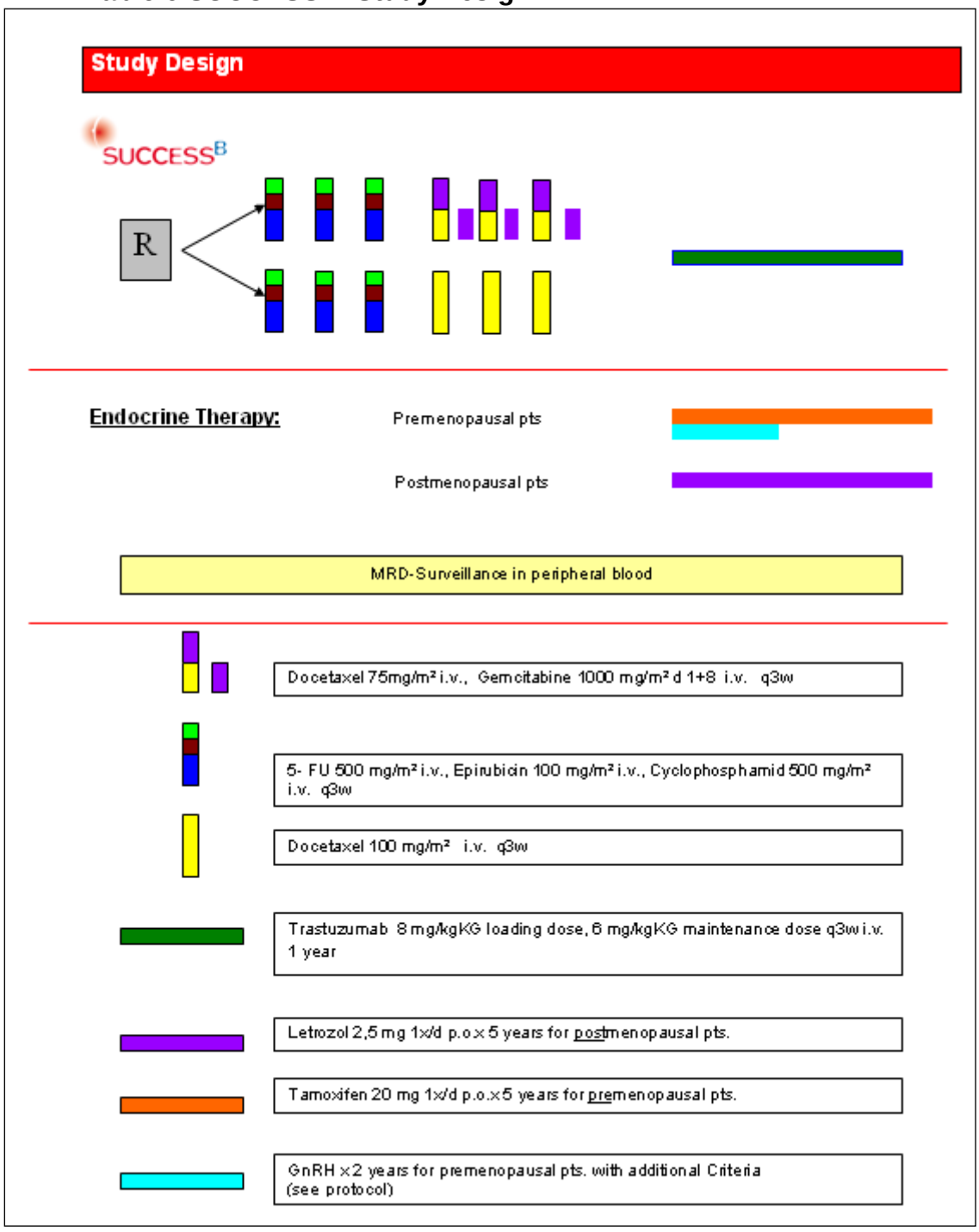
Randomization

A: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3 cycles of Docetaxel 75 mg/m² body surface area i.v. (D), and Gemcitabine 1000 mg/m² i.v. (30 min infusion) (G), administered on day 1, followed by Gemcitabine 1000 mg/m² i.v. (30 min infusion) on day 8, repeated on day 22

B: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3

cycles of Docetaxel 100 mg/m² body surface area i.v. (D), administered on day 1, repeated on day 22

Table 6 SUCCESS B Study Design



After the end of chemotherapy all patients premenopausal patients received received biological anti-HER2 treatment with Trastuzumab (Herceptin®). Loading dose: 8 mg/kg body weight for the first

application (route i.v. , 90 min.) , Maintenance dose: 6 mg/kg body weight for all further applications (route i.v. 90 min.) day 1 q day 21 for 52 weeks.

Postmenopausal patients with positive hormone receptor status (≥ 10 % positively stained cells for estrogen and/or progesterone) of the primary tumor were treated subsequently with letrozole (Femara®) 2,5 mg p.o. for 5 years, Tamoxifen treatment for 5 years after the end of chemotherapy. In addition to tamoxifen, all patients with positive hormone receptor status of the primary tumor and under the age of 40 or restart of menstrual bleeding within 6 months after the completion of cytostatic treatment or with premenopausal hormone levels as defined below will receive goserelin (Zoladex®) 3.6 mg subcutaneously every 4 weeks over a period of 2 years. (1, 2) Premenopausal endocrine status will be assumed, if the following serum levels are met: LH < 20 mIE/ml, FSH < 20 mIE/ml and E2 > 20 pg/ml. Endocrine therapy will start after the end of chemotherapy. All patients with breast conserving therapy or at least 4 axillary lymph node metastases received adjuvant radiotherapy following the completion of the systemic cytotoxic treatment. If necessary to meet patients' needs or for logistic reasons, it was also allowed to administer the radiotherapy intermittently following the completion of 50 % of the cytotoxic treatment.

Each patient remained in the study until either the patient or the investigator determine discontinuation to patient's best interest. The treatment had to be discontinued in any case of intolerable toxicity.

Each patient's treatment modality was unknown until the time of randomization. Randomization was stratified on the baseline prognostic variable of metastatic axillary lymph node involvement and the hormone receptor status of the primary tumor, histopathological grading, menopausal status, as well as the study center.

For each factor the following strata were formed:

- Metastatic axillary lymph node involvement:
    - No evidence of metastatic axillary lymph nodes or unknown axillary status
    - 1-3 metastatic axillary lymph nodes

  - 4-9 metastatic axillary lymph nodes vs.
  - ≥ 10 metastatic axillary lymph nodes
- Hormone receptor status (≥ 10 % positively stained cells for estrogen and/or progesterone) of the primary tumor:
  - negative vs.
  - positive
- Histopathological grading:
  - G1 vs.
  - G 2-3
- Menopausal status
  - Premenopausal
  - Postmenopausal

Docetaxel medication for node negative patients was provided by the manufacturer Sanofi-Aventis at his own cost. Gemcitabine medication for all patients in treatment arm B was provided by the manufacturer Lilly at his own cost.

Peripheral blood samples were drawn from each patient before starting chemotherapy, after the completion of the chemotherapy and in case of disease recurrence. In case of considerable risk for subsequent relapse, as defined in the relevant chapter of the translational research program, patients were subjected to intensified follow-up, including chest x-ray, liver sonography and bone scan.

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

A study design without controls is appropriate for the goals of this study because all treatment regimes have been proven to be effective in the treatment of patients with

breast cancer as described in the background section. The study population has been restricted to patients with high-risk for relapse. The high risk for relapse in these patients is defined by evidence of axillary lymph nodes status or by additional tumor biological characteristic, as defined in the inclusion criteria. Because of the potential toxicity of therapy regimens, those patients with a low risk for recurrence will be excluded from the study.

9.3 Selection of Study Population

An informed consent was obtained from each patient after the nature of the study was explained.

9.3.1 Criteria for enrollment

The terminology for the criteria for enrollment were defined as:

Enter The act of obtaining informed consent for participation in a clinical study from individuals deemed potentially eligible to participate in the SUCCESS B-Trial. Individuals ***entered*** into the study are those for whom informed consent documents for the study have been signed by the potential study participants.
Adverse events are reported for each individual who has entered the SUCCESS B-Trial, even if the individual is never assigned to a treatment group.

Enroll The act of assigning an individual to a treatment group. Individuals who are ***enrolled*** in the SUCCESS B-Trial are those who have been assigned to a treatment group.
A person who has been entered into the SUCCESS B-Trial is potentially eligible to be enrolled in the study, but must meet all criteria for enrollment specified in the protocol before being enrolled (assigned to a treatment group).
Individuals who are entered into the SUCCESS B-Trial but fail to meet the criteria for enrollment are not eligible to participate in the study and will not be enrolled.
Adverse events are reported for all individuals who have entered the study and all individuals who are enrolled in the SUCCESS B-Trial (assigned to treatment groups).

The numbering system used for inclusion and exclusion criteria provides a unique number for each criterion and allows for efficiency in data collection.

9.3.2 Inclusion criteria

Patients may be included in the study only if they meet all the following criteria:

[1.] Primary epithelial invasive carcinoma of the breast pT1-4, pN0-3, M0

[2.] Evidence of HER2-neu overexpressing (IHC +++) or amplifying (FISH +) tumor

[3.] Histopathological proof of axillary lymph node metastases (pN1-3) or high risk node negative, defined as at least one criterion of the following: 'pT≥1c, histopathological grade ≥2 , age ≤ 35, negative hormone receptor', if chemotherapy is indicated.

[4.] Complete resection of the primary tumor with margins of resection free of invasive carcinoma not more than 6 weeks ago

[5.] Females ≥ 18 years of age

[6.] Performance Status <2 on ECOG-Scale

[7.] Adequate bone marrow reserve: leucocytes ≥ 3.0 x 109/l and platelets ≥ 100 x 109/l

[8.] Bilirubin within one fold of the reference laboratory's normal range, ASAT (SGOT), ALAT (SGPT) and AP within 1,5 fold of the reference laboratory's normal range for patients

[9.] Intention of regular follow up visits for the duration of the study

[10.] Ability to understand the nature of the study and to give written informed consent

[11.] Women of childbearing potential must agree to use an effective method of contraception (Pearl-Index < 1, e.g. , intrauterine devices or sterilization) during treatment and for at least 6 months thereafter.

9.3.3 Exclusion criteria

Patients will be excluded from the study for any of the following reasons:

[12.] Inflammatory breast cancer

[13.] Previous or concomitant cytotoxic or other systemic antineoplastic treatment which is not part of this study

[14.] A second primary malignancy (except in situ carcinoma of the cervix or adequately treated basal cell carcinoma of the skin)

[15.] Cardiomyopathy with impaired ventricular function (NYHA > II), cardiac arrhythmias influencing LVEF and requiring medication, history of myocardial

infarction or angina pectoris within the last 6 months, or arterial hypertension not being controlled by medication

[16.] Any known hypersensitivity reaction against docetaxel, epirubicin, cyclophosphamide, gemcitabine or any other medication included in the study protocol. The contraindication, warning notices and measures of precaution of the products, as notified in the product information, have to be respected

[17.] Instable diabetes mellitus, out of sufficient medical control

[18.] Use of any investigational agent within 3 weeks prior to inclusion

[19.] Patients in pregnancy or breast feeding (in premenopausal women anticonception has to be assured)

9.3.4 Violation of Criteria for Enrollment

The criteria for enrollment must be followed explicitly. If there was an inadvertent enrollment of individuals who did not meet enrollment criteria, these individuals hadto be discontinued from the study. Such individuals could remain in the study only if there were ethical reasons to have them continue. In these cases, the investigator had to obtain approval from the sponsor for the study participant to continue in the study.

9.3.5 Disease Diagnostic Criteria

Patients were required to have histopathological proof of a HER2-neu positive tumor and: axillary lymph node metastases (pN1-3) or high risk node negative, defined as: ‘pT ≥ 1c or histopathological grade ≥ 2, or age ≤ 35 or negative hormone receptor’, if chemotherapy was indicated, but were not allowed to have evidence of distant disease. The complete resection of the primary tumor with margins of resection free of invasive carcinoma had to be verified by histopathological examination. The estrogen receptor status had to be evaluated by immunohistochemistry scoring semi-quantitatively the intensity of staining. Distant metastatic disease had to be excluded by chest x-ray, ultrasonography of the liver and whole body bone scan.

Note that the AJCC TNM-Classification Breast Cancer 6th edition had to be used as of July 1st 2003. The following paragraph summarizes the changes, compared to the previous edition of the classification system. (121)

Summary of Changes AJCC TNM-Classification Breast Cancer 5th vs. 6th edition

- Micrometastases are distinguished from isolated tumor cells on the basis of size and histologic evidence of malignant activity.
- Identifiers have been added to indicate the use of sentinel lymph node dissection and immunohistochemical or molecular techniques.
- Major classifications of lymph node status are designated according to the number of involved axillary lymph nodes as determined by routine hematoxylin and eosin staining (preferred method) or by immunohistochemical staining.
- The classification of metastasis to the infra-clavicular lymph nodes has been added as N3.
- Metastasis to the internal mammary nodes, based on the method of detection and the presence or absence of axillary nodal involvement, has been reclassified. Microscopic involvement of the internal mammary nodes detected by sentinel lymph node dissection using lymphoscintigraphy, but not by imaging studies or clinical examination, is classified as N1. Macroscopic involvement of the internal mammary nodes as detected by imaging studies (excluding lymphoscintigraphy) or by clinical examination is classified as N2 if it occurs in the absence of metastases to the axillary lymph nodes, or as N3 if t occurs in the presence of metastases to the axillary lymph nodes.
- Metastasis to the supraclavicular lymph nodes has been reclassified as N3 rather than M1.

9.4 Treatment

9.4.1 Docetaxel

Docetaxel is one of two currently available taxanes. Taxanes are a member of the plant alkaloid group, which also comprises vinca alkaloids and epipodophyllotoxins. The drug is derived from the Pacific yew (Taxus brevifolia). It functions by stabilizing microtubules and thereby preventing their disassembly.

The incidence of treatment-related mortality associated with docetaxel therapy is increased in patients with abnormal liver function. Patients with elevations of bilirubin or abnormalities of transaminase concurrent with alkaline phosphatase are at increased risk for the development of grade 4 neutropenia, febrile neutropenia, infections, severe thrombocytopenia, severe stomatitis, severe skin toxicity and toxic

death. For this reason, Bilirubin had to be within one fold of the reference laboratory's normal range, ASAT (SGOT), ALAT (SGPT) and AP within 1,5 fold of the reference laboratory's normal range for patients to be included into this study.

Taxane infusions are frequently associated with hypersensitivity reactions manifested initially by hypotension, bronchospasm, and urticaria. The risk for hypersensitivity reactions seems to be lower in docetaxel than in paclitaxel. However, extensive premedication is advised in patients with a history or disposition for hypersensitivity reactions and significantly abrogate this problem. Bradyarrhythmias, especially AV block, atypical chest pain, and rarely more severe cardiac problems have also been associated with taxane infusions. Bone marrow suppression with neutropenia is the dose-limiting toxicity. For this reason, only patients with adequate bone marrow reserve (neutrophils $\geq$ 1.5 x 109/l and platelets $\geq$ 100 x 109/l) were included into the study. Peripheral neuropathy with paresthesias in a glove-stocking distribution also is common. Peripheral neuropathy is only in part reversible and there is no advisable precaution to prevent this complication to date. Severe fluid retention occurs in approximately 6.5 % of the patients despite the use of a 3-day dexamethasone premedication, as planned with this protocol. The severe fluid retentions may comprise poorly tolerated peripheral edema, generalized edema, pleural effusion requiring urgent drainage, dyspnoea at rest, cardiac tamponade, or pronounced abdominal distention due to ascites. Patients with pre-existing effusions should be closely monitored from the first dose for the possible exacerbation of the effusions. When fluid retention occurs, peripheral edema usually starts in the lower extremities and may become generalized with a median weight gain of 2 kg. Other toxicities include mucositis, myalgias, and alopecia.(122)

9.4.2 Cyclophopshamide

Cyclophosphamide is member of the alkylating agent family, one of the most widely used antitumor agents. These drugs lead to inhibition of DNA synthesis by forming covalent bonds with nucleic acids. Most alkylating agents are bifunctional and are efficient at cross-linking DNA with subsequent strand breakage and ultimately cell death. These agents add alkyl groups to the N-7 guanine in addition to other nitrogen or oxygen positions in adenine or cytidine. Although alkylation of DNA can occur at any phase of the cycle, cytotoxicity is greatest in those cells that are progressing

through the cell cycle. Cyclophosphamide is only active after microsomal liver metabolism to 4-hydroxycyclophosphamide. It is further metabolized in peripheral tissues to phosphoramide mustard and to acrolein.

The most common dose-limiting toxicity of cyclophosphamide is myelosuppression. The severity and duration varies with the individual drugs, but is moderate with cyclophosphamide. This agent is also quite emetogenic and requires extensive premedication. Cyclophosphamide therapy may be complicated by hemorrhagic cystitis, believed to be due to the metabolite acrolein, which is excreted unchanged in the urine. Adequate hydration and administration of the bladder protectant mesna, can prevent this complication and were planned in the study protocol. Cyclophosphamide is also associated with a syndrome of inappropriate antidiuresis due primarily to a distal renal tubular effect. Amenorrhea and ovarian atrophy, sometimes permanent, have been associated with alkylating agent therapy in women. Because of the possible resumption of normal menstrual cycles, which is inversely related to the age of the patient and the cumulative dose received, ovarian ablation had been included into this protocol for all patients under the age of 40.

A serious long-term complication of alkylating agent chemotherapy is the development of secondary leukemias. In patients who have received an alkylating agent as part of combination chemotherapy, the incidence of secondary acute myeloid leukemia may be as high as 5 to 10 %. This data, however, originate from patients treated for lymphomas. In breast cancer patients the risk for the development of secondary leukemias is presumably lower because of lower doses of the alkylating agent and because of a lower baseline risk for leukemias in these patients compared to patients with lymphomas. (122)

9.4.3 Flurouracil

5- Fluorouracil (5-FU) is a fluorinated pyrimidine-analogue antimetabolite. For cytotoxicity, fluorouracil requires intracellular activation to one of several metabolites. Fluordeoxyuridine monophosphate is a potent inhibitor of thymidylate synthase, an enzyme necessary for the synthesis of dTTP and ultimately DNA. Fluorouridine triphosphate incorporates into RNA and interferes with its processing and function. Fluorodeoxyuridine triphosphate is incorporated into DNA and eventually leads to DNA strand breakage. The importance of each of these mechanisms to fluorouracil-

induced cytotoxicity has not been fully delineated. Studies suggest that tumor cells may be deficient in reduced folates, so leucovorin has been administered fluorouracil in an attempt to increase antitumor activity.
Like most antimetabolites, fluorouracil's toxicity is schedule dependent. With bolus infusion, bone marrow suppression predominates whereas with continuous infusion therapy gastrointestinal toxicity may be more limiting. For this reason, an infusion interval of 15 minutes had been decided for this study. Fluorouracil has been infrequently associated with myocardial ischemic syndrome characterized by chest pain, ECG and isoenzyme changes. For this reason, patients with cardiomyopathy and impaired ventricular function (NYHA > II), cardiac arrhythmias influencing LVEF and requiring medication, and patients with a history of myocardial infarction or angina pectoris within the last 6 months, or arterial hypertension not being controlled by medication were be excluded from this study. Neurological symptoms, usually reversible, have been reported that include headaches, cerebellar ataxia and somnolence. Dermatological complaints are not uncommon and include dermatitis, hyperpigmentation, and skin atrophy.(122)

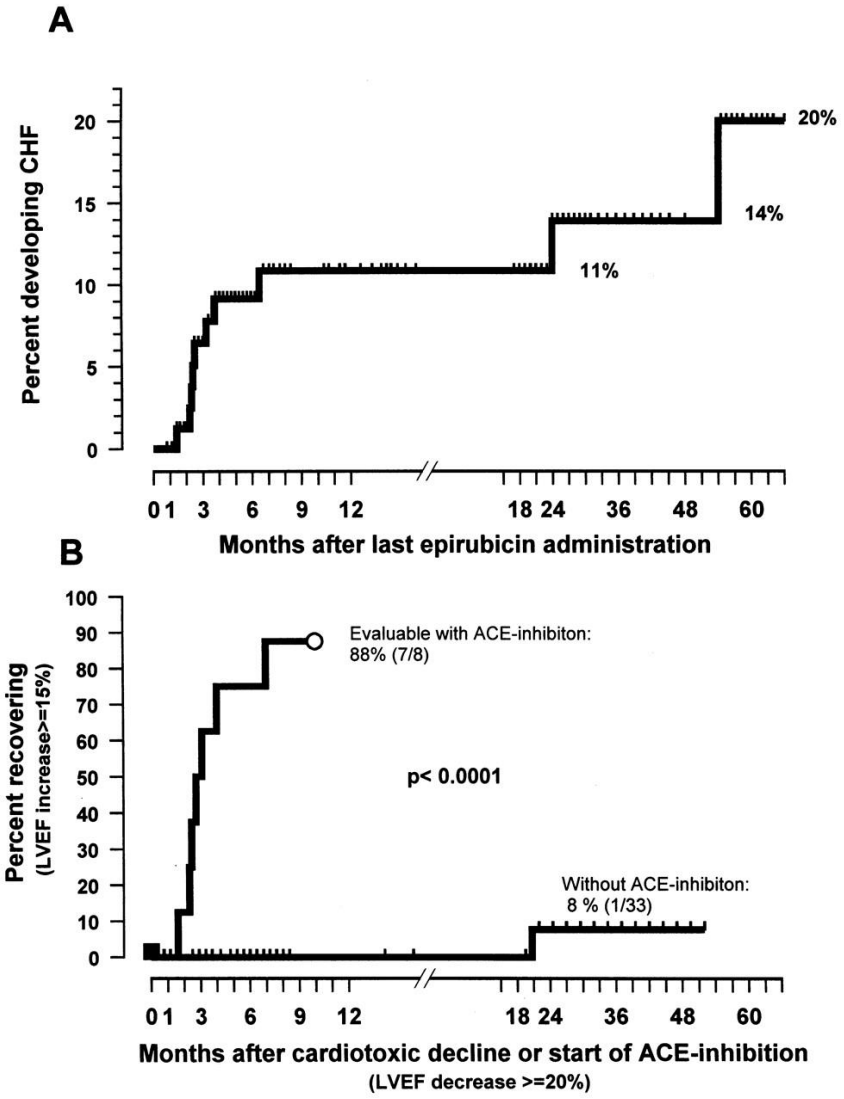
9.4.4 Epirubicin

Epirubicin is member of the anthracycline group of antitumor antibiotics. The anthracyclines are cell-cycle active and phase nonspecific but have pleiotropic actions upon the cell. Although they are classic DNA intercalating agents, their mechanism of cytotoxicity is likely related to interaction with the enzyme topoisomerase II with production of double-stranded DNA breaks. Other data suggest that the anthracyclines undergo one- and two-electron reductions generating intracellular free radicals, particularly the hydroxyl radical, which is highly cytotoxic. Epirubicin is highly myelosuppressive. For this reason, only patients with adequate bone marrow reserve (neutrophils ≥ 1.5 x 109/l and platelets ≥ 100 x 109/l) were included into the study. White cell and platelet count nadirs will occur 10 to 14 days after treatment. The anthracyclines cause gastrointestinal toxicity including acute nausea and vomiting and mucositis later. These agents are severe vesicants. Extravasations during infusion can lead to local tissue necrosis. In extreme cases skin grafting or even amputation may be required. For these reason, great caution as to the infusion sight should be given. The i.v. line has to be tested with saline

infusions cautiously before administering epirubicin. Anthracyclines are cleared predominantly by liver metabolism. For this reason, SGOT, SGPT, Alkaline Phosphatase, Bilirubin, Albumin have to be within 1.5 fold of the reference laboratory's normal range for patients to be included into this study. Long-term administration of anthracyclines is limited by cumulative dose-dependent cardiotoxicity. Irreversible cardiomyopathy with serious congestive heart failure is a significant risk in patients who have received doses in excess of 900 mg epirubicin/m2 body surface area. The cumulative epirubicin dose will be 360 mg/m2 body surface area in treatment Arm A and 720 mg/m2 body surface area in the mainly anthracycline-based treatment Arm B. For this reason, great caution should be taken when anthracycline containing second line chemotherapies will be considered in these patients.(122)

Recent publications demonstrate that the incidence of cardiomyopathy increases significantly within the first 6 months of the last application of epirubicin. (123, 124) A common property of cardiac toxicity associated with cardiac matrix alterations, including anthracycline cardiotoxicity, is the salutary effect of prolonged ACE inhibition. Without ACE inhibition the prognosis of anthracycline-induced CHF is grave , resembling the general prognosis of CHF and idiopathic cardiomyopathy with a mortality rate of about 50% within 2 years of diagnosis.(123, 125, 126) In a current prospective study with ACE inhibition only 1 of 10 patients with severe heart failure (NYHA class III–IV) died of CHF. The patients with a decreased cardiac function did not spontaneously recover during the observation period but function could only be reversed by ACE inhibition for several months. The investigators have successfully treated a total of more than 60 patients with severe CHF after anthracycline therapy with ACE inhibition, with a remarkably long-lasting recovery evaluated clinically and by LVEF determination.(123) This corresponds to trials with ACE inhibition documenting the necessity of ACE inhibitor therapy lasting years in heart failure,(127, 128) and this should probably also be the case after anthracycline-induced CHF.

Figure 17 Risk of epirubicin-induced congestive heart failure (CHF) (A) and recovery after angiotensin-converting enzyme (ACE) inhibition (B)(123)



For the reasons mentioned above, LVEF monitoring by ultrasoundcardiography was performed whenever cardiac symptoms occurred, which justify this examination.

9.4.5 Tamoxifen

Tamoxifen, a first generation selective estrogen receptor modulator, has been studied most extensively. It can be employed both as an adjuvant in estrogen receptor positive women and as palliative therapy for metastatic disease to the estrogen receptor and appears to function as a weak agonist/antagonist. The cellular actions of tamoxifen are not completely understood, but it appears that the drug's antiproliferative effects are mediated primarily by inhibition of the activities of

estrogen through binding to estrogen receptors. It has a long plasma half-life and requires 4 weeks or longer to achieve steady-state levels. Tamoxifen can cause amenorrhea, hot flashes, and occasionally nausea and vomiting. It has been reported to modestly increase the risk of thromboembolic phenomenon. The risk for endometrial cancer among women with breast cancer might increase following use of tamoxifen, recently classified as a carcinogen of the human endometrium. However, this risk is small and, by far, outweight by the antitumoral effects of tamoxifen.(129, 130) Changes in serum lipid profiles also have been noted. (122)

9.4.6 Letrozole

Aromatase inhibitors inhibit several enzymes responsible for the conversion of androgens to estrogens in the peripheral tissues. There are two types of aromatase inhibitors, irreversible steroidal activators and reversible nonsteroidal imidazole-based inhibitors. Although both types interfere with the final step in estrogen biosynthesis, they do so by different mechanisms. Steroidal agents, such as exemestane, have an androgen structure and compete with the natural aromatase substrate androstenedione; they bind irreversibly to the catalytic site of aromatase causing loss of enzyme activity, and more aromatase enzyme must be produced before estrogen biosynthesis can resume. Therefore, steroidal agents are often referred to as suicide inhibitors. (131)

Letrozole is an oral, active highly selective, non-steroidal inhibitor of the aromatase enzyme system. It blocks the aromatase enzyme, consequently lowering estrogen levels and thereby depriving the tumor of its growth stimulus. Letrozole effectively inhibits the conversion of androgens to estrogens in both in vitro and in vivo. (132)

Large randomized controlled multinational trials were conducted in postmenopausal women with advanced breast cancer who had progressed despite anti-estrogen therapy (e.g. AR/BC2, AR/BC3). These studies demonstrated that Letrozole was statistically superior to Megestrolacetate in tumor response (study AR/BC2), superior to Megestrolacetate and AG in TTP (both studies) and superior to Megestrolacetate and Aminoglutethimide in TTF (studies AR/BC2 and AR/BC3).

In the BIG 1-98 investigated Letrozole in the early adjuvant setting in comparison to Tamoxifen. Postmenopausal patients with hormone receptor positive primary breast

cancer were randomized after complete surgery to receive Letrozole or Tamoxifen for 5 years. Two additional study arms explore the sequences Tamoxifen for 2 years followed by Letrozole vs. Letrozole for 2 years followed by Tamoxifen. The first analysis of the study compares the efficacy and safety of the monotherapy arms, e.g. Letrozole alone for 5 years versus Tamoxifen alone for 5 years. The results show, that Letrozole decreased the risk of recurrence significantly compared to Tamoxifen (HR = 0.81, p = 0.003). Also for patients with increased risk of recurrence (patients with nodal involvement and after adjuvant chemotherapy) Letrozole reduced the risk of recurrence by 29% in both patient groups compared to Tamoxifen (HR = 0.79). Specifically, Letrozole reduced the risk of distant metastases, an accepted proxy for survival, by 27% (HR = 0.73, p=0.0012). Cardiovascular events, in particular hypercholesterolemia and myocardial infarction, occurred significantly more often with Letrozole than with Tamoxifen (133).

The MA-17 trial, which randomized patients to Letrozole or placebo after 5 years of Tamoxifen, had been closed before the start of the SUCCESS B trial after a significant benefit in favor of the aromatase inhibitor. (134) At final analysis a significant 4.9% difference in estimated 4-year disease-free survival was seen after a total of 247 events. The benefit in favor of Letrozole was seen regardless of axillary nodal status. The final analysis revealed also a survival benefit in nodal positive patients of 39%. In MA17 Letrozole not only proved to be highly effective in prolonging the DFS even after 5 years pretreatment with Tamoxifen, this efficacy was paralleled by a safety profile comparable to Placebo with no increase of infarction risk under Letrozole and a small increase in self reported osteoporosis. In general Letrozole was safe and well tolerated. The percentage of patients with serious adverse events was almost identical in both arms. Thromboembolic events and endometrial hyperplasia occurred significantly more often with placebo than with Letrozole.

Taken together, the results and facts summarized here led to the approval of Letrozole for the extended adjuvant treatment of hormone dependent early breast cancer in postmenopausal women who have received prior standard adjuvant Tamoxifen therapy for 5 years, for the 1st-line treatment in postmenopausal women with hormone-dependent advanced breast cancer and for the treatment of advanced breast cancer in women with natural or artificially induced postmenopausal status

after relapse or disease progression, who have previously been treated with antiestrogens. Furthermore the International Concensus Conference on the optimal therapy of early breast cancer 2005 in St. Gallen recommended updating their guidelines to consider using an aromatase inhibitor such as Letrozole as part of the adjuvant therapy for postmenopausal women with breast cancer. The panel specifically suggested updating the guideline to reflect that Letrozole is a viable option after 5 years of Tamoxifen therapy (extended adjuvant) in early breast cancer patients. Also the 2003 American Society of Clinical Oncology Technology assessment on adjuvant use of aromatase inhibitors was updated taking into account the adjuvant data on Letrozole. Aromatase inhibitors are appropriate as initial treatment for women with contraindications to Tamoxifen. For all postmenopausal women, treatment options include 5 years of aromatase inhibitors treatment or sequential therapy consisting of Tamoxifen (for either 2 to 3 years or 5 years) followed by aromatase inhibitors for 2 to 3, to 5 years. Patients intolerant of aromatase inhibitors should receive Tamoxifen.

9.4.7 Goserelin

Goserelin is a gonadotropin-releasing hormone (GnRH) agonist. Continuous pituitary stimulation by GnRH, normally under pulsatile control, leads to an eventual downregulation of LH and FSH secretion with subsequent diminution of androgen levels. During the first weeks of therapy an initial LH and FSH release may occur. Goserelin lowers serum estradiol to postmenopausal levels. Goserelin is well tolerated both locally and systemically. It produced effective castration and the objective response rates and duration of remission are at least comparable to those seen following oophorectomy; however, the side effects are less. The use of depot goserelin avoids the psychological trauma and operative morbidity of the irreversible operative castration.(135) Amenorrhea and hot flashes (75 %) are common adverse effects of goserelin therapy. Peripheral edema with worsening congestive heart failure have been described.(136, 137)

9.5 Treatment administered

After surgery, leading to R0 resection of the invasive and intraductal components of the primary tumor, all patients were randomized to one of the following treatments:

Randomization

A: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3 cycles of Docetaxel 75 mg/m² body surface area i.v. (D), and Gemcitabine 1000 mg/m² i.v. (30 min infusion) (G), administered on day 1, followed by Gemcitabine 1000 mg/m² i.v. (30 min infusion) on day 8, repeated on day 22

B: 3 cycles of 5-Fluorouracil 500 mg/m² i.v. body surface area and Epirubicin 100 mg/m² i.v. and Cyclophosphamide 500 mg/m² i.v., (FEC100), each administered on day 1, repeated on day 22, subsequently followed by 3 cycles of Docetaxel 100 mg/m² body surface area i.v. (D), administered on day 1, repeated on day 22

Each patient's treatment modality was unknown until the time of randomization.

The randomization was performed by fax or electronically via Internet by the appointed CRO.

9.6 Investigational products

9.6.1 Gemcitabine

Gemcitabine [2'-deoxy-2',2'-difluorocytidine monohydrochloride (beta isomer); dFdC] is a novel deoxycytidine analogue which was originally investigated for its antiviral effects but has since been developed as an anticancer therapy. It is a pro-drug and, once transported into the cell, must be phosphorylated by deoxycytidine kinase to an active form. Gemcitabine is phosphorylated intracellularly to difluorodeoxycytidine triphosphate, which terminates DNA-chain elongation and competitively inhibits DNA polymerase and ribonucleotide reductase. After i.v. administration, gemcitabine is

rapidly distributed into total body water. The drug is deaminated in the plasma to inactive difluorodeoxyuridine; both gemcitabine and difluorodeoxyuridine are primarily renally eliminated. (138) Both gemcitabine diphosphate (dFdCTP) and gemcitabine triphosphate (dFdCTP) inhibit processes required for DNA synthesis. Incorporation of dFdCTP into DNA is most likely the major mechanism by which gemcitabine causes cell death. After incorporation of gemcitabine nucleotide on the end of the elongating DNA strand, one more deoxynucleotide is added and thereafter, the DNA polymerases are unable to proceed. This action ("masked termination") apparently locks the drug into DNA as the proofreading enzymes are unable to remove gemcitabine from this position. Furthermore, the unique actions that gemcitabine metabolites exert on cellular regulatory processes serve to enhance the overall inhibitory activities on cell growth. This interaction is termed "self-potentiation" and is evidenced in very few other anticancer drugs.(139)

Gemcitabine monotherapy produced an objective tumor response in 18 to 26% of patients with advanced non-small cell lung cancer (NSCLC) and appears to have similar efficacy to cisplatin plus etoposide. Objective response rates ranging from 26 to 54% were recorded when gemcitabine was combined with cisplatin, and 1-year survival duration after such treatment ranged from 35 to 61%. Improvements in a range of NSCLC disease symptoms and/or in general performance status occurred in many patients who received gemcitabine, with or without cisplatin, in 3 clinical trials. Gemcitabine appears to be cost effective compared with best supportive care for NSCLC. In addition, direct costs associated with administration of gemcitabine monotherapy may be lower than those for some other NSCLC chemotherapy options, according to retrospective cost-minimisation analyses. The combination of gemcitabine plus cisplatin was associated with a lower cost per tumor response than cisplatin plus etoposide or cisplatin plus vinorelbine, according to a retrospective cost-effectiveness analysis. In a single comparative study in patients with advanced pancreatic cancer, gemcitabine was more effective than fluorouracil with respect to survival duration and general clinical status. It also showed modest antitumor and palliative efficacy in patients refractory to fluorouracil. Gemcitabine appears to be well tolerated, although further comparisons with other chemotherapy regimens are required.(140)

In breast cancer, as a single agent, gemcitabine yields response rates ranging from 14%-37% as first-line therapy for advanced breast cancer and 23%-42% as salvage therapy. However, these were small studies with large confidence intervals around all the indices of benefit including response rate, response duration, and time to disease progression. Gemcitabine is associated with higher response rates when used in combination with other agents.(27) The combination of gemcitabine and anthracyclines-containing double- and triple-drug combinations used to treat patients with early-stage and advanced breast cancer were promising, with good tolerability and overall response rates ranging from 33%-89% in advanced disease and up to 95% in the neoadjuvant treatment of early-stage disease. (50) Numerous phase II clinical studies have combined gemcitabine with other active agents such as the taxanes, vinorelbine, vindesine, cisplatin, 5-fluorouracil, as well as anthracyclines across various regimens and conditions of pretreatment. Most of these two-drug combinations have consistently demonstrated higher efficacy than either single agent, particularly in pretreated patients. Even higher efficacy has been obtained with triple-drug regimens including gemcitabine, anthracyclines (epirubicin or doxorubicin), and paclitaxel; these regimens have yielded overall response rates of 58-92% as first-line treatment.(49)

In an early review, the toxicity profile of gemcitabine was analyzed in a large group of patients (up to 790) from pivotal phase II studies, in which the drug was given intravenously as a 30 min infusion, in a schedule once a week for 3 weeks followed by a week of rest. The safety profile of gemcitabine is unusually mild for such an active agent in solid tumors. Haematological toxicity is mild and short-lived with modest WHO grades 3 and 4 for haemoglobin (6.4% and 0.9% of patients), leukocytes (8.1% and 0.5%), neutrophils (18.7% and 5.7%) and platelets (6.4% and 0.9%). The incidence of grade 3 and 4 infection associated with this level of myelosuppression was low (0.9% and 0.2%). Transaminase elevations occurred frequently, but they were usually mild, and rarely dose limiting. Mild proteinuria and haematuria were seen but were rarely clinically significant. There was no evidence of cumulative hepatic or renal toxicity. Nausea and vomiting was mild, rarely dose limiting, and generally well controlled with standard antiemetics. Flu-like symptoms were experienced in a small proportion of patients but were of short duration. Where oedema/peripheral oedema was experienced there was no evidence of any

association with cardiac, hepatic or renal failure. Hair loss was rare, with WHO grade 3 alopecia reported in 0.5% of patients. There was no grade 4 alopecia. Furthermore, gemcitabine displayed minimal toxicity in elderly patients, and the side-effect profile does not seem to be affected by patient age. The adverse events typically experienced with cytotoxic agents, namely myelosuppression, nausea and vomiting and alopecia, are not seen to such a degree with gemcitabine, and this nonoverlapping toxicity profile suggests that gemcitabine is a promising agent for incorporation into combination chemotherapy regimens.(141)

9.6.2 Trastuzumab

As Trastuzumab is a monoclonal antibody against the HER2-receptor, it's an important agent in targeted therapy of HER2 positive patients. Trastuzumab was given every 3 weeks with a loading dose of 8 mg/kg body weight and a maintenance dose of 6 mg /kG body weight. The dose had to be recalculated at each cycle according to the patients body weight. The total treatment duration was one year following adjuvant chemotherapy. Since Trastuzumab can cause anaphylactic reactions, patients were observed for at least six hours after the start of the first dose of Trastuzumab and for at least two hours during the following infusions. According to the current guidelines, a cardiac ultrasound examination with measurement of LVEF was performed before the first application of Trastuzumab and regulary during treatment (see schedule for necessary examinations).

9.6.3 Treatment assignment

All patients were randomized to receive either 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy or 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy. Randomization was stratified and performed as described previously.

9.6.4 Selection of dose

Accountability for the investigational products lied with the investigator. The investigator had the responsibility to explain the correct use of the investigational

products and to check at appropriate intervals that each study participant is following instructions properly.

9.6.5 Dispensing, supply and tracking the study treatment

During the SUCCESS-B trial the study medication was guideline conform standard treatment for primary breast cancer. Therefore it could be ordered and preceded as for any other breast cancer patient. The medication was ordered if a patient`s blood values, history and examination provided no contraindications for the application of chemotherapy treatment. Further details are described in the respective parts of the study protocol. The day of treatment the medication was dispensed according to the patients characteristics by the hospital`s pharmacy.

Excluded from this are the agents Gemcitabine and Docetaxel.

Docetaxel medication for node negative patients was provided by Sanofi-Aventis at its own expenses. Gemcitabine medication for all patients in treatment arm A was provided by Lilly at its own expenses.

Hence these agents were ordered automatically from the company within the randomization process. The study treatment was straightly shipped to the allocated pharmacy. Docetaxel was progressed like it was normal treatment. For Gemcitabine an amount sufficient for two patients was provided with the first randomization of a patient into the AB-Arm, therefore the drugs had to be stored at the site`s pharmacy. Hence, the shipment receipt, drug coverage and consumption had to be recorded in the Gemcitabine drug accountability log. In case study medication
expired at the site`s pharmacy the medication was to be destroyed at the site.

9.6.6 Instructions for prescribing and taking study treatment

The chemotherapy and Trastuzumab were applied intravenously. The patients had to come to the study site to receive treatment. The application was performed by the site`s
physician via port or intravenous line. After checking the right location by flushing with NaCl the study drug application was monitored by the skilled staff of the SUCCESS B study sites. The patients with positive hormone receptor status received antihormonal treatment after the end of chemotherapy, which they took orally.

9.6.7 Hematological Toxicity

Hematological toxicities were graded according to the Common Toxicity Criteria of the National Cancer Institute (CTC, Version 3.0).

The WBC (white blood count) had to be ≥ 3.0 x 109/l and the platelet count ≥ 100 x 109/l prior to the beginning of the treatment. In case of required delay, the patient should have had blood count checked at least twice per week and retreated as soon as sufficient recovery had been achieved.

Primary prophylactic application of G-CSF was no more recommended for the regimens in this trial today. (142) However, in order to achieve sufficient dose intensity and to prevent infections, G-CSF (Granocyte®) should be applied as secondary prophylaxis in the following cases:

- Febrile neutropenia (i.e. temperature >38.5 °C, ANC < 0.5 x 109/l, requiring hospitalization and intravenous antibiotics)
- neutropenia (ANC < 0.5 x 109/l) for more than 5 days
- severe neutropenia (ANC < 0.1 x 109/l)
- prolongation of the time interval due to insufficient leucocytes or neutrophils

G-CSF as secondary prophylaxis should be given in all the following courses on day 5 – 10, with no application on day 8 in patients with gemcitabine medication, or until leucocytes have reached 5.0 x 109/l after crossing the nadir.

In case of the following hematological toxicities despite secondary prophylactic application of G-CSF, doses was reduced successively by one dose level in case of

- ANC < 0.5 x 109/l for more than 5 days
- Febrile neutropenia (i.e. temperature >38.5 °C, ANC < 0.5 x 109/l, requiring hospitalization and intravenous antibiotics)
- Thrombocytopenia grade 4
- Prolongation of the time interval due to insufficient leucocytes or neutrophils and/or platelets

9.6.8 Non-hematological Toxicity

Non-hematologic toxicities was graded according to the Common Toxicity Criteria of the National Cancer Institute (CTC, Version 3.0). Patients with a grade 0-2 toxicity other than a hematologic toxicity received the full dose of therapy on time.

Neurological toxicity:	NCI-grade 2: dose reduction 1 level
	NCI-grade >2: removal from study

Gastrointestinal toxicity:

Mucositis:	NCI-grade 3: dose reduction 1 level and treatment according to recommendations in section 8.1.6
Mucositis or vomiting:	NCI-grade 4: removal from study and treatment according to recommendations in section 8.1.6

Hepatic toxicity (under treatment with Docetaxel):

Elevated liver enzymes /ALT, AST) > 1,5 x UNL or alkaline phosphatase > 2,5 x UNL: dose reduction 1 level

Bilirubin > 1 x UNL or elevated liver enzymes > 3,5 x UNL or alk. Phosphatase > 6 x UNL : removal from study

Cardiac toxicity:

AV-bockage 1st grade, asymptomatic bradycardia, isolated asymptomatic ventricular extrasystoles: treatment continuation under cardiac monitoring

Arrythmias requiring treatment, AV-blockage 2nd or 3rd grade, reduction of LVEF of >20%/or >10% and below the clinic's normal range: removal from study

Pulmonary Toxicity (in special respect to Gemcitabine):

If pneumonitis grade 2 or higher develops in a given cycle and is related to gemcitabine, gemcitabine should be promptly discontinued and the patient should be removed from protocol treatment. Treatment with corticosteroids should be given according to established guidelines.

For patients who developed other grade 3 non-hematologic toxicity, the decision to have their treatment reduced to 75 % or withheld depended on the course that is medically most sound in the judgment of the investigator.

Patients who developed a grade 4 non-hematologic toxicity or treatment resistant uncontrolled diabetes mellitus that is judged to be life-threatening had to be removed from the study.

For subsequent cycles, doses had to be reduced successively by one dose level in case of the above mentioned toxicities.

A patient had to be discontinued from the study if the beginning of a given cycle had to be postponed due to toxicity for more than 2 weeks, unless approved by the study coordinator group. In this case, the dose had to be reduced by one dose level.

9.6.9 Permitted dose adjustments

The following dosage adjustment levels were considered:

Table 7 Dose adjustments

Dose Level	0	-1	-2
Fluorouracil	500 mg/m²	400 mg/m²	300 mg/m²
Epirubicin	100 mg/m²	80 mg/m²	60 mg/m²
Cyclophosphamide	500 mg/m²	400 mg/m²	300 mg/m²
Docetaxel (Arm A)	75 mg/m²	60 mg/m²	45 mg/m²
Docetaxel (Arm B)	100 mg/m²	80 mg/m²	60 mg/m²
Gemcitabine (each day)	1000 mg/m²	800 mg/m²	600 mg/m²

Further dose reductions would result in an ineffective therapy. Therefore, patients requiring further dose reductions had to be discontinued from the study.

9.6.10 Interval Modification

The cytostatic treatment of a patient could be postponed for up to 2 weeks if she had not recovered from hematological and/or non-hematological toxicity at the beginning of a cycle (day 1).

The following items must be fulfilled:

- neutrophiles ≥ 1.5 x 109/l or leucocytes ≥ 3.0 x 109/l
- platelets ≥ 100 x 109/l

The treatment was restarted immediately after recovery.

A patient was discontinued from the study if the beginning of a given cycle has to be postponed due to toxicity for more than 2 weeks, unless approved by the study coordinators.

In this case, the dose was reduced by one dose level.

In case these requirements were not achieved, frequent laboratory controls were recommended as well as interval prolongation for up to 1 week (no application of G-CSF immediately before chemotherapy!).

If the application of gemcitabine on day 8 had to be postponed for less than one week, the next cycle of chemotherapy was given as scheduled. If day 8 was postponed for more than one week, the next cycle was postponed accordingly.

9.6.11 Compliance

Epirubicin, docetaxel, fluoruracil, Gemcitabine, cyclophphoshamide and trastuzumab and therefore all main investigational drugs were administered intravenously only at the investigational sites. Therefore patient compliance was ensured. Patients who returned for follow-up visits received study drugs unless they were encountering toxicity problems or their diseases had progressed.

9.7 Efficacy and safety variables

A log-rank test was used to perform confirmatory testing on the primary objective, the difference in the disease free survival after randomisation in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy in patients with early primary breast cancer. The global two-sided significance level is set to α=0. 04507 for the final analysis. (143)

On a 5%-significance level, additional exploratory testing was performed on the difference between the two treatment groups according to the secondary objectives of this study:

- Overall survival time after randomization
- Toxicity
- Changes in quality of life over time as defined by EORTC QLQ-C30 and BR23 questionnaires
- The predictive and prognostic value of markers in peripheral blood, as specified in the translational research program
- Effect on the presence of disseminated tumor cells in bone marrow in peripheral blood
- Subgroup analyses for the above mentioned criteria in terms of tumor size, axillary lymph node status, histopathological grading, HER2-status, menopausal status and interaction between the randomizations
- Additional analyses as regarded necessary and informative

9.7.1 Efficacy and safety measurements assessed and flow chart

A panel of independent experts evaluated the response of each enrolled patient by applying standard oncological criteria. (144) The measurability of a tumor is defined as follows:

Objective status (to be recorded at each evaluation)

To confirm any of the following disease assessments, repetition of the respective examination and determination of the status is required after 4 weeks, i.e. all responses must be documented to last at least 4 weeks to be considered as valid.

- ***Free of Recurrence:*** There are no clinical or radiological signs of tumor growth either in the region of the primary tumor or at any distant sight.
- Local Recurrence: Local recurrence is defined as any relapse in the area of surgery between the sternum and the anterior axillary line, below the inferior clavicular fossa, and above the 7th rib. Tumor recurrence at one of the pectoral muscles or at the fascias of the serratus lateralis muscle or the oblique externus muscle is also considered as local recurrence.
- Regional Recurrence: Relapse infiltrating the skin and/or involving the axillary lymph nodes, or the metastatic infiltration of the nodules in the infraclavicular fossa, is considered a regional recurrence. (145)
- Distant Metastases: Patients with relapsing tumor outside the above mentioned sights are considered to have distant disease.
- Cancer Associated Death: Only death which can be clearly associated with conditions attributed to the malignant disease, such as distant disease, will be considered as cancer associated death.

The analyses were performed as an intention to treat analyses, not excluding those patients who discontinue from the study for any given reason.

Examinations during the study

No more than 2 weeks before enrolling into the study, the disease status of each patient was assessed with the following procedures:

- Medical history and physical examination, including measurements of height and weight.
- Evaluation of performance status (ECOG scale).

- Quality of Life (Qol) questionnaires EORTC QLQ-C30 and QLQ-BR23 completed by the patient, not to be repeated before the first cycle of therapy.
- Electrocardiography (ECG)
- LVEF
- Laboratory tests:
  - Hemoglobin, WBC, platelets and neutrophils (differencial blood count)
  - Bilirubin, liver enzymes (GOT,GPT), gamma-GT, AP, creatinine, potassium, sodium, PTT, INR, albumine and protein.
  - Pregnancy test in premenopausal women
  - Blood-Sample for translational research program

No more than 5 weeks before enrolling into the study, each patient was assessed by the following radiological tests:

- Chest x-ray (two dimensional)
- Whole body bone scan
- Ultrasound of the liver

At the stated intervals during the study, efficacy was examined in each patient by the following evaluations:

- 1- 2 x per week (whole duration of chemotherapy):
  - white blood count (differential blood count)

  Before every therapy cycle:
  - Limited medical history and physical examination, including toxicity assessment.
  - Weight measurements.
  - Clinical laboratory tests mentioned in points 1-2 above (exclusive PTT, INR, albumine and protein, if not clinically relevant)
  - Type and number of units required for transfusions at every cycle
  - Toxicity rating using the NCI CTX scale
  - Performance status evaluation (ECOG scale)..
  - QoL EORTC QLQ-C30 and BR23 by the patient before first and fourth *chemotherapy cycle is administered*
  - 4 weeks after the last application of chemotherapy:
    - Clinical laboratory tests mentioned in points 1-2 above and pregnancy test in premenopausal women

  - QoL EORTC QLQ-C30 and BR23 by the patient
  - Limited medical history and physical examination, including toxicity assessment.
  - Evaluation of performance status (ECOG scale).
  - ECG
  - LVEF
  - Blood Sampling for the translational research program
  - Toxicity (NCI)
- 6 weeks after the end of radiotherapy:
  - QoL EORTC QLQ-C30 and BR23 by the patient
  - Limited medical history and physical examination, including toxicity assessment.
  - Evaluation of performance status (ECOG scale).
  - Tox (NCI)
- Tri-monthly Follow-Ups:
  - QoL EORTC QLQ-C30 and BR23 by the patient
  - Limited medical history and physical examination, including toxicity assessment.
  - Evaluation of performance status (ECOG scale).
  - Tox (NCI)
  - Blood Sampling for the translational research program (for details see schedule in the appendix of this protocol)

Table 8 Schedules of visits

Time **Examination**	**Before treatment**	**Before every cycle**	**28 days after last chemotherapy**	**6 weeks after last radiotherapy**	**Follow-up (every 3 months for 2 ys, every 6 mths. for 3 ys.)**
Demographic Data	***X***				
Criteria for inclusion/ exclusion	***X***				
Signed Content Form	***X***				
Registration/Randomisa-tion	***X***				
Medical history	***X***	***X***	***X***	***X***	***X***
Clinical exam	***X***	***X***	***X***	***X***	***X***
Weight, Hight	***X***	***X***	***X***	***X***	***X***

Concomittent medication	***X***	***X***	***X***	***X***	***X***
Concomittent diseases	***X***	***X***	***X***	***X***	***X***
Status of activity (ECOG)	***X***	***X***	***X***	***X***	***X***
EORTC QLQ-C30 + BR23 Quality of Life-questionn.	***X***	Before cycle 4	***X***	***X***	***X***
X-ray of the lung, bone scan, sonography of liver	***X***				If needed
Peripheral Blood Sampling (MRD Surveillance)	***X***		***X***		in case of disease recurrence
Translational research within the Altto-Trial (in case of participation)			***X***		Week 13 and month 18 after beginning of altto-trial and in case of disease recurrence
White blood count and Differential blood count	***X***	As indicated, appr. 1 – 2/week	***X***		If needed
Creatine	***X***	***X***	***X***		If needed
Sodium, Potassium	***X***	***X***	***X***		If needed
Bilirubin, GOT, GPT, γGT, AP	***X***	***X***	***X***		If needed
Albumine, Protein	***X***		***X***		
Pregnancy test (premenopausal)	***X***		***X***		
INR, PTT	***X***		***X***		
EKG, LVEF	***X***		***X***		2 ys after end of chemotherapy
Toxicity (NCI)		***X***	***X***	***X***	***X***
Status of survival/ recurrence			***X***	***X***	***X***
Mammography	***X***				Each 6/12 month Accord. to guidelines

9.7.2 Primary efficacy variable(s)

Progression free survival (PFS) is defined as the time from the date of randomization into the study to the date of diagnosis of loco-regional recurrence or distant metastases. Loco-regional recurrence is defined as any relapse in the area of primary surgery and/or ipsilateral regional axillary lymph nodes including the nodules of the infra- or supraclavicular fossa. Any other tumor manifestation is defined as distant disease.

Additionally, analyses for overall survival (OS), the time from the date of randomization to the date of death associated with cancer related causes as well as from any other cause were performed, as well as the evaluation of all other secondary study objective.

9.8 Data Quality Assurance

There were several measuring methods by which quality of documented data was ensured.

Throughout the trial, according to GCP, study monitor visits took place regular directly at the sites. The monitors contacted study staff personally to resolve problems immediately and regularly checked site`s source data. They additionally acted as contact persons for questions concerning documentation.

At a second step, at the end of the clinical trial documented data were reviewed by data management.

Furthermore, there were several boards consisting of different persons not directly involved in the clinical trial which were responsible for overall data management and conduct of the clinical trial.

9.8.1 Monitoring

Initiation visit – Before the start of SUCCESS-B, an initiation visit was performed in each participating center. For initiation at least one physician and study nurse had to take part. Detailed information concerning the conduct of the trial, inclusion and exclusion criteria, objectives, eCRF and handling of SAEs was given. The Investigator site file (ISF) with the necessary documents and information was handed over.

Monitoring visits

Quality of study data were assured by monitoring of investigational sites, provision of appropriate training for the site`s team, and use of data management procedures. Completeness and plausibility were monitored by visits on-site as well as review of the eCRF.

Monitoring was planned according to the risk-adapted Adamon-concept. The hazard risk of the clinical trial was classified as comparable to standard treatment. Monitoring was planned according to the monitoring risk class K3, lowest risk class. Each active

center had at least one monitoring visit carried out by CRO monitors during the course of the clinical trial. The order of the visited centers was randomly determined. The number of the patients who had to be monitored to 100% was randomly selected by the project management of the CRO.

Source data verification was performed according to the following specification:

Inclusion-/exclusion criteria:	100%
Progression free survival (primary study objective):	100%
Overall survival:	100%
Adverse events / serious adverse events:	30%
Quality of life:	30%
Others:	20%

In addition, the following issues were addressed at each monitoring visit:

- Protocol deviations
- Up-to-dateness of the documentation / clarification of questions concerning the documentation
- Examination of the investigator site file and updating if necessary

Depended on the number of enrolled patients the number of monitoring visits per center ranged 1 to 5.

At each contact the clinical monitor addressed, if AEs and SAEs have occurred, if Informed consent of patients was checked and if Source data verification was performed according to monitoring plan and deviations from study protocol were listed. ISF was controlled and up-dated if necessary.

Not all patients were monitored 100% on-site, but all eCRFs were checked for completeness.

Close out-visits

After “last patient last visit” each center had a close out visit. At this visit ISF was updated.

9.8.2 Electronic data checks (eChecks)

Electronic edit checks (eChecks) were performed automatically and immediately when entering data in the eCRF. The user was made aware of missing data entries or any implausibility. This was done by warning and error messages. Certain

triggered error messages did not allow the user to saving the eCRF form until the error was corrected.

9.8.3 Manuel data checks (mChecks)

Manuel corrections were performed by the Data management, whenever electronic checks couldn`t be done.

To verify the correctness, validity and completeness of the data, as well as to find protocol violations, plausibility- or cross-checks were performed. Additionally free text entries had to be checked on plausibility or any "hidden" information (E.g. AEs or SAEs) or "prohibited" information (such as patient's full name or documented data after a withdrawal of consent).

9.8.4 Checks by Pharmacovigilance

Pharmacovigilance was monitored by the SUCCESS trial center in Munich. Adverse events (AEs) were documented by the sites directly into the eCRFs.

AEs that met the following criteria had to be classified as a serious adverse event (SAE):

- Death of the patient
- serious deterioration of patient`s health resulting in life-threatening illness
- permanent impairment
- in-patient hospitalization or prolongation of hospitalization
- medical or surgical intervention to prevent life-threatening illness or injury
- fetal distress
- fetal death
- fetal congenital abnormality
- fetal birth defect

The study sites had to report SAEs within 24 hours by filling in an online SAE-template provided by the patient`s eCRF. They needed to assess the degree of likelihood that the SAE was related to the administered study medication.

The processed online document automatically was faxed to the SUCCESS trial center in Munich and the involved pharmaceutical company.

After receiving notice of the SAE the sponsor also had to assess immediately whether the SAE was unexpected (side effect not listed in the investigator`s brochure) and very likely (1-3) caused by the study medication administered. SAE that met those criteria were classified as Suspected Unexpected Serious Adverse Reaction (SUSAR) and had to be reported to the BfArm and ethical committee. Furthermore the SUSAR was reported according to the GCP-guidelines within 15 days to all study sites, except the AE/SAE caused a life threating condition. In this case it had to be reported within 7 days.
In order to clarify all circumstances the study site was requested to provide all information available. The receipt of the BfArm and all documents on the SAE/SUSAR were documented according to the SAE/SUSAR-SOP and Checklist attached in the appendix. After having collected all information on the SAE/SUSAR, the documents were deposed anonymized in the SUCCESS trial center in Munich and can also be found in the attachment.

Table 9 Degree of relation between SAE and expected side effects.

1	Very likely
2	Likely
3	Possible
4	Unlikely
5	No relation

9.8.5 Study Coordinator Board

The study coordinator board had to ensure that the correct, complete and reliable data were taken. Therefore it had to:

- Provide instructional material to each study site (following GCP)
- Perform and sponsor study meetings before the start of the study and at least
- once a year following. These meetings included instruction in all sections of the protocol, the completion of the clinical report forms and general study procedures
- Be available at any time for consultation and stay in personnel contact with the study site by mail, email, telephone or fax
- Review and evaluate clinical report data and use standard computer edits to
- detect errors in data collection

9.8.6 Independent Data and Safety Monitoring Board (DSMB)

An Independent Data and Safety Monitoring Committee was attached to the randomized clinical trial and charged with the responsibility of monitoring performance of the trial, safety of the participants, and efficacy of the treatments being tested. (146, 147)The necessity of a DSMB stems from the ethical imperative to dissociate the treating physician from the accruing data in order to maintain a legitimate “state of equipoise” regarding the therapies being studied and to remove those with vested interest in specific treatment from deciding whether the trial should continue.

The duties of the Independent Data and Safety Monitoring Committee comprised monitoring the following:

- Recruitment, retention
- Protocol violations
- Identify need for additional data to clarify endpoints
- Identify problems with the study assumptions used for planning and design (e.g., sample size reviews)
- If interim data indicate an intervention is harmful, the trial may be stopped.
- If interim data demonstrate a clear benefit from an intervention, the trial may be stopped.

The Independent Data and Safety Monitoring Committee compromised a Chair and multidisciplinary member, including clinician(s), statistician(s), an ethicist, a clinical trialist and an executive secretary.

9.8.7 Statistical Methods Planned in the Protocol

Efficacy interim analyses were planned to be performed after 50% and 75% of the expected events of recurrence for the primary objective, the difference in the disease free survival after randomisation in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine-Docetaxel(DG)-chemotherapy have emerged.

The level of significance was planned to be adjusted according to a group sequential design with two efficacy interim analyses after occurrence of 50% and 75%, with the alpha spending function as proposed by Lan – DeMets (148) and with alpha-levels for the 1st, 2nd and final analysis as proposed by O'Brien and Fleming (149). The respected adjusted alpha-levels for the 1st, 2nd and final analysis were be set to alpha1=0.00052, alpha2=0.01411 and alpha3=0.04507.

9.8.8 Determination of sample size

The sample size for the previous Success A-Study was calculated to be 3.658 for patients with HER2 negative and HER2-positive disease. Given the rapid changes in targeted therapy based on the HER2-neu antigen (e.g. trastuzumab, lapatinib), optimizing cytostatic treatment in the HER2-neu positive patient group becomes increasingly challenging and relevant.

The following assumptions were made:

- The DFS at 5 years of patients receiving FU/Epirubicin/Cyclophosphomide followed by Docetaxel is 78,3%.
- There will be an absolute of 4% improvement in 5-year DFS (i.e. an increase from 78,3% to 82,3%) for patients receiving FU/Epirubicin/Cyclophosphomide followed by Docetaxel/Gemcitabine.
- The error rate for a false positive outcome (α) is set to 5%, using two-sided significance tests.
- The error rate for a false negative outcome (ß) is set to 20%, i.e. the power of the trial is set to 80%.
- 945 patients withHER2-neu positive breast cancer have been recruited into the Success A-Trial
- The common exponential drop out rate over whole duration of the study is 10%.
- The accrual period during which patients enter the study is 60 months (5 years).

The follow-up period from the end of accrual until the analysis of the data is 36 months (3 years).

To confirm the absolute increase of DFS-rates at 5 years by 4% from 78,3% to 82,3% for patients from therapy arms by a two-sided log rank tests, a total of N=297 events were needed. The total number of patients to be included into the final analysis of HER2-neu positive patients are equal to N=1723 (i.e. 783 patients per arm, assuming both 1:1 randomization and common exponential drop-out rate over whole duration of the study of 10%).
An overview on the relation of total sample size calculated to different values of clinically significant difference (4% - 6%) and to the length of the follow-up period (3, 5, 8 years) is given in the following table.

Based on the number of 945 HER2-neu positive patients already recruited into the original SUCCESS A Trial, an additional number of 778 patients had to be recruited into the SUCCESS B-Trial on the premises given above.

α =0,05% and β=20% (Power = 80%), expected drop-out rate=10%, accrual = 5 years

Two-sided log-rank test (preferred by regulatory authorities)

Sample Size	+ FU = 3 years	+ FU = 5 years	+ FU = 8 years
Δ DFS = 4% (78,3% → 82,3%)	# events = 743 # pat = 2 x 1829	# events = 743 # pat = 2 x 1397	# events = 743 # pat = 2 x 1060
Δ DFS = 5% (78,3% → 83,3%)	# events = 444 # pat = 2 x 1131	# events = 444 # pat = 2 x 863	# events = 444 # pat= 2 x 654
Δ DFS = 6% (78,3% → 84,3%)	# events = 297 # pat = 2 x 783	# events = 297 # pat = 2 x 596	# events = 297 # pat = 2 x 451

All sample size calculations were obtained using the software nQuery Advisor ® 5.0.

9.8.9 Changes in the Conduct of the Study or Planned Analyses

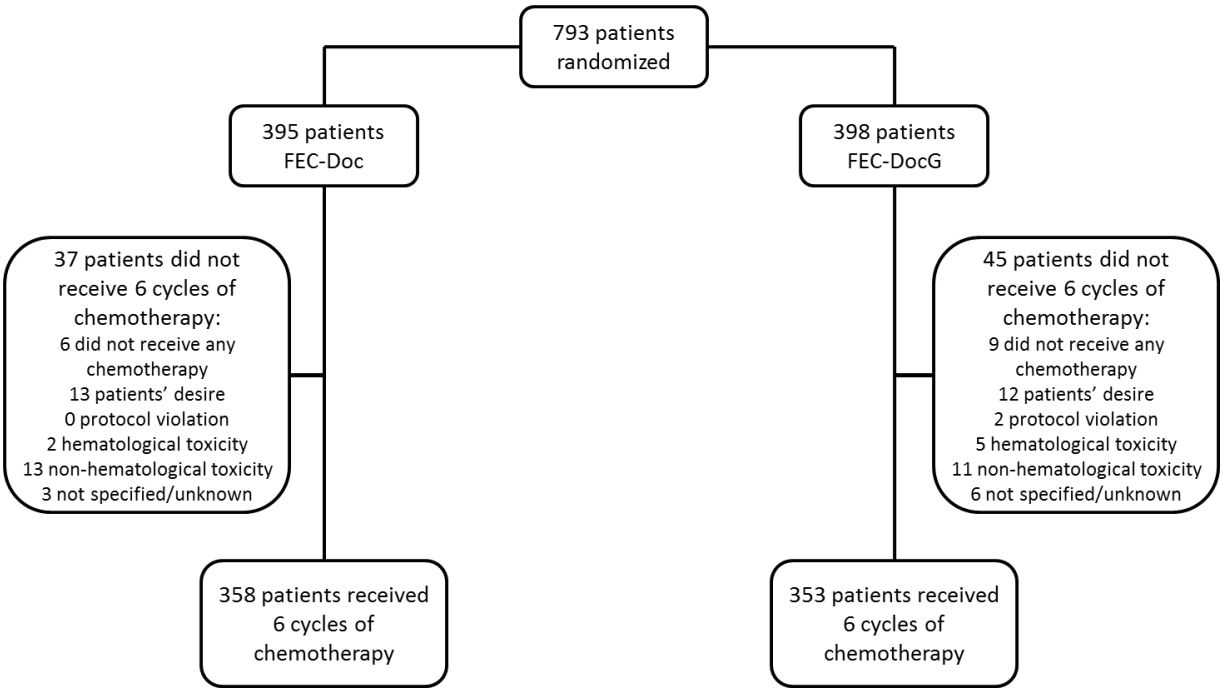
There was an amendment necessary for the SUCCESS-B trial – extension of the inclusion criterias from pT2 to pT1c, Extension of targeted therapy for all patients, extension of the recruitment period to 5 years, docetaxel no more study product due to approval for nodal negative patients, Monitoring of cardiac safety throughout the study, updating the recommendation for adjuvant radiotherapy, additional blood samples during the conduct of the study. All points were submitted to the lead ethical board and approved in 2010.

10 Study patients

10.1 Disposition of patients

Overall, 793 patients were randomized in the SUCCESS B trial (see CONSORT diagram in Fig. 17). 395 patients were assigned to FEC-Doc chemotherapy and 398 patients were assigned to FEC-DocG chemotherapy.

Figure 18 Consort diagram showing patient disposition in the SUCCESS B trial



10.2 Protocol deviations

10.2.1 Protocol deviations inclusion and exclusion criteria

Violations of the inclusion and exclusion criteria were observed in 49 (6.2%) patients (in three patients, more than one inclusion or exclusion criterion was violated), with the violation of the inclusion criterion 8 (Bilirubin, ASAT (SGOT), ALAT (SGPT) and/or Alkaline phosphatase within 1.5 fold of the reference laboratory's normal range for patients) in 41 patients (5.2%). All other violations of in- and exclusion criteria concerned less than 1.0% of patients. Further details are shown in Table 10 below.

Table 10 Violation of inclusion and exclusion criteria

Number of Cycles	Number of patients n (%)
IC1	1 (0.1)
IC2	7 (0.9)
IC7	2 (0.3)
IC8	41 (5.2)
EC3	2 (0.3)
EC4	2 (0.3)
Total	793 (100.0)

10.2.2 Protocol deviations during chemotherapy

Dose modifications were documented in 5.3% (n = 238) of all chemotherapy cycles (Table 11). 77.7% of these were dose reductions necessary due to hematologic or non-hematologic toxicity according to the requirements of the study protocol. In 8 cycles (3.4%) dose reductions were performed because of patients request and in 45 (18.9%) cycles dose modifications were performed because of other reasons (Table 12).

Table 11 Frequency of chemotherapy dose modifications for single cycles according to treatment arm

Cycle	Both randomization arms		FEC-Doc		FEC-DocG	
	Number of chemotherapy cycles applied	Dose modifications % (n)	Number of chemotherapy cycles applied	Dose modifications % (n)	Number of chemotherapy cycles applied	Dose modifications % (n)
Cycle 1	778	0.0 (0)	389	0.0 (0)	389	0.0 (0)
Cycle 2	771	1.8 (14)	385	2.1 (8)	386	1.6 (6)
Cycle 3	764	2.2 (17)	384	1.8 (7)	380	2.6 (10)
Cycle 4	756	5.3 (40)	381	2.1 (8)	375	8.5 (32)
Cycle 5	732	12.6 (92)	368	7.9 (29)	364	17.3 (63)
Cycle 6	711	10.5 (75)	358	6.1 (22)	353	15.0 (53)
Total	4512	5.3 (238)	2265	3.3 (74)	2247	7.3 (164)

Table 12 Reasons for chemotherapy dose modifications according to treatment arm

Reason	Both randomization arms % (n)	FEC-Doc % (n)	FEC-DocG % (n)
Hematological toxicity	39.9 (95)	36.5 (27)	41.5 (68)
Non-hematological toxicity	37.8 (90)	41.9 (31)	36.0 (59)
Patient request	3.4 (8)	1.4 (1)	4.3 (7)
Other	18.9 (45)	20.3 (15)	18.3 (30)
Total	100.0 (238)	100.0 (74)	100.0 (164)

According to the study protocol, the application of the following chemotherapy cycle should not be delayed longer than two weeks from the preplanned infusion date. In fact, chemotherapy applications were delayed for more than two weeks after the preplanned date in only 0.4% of chemotherapy cycles (Table 13). The majority of these 19 delays of more than two weeks was attributed to hematologic or non-hematologic toxicity (see Table 14).

Table 13 Frequency of chemotherapy treatment delays of more than two weeks for single cycles according to treatment arm

	Both randomization arms		FEC-Doc		FEC-DocG	
Cycle	Number of chemotherapy cycles applied	Treatment delays % (n)	Number of chemotherapy cycles applied	Treatment delays % (n)	Number of chemotherapy cycles applied	Treatment delays % (n)
Cycle 1	778	0.0 (0)	389	0.0 (0)	389	0.0 (0)
Cycle 2	771	0.4 (3)	385	0.0 (0)	386	0.8 (3)
Cycle 3	764	0.1 (1)	384	0.0 (0)	380	0.3 (1)
Cycle 4	756	0.3 (2)	381	0.3 (1)	375	0.3 (1)
Cycle 5	732	0.7 (5)	368	0.3 (1)	364	1.1 (4)
Cycle 6	711	1.1 (8)	358	0.0 (0)	353	2.3 (8)
Total	4512	0.4 (19)	2265	0.1 (2)	2247	0.8 (17)

Table 14 Reasons for chemotherapy treatment delays of more than two weeks according to treatment arm

Reason	Both randomization arms % (n)	FEC-Doc % (n)	FEC-DocG % (n)
Hematological toxicity	26.3 (5)	50.0 (1)	23.5 (4)
Non-hematological toxicity	42.1 (8)	50.0 (1)	41.2 (7)
Patient request	5.3 (1)	0.0 (0)	5.9 (1)
Technical reasons	10.5 (2)	0.0 (0)	11.8 (2)
Other	15.8 (3)	0.0 (0)	17.6 (3)
Total	100.0 (19)	100.0 (2)	100.0 (17)

11 Efficacy evaluation

11.1 Data sets analyzed

The primary efficacy analysis was performed on the ITT Population: This population includes all patients who gave informed consent to take part in this study AND who were randomized into one of the chemotherapy treatment arm (A or B). A total of 793 patients were included into this analysis.

11.2 Analysis of Chemotherapy randomization

11.2.1 Primary Study Aim

The primary study aim was the comparison of disease-free survival (DFS) in patients treated with FEC-chemotherapy followed by Docetaxal chemotherapy (FEC-Doc arm) and in patients treated with FEC-chemotherapy followed by Docetaxal and gemcitabine chemotherapy (FEC-DocG arm).

11.2.2 Secondary Efficacy Study Aim

Secondary study aim with regard to efficacy was the comparison of overall survival (OS) between treatment arms FEC-Doc and FEC-DocG.

11.2.3 Statistical Method / Statistical Analysis

Disease-free survival (DFS) was defined as the time from the date of randomisation to the earliest date of disease progression (distant metastasis, local recurrence, death from any cause) or the date of censoring. Patients who were lost to follow-up before the maximal observation time of 8 years (accrual period, 5 years; follow-up period, 3 years) or were disease-free after the maximal observation time were censored at the last date they were known to be disease-free or at the maximal observation time. Overall survival was defined in a similar fashion.

The primary objective was to compare disease-free survival between the two treatment arms; secondary objective was to compare overall survival. Survival rates were estimated by the Kaplan-Meier product limit method. Simple Cox proportional hazards regression models with treatment arm as predictor were fitted to estimate hazard ratios (HRs) with 95% confidence intervals and corresponding p-values. Adjusted HRs were estimated using multivariable Cox regression models with age at randomisation (continuous), estrogen receptor status (categorical; negative, positive) and lymph node stage (categorical; pN0, pN+) as additional predictors.

All of the tests were two-sided, and a P value of < 0.05 was regarded as statistically significant. Calculations were carried out using the R system for statistical computing (version 3.4.0; R Development Core Team, Vienna, Austria, 2017)

11.2.4 Demographic and other baseline characteristics

Patient chacarcteristics are described in Table 15. The distribution of patient characteristics was within expected ranges according to the inclusion and exclusion criteria of the study. The patient and tumor characteristics were well balanced between the two treatment arms.

Table 15 Patient Characteristics according to treatment arm

Characteristics		FEC-Doc Mean or n	FEC-Doc SD or %	FEC-DocG Mean or n	FEC-DocG SD or %
Age		55,1	10,3	54,4	10
BMI		26,1	4,8	26,2	5,3
Tumor Stage	pT1	201	50,9	193	48,5
	pT2	175	44,3	183	46
	pT3	18	4,6	12	3
	pT4	1	0,3	10	2,5
Grading	1	4	1	6	1,5
	2	162	41	145	36,4
	3	229	58	247	62,1
Nodal status	pN0	210	53,2	217	54,5
	pN+	185	46,8	181	45,5
Tumor type	invasive ductal	142	35,9	152	38,2
	invasive lobular	16	4,1	10	2,5
	others	237	60	236	59,3
ER	negative	146	37	153	38,4
	positive	249	63	245	61,6
PR	negative	176	44,6	175	44
	positive	219	55,4	223	56
Menopauasal status	pre	149	37,7	145	36,4
	post	246	62,3	253	63,6

11.2.5 Efficacy results

A total of 793 patients were included in the analysis. The percentage of missing values in each variable was below 1.0%. Events per treatment arm and observation time are shown in Table 16.

Table 16 2 and 5 year desease free and overall survival rate according to treatment arm with 95% confidence intervals in the brackets

Arm	DFS Events	DFS 2-year-rate	DFS 5-year-rate	OS Events	OS 2-year-rate	OS 5-year-rate
FEC-Doc	27	0.98 (0.96, 0.99)	0.90 (0.86, 0.94)	15	0.99 (0.98, 1.00)	0.95 (0.92, 0.97)
FEC-DocG	26	0.96 (0.94, 0.98)	0.91 (0.88, 0.95)	14	0.99 (0.97, 1.00)	0.96 (0.93, 0.98)

Disease-free survival

The study did not show a significant difference between the treatment arms (HR = 0.96, 95% CI: 0.56 to 1.65, p = 0.89) with 27 events in the FEC-Doc group and 26 events in the FEC-DocG group. Survival rates are shown in Table 16 and Figure 19. The adjusted HR was 0.99 (95% CI: 0.58 to 1.70, p = 0.97).

Overall Survival

The study did not show a significant difference between the treatment arms (HR = 0.89, 95% CI: 0.43 to 1.85, p = 0.75) with 15 events in the FEC-Doc group and 14 events in the FEC-DocG group. Survival rates are shown in Table 16 and Figure 20. The adjusted HR was 0.95 (95% CI: 0.46 to 1.98, p = 0.90).

Figure 19 Kaplan-Meier curves for disease-free survival according to treatment arm.

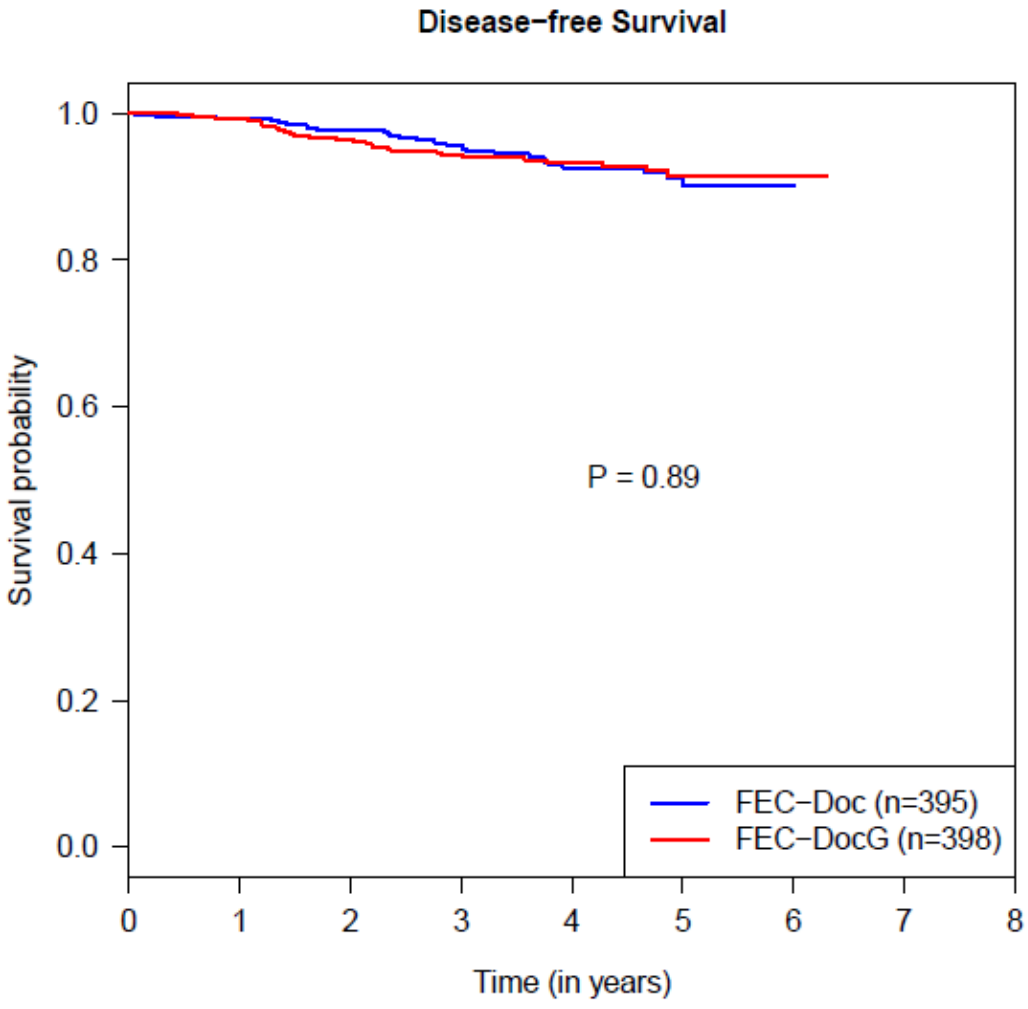
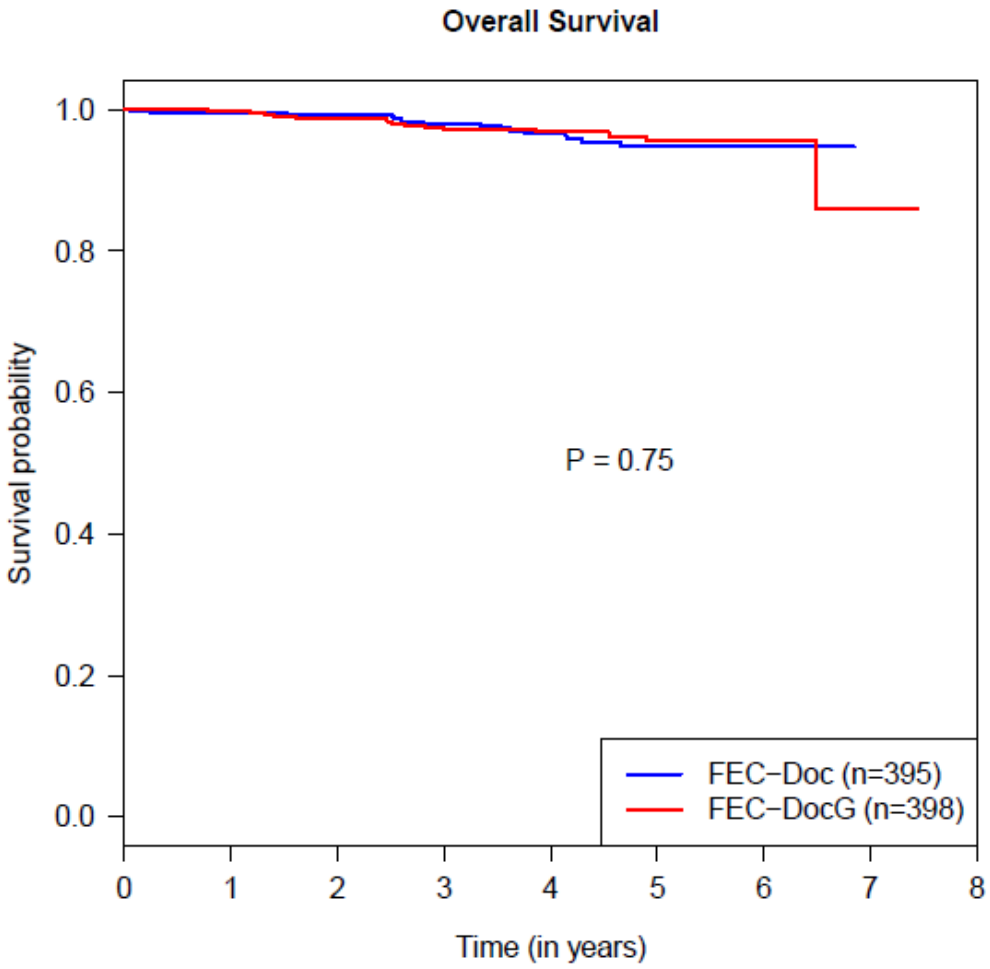


Figure 20 Kaplan-Meier curves for overall survival according to treatment arm.



11.2.9 Efficacy conclusion Chemotherapy

The primary study aim of SUCCESS-B was to compare the disease-free survival (DFS) in patients treated with FEC-chemotherapy followed by docetaxel chemotherapy (FEC-Doc arm) to the DFS in patients treated with FEC-chemotherapy followed by docetaxel and gemcitabine chemotherapy (FEC-DocG arm), followed by targeted therapy for all patients. Therefore all events of 793 were included, whereby 395 were randomized into FEC-Doc and 398 into the FEC-DocG arm. The final survival analysis revealed no significant difference in DFS between the two treatment arms (HR = 0.96, 95% CI: 0.56 to 1.65, p = 0.89); thus, the addition of gemcitabine to FEC-Docetaxel chemotherapy in early breast cancer patients did not significantly improve DFS.

12 Safety evaluation

The safety evaluation regarding the chemotherapy treatment (FEC-Doc or FEC-DocG) was performed in the Safety Population, defined as all patients who were treated with at least one cycle of FEC chemotherapy (n=778).

12.1 Brief summary of adverse events

For the safety evaluation, all adverse events observed within cycles 1 to 6 of the cytostatic treatments were analyzed. As the first three chemotherapy cycles comprised the same regimen (FEC) in both randomization arms, while the chemotherapy cycles 4 to 6 comprised different chemotherapy regimen in the two randomization arms (Doc vs. DocG), some of the following analyses were performed separately for chemotherapy cycles 1 to 3 and chemotherapy cycles 4 to 6.

The most common adverse events of any grade (more than 30% of patients) recorded during the first three cycles of chemotherapy treatment (FEC in both randomization arms) were blood and lymphatic system disorders (leukopenia, anemia, neutropenia), alopecia, fatigue and nausea. Leukopenia and neutropenia were also by far the most common grade 3 to 4 adverse events during the first three cycles of chemotherapy treatment, while no other type of grade 3 to 4 adverse event was observed in more than 3% of patients.

In cycles 4 to 6 of chemotherapy treatment (Doc or DocG) the most common adverse events of any grade (more than 30% of patients) observed in both treatment arms were blood and lymphatic system disorders (leukopenia, anemia, neutropenia), while in addition also SGPT elevation was frequently observed in the DocG arm. In both treatment arms, the most common grade 3 to 4 adverse events were leukopenia (more than 45% in both treatment arms) and neutropenia (more than 20% in both treatment arms), while febrile neutropenia was observed in about 3% of patients in both treatment arms. Types and frequencies of adverse events observed in the SUCCESS B study are as expected for cytostatic chemotherapy regimens containing anthracyklines and taxanes.

12.1.1 Adverse events

Frequencies of the main adverse events according to treatment arms, treatment cycles, and grades are shown in Tables 17 – 21. In addition, frequencies of G-CSF and antibiotic treatment are presented in Tables 22- 23.

Table 17 Most common adverse events for FEC treatment (cycles 1-3) according to CTCAE V 3.0 (n = 778 patients in the safety population)

Adverse event	**FEC (cycles 1-3; all grades) % (n)**		**FEC (cycles 1-3; grades 3 and 4) % (n)**	
Leukopenia	67.2	(523)	41.1	(320)
Alopecia	65.9	(513)	0.0	(0)
Nausea	54.6	(425)	2.6	(20)
Anemia	43.7	(340)	0.1	(1)
Neutropenia	38.9	(303)	29.8	(232)
Fatigue	38.3	(298)	2.1	(16)
SGPT elevation	27.0	(210)	1.4	(11)
Vomiting	20.8	(162)	2.1	(16)
Constipation	19.4	(151)	0.5	(4)
Stomatitis	18.0	(140)	0.3	(2)
Diarrhea	12.5	(97)	0.4	(3)
Thrombopenia	12.1	(94)	0.9	(7)
SGOT elevation	11.3	(88)	0.1	(1)
Headache	10.5	(82)	0.6	(5)
Bone pain	8.9	(69)	0.0	(0)
Neuropathy	8.2	(64)	0.3	(2)
Arthralgia	3.3	(26)	0.1	(1)
Nail changes	2.6	(20)	0.0	(0)
General pain	2.3	(18)	0.0	(0)
Gastrointestinal disorder	1.7	(13)	0.1	(1)
Febrile neutropenia	1.7	(13)	1.7	(13)

Table 18 Most common adverse events for cycles 4-6 all grades CTCAE V3.0 (n = 381 patients that received at least one cycle of Doc; n = 375 patients that received at least one cycle of DocG)

Adverse event	Doc (cycles 4-6; all grades) % (n)		DocG (cycles 4-6; all grades) % (n)	
Leukopenia	61.7	(235)	70.7	(265)
Anemia	36.2	(138)	52.8	(198)
Neutropenia	32.8	(125)	35.5	(133)
Bone pain	25.5	(97)	13.9	(52)
Neuropathy	23.4	(89)	18.7	(70)
Fatigue	22.6	(86)	26.7	(100)
Diarrhea	20.2	(77)	27.5	(103)
SGPT elevation	19.7	(75)	36.3	(136)
Stomatitis	17.8	(68)	20.3	(76)
Nail changes	17.3	(66)	13.6	(51)
Nausea	13.6	(52)	21.9	(82)
SGOT elevation	11.5	(44)	25.9	(97)
Arthralgia	11.5	(44)	9.3	(35)
Constipation	10.8	(41)	12.8	(48)
Vomiting	5.5	(21)	8.5	(32)
Headache	5.0	(19)	6.1	(23)
Febrile neutropenia	3.7	(14)	3.2	(12)
Thrombopenia	3.1	(12)	18.7	(70)
Alopecia	2.6	(10)	2.1	(8)
General pain	2.6	(10)	2.4	(9)
Gastrointestinal disorder	2.6	(10)	2.7	(10)

Table 19 Most common adverse events for cycles 4-6 grades 3-4 CTCAE V3.0 (n = 381 patients that received at least one cycle of Doc; n = 375 patients that received at least one cycle of DocG)

Adverse event	**Doc (cycles 4-6; grades 3-4) % (n)**		**DocG (cycles 4-6; grades 3-4) % (n)**	
Leukopenia	46.5	(177)	52.0	(195)
Neutropenia	27.6	(105)	24.8	(93)
Febrile neutropenia	3.7	(14)	3.2	(12)
Alopecia	0.0	(0)	0.0	(0)
Nausea	0.3	(1)	2.1	(8)
Anemia	0.5	(2)	1.9	(7)
Fatigue	1.3	(5)	3.7	(14)
Vomiting	0.5	(2)	0.5	(2)
Stomatitis	1.6	(6)	1.9	(7)
SGPT elevation	1.0	(4)	6.9	(26)
Constipation	0.3	(1)	0.0	(0)
Thrombopenia	0.3	(1)	1.9	(7)
SGOT elevation	0.3	(1)	1.3	(5)
Diarrhea	1.6	(6)	1.3	(5)
Headache	0.5	(2)	0.8	(3)
Neuropathy	0.8	(3)	0.5	(2)
General pain	0.0	(0)	0.5	(2)
Gastrointestinal disorder	0.0	(0)	0.0	(0)
Arthralgia	0.3	(1)	0.5	(2)
Bone pain	1.8	(7)	0.3	(1)
Nail changes	0.3	(1)	0.5	(2)

Table 20 Combined frequency of grade 3 or 4 leukopenia, neutropenia or febrile neutropenia for cycles 1-3 combined and for cycles 4-6 combined

Cycle	Number of patients	FEC (both randomization arms) % (n)		Doc % (n)		DocG % (n)	
Cycles 1-3 combined	778	52.2	(406)	-	-	-	-
Cycle 4-6 combined	381/375	-	-	51.4	(196)	55.2	(207)

Table 21 Frequency of G-CSF treatment performed for each single cycle, for cycles 1-3 combined and for cycles 4-6 combined

Cycle	Number of chemotherapy cycles received	FEC (both randomization arms) % (n)		Doc % (n)		DocG % (n)	
Cycle 1	778	11.2	(87)	-	-	-	-
Cycle 2	771	18.4	(142)	-	-	-	-
Cycle 3	764	22.3	(170)	-	-	-	-
Cycles 1-3 combined		26.1	(203)	-	-	-	-
Cycle 4	381/375	-	-	32.3	(123)	32.8	(123)
Cycle 5	368/364	-	-	35.3	(130)	42.0	(153)
Cycle 6	358/353	-	-	36.3	(130)	44.2	(156)
Cycle 4-6 combined		-	-	44.4	(169)	53.3	(200)

Table 22 Frequency of oral antibiotic therapy performed for each single cycle, for cycles 1-3 combined and for cycles 4-6 combined

Cycle	Number of chemotherapy cycles received	FEC (both randomization arms) % (n)		Doc % (n)		DocG % (n)	
Cycle 1	778	13.2	(103)	-	-	-	-
Cycle 2	771	8.4	(65)	-	-	-	-
Cycle 3	764	8.1	(62)	-	-	-	-
Cycles 1-3 combined		19.8	(154)	-	-	-	-
Cycle 4	381/375	-	-	14.7	(56)	14.4	(54)
Cycle 5	368/364	-	-	14.1	(52)	10.4	(38)
Cycle 6	358/353	-	-	10.1	(36)	9.9	(35)
Cycle 4-6 combined		-	-	24.9	(95)	25.6	(96)

Table 23 Frequency of intravenous antibiotic therapy performed for each single cycle, for cycles 1-3 combined and for cycles 4-6 combined

Cycle	Number of chemotherapy cycles received	FEC (both randomization arms) % (n)		Doc % (n)		DocG % (n)	
Cycle 1	778	2.2	(17)	-	-	-	-
Cycle 2	771	1.0	(8)	-	-	-	-
Cycle 3	764	0.9	(7)	-	-	-	-
Cycles 1-3 combined		3.2	(25)	-	-	-	-
Cycle 4	381/375	-	-	2.9	(11)	1.6	(6)
Cycle 5	368/364	-	-	1.9	(7)	2.5	(9)
Cycle 6	358/353	-	-	0.8	(3)	2.5	(9)
Cycle 4-6 combined		-	-	5.0	(19)	4.8	(18)

12.1.2 Analysis of adverse events

The most common adverse events (all grades) in the first three treatment cycles (FEC in both randomization arms; Table 17) affecting more that 20% of all patients were in descending order of frequency leukopenia (67.2%), alopecia (65.9%), nausea (54.6%), anemia (43.7%), neutropenia (38.9%), fatigue (38.3%), elevation of SGPT (27.0%), and vomiting (20.8%). The most common adverse events of grades 3 or 4 in the first three FEC cycles were leukopenia (41.1%) and neutropenia (29.8%). No other type of grade 3 or 4 adverse events was observed in more than 3% of patients (Table 18).

In the treatment cycles 4 to 6, the patients received either Doc (FEC-Doc group) or DocG (FEC-DocG group). The most common adverse events affecting more that 20% of all patients (all grades) observed in cycles 4 to 6 (Table 19) in the FEC-Doc group were in descending order of frequency leukopenia (61.7%), anemia (36.2%), neutropenia (32.8%), bone pain (25.5%), neuropathy (23.4%), fatigue (22.6%) and diarrhea (20.2%). In the FEC-DocG group, the most common adverse events were leukopenia (70.7%), anemia (52.8%), SGPT elevation (36.3%), neutropenia (35.5%), diarrhea (27.5%), fatigue (26.7%), SGOT elevation (25.9%), nausea (21.9%), and stomatitis (20.3%). Patients in the FEC-DocG group experienced considerably more adverse events of any grade during cycles 4 to 6 with regard to leukopenia, nausea, anemia, fatigue, SGPT elevation, thrombopenia, SGOT elevation, and diarrhea, while patients in the FEC-Doc group had considerably more often adverse events of any grade in terms of neuropathy and bone pain (Table 18). The most common grade 3 or 4 adverse events in cycles 4 to 6 observed in the FEC-Doc and the FEC-DocG group are listed in Table 18. The most common grade 3 or 4 adverse events in cycles 4 to 6 in the FEC-Doc group were leukopenia (46.5%), neutropenia (27.6%) and febrile neutropenia (3.7%); the most common grade 3 or 4 adverse events in cycles 4 to 6 in the FEC-DocG group were leukopenia (52.0%), neutropenia (24.8%), SGPT elevation (6.9%), fatigue (3.7%) and febrile neutropenia (3.2%). Patients in the FEC-DocG group experienced considerably more grade 3 or 4 adverse events during cycles 4 to 6 with regard to leukopenia, fatigue, nausea, anemia, SGPT elevation, thrombopenia and SGOT elevation, while patients in the FEC-Doc group had considerably more often grade 3 or 4 bone pain (Table 19).

We further analyzed the combined frequency of the three most common grade 3 or 4 adverse events leukopenia, neutropenia and febrile neutropenia in cycles 1 to 3 combined (FEC in both randomization arms) and in cycles 4 to 6 combined (Doc or DocG). As shown in Table x4, 52.2% of patients had at least one occurrence of grade 3 or 4 leukopenia, neutropenia and/or febrile neutropenia during the first three cycles. Overall, patients in the FEC-DocG group had more often at least one occurrence of grade 3 or 4 leukopenia, neutropenia or febrile neutropenia during cycles 4 to 6 than patients in the FEC-Doc group (55.2% vs. 51.4%; Table 19).

During the first three cycles, 11.2% (cycle 1) to 22.3% (cycle 3) of patients were treated with G-CSF (Table 20); overall, 26.1% of patients received G-CSF treatment during the first three cycles. A similar proportion of patients in the FEC-DocG group and the FEC-Doc group received G-CSF treatment in cycle 4 (32.8% vs. 32.3%), but patients in the FEC-DocG group received considerably more often G-CSF treatment than patients in the FEC-Doc group in cycle 5 (42.0% vs. 35.3%), cycle 6 (44.2% vs. 36.3%) and overall at least once during cycles 4 to 6 (53.3% vs. 44.4%).

Oral antibiotic therapy was given to 13.2%, 8.4% and 8.1% of patients in cycle 1, 2 and 3 respectively, and 19.8% of patients received at least one oral antibiotic treatment during the first three cycles (Table 21). The proportion of patients receiving oral antibiotic therapy in each of the cycles 4 to 6 ranged from 9.9% to 14.7% and was similar in the FEC-DocG and FEC-Doc group. In addition, the two groups were also similar with regard to the proportion of patients that received oral antibiotic treatment at least once during cycles 4 to 6 (25.6% vs. 24.9%; Table 22).

Only a small proportion of patients needed intravenous antibiotic treatment (Table 22). During the first three cycles, 2.2% (cycle 1), 1.0 % (cycle 2) and 0.9% (cycle 3) received intravenous antibiotic therapy, and 3.2% of patients were treated with intravenous antibiotics at least once during cycles 1 to 3. The proportion of patients receiving intravenous antibiotic therapy in each of the cycles 4 to 6 ranged from 0.8% to 2.9% and was similar in the FEC-DocG and FEC-Doc group; the two groups were also similar with regard to the proportion of patients that received intravenous antibiotics at least once during cycles 4 to 6 (4.8% vs. 5.0%; Table 22).

12.1.3 Listing of adverse events by patient

For a line listing of all adverse events (n=18048) by patients please see electronic appendix (Supplementary Table S12.1).

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

12.2.1 Deaths

Overall, 31 deaths were reported within the follow-up period (Table 24); the exact date of death was known for 29 deaths and unknown for two deaths. Most of the deaths (26; 83.9%) were breast cancer related, three deaths (9.7%) were regarded as not being tumor-related and for two deaths (6.5%) the cause of death was unknown.

Table 24 Deaths and causes of deaths within the follow-up period

Cause of death	n (%)	
Death breath cancer related	26	(83.9)
Death not tumor related	3	(9.7)
Cause of death unknown	2	(6.5)
Total	31	(100.0)

The line listing of all deaths (n = 31) is attached in the appendix of the study report (Supplementary Table S12.2).

12.2.2 Serious adverse events

Overall, 453 SAE's were observed during the SUCCESS B study, with 256 SAE's in the FEC-DocG arm and 197 SAE's in the FEC-Doc arm (Table 25). Most common SAE's were blood and lymphatic system disorders (mainly leukopenia, neutropenia, and febrile neutropenia), gastrointestinal disorders (mainly diarrhea, nausea, and

vomiting), and general disorders (mainly fatigue, general physical health deterioration, and fever). Types and frequencies of SAE's were as expected according to the relevant Summaries of Product Characteristics (SPC's).
Table x9 gives an overview of SAEs according to the MedDRA System Organ Class (SOC) terms; the line listing of all serious adverse events is attached in the appendix of the study report (see Supplementary Table S12.3).

Table 25 Overview of SAEs that occurred in the SUCCESS B study

System Organ Class Term	PT Term	Arm				Total
		A (FEC-DocG)		B (FEC-Doc)		
		N	%	N	%	N
Blood and lymphatic system disorders	Agranulocytosis		0.0	1	0.5	1
	Anaemia	1	0.4	2	1.0	3
	Febrile neutropenia	13	5.1	17	8.6	30
	Leukocytosis	1	0.4		0.0	1
	Leukopenia	19	7.4	18	9.1	37
	Lymphadenocyst		0.0	1	0.5	1
	Lymphadenopathy	1	0.4		0.0	1
	Neutropenia	6	2.3	10	5.1	16
	Thrombocytopenia	4	1.6		0.0	4
	White blood cell disorder	1	0.4	4	2.0	5
Blood and lymphatic system disorders - Total		*46*	*18.0*	*53*	*26.9*	*99*
Cardiac disorders	Angina pectoris	1	0.4		0.0	1
	Arrhythmia supraventricular		0.0	1	0.5	1
	Atrial fibrillation		0.0	1	0.5	1
	Atrial tachycardia		0.0	2	1.0	2
	Cardiac failure	1	0.4	1	0.5	2
	Coronary artery disease		0.0	1	0.5	1
	Nodal arrhythmia		0.0	1	0.5	1
	Palpitations		0.0	1	0.5	1
	Tachycardia paroxysmal		0.0	2	1.0	2
	Tachyarrhythmia	1	0.4		0.0	1
	Left ventricular dysfunction	1	0.4	3	1.5	4
Cardiac disorders - Total		*4*	*1.6*	*13*	*6.6*	*17*

Table 24 *- continued*

System Organ Class Term	PT Term	Arm				Total
		A (FEC-DocG)		B (FEC-Doc)		
		N	%	N	%	N
Ear and labyrinth disorders	External ear pain		0.0	1	0.5	1
	Vertigo	3	1.2		0.0	3
Ear and labyrinth disorders - Total		***3***	***1.2***	***1***	***0.5***	***4***
Eye disorders	Uveitis		0.0	1	0.5	1
Eye disorders - Total		***0***	***0.0***	***1***	***0.5***	***1***
Gastrointestinal disorders	Abdominal distension		0.0	1	0.5	1
	Abdominal pain	2	0.8	2	1.0	4
	Abdominal pain upper	1	0.4		0.0	1
	Anal haemorrhage		0.0	1	0.5	1
	Ascites	1	0.4		0.0	1
	Diarrhoea	8	3.1	3	1.5	11
	Dyspepsia		0.0	1	0.5	1
	Inguinal hernia		0.0	1	0.5	1
	Nausea	8	3.1	8	4.1	16
	Pancreatitis		0.0	2	1.0	2
	Stomatitis	1	0.4	2	1.0	3
	Vomiting	8	3.1	9	4.6	17
Gastrointestinal disorders - Total		***29***	***11.3***	***30***	***15.2***	***59***
General disorders and administration site conditions	Asthenia	2	0.8		0.0	2
	Chest discomfort	1	0.4		0.0	1
	Chest pain	2	0.8	1	0.5	3
	Chills	1	0.4		0.0	1
	Disease progression	2	0.8	1	0.5	3
	Fatigue	6	2.3	2	1.0	8
	General physical health deterioration	14	5.5	5	2.5	19
	Impaired healing	2	0.8		0.0	2
	Malaise	1	0.4		0.0	1
	Mucosal inflammation	5	2.0	1	0.5	6
	Necrosis	1	0.4		0.0	1
	Oedema	2	0.8		0.0	2
	Pain	1	0.4	1	0.5	2
	Performance status decreased	1	0.4		0.0	1
	Pyrexia	18	7.0	9	4.6	27
	Sudden death		0.0	1	0.5	1
General disorders and administration site conditions - Total		***59***	***23.0***	***21***	***10.7***	***80***
Immune system disorders	Hypersensitivity	2	0.8		0.0	2
Immune system disorders - Total		***2***	0.8	***0***	***0.0***	***2***
Infections and infestations	Appendicitis		0.0	1	0.5	1
	Bacterial sepsis		0.0	1	0.5	1
	Breast abscess		0.0	2	1.0	2
	Bronchitis	2	0.8		0.0	2
	Cellulitis		0.0	1	0.5	1
	Cystitis	1	0.4	1	0.5	2
	Device related infection	1	0.4	3	1.5	4
	Enterocolitis infectious		0.0	1	0.5	1
	Erysipelas	3	1.2		0.0	3
	Gastrointestinal infection		0.0	1	0.5	1
	Herpes zoster	1	0.4	1	0.5	2
	Infection	1	0.4		0.0	1
	Influenza	2	0.8		0.0	2
	Lip infection	1	0.4		0.0	1
	Mastitis	1	0.4		0.0	1
	Pneumonia	3	1.2	1	0.5	4
	Pyelonephritis	1	0.4		0.0	1
	Sepsis	1	0.4		0.0	1
	Urinary tract infection	1	0.4		0.0	1
	Wound infection	2	0.8	1	0.5	3
Infections and infestations - Total		***21***	***8.2***	***14***	***7.1***	***35***

Table 24 *- continued*

System Organ Class Term	PT Term	Arm				Total
		A (FEC-DocG)		B (FEC-Doc)		
		N	%	N	%	N
Injury, poisoning and procedural complications	Fracture	1	0.4	3	1.5	4
	Procedural complication	2	0.8		0.0	2
	Rib fracture	1	0.4		0.0	1
	Seroma	1	0.4		0.0	1
	Suture related complication		0.0	1	0.5	1
	Wound complication		0.0	1	0.5	1
	Wound dehiscence	2	0.8		0.0	2
Injury, poisoning and procedural complications - Total		***7***	***2.7***	***5***	***2.5***	***12***
Investigations	Alanine aminotransferase	1	0.4		0.0	1
	C-reactive protein increased	1	0.4		0.0	1
	Ejection fraction decreased	1	0.4		0.0	1
	Granulocytes abnormal	1	0.4		0.0	1
	Haemoglobin decreased		0.0	1	0.5	1
	Neutrophil count		0.0	1	0.5	1
	White blood cell count increased	1	0.4		0.0	1
Investigations - Total		***5***	***2.0***	***2***	***1.0***	***7***
Metabolism and nutrition disorders	Decreased appetite	2	0.8		0.0	2
	Dehydration	2	0.8		0.0	2
	Hyperglycaemia	2	0.8		0.0	2
	Type 2 diabetes mellitus		0.0	1	0.5	1
Metabolism and nutrition disorders - Total		***6***	***2.3***	***1***	***0.5***	***7***
Musculoskeletal and connective tissue disorders	Back pain	3	1.2	5	2.5	8
	Flank pain	1	0.4		0.0	1
	Musculoskeletal pain	1	0.4	1	0.5	2
	Myalgia	1	0.4	3	1.5	4
	Neck pain		0.0	1	0.5	1
	Osteoarthritis	1	0.4		0.0	1
	Osteoporosis		0.0	1	0.5	1
	Osteoporotic fracture	1	0.4		0.0	1
Musculoskeletal and connective tissue disorders - Total		***8***	***3.1***	***11***	***5.6***	***19***
Neoplasms benign, malignant and unspecified (incl cysts and polyps)	Lymphangiosis carcinomatosa	1	0.4		0.0	1
	Metastases to bone	2	0.8		0.0	2
	Metastases to liver	1	0.4	1	0.5	2
	Metastases to lung		0.0	1	0.5	1
	Metastases to lymph nodes	1	0.4		0.0	1
	Metastases to spine	1	0.4		0.0	1
	Neoplasm progression		0.0	1	0.5	1
	Ovarian cancer	1	0.4		0.0	1
	Second primary malignancy	1	0.4	1	0.5	2
	Uterine cancer		0.0	1	0.5	1
Neoplasms benign, malignant and unspecified (incl cysts and polyps) - Total		***8***	***3.1***	***5***	***2.5***	***13***
Nervous system disorders	Dizziness	1	0.4	1	0.5	2
	Epilepsy		0.0	1	0.5	1
	Headache	3	1.2	1	0.5	4
	Lethargy	1	0.4		0.0	1
	Loss of consciousness		0.0	1	0.5	1
	Migraine	1	0.4		0.0	1
	Muscle spasticity		0.0	1	0.5	1
	Neuralgia		0.0	1	0.5	1
	Presyncope		0.0	1	0.5	1
	Speech disorder		0.0	1	0.5	1
	Subarachnoid haemorrhage		0.0		0.0	0
	Syncope	1	0.4		0.0	1
Nervous system disorders - Total		***7***	***2.7***	***8***	***4.1***	***15***
Product issues	Device malfunction	1	0.4		0.0	1
	Thrombosis in device		0.0	1	0.5	1
Product issues - Total		***1***	***0.4***	***1***	***0.5***	***2***

Table 24 *- continued*

System Organ Class Term	PT Term	Arm				Total
		A (FEC-DocG)		B (FEC-Doc)		
		N	%	N	%	N
Psychiatric disorders	Anxiety	1	0.4	1	0.5	2
	Depression	2	0.8	1	0.5	3
	Fear		0.0	1	0.5	1
	Mood altered	1	0.4	1	0.5	2
Psychiatric disorders - Total		***4***	***1.6***	***4***	***2.0***	***8***
Renal and urinary disorders	Pollakiuria		0.0	1	0.5	1
	Renal failure		0.0	3	1.5	3
	Urethral caruncle		0.0	1	0.5	1
Renal and urinary disorders - Total		***0***	***0.0***	***5***	***2.5***	***5***
Reproductive system and breast	Breast mass	1	0.4		0.0	1
	Menorrhagia	1	0.4		0.0	1
Reproductive system and breast disorders - Total		***2***	***0.8***	***0***	***0.0***	***2***
Respiratory, thoracic and mediastinal disorders	Alveolitis	1	0.4		0.0	1
	Bronchial hyperreactivity	1	0.4		0.0	1
	Cough	1	0.4	1	0.5	2
	Dyspnoea	5	2.0	2	1.0	7
	Lung infiltration	1	0.4	1	0.5	2
	Pleural effusion	1	0.4	1	0.5	2
	Pneumonitis	1	0.4	1	0.5	2
	Pneumothorax	1	0.4		0.0	1
	Pulmonary congestion	1	0.4		0.0	1
	Pulmonary embolism	4	1.6	1	0.5	5
	Pulmonary infarction	1	0.4		0.0	1
	Pulmonary oedema	1	0.4		0.0	1
	Tracheal inflammation	1	0.4		0.0	1
Respiratory, thoracic and mediastinal disorders - Total		***20***	***7.8***	***7***	***3.6***	***27***

System Organ Class Term	PT Term	Arm				Total
		A (FEC-DocG)		B (FEC-Doc)		
		N	%	N	%	N
Skin and subcutaneous tissue disorders	Dermatitis allergic	2	0.8		0.0	2
	Erythema	2	0.8		0.0	2
	Erythema multiforme	1	0.4		0.0	1
	Skin discouloration	1	0.4		0.0	1
Skin and subcutaneous tissue disorders - Total		***6***	***2.3***	***0***	***0.0***	***6***
Surgical and medical procedures	Breast operation		0.0	1	0.5	1
	Breast prosthesis implantation	1	0.4	1	0.5	2
	Breast reconstruction		0.0	1	0.5	1
	Hip surgery	1	0.4		0.0	1
	Mammoplasty	2	0.8		0.0	2
	Mastectomy		0.0	1	0.5	1
	Medical device implantation	2	0.8		0.0	2
	Medical device removal	1	0.4		0.0	1
	Ovarian operation	1	0.4		0.0	1
	Thyroidectomy	1	0.4		0.0	1
Surgical and medical procedures - Total		***9***	***3.5***	***4***	***2.0***	***13***
Vascular disorders	Circulatory collapse	2	0.8	1	0.5	3
	Embolism		0.0	2	1.0	2
	Hypertension	1	0.4		0.0	1
	Hypertensive crisis		0.0	1	0.5	1
	Hypotension		0.0	1	0.5	1
	Orthostatic hypotension	1	0.4		0.0	1
	Subclavian vein thrombosis	1	0.4		0.0	1
	Thrombophlebitis superficial		0.0	1	0.5	1
	Thrombosis	4	1.6	5	2.5	9
Vascular disorders - Total		***9***	***3.5***	***11***	***5.6***	***20***
TOTAL		**256**	**100.0**	**197**	**100.0**	**453**

12.2.3 Suspected unexpected serious adverse reactions

During the course of the SUCCESS B study, two SUSARs were reported (Table 25). One SUSAR, “Collapse in presence of bronchitis”, occurred under gemcitabine treatment, and causality was given as “possible”. The patient was transferred to an intensive care unit and received artificial respiration (diagnosis: pneumonia with respiratory insufficiency, sepsis, and acute renal failure); finally the patient recovered. The second SUSAR, “kidney failure”, occurred under FEC treatment, and causality was given as “probably”. The patient received gastroenteritis therapy and infusions, but eventually died because of kidney failure.

Table 26 SUSARS in the SUCCESS B study

SUSAR ID	Patient ID	Description of event	Date of randomization	Randomization arm	Date SUSAR	Treatment	Causality	Reason	Outcome
1	269	Collapse in presence of bronchitis	13.11.2009	A (FEC-DocG)	25.11.2010	Gemcitabine	possibly	hospitalization, life-threatening	recovered
2	504	Kidney Failure	30.11.2010	B (FEC-Doc)	17.05.2011	FEC	probably	hospitalization	death

12.3 Summary of safety results

All adverse events, serious adverse events and serious unexpected adverse events represent the well-known spectrum of side effects associated with a cytotoxic breast cancer treatment based on an anthracycline-taxane-based chemotherapy regimen. The most common adverse events that occurred in cycles 4 to 6 in the FEC-Doc group were leukopenia (61.7%), anemia (36.2%), neutropenia (32.8%). In comparison, in the FEC-DocG group, the most frequent adverse events that occurred in cycles 4 to 6 were again leukopenia (70.7%), anemia (52.8%), SGPT elevation (36.3%) and neutropenia (35.5%). In general, patients in the FEC-DocG group experienced more adverse events of any grade during cycles 4 to 6 with regard to leukopenia, nausea, anemia, fatigue, SGPT elevation, thrombopenia, SGOT elevation and diarrhea, while patients in the FEC-Doc group had considerably more often adverse events of any grade in terms of neuropathy and bone pain (Table 18). In addition, patients in the FEC-DocG group experienced considerably more grade 3 or 4 adverse events during cycles 4 to 6 with regard to leukopenia, fatigue, nausea, anemia, SGPT elevation, thrombopenia and SGOT elevation, while patients in the FEC-Doc group had considerably more often grade 3 or 4 bone pain (Table 19). During the first three cycles (FEC), 52.2% of patients had at least one occurrence of grade 3 or 4 leukopenia, neutropenia or febrile neutropenia. The combined frequency of grade 3 or 4 leukopenia, neutropenia or febrile neutropenia during cycles 4 to 6

was higher in the FEC-DocG group compared to the FEC-Doc group (55.2% vs. 51.4%). This effect was also apparent with regard to the need for application of concomitant medication, as patients in the FEC-DocG group received more often at least once G-CSF treatment during cycles 4 to 6 than patient in the FEC-Doc group (53.3% vs. 44.4%). However, no large differences were observed with regard to antibiotic treatment: 25.6% and 24.9% of patients received at least once oral antibiotic treatment during cycles 4 to 6 in the FEC-DocG and FEC-Doc group, respectively, while 4.8% and 5.0% of patients received at least once intravenous antibiotic treatment during cycles 4 to 6 in the FEC-DocG and FEC-Doc group, respectively.

In conclusion, in the SUCCESS B study the FEC-DocG regime in general caused more toxicity than the FEC-Doc regime. Overall, the results regarding the toxicity and safety profile of the two chemotherapy regimen (FEC-DocG and FEC-Doc) are similar to the ones observed in the SUCCESS A study (EuDURA-CT number 2005-000490-21), in which the same chemotherapy regimen were compared.

13 Discussion and overall conclusions

The SUCCESS-B study compared the disease free survival after randomization in patients treated with 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC)-chemotherapy, followed by 3 cycles of Docetaxel(D)-chemotherapy versus 3 cycles of Epirubicin-Fluorouracil-Cyclophosphamide(FEC), followed by 3 cycles of Gemcitabine- Docetaxel(DG) chemotherapy. All patients were required to have HER2-positive disease and therefore both groups received biological anti-HER2 treatment with Trastuzumab according to the general therapy guidelines.

Postmenopausal patients with positive hormone receptor status of the primary tumor received Letrozole treatment for 5 years, after the end of chemotherapy. Premenopausal patients received Tamoxifen treatment. In addition to Tamoxifen, all patients with positive hormone receptor status of the primary tumor and under the age of 40 or restart of menstrual bleeding within 6 months after the completion of cytostatic treatment or with premenopausal hormone levels as defined below received Goserelin 3.6 mg subcutaneously every 4 weeks over a period of 2 years following chemotherapy (1, 2).

The multicenter SUCCESS-B study was conducted in 251 study centers that comprised academic and non-academic cancer centers, specialist hospitals and outpatient clinics in all regions of Germany. Aim of the study was to evaluate the efficacy of the addition of gemcitabine to the chemotherapy treatment of high risk early breast cancer patients.

In conclusion, none of the agents under investigation in this trial did show any unknown or unexpected side effects compared to the so far published literature. The addition of gemcitabine to FEC-D adjuvant chemotherapy increases toxicity moderately. However, adjuvant Gemcitabine does not improve the efficacy of FEC-Doc chemotherapy for high risk breast cancer patients, with regard to the risk-benefit ratio, we do not recommend adjuvant Gemcitabine for the adjuvant treatment of high risk breast cancer patients.

14 Literature

1. Taylor CW GS, Dalton WS, Martino S, Rector D, Ingle JN et al. . Multicenter randomized clinical trial of goserelin versus surgical ovariectomy in premenopausal patients with receptor-positive metastatic breast cancer: an intergroup study. *J Clin.Oncol.* 1998;16:994-9. *J ClinOncol* 1998;16:994-9.
2. Davidson N, O'Neill, A., Vukov, A., Osborne, C. K., Martino, S., White, D., and Abeloff, M. . Chemohormonal therapy in premenopausal node-positive, receptor-positive, breast cancer: an Eastern Cooperative Oncology Group Phase III Intergroup trial (E5188,INT-0101). 2003.
3. Black RJ BF, Ferlay J, Parkin DM. Cancer Incidence and Mortality in the European Union: Cancer Registry Data and Estimates of National Incidence for 1990. European Journal of Cancer. 1997;33:1075-107.
4. Bonadonna G VP, Brambilla C, Ferrari L, Moliterni A, Terenziani M et al. . Primary chemotherapy in operable breast cancer: eight-year experience at the Milan Cancer Institute. . *JClin Oncol* 1998:16:93-100.
5. HD. B. Zur Strahlentherapie bei Mamma-Karzinom (Radiation therapy in breast carcinoma). . *ZentralblGynakol*. 1991;113:1147-61.
6. Blichert TM RC, Andersen JA, Overgaard M, Axelsson CK, Andersen KW et al. Danish Breast Cancer Cooperative Group. . Danish randomized trial comparing breast conservation therapy with mastectomy: six years of life-table analysis. . *J NatlCancer InstMonogr*. 1992;11:19-25.
7. Fisher B RC, Poisson R, Margolese R, Wolmark N, Wickerham L et al. . Eight-year results of a randomized clinical trial omparing total mastectomy and

lumpectomy with or without irradiation in the treatment of breast cancer. *NEnglJ Med*. 1989;320:822-8.
8. Jacobson JA DD, Cowan KH, d'Angelo T, Steinberg SM, Pierce L et al. Ten-year results of a comparison of conservation with mastectomy in the treatment of stage I and II breast cancer. *NEnglJ Med*. 1995(332)::907-11.
9. Sarrazin D LM, Arriagada R, Contesso G, Fontaine F, Spielmann M et al. . Ten-year results of a randomized trial comparing a conservative treatment to mastectomy in early breast cancer. . *RadiotherOncol*. 1989;14:177-84.
10. Van-Dongen JA BH, Fentiman IS, Lerut T, Mignolet F, Olthuis G et al. Randomized clinical trial to assess the value of breast-conserving therapy in stage I and II breast cancer, EORTC 10801 trial. *J NatlCancer InstMonogr* 1992:15-8.
11. Veronesi U SB, Luini A, Greco M, Saccozzi R, Del-Vecchio M et al. . Breast conservation is a safe method in patients with small cancer of the breast. Long-term results of three randomised trials on 1,973 patients. *EurJ Cancer*. 1995;31A:1574-9.
12. Meijer-van-Gelder ME LM, Bolt d, V, Peters HA, Klijn JG, Foekens JA. . Breast-conserving therapy: proteases as risk factors in relation to survival after local relapse. *JClinOncol*. 1999;17:1449-57.
13. Osborne MP ON, Harmer CL, McKinna JA, Baker J, Greening WP. Breast conservation in the treatment of early breast cancer. A 20-year follow-up. *Cancer* 1984;53:349-55.
14. Veronesi U ME, Del-Vecchio M, Manzari A, Andreola S, Greco M et al. Local recurrences and distant metastases after conservative breast cancer treatments: partly independent events. *J NatlCancer InstMonogr*. 1995;87:19-27.
15. Goldhirsch A WW, Gelber RD, Coates AS, Thurlimann B, Senn HJ. . Meeting highlights: updated international expert consensus on the primary therapy of early breast cancer. . *JClinOncol*. 2003;21:3357-65.
16. Goldhirsch A WW, Gelber R, Coates A, Thurlimann B, Senn HJ et al.expert Progress and promise: highlights of the international consensus on the primary therapy of early breast cancer. *AnnOncol* 2007. 2007;18:1133-44.
17. Arnesson LG SS, Fagerberg G. Recurrence-free survival in patients with small breast cancer. An analysis of cancers 10 mm or less detected clinically and by screening. *EurJ Surg*. 1994;160:271-6.
18. Bonadonna G BE, Valagussa P, Rossi A, Brugnatelli L, Brambilla C et al. . Combination chemotherapy as an adjuvant treatment in operable breast cancer. *NEnglJ Med* 1976;294:405-10.
19. Bonadonna G VP, Moliterni A, Zambetti M, Brambilla C. Adjuvant cyclophosphamide, methotrexate, and fluorouracil in node-positive breast cancer: the results of 20 years of follow-up *NEnglJ Med*. 1995(332):901-6.
20. Lenaz L PJ. Cardiotoxicity of adriamycin and related anthracyclines. *Cancer TreatRev*. 1976(3):111-20.

21. Group EBCTC, editor Analysis overview results. Fifth Meeting of the Early Breast Trialists' Collaborative Group 21-9-2000; Oxford.
22. Levine MN BV, Pritchard KL, Norris B, Shepherd LE, Abu ZH et al. Randomized Trial of Intensive Cyclophosphamide, Epirubicin and Fluorouracil Chemotherapy Compared with Cyclophosphamide, Methothrexat and Fluorouracil in Premenopausal Women With Node-Positive Breast *J ClinOncol* Cancer. 1998;16:2651-8.
23. EBCTCG. Polychemotherapy for early breast cancer: an overview of the randomised trials. Early Breast Cancer Trialists' Collaborative Group. *Lancet* 1998;352:930-42.
24. Coombes RC BJ, Wils J, Morvan F, Espie M, Amadori D et al. The International Collaborative Cancer Group. Adjuvant cyclophosphamide, methotrexate, and fluorouracil versus fluorouracil, epirubicin, and cyclophosphamide chemotherapy in premenopausal women with axillary node-positive operable breast cancer: results of a randomized trial. *JClin Oncol* 1996;14:35-45.
25. Fisher B BA, Dimitrov NV, Poisson R, Redmond C, Margolese RG et al. Two months of doxorubicin-cyclophosphamide with and without interval reinduction therapy compared with 6 months of cyclophosphamide, methotrexate, and fluorouracil in positive-node breast cancer patients with tamoxifen-nonresponsive tumors: results from the National Surgical Adjuvant Breast and Bowel Project B-15. *JClin Oncol*. 1990;8:1483-96.
26. Bonneterre J, Roche, H., Kerbrat, P., Bremond, A., Fumoleau, P., Namer, M., Goudier, M. J., Schraub, S., Fargeot, P., and Charpelle-Margillac, I. . French Adjuvant Study Group 05 trial (FEC 50 vs FEC 100): 10-year update of benefit/risk ratio after adjuvant chemotherapy (CT) in node-positive (N+), early breast cancer (EBC) patients (pts). Proc ASCO 22, 24.
. Ref Type: Abstract. 2003.
27. cancer. TDOgas-atfab. *Clin.Breast Cancer* 3
2002;3 Suppl 1:8-11.
28. Fisher B AS, Wickerham DL, DeCillis A, Dimitrov N, Mamounas E et al. Increased intensification and total dose of cyclophosphamide in a doxorubicin-cyclophosphamide regimen for the treatment of primary breast cancer: findings from National Surgical Adjuvant Breast and Bowel Project B-22. *JClin Oncol*. 1997;15:1858-69.
29. Fisher B AS, DeCillis A, Dimitrov N, Atkins JN, Fehrenbacher L et al. Project B-25. Further evaluation of intensified and increased total dose of cyclophosphamide for the treatment of primary breast cancer: findings from National Surgical Adjuvant Breast and Bowel *JClin Oncol*. 1999;17:3374-88.
30. Henderson CI, Berry, D., Demetri, G., and et al. . Improved disease free and overall survival from the addition of sequential paclitaxel but not from the

escalation of doxorubicin dose level in the adjuvant chemotherapy of patients with node-positive cancer. Proc Am Soc Clin Oncol A 1998;17(101).
31. Cersosimo RJ HW. Epirubicin: a review of the pharmacology, clinical activity, and adverse effects of an adriamycin analogue. . *J ClinOncol* 1986;4:425-39.
32. Lopez M PP, Di-Lauro L, Vici P, Carpano S, Conti EM. 5-Fluorouracil, adriamycin, cyclophosphamide (FAC) vs. 5-fluorouracil, epirubicin, cyclophosphamide (FEC) in metastatic breast cancer. . *Oncology* 1989;46:1-5.
33. Vermorken JB BHW, Mandjes IA, Postma TJ, Huizing MT, Heimans JJ et al. High-dose paclitaxel with granulocyte colony-stimulating factor in patients with advanced breast cancer refractory to anthracycline therapy: a European Cancer Center trial. . *SeminOncol*. 1995;22:16-22.
34. Chan S FK, Noel D, Pinter T, Van Belle S, Vorobiof D et al. . Prospective randomized trial of docetaxel versus doxorubicin in patients with metastatic breast cancer. The 303 Study Group. *JClin Oncol*. 1999;17:2341-54.
35. Nabholtz JM SH, Bezwoda WR, Melnychuk D, Deschenes L, Douma J et al. . Prospective randomized trial of docetaxel versus mitomycin plus vinblastine in patients with metastatic breast cancer progressing despite previous anthracycline-containing chemotherapy. 304 Study Group. *JClin Oncol*. 1999(17):1413-24.
36. Nabholtz JM NS, Smylie M, Mackey J, Au HJ, Au R et al. . Docetaxel (Taxotere) in combination with anthracyclines in the treatment of breast cancer. *SeminOncol* 2000;27:11-8.
37. Nabholtz JM, Falkson, G., and Campos, D. . A phase III trial comparing adriamycin and docetaxel to adriamycin and Acyclophosphamide as fist line chemotherapy for metastatic breast cancer. Proc Am Soc Clin Oncol 1999;18.
38. Bonneterre J DV, Tubiana-Hulin M, Bougnoux P, et al. 6 Cycles of Epirubicin/Docetaxel (ET) Versus 6 Cycles of 5FU Epirubici/Cyclophosphamide (FEC) as First Line Metastatic Breast Cancer (MBC) Treatment. *Proc Am Soc Clin Oncol*. 2001;20.
39. Nabholtz JM, Paterson, A., Dirix, L., Dewar, J., Chap, L., Chan, S., and et al. . A Phase III Randomized Trial Comparing Docetaxel, Doxorubicin and Cyclophphosphamide to FAC as First Line Chemotherapy for Patients with Metastatic Breast Cancer. Proc Am Soc. Clin Oncol 20. 2001.
40. Henderson IC BD, Demetri GD, Cirrincione CT, Goldstein LJ, Martino S et al. . Improved outcomes from adding sequential Paclitaxel but not from escalating Doxorubicin dose in an adjuvant chemotherapy regimen for patients with node-positive primary breast cancer. *J Clin Oncol*. 2003;21:976-83.
41. Mamounas E.P. B, J., Lembersky, C., Fisher, B., Atkins, J. N., Fehrenbacher, L., Raich, P. C., Yother, G., Soran, A., and Wolmark, N. , editor Paclitaxel (T) following doxorubicin/cyclophosphamide (AC) as adjuvant chemotherapy for node-positive breast cancer: Results from NSABP B-28.

. ASCO 22, 4; 2003.
42. Nabholtz JM, Pienkowsky, T., Mackey, J., Pawlicky, M., Guastalla, J., Vogel, C., Weaver, C., Walley, B, Martin, M., Chap, L., Tomiak, E. M., Juhos, E., Guevin, R., Howell, A., Hainsworth, J., Fornander, T., Blitz, S., Gazel, S., Loret, C., and Riva, A. . Phase III Trial comparing TAC (Docetaxel, Doxorubicin, Cyclophosphamide) with FAC (5-Fluorouracil, Doxorubicin, Cyclophosphamide) in the adjuvant treatment of node positive breast cancer patients: interim analysis of the BCIRG 001 Study. Proc Am Soc Clin Oncol 2002.
43. Martin M. PT, Mackey J., Pawlicki M., Guastalla J.P., Weaver C.H., Tomiak E., Al-Tweigeri T., Chap L., Juhos E., Guevin R., Howell A., Fornander T., Hainsworth T., Coleman R., Vinholes J., Modiano M., Pinter T., Hugh J., Nabholtz J.M., Loret C., Rupin M., Blitz S., Riva A., and Vogel, C. L. . TAC Improves DFS and OS Over FAC in Node Positive Early Breast Cancer Patients,BCIRG001: 55 Months Follow-up. Breast Cancer ResTreat 67 ((Suppl) 1) 2003.
44. Roche H, Fumoleau, P., Spielmann M., Canon, J. L., Delozier, T., Kerbrat, P., Serin, D., Lortholary, A., de Ghislain, C., Viens, P., Bergerat, J. P., Geneve, J., Martin, A. L., and Asselain, B. Five years analysis of the PACS 01 trial: 6 cycles of FEC100 vs 3 cycles of FEC100 followed by 3 cycles of docetaxel (D) for the adjuvant treatment of node positive breast cancer. . Breast Cancer Res Treat2004.
45. Bria E NC, Cuppone F, Carlini P, Ciccarese M, Milella M et al. . Benefit of taxanes as adjuvant chemotherapy for early breast cancer: pooled analysis of 15,500 patients. *Cancer*. 2006;106:2337-44.
46. Lohrisch C DLA, Piccart MJ. In: Wood WC, Henderson CI, Piccart MJ. Optimal Adjuvant Cytotoxic Therapy for Breast Cancer. Adjuvant Therapy for Breast Cancer. ASCO2001. p. 61-70.
47. Davidson N. The use of anthracyclines and taxanes for adjuvant therapy of breast cancer. The Breast. 2001;10:1.
48. D. T. Overview: gemcitabine as single-agent therapy for advanced and metastatic breast cancer. . Oncology 2003;64 191-206.
49. Heinemann V. Role of gemcitabine in the treatment of advanced and metastatic breast cancer. . *Oncology* 2003;64:191-206.
50. CC. Z. Gemcitabine/anthracycline combinations in metastatic breast cancer. . *ClinBreast Cancer*. 2002;3 Suppl 1:30-3.
51. Yardley D. Gemcitabine and taxanes as new szandard if care in breast cancer Clin Breast Cancer. 2004;4(Suppl. 1):30-3.
52. Laufman LR SC, Pritchard J, Roach R, Zangmeister J, Larrimer N et al. . Monthly docetaxel and weekly gemcitabine in metastatic breast cancer: a phase II trial. . *AnnOncol 2001*. 2001;12:1259-64.

53. Vici P BF, Di Lauro L, Amodio A, Conti F, Foggi P et al. . Docetaxel in patients with anthracycline-resistant advanced breast cancer. *Oncology* 2001. 2001;60:60-5.
54. Fountzilas G NC, Bafaloukos D, Kalogera-Fountzila A, Kalofonos H, Samelis G et al. . Docetaxel and gemcitabine in anthracycline-resistant advanced breast cancer: a Hellenic Cooperative Oncology Group Phase II study. . *Cancer Invest*. 2000;18:503-9.
55. Schneeweiss A HJ, Sinn HP, von Fournier D, Rudlowski C, Beldermann F et al. Gemcitabine, epirubicin and docetaxel as primary systemic therapy in patients with early breast cancer: results of a multicentre phase I/II study. . *EurJCancer*. 2004;40:2432-8.
56. Mavroudis D MN, Alexopoulos A, Kourousis C, Agelaki S, Sarra E et al. Salvage chemotherapy in anthracycline-pretreated metastatic breast cancer patients with docetaxel and gemcitabine: a multicenter phase II trial. Greek Breast Cancer Cooperative Group. . *AnnOncol* 1999. 1999;10:211-5.
57. Alexopoulos A TD, Karamouzis MV, Gerasimidis G, Karydas I, Kandilis K et al. b. . Evidence for in vivo synergism etween docetaxel and gemcitabine in patients with metastatic breast cancer. *AnnOncol* 2004. 2004(15):95-9.
58. Brandi M VP, Lopez M, Valerio MR, Giotta F, Gebbia N et al. Novel association with gemcitabine and docetaxel as salvage chemotherapy in metastatic breast cancer previously treated with anthracyclines: results of a multicenter phase II study. . *SeminOncol* 2004. 2004;31:13-9.
59. Murad AM GR, Aragao BC, Scalabrini-Neto AO, Rodrigues VH, Garcia R. *AmJClinOncol* 2001. Phase II trial of the use of paclitaxel and gemcitabine as a salvage treatment in metastatic breast cancer. ;24:264-8.
60. Delfino C CG, Gonzales LR, Mickiewicz E, Rodger J, Balbiani L et al. . Gemcitabine plus paclitaxel as first-line chemotherapy for patients with advanced breast cancer. *Oncology*. 2004;66:18-23.
61. Genot JYaT-H, M. . Gemcitabine and paclitaxel in metastatic breast cancer: a phase II study in the first line setting. . Proc ASCO2002.
62. Palmeri S, Vaglica, M., Accurso, V., and Gebbia, V. Weekly docetaxel (doc)+ gemcitabine (gem) as first line therapy for advanced breast cancer (ABC): a multicenter phase II study. . Proc ASCO2002.
63. Colomer R LA, Lluch A, Ojeda B, Barnadas A, Caranana V et al. Paclitaxel/gemcitabine administered every two weeks in advanced breast cancer: preliminary results of a phase II trial. . *SeminOncol* 2000;27:20-4.
64. Sanchez-Rovira P JA, Gonzalez E, Porras I, Duenas R, Medina B et al. . Biweekly gemcitabine, doxorubicin, and paclitaxel as first-line treatment in metastatic breast cancer. Final results from a phase II trial. . *Oncology (Huntingt)*. 2001;15:44-7.
65. Kornek GV HK, Kwasny W, Raderer M, Schull B, Payrits T et al;. Treatment of advanced breast cancer with docetaxel and gemcitabine with and

without human granulocyte colony-stimulating factor. . *ClinCancer Res* 2002;8:1051-6.
66. Pelegri A CL, Mayordomo JI, Florian J, Vazquez S, Arcusa A et al. Gemcitabine plus docetaxel administered every other week as first-line treatment of metastatic breast cancer: preliminary results from a phase II trial. . *SeminOncol*. 2004;31:20-4.
67. Llombart A, Lluch, A., Ojeda, B., Barnadas, A., Caranana, V., and Fernandez, Y. Paclitaxel and Gemcitabine Administered Every Two Weeks in Advanced Breast Cancer. Proc ASCO2000.
68. O'Shaughnessy J, Nag, S., Calderillo-Ruiz, G., Jordaan, J., Llombart, A., Pluzanska, A., Pawlicki M., Reyes, M., Sekhon, J., and Albain, K. . Gemcitabine plus paclitaxel (GT) versus paclitaxel (T) as first-line treatment for anthracycline pre-treated metastatic breast cancer (MBC): Interim results of a global phase III study. Proc ASCO 2003.
69. Jaiyesimi IA BA, Decker DA, Hortobagyi GN. Use of tamoxifen for breast cancer: twenty-eight years later. *J ClinOncol*. 1995;13:513-29.
70. Pritchard K. Endocrine therapy of advanced disease: analysis and implications of the existing data. *ClinCancer Res*. 2003;9:460S-7S.
71. J. C. The ATAC (`Arimidex', Tamoxifen, Alone or in Combination) trial in postmenopausal women with early breast cancer - Updated efficacy results based on a median follow-up of 47 months. The Breast 2003;12(Suppl. 1):47.
72. ATAC HA. ('Arimidex', Tamoxifen, Alone or in Combination) completed treatment analysis: Anastrozole demonstrates superior efficacy and tolerability compared with tamoxifen. Breast Cancer Res Treat Suppl 1. 2004.
73. Baum M BA, Cuzick J, Forbes J, Houghton JH, Klijn JG et al. . Anastrozole alone or in combination with tamoxifen versus tamoxifen alone for adjuvant treatment of postmenopausal women with early breast cancer: first results of the ATAC randomised trial. *Lancet*. 2002;359:2131-9.
74. Group EBCTC. Analysis overview results. Fifth Meeting of Early Breast Trialists' Collaborative Group; Oxford2000
75. Wood W. Who Doesn't Need Systemic Adjuvant Therapy, and What is Optimal Endocrine Therapy. In: . Wood WC Adjuvant Therapy for Breast Cancer 2001:;ASCO:49-51.
76. Davidson N. Consensus Development Conference on Adjuvant Therapy for Breast Cancer. Bethesda, National Institue of NIH Health. 2000:63-6.
77. Davidson N, O'Neill, A., Vukov, A., Osborne, C. K., Martino, S., White, D., and Abeloff. Chemohormonal therapy in remenopausal node-positive, receptor-positive, breast cancer: an Eastern Cooperative Oncology Group Phase III Intergroup trial (E5188,INT-0101). . pProc Am Soc Clin Oncol 22. 2003.
78. Jonat W. Zoladex (goserelin) vs. CMF as adjuvant therapy in Pre-/perimenopausal early (node positive) breast cancer: Preliminary efficacy -

QOL, and BMD results form the ZEBRA study. Breast Cancer Res Treat. 2000;64((13)).
79. Henderson C. Endocrine Therapy, Chemotherapy, or Both as Adjuvant Systemic Treatment of Patients with Early Breast Cancer. Wood WC Adjuvant Therapy for Breast Cancer ASCO. 2001.
80. Taylor CW GS, Dalton WS, Martino S, Rector D, Ingle JN et al. Multicenter randomized clinical trial of goserelin versus surgical ovariectomy in premenopausal patients with receptor-positive metastatic breast cancer: an intergroup study. *J ClinOncol*. 1998;16:994-9.
81. Cross M DT. Growth factors in development, transformation, and tumorigenesis. *Cell* 1991;64:271-80.
82. Popescu NC KC, Kraus MH. . Localization of the human erbB-2 gene on normal and rearranged chromosomes 17 to bands q12-21.32. *Genomics*. 1989;4:362-6.
83. Hynes NE SD. The biology of erbB-2/neu/HER-2 and its role in cancer. *BiochimBiophysActa*. 1994;1198:165-84.
84. Lemoine NR SS, Dickson C, Barnes DM, Gullick WJ. Absence of activating transmembrane mutations in the c-erbB-2 proto-oncogene in human breast cancer. *Oncogene* 1990;5:237-9.
85. Pauletti G GW, Press MF, Slamon DJ. . Detection and quantitation of HER-2/neu gene amplification in human breast cancer archival material using fluorescence in situ hybridization. . *Oncogene*. 1996;13:63-72.
86. Slamon DJ GW, Jones LA, Holt JA, Wong SG, Keith DE et al. Studies of the HER-2/neu proto-oncogene in human breast and ovarian cancer. *Science*. 1989;244:707-12.
87. Slamon DJ CG, Wong SG, Levin WJ, Ullrich A, McGuire WL. . Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. *Science*. 1987;235:177-82.
88. Bargmann CI HM, Weinberg RA. Multiple independent activations of the neu oncogene by a point mutation altering the transmembrane domain of p185. *Cell*. 1986;45:649-57.
89. Bargmann CI WR. Oncogenic activation of the neu-encoded receptor protein by point mutation and deletion. . *EMBO J*. 1988;7:2043-52.
90. Di Fiore PP PJ, Kraus MH, Segatto O, King CR, Aaronson SA. . erbB-2 is a potent oncogene when overexpressed in NIH/3T3 cells. *Science*. 1987;237:178-82.
91. Hudziak RM SJ, Ullrich A. Increased expression of the putative growth factor receptor p185HER2 causes transformation and tumorigenesis of NIH 3T3 cells. . *Proc NatlAcad Sci USA* 1987(84):7159-63.
92. Chazin VR KM, Miller AD, Slamon DJ. . Transformation mediated by the human HER-2 gene independent of the epidermal growth factor receptor. *Oncogene*. 1992;7:1859-66.

93. Muller WJ SE, Pattengale PK, Wallace R, Leder P. Single-step induction of mammary adenocarcinoma in transgenic mice bearing the activated c-neu oncogene. *Cell*. 1988;54:105-15.
94. Guy CT WM, Schaller M, Parsons TJ, Cardiff RD, Muller WJ. Expression of the neu protooncogene in the mammary epithelium of transgenic mice induces metastatic disease. . *Proc NatlAcad Sci USA*. 1992;89:10578-82.
95. Drebin JA LV, Greene MI. Monoclonal antibodies reactive with distinct domains of the neu oncogene-encoded p185 molecule exert synergistic anti-tumor effects in vivo. *Oncogene*. 1988;2:273-7.
96. Drebin JA LV, Greene MI. . Monoclonal antibodies specific for the neu oncogene product directly mediate anti-tumor effects in vivo. *Oncogene*. 1988;2:387-94.
97. Fendly BM WM, Hudziak RM, Lipari MT, Napier MA, Ullrich A. . Characterization of murine monoclonal antibodies reactive to ither the human epidermal growth factor receptor or HER2/neu gene product. e *Cancer Res* 1990;50:1550-8.
98. Gusterson BA GR, Goldhirsch A, Price KN, Save SJ, Anbazhagan R et al. Prognostic importance of c-erbB-2 expression in breast cancer. International (Ludwig) Breast Cancer Study Group. *JClinOncol* 1992;10:1049-56.
99. Press MF BL, Thomas PA, Meisner LF, Zhou JY, Ma Y et al. HER-2/neu gene amplification characterized by fluorescence in situ hybridization: poor prognosis in node-negative breast carcinomas. . *JClinOncol*. 1997;15:2894-904.
100. Revillion F BJ, Peyrat JP. ERBB2 oncogene in human breast cancer and its clinical significance. . *EurJCancer*. 1998;34:791-808.
101. Ross JS FJ. The HER-2/neu oncogene in breast cancer: prognostic factor, predictive factor, and target for therapy. *tem Cells* 1998;16:413-28.
102. Baselga J SA, Rosen PP, Norton L. . HER2 overexpression and paclitaxel sensitivity in breast cancer: therapeutic implications. . *Oncology Huntingt*. 1997;11:43-8.
103. Benz CC SG, Sarup JC, Johnson RM, Tripathy D, Coronado E et al. . Estrogen-dependent, tamoxifen-resistant tumorigenic growth of MCF-7 cells transfected with HER2/neu. . *Breast Cancer Res Treat*. 1993;24:85-95.
104. Carlomagno C PF, Gallo C, De Laurentiis M, Lauria R, Morabito A et al. c-erb B2 overexpression decreases the benefit of adjuvant tamoxifen in early-stage breast cancer without axillary lymph node metastases. . *JClinOncol*. 1996;14:2702-8.
105. Giai M RR, Ponzone R, De Bortoli M, Dati C, Sismondi P. . Prognostic and predictive relevance of c-erbB-2 and ras expression in node positive and negative breast cancer. . *Anticancer Res*. 1994;14:1441-50.
106. Muss HB TA, Berry DA, Kute T, Liu ET, Koerner F et al. c-erbB-2 expression and response to adjuvant therapy in women with node-positive early

breast cancer [published erratum appears in N Engl J Med 1994 Jul 21;331(3):211]. . *NEnglJMed*. 1994;330:1260-6.
107. Newby JC JS, Smith IE, Dowsett M. Expression of epidermal growth factor receptor and c-erbB2 during the development of tamoxifen resistance in human breast cancer. . *ClinCancer Res* 1997;3:1643-51.
108. Paik S BJ, Park C, Fisher B, Tan CE, Hyams D et al. . erbB-2 and response to doxorubicin in patients with axillary lymph node-positive, hormone receptor-negative breast cancer *JNatlCancer Inst*. 1998;90:1361-70.
109. Thor AD BD, Budman DR, Muss HB, Kute T, Henderson IC et al. erbB-2, p53, and efficacy of adjuvant therapy in lymph node-positive breast cancer. *JNatlCancer Inst* 1998;90:1346-60.
110. Yu D LB, Tan M, Li J, Wang SS, Hung MC. Overexpression of c-erbB-2/neu in breast cancer cells confers increased resistance to Taxol via mdr-1-independent mechanisms. *Oncogene*. 1996;13:1359-65.
111. Lewis GD FI, Fendly B, Wong WL, Carter P, Gorman C et al. . Differential responses of human tumor cell lines to anti-p185HER2 monoclonal antibodies. *Cancer ImmunolImmunother*. 1993;37:255-63.
112. Shepard HM LG, Sarup JC, Fendly BM, Maneval D, Mordenti J et al. Monoclonal antibody therapy of human cancer: taking the HER2 protooncogene to the clinic. . *JClinImmunol*. 1991;11:117-27.
113. Carter P PL, Gorman CM, Ridgway JB, Henner D, Wong WL et al. Humanization of an anti-p185HER2 antibody for human cancer therapy. . *Proc NatlAcad Sci USA* 1992;89:4285-9.
114. Cobleigh MA VC, Tripathy D, Robert NJ, Scholl S, Fehrenbacher L et al. . Multinational study of the efficacy and safety of humanized anti-HER2 monoclonal antibody in women who have HER2-overexpressing metastatic breast cancer that has progressed after chemotherapy for metastatic disease. *JClinOncol*. 1999;17:2639-48.
115. Liebermann G, Burchmore, M. J., Ferhenbacher, L., and Cobleigh, M. A. . Health-related quality of life of patients with HER2-overexpressing, metastatic breast cancer treated with Herceptin (tratuzumab) as single agent. . Proc Am Soc Clin Oncol. 1999;18(417).
116. Vogel CL CM, Tripathy D, Gutheil JC, Harris LN, Fehrenbacher L et al. . First-line, single-agent Herceptin (tratuzumab) in metastatic breast cancer: a priliminary report. . *EurJCancer*. 2001;37:25-9.
117. Slamon DJ L-JB, Shak S, Fuchs H, Paton V, Bajamonde A et al. Use of Chemotherapy plus a Monoclonal Antibody against HER2 for Metastatic Breast Cancer That Overexpresses HER2. . *NEnglJMed*. 2001;344:783-92.
118. Mass R, Press, M. F., Anderson, S., and Slamon, D. J. Improved survival benefit from Herceptin (trastuzumab) and chemotherapy in patients selected by fluorescence in situ hybridization. Breast Cancer Res Treat. . 2001;65(1).

119. Fyfe G, Mass, R., and Slamon, D. J. Survival benefit of Herceptin (trastuzumab) and chemotherapy in older (age>60) patients. . Breast Cancer Res Treat. 2001;65(1).
120. Sauer R SK, Hellriegel KP. [German Society of Senology]. Radiation therapy after mastectomy--interdisciplinary consensus puts and end to a controversy. . *StrahlentherOnkol* 2001;177:1-9.
121. Singletary ES AC, Ashley P., Bassett LW, Berry D, Bland KI et al. Revision of the American Joint Committee on Cancer Staging System for Breast Cancer. *JClinOncol*. 2002;20:3628-36.
122. Slapak CA KD, editor. Harrison's Principle of Internal Medicine,. 13 ed ed. New York: McGraw-Hill1994.
123. Jensen BV ST, Nielsen D. . Functional monitoring of anthracycline cardiotoxicity: a prospective, blinded, long-term observational study of outcome in 120 patients. . *Annals of Oncology*. 2002;13:699-709.
124. Nyberg M ND, Skovsgaard T, Hansen J, Jensen BV, Dombernowsky P. Epirubicin cardiotoxicity: an analysis of 469 patients with metastatic breast cancer. JClin Oncol. 1998;16:3502-8.
125. Fuster V GB, Giuliani ER, Tajik AJ, Brandenburg RO, Frye RL. . The natural history of idiopathic dilated cardiomyopathy. . Am JCardiol. 1981;47:525-31.
126. Wilson JR SJ, Sutton MS, Ferraro N, Horowitz LN, Reichek N Prognosis in severe heart failure: relation to hemodynamic measurements and ventricular ectopic activity. *JAm CollCardiol*. 1983;2:403-10.
127. Flather MD YS, Kober L, Pfeffer M, Hall A, Murray G et al.. Long-term ACE-inhibitor therapy in patients with heart failure or left-ventricular dysfunction: a systematic overview of data from individual patients. ACE-Inhibitor Myocardial Infarction Collaborative Group Lancet 2000;355:1575-81.
128. Jensen BV NS. Losartan versus captopril in elderly patients with heart failure. . Lancet 1997;349:1473.
129. Katase K SY, Hasumi K, Yoshimoto M, Kasumi F. :. The incidence of subsequent endometrial carcinoma with tamoxifen use in patients with primary breast carcinoma. Cancer 1998;82:1698-703.
130. Adami HO BR, Weiderpass E, Persson I, Barlow L, McLaughlin JK. . Risk for endometrial cancer following breast cancer: a prospective study in Sweden. Cancer CausesControl. 1997;8:821-7.
131. Campos S. Aromatase inhibitors for breast cancer in postmenopausal women. Oncologist. 2004;9:126-36.
132. Goss PE. Letrozole in the extended adjuvant setting: MA.17. Breast Cancer Res Treat. 2007;105:45–53.
133. Brian Leyland-Jones KPG, Mark Abramovitz, Mark Bouzyk Brandnin Young, Bradley Kong, Roswitha Kamler, Patrizia Dell' Orto, Maria Olivia

Biasi, Beat Thürliman, Vernon Harvey, Patrick Neven, Laurent Arnould, Rudolf Maibach, Karen N. Price, Alan S. Coates, Aron Goldhrisch, Richard D. Gelber, Olivia Pagani, Giuseppe Viale, James M. Rae, Meredith Regan BIG 1-98 Collaborative Group. ESR1 and ESR2 polymorphisms in the BIG 1-98 trial comparing adjuvant letrozole versus tamoxifen or their sequence for early breast cancer. Breast Cancer Res Treat 2015;154:543-55.

134. Alan S. Coates AK, Beat Thu¨rlimann, Henning Mouridsen, Louis Mauriac, John F. Forbes,, Robert Paridaens MC-G, Richard D. Gelber, Marco Colleoni, Istva´n La´ng,, Lucia Del Mastro IS, Jacquie Chirgwin, Jean-Marie Nogaret, Tadeusz Pienkowski, Andrew Wardley,, Erik H. Jakobsen KNP, and Aron Goldhirsch. Five Years of Letrozole Compared With Tamoxifen As Initial
Adjuvant Therapy for Postmenopausal Women With
Endocrine-Responsive Early Breast Cancer: Update of Study
BIG 1-98. JOURNAL OF CLINICAL ONCOLOGY. 2007;25(5):486-73.

135. Kaufmann M JW, Kleeberg U, Eiermann W, Janicke F, Hilfrich J et al. Goserelin, a depot gonadotrophin-releasing hormone agonist in the treatment of premenopausal patients with metastatic breast cancer. German Zoladex Trial Group *J ClinOncol*. 1989;7:1113-9.

136. Taylor CW GS, Dalton WS, Martino S, Rector D, Ingle JN et al. . Multicenter randomized clinical trial of goserelin versus surgical ovariectomy in premenopausal patients with receptor-positive metastatic breast cancer: an intergroup study. . *J ClinOncol*. 1998;16:994-9.

137. De Luca A PS, Cassamassimi A, D' Antonio A, Gridelli C, Rossi A et al. . Detection of citculating tumor cells in carcinoma patients by a novel epidermal growth factor receptor reverse transcription-PCR assay. Clin Cancer Res 2000 6:1439-44.

138. Hui YF RJ. Gemcitabine: a cytidine analogue active against solid tumors. *AmJHealth SystPharm*. 1997;54:162-70.

139. Plunkett W HP, Xu YZ, Heinemann V, Grunewald R, Gandhi V. . Gemcitabine: metabolism, mechanisms of action, and self-potentiation. *SeminOncol*. 1995;22:3-10.

140. Noble S GKG. A review of its pharmacology and clinical potential in non-small cell lung cancer and pancreatic cancer. . *Drugs* 1997;54:447-72.

141. Tonato M MA, Martin C. . Safety profile of gemcitabine. *Anticancer Drugs*. 1995;6 Suppl 6:27-32.

142. Ozer H AJ, Bennett CL, Crawford J., Demetri GD, Pizzo PA et al. . Update of Recommendations for the Use of Hematopoietic Colony-Stimulating Factors: Evidence-Based, Clinical Practice Guidelines. *J ClinOncol* 2000. 2000;18:3558-85.

143. O'Brien PCaF, T. R. A multiple testing procedure for clinical trials. Biometrics. 1979(35):349-556.

144. Green S WG. Southwest Oncology Group standard response criteria, endpoint definitions and toxicity criteria. *Invest New Drugs*. 1992;10:239-53.
145. Genz Th SG. Das Rezidiv beim Mammakarzinom. *Krebsmedizin* 1990;11:15-8.
146. O'Shaughnessy JA WR, Burke G, Friedman MA, Johnson JR, Niederhuber JE et al. . Commentary concerning demonstration of safety and efficacy of investigational anticancer agents in clinical trials. *JClinOncol*. 1991(9):2225-32.
147. know. WRHwkwwtw. *JClinOncol*. 1986;4:827-9.
148. DeMets DL LK. Interim analysis: the alpha spending function approach. *StatMed*. 1994;13:1341-52.
149. O'Brien PC FT. A multiple testing procedure for clinical trials. *Biometrics* 1979;35:549-56.