Name of sponsor/company: AIO-Studien-gGmbH Kuno-Fischer-Str. 8 14057 Berlin Germany		
Name of finished product: Binimetinib Encorafenib Cetuximab		
Name of active ingredient: Binimetinib Encorafenib Cetuximab		

Title of study:
Neoadjuvant encorafenib, binimetinib and cetuximab for patients with BRAF V600E mutated/pMMR localized colorectal cancer (NeoBRAF)

Neoadjuvante Behandlung mit Encorafenib, Binimetinib und Cetuximab bei Patienten mit BRAFV600E mutiertem/pMMR lokalisiertem kolorektalem Karzinom - AIO Phase II Studie (NeoBRAF)

Studied period (years):	approx. 4.5 years	**Phase of development:**
Date of first enrolment:	03-Apr-2023	II
Date of last completed:	09-Feb-2024	

Objectives:

Primary objective:

The primary clinical objective was to determine the clinical efficacy of the targeted triplet combination of encorafenib, binimetinib and cetuximab in BRAF V600E mutant/pMMR localized colorectal cancer patients in terms of tumor regression (TRG2-4).

Secondary objectives:

The main secondary objective was to determine safety, tolerability, and feasibility of the regimen in the neoadjuvant setting. Further secondary objectives were to determine the efficacy of the experimental regimen in terms of overall response rate and disease-free survival. Translational data will inform about molecular mechanisms of response/resistance to triplet combination and the potential utility of liquid biopsy monitoring during treatment.

Methodology:
Open label, single arm, multicenter phase II trial

Number of patients (planned and analyzed):

Planned: 48 Analyzed: 2

Diagnosis and main criteria for inclusion:

Trial indication:

Patients with unresected BRAF V600E mutated/pMMR localized colorectal cancer (CRC).

Inclusion criteria:

1. Biopsy-confirmed adenocarcinoma of the colon or upper rectum if too high for radiotherapy.
2. Radiologically (CT/MRI) staged disease as: T3-4 (as invasion of surrounding tissue structures or organs) and/or nodal positive (N+ defined as regional lymph node(s) without fat hilus and short axis diameter of ≥1 cm), M0.
3. BRAF V600E mutation and pMMR or MSS (as determined by a validated test, preferably PCR or NGS).
4. ECOG performance status ≤ 1.
5. Age ≥ 18 years.
6. Adequate hematologic function at screening as follows: ANC ≥ 1.5 x 109/L, platelets ≥ 100 x109/L, hemoglobin ≥ 9.0 g/dL.
7. Adequate liver function at screening as measured by serum transaminases (AST & ALT) ≤ 2.5 x ULN and total bilirubin ≤ 1.5 x ULN. Patients with known Gilbert disease who have serum bilirubin level ≤ 3 × ULN may be enrolled.
8. Adequate renal function at screening: serum creatinine □ 1.5 x ULN.
9. Adequate serum electrolytes at screening defined as serum potassium and magnesium levels within institu¬ti¬onal normal limits (Note: replacement treatment to achieve adequate electrolytes will be allowed.
10. Adequate cardiac function at screening characterized by left ventricular ejection fraction (LVEF) ≥ 50% as determined by ECHO and QT interval corrected for heart rate using Fridericia's formula (QTcF) value ≤ 480 msec.
11. Negative serum pregnancy test at screening for women of childbearing potential.
12. Highly effective contraception for both male and female subjects if the risk of conception exists. (Note: The effects of the trial drugs on the developing human fetus are unknown; thus, women of childbearing potential and men able to father a child must agree to use highly effective contraception, defined as methods with a failure rate of less than 1 % per year, containing at least 1 form of non-hormonal contraception. Highly effective contraception is required at least 28 days prior, throughout and for at least 6 months after interventional study treatment (encorafenib, binimetinib and cetuximab).
13. Signed and dated written informed consent.
14. Ability to take oral medication.
15. Ability to comply with the protocol for the duration of the study, including hospital/office visits for treatment and scheduled follow-up visits and examinations.

Exclusion criteria:

1. Any prior systemic therapy, surgery or radiotherapy of the colorectal cancer disease.
2. History or current evidence of retinal vein occlusion (RVO) or current risk factors for RVO (e.g., uncontrolled glaucoma or ocular hypertension, history of hyperviscosity or hypercoagulability syndromes).
3. Malignancies other than disease under study within 5 years prior to inclusion, with the exception of those with a negligible risk of metastasis or death (e.g., expected 5-year OS > 90%) treated with expected curative outcome (such as adequately treated carcinoma in situ of the cervix, localized prostate cancer treated surgically with curative intent, ductal carcinoma in situ treated surgically with curative intent).
4. Known severe hypersensitivity reactions to monoclonal antibodies or BRAF-/MEK-inhibitors (grade ≥ 3 NCI-CTCAE v 5), any history of anaphylaxis, or uncontrolled asthma (that is, 3 or more features of partially controlled asthma).
5. Pregnancy or lactation.
6. Known alcohol or drug abuse.
7. Clinically significant (i.e., active) cardiovascular disease: cerebral vascular

accident/stroke
(≤ 6 months prior to enrolment); myocardial infarction (≤ 6 months prior to enrolment), acute coronary syndromes [including unstable angina, coronary artery bypass graft (CABG), coronary angioplasty or stenting) ≤ 6 months prior to enrolment]; congestive heart failure (≥ New York Heart Association Classification Class II); or history or current evidence of clinically significant arrhythmia and/or conduction abnormality (≤ 6 months prior to enrolment), except rate controlled atrial fibrillation and paroxysmal supraventricular tachycardia.

8. Uncontrolled hypertension defined as persistent elevation of systolic blood pressure ≥ 150 mmHg or diastolic blood pressure ≥ 100 mmHg despite current therapy.
9. Preexisting interstitial lung disease.
10. Impaired GI function or disease that may significantly alter the absorption of encorafenib or binimetinib (e.g., ulcerative diseases, uncontrolled vomiting, malabsorption syndrome, small bowel resection with decreased intestinal absorption).
11. History of thromboembolic or cerebrovascular events ≤ 6 months prior to enrolment, including transient ischemic attacks, cerebrovascular accidents, deep vein thrombosis or pulmonary emboli.
12. Concurrent neuromuscular disorder that is associated with the potential of elevated CK (e.g., inflammatory myopathies, muscular dystrophy, amyotrophic lateral sclerosis, spinal muscular atrophy).
13. Known human immunodeficiency virus (HIV) infection or active hepatitis B or C infection.
14. All other significant diseases, which, in the opinion of the Investigator, might impair the subject's tolerance of trial treatment.
15. Any psychiatric condition that would prohibit the understanding or rendering of informed consent.
16. Any approved anticancer therapy, including chemotherapy, hormonal therapy or radiotherapy, within 5 half-lives or 4 weeks (the longer period applies) prior to initiation of study treatment.
17. Current treatment with a non-topical medication or current intake of herbal preparations / supplements / foods known to be a strong inhibitor of CYP3A4. However, patients who either discontinue such treatment/intake or switch to another medication at least 7 days prior to starting study treatment are eligible.
18. Concomitant use of St. John's Wort (hypericum perforatum).

Test product, dose and mode of administration, batch number:

<u>Test product and mode of administration:</u>

Premedication:

For prevention of allergic reaction concerning cetuximab an IV antihistaminic (e.g. clemastine 2 mg or diphenhydramine 50 mg) should be administered before all cetuximab infusions. In the first cycle a corticosteroid (e.g. dexamethasone 8 mg) should be administered before cetuximab as well. The following cycles cetuximab may be administered without steroids. For delayed nausea and vomiting metoclopramide or dimenhydrinate may be used at the discretion of the Investigator.

Binimetinib:

45 mg twice daily (BID) per os. Patients should be instructed to take binimetinib twice daily 12 +/- 2 hours apart with a large glass of water (250 ml) in the morning and the evening at appro¬ximately the same time every day. Doses of binimetinib that are omitted for AEs or any other reason should not be made up later in the day, or at the end of the dosing period.

Encorafenib:

300 mg once daily (QD) per os. Patients should be instructed to take encorafenib daily with a large glass

of water (250 ml) in the morning at approximately the same time every day. Doses of encorafenib that are omitted for AEs or any other reason can be taken up to 12 hours prior to the next dose.

Cetuximab:
at a dose of 500 mg/m^2 IV over 120 min (every two weeks). The infusion rate should not exceed 5 mg/min for the first infusion and 10mg/min for subsequent infusions. Close monitoring is required during the infusion and for at least 1 hour after the end of the infusion.

Batch numbers:
Encorafenib (42) / Braftovi®: G02034 (exp. 06-2024)
Encorafenib (168) / Braftovi®: G02035 (exp. 06-2024)
Bimetinib (84) / Mektovi®: G01017 (exp. 04-2024)
Bimetinib (168) / Mektovi®: G99003 (exp. 10-2024)
Cetuximab 20 mL / Erbitux®: 4636902-01 (exp. 09-2023), 4636902-02 (exp. 10-2025)

Duration of treatment:

Treatment with binimetinib, encorafenib and cetuximab were administered for a maximum of 8 weeks preoperatively and for up to 16 weeks postoperatively in case of adequate pathohistological response (TRG>1). Treatment should be stopped in case of clinical signs of relapse / progression (to be confirmed by imaging), intolerable toxicity or withdrawal of consent.

Reference therapy, dose and mode of administration, batch number:
Not applicable. This was a single arm study.

Criteria for evaluation:

Efficacy:
Primary endpoint:
The primary endpoint in this study was the Tumor Regression Grade (TRG).

Secondary endpoints:
- safety and tolerability (acc. to NCI CTC AE v5.0) incl. vital signs, clinical parameters and overall feasibility of the regimen
- Perioperative morbidity and mortality
- R0-resection rate
- Overall response rate (according to RECIST v1.1)
- Disease free survival
- Overall survival
- Correlation of quantitative BRAF V600E levels (measured by ddPCR) with TRG
- Evaluation of mechanism of relative resistance in patients with less response (evaluated by tumor and liquid biopsy NGS profiling at baseline and after treatment)
- Comparison of ctDNA clearance and TRG with a BRAF mutant/pMMR cohort from the planned neoadjuvant PROTECTOR study receiving neoadjuvant chemotherapy

Safety:
Safety assessments included physical examinations including visual and skin assessment as well as vital signs (blood pressure, heart rate, respiratory rate), performance status (ECOG), clinical laboratory profile, 12-lead ECG and ECHO, ophthalmologic assessment, concomitant medication, therapies, procedures and adverse events.

Statistical methods:

The present trial was terminated prematurely with only two patients enrolled. No statistical methods were used. Results are only reported descriptively.

Primary endpoint:
The primary endpoint was calculated as the number with TRG of at least 2 divided by the total number of patients in the ITT population, defined as all patients having received at least one dose of protocol treatment.

Summary – Conclusions:

The AIO-KRK-0420 NeoBRAF trial was terminated early on 09-Feb-2024. The reason for the early termination was a lack of eligible patients. Of 48 patients planned, only two patients were enrolled within 18 months. No patients received treatment at the time of early termination.

Efficacy Results:

This trial investigated the clinical efficacy of the targeted triplet combination of encorafenib, binimetinib and cetuximab in BRAF V600E mutant/pMMR localized colorectal cancer patients in terms of tumor regression (TRG2-4). The subjects were a male and a female patient, with the age of 59 and 69 years and had a diagnosis of a BRAF V600E mutated (pMMR/MSS) localized colorectal cancer and an ECOG of 0 and 1. Patient 4901-001 received two cycles of study treatment, withdrew from treatment (due to SAE colitis) and subsequently underwent tumor surgery (R0). Patient 4901-002 received four cycles of neoadjuvant study treatment, underwent tumor surgery (R0) and subsequently received eight cycles of adjuvant therapy. Central review analysis of the resected tumor samples for the primary endpoint resulted in grade 2 according to the grading system developed by Dworak et al (Dworak, Keilholz et al. 1997) for both samples. The result for the secondary efficacy endpoint of best tumor response according to RECIST 1.1 was not evaluable for the first and stable disease for the second patient. The first patient had recurrence 6 months after surgery. The second patient remains disease free.

Safety results:

Each patient was affected by several adverse events (patient 4901-001: three, patient 4901-002: ten). Two out of 13 adverse events were serious. One adverse event (colitis) was assessed as possibly related to study treatment and lead to the withdrawal from study treatment; its maximum severity grade was reported as 4 and the patient recovered/resolved within eight days of occurrence.

Table 1: Cumulative Summary Tabulation of AEs and SAEs Reported in the AIO-KRK-0420 NeoBRAF Trial since Study Start

System Organ Class MedDRA Preferred Term	**Incidence AE**	**Incidence SAE**
Gastrointestinal disorders	**5**	**1**
Colitis	1	1
Diarrhoea	4	0
General disorders and administration site conditions	**1**	**0**
Fatigue	1	0
Infections and infestations	**1**	**0**
Urinary tract infection	1	0
Investigations	**1**	**0**
Alanine aminotransferase increased	1	0
Nervous system disorders	**1**	**1**
Cerebrovascular accident	1	1

Skin and subcutaneous tissue disorders	**4**	**0**
Erythema	1	0
Pruritus	1	0
Rash	2	0
TOTAL	**13**	**2**

Conclusion:
While the treated patient cohort is small, strongly limiting the significance of the observations, no conclusive efficacy signals were detected. Tumors of both subjects maintained resectability during study treatment and a response in terms of tumor regression (TRG according to Dworak) could be observed. In terms of response rate (RECIST) the patients had progressive and stable disease respectively. Safety results revealed no unexpected findings, but, like the efficacy results, the generated data set is too small for any meaningful conclusion.

Date of report:
03-Mar-2025

Substantial protocol amendments:
Clinical Study Protocol (CSP) Version 3.0 was the first CSP version approved by ethics committee and national health authority (Paul-Ehrlich-Institut). After initial approval, one further amended CSP (Version 4.0) became effective. Changes to the substantially amended CSP Version 4.0 were made to remove inconsistencies between flow chart (figure 1) and the description of assessments during treatment.