Gastro Intestinal Tumors

Gastrointest Tumors 2014;1:180–194
DOI: 10.1159/000380786
Published online: March 27, 2015

2296–3774/15/0014–0180$39.50/0
www.karger.com/gat

**Mini-Review**

# Gastric Cancer: New Drugs – New Strategies

Nadine Schulte Matthias P. Ebert Nicolai Härtel

Department of Medicine II, University Hospital Mannheim, University of Heidelberg, Mannheim, Germany

**Key Words**
c-Met inhibitors · Epidermal growth factor pathway · Gastric cancer · Human epidermal growth factor receptor 2 protein · Molecular targeting therapy · Vascular endothelial growth factors/vascular endothelial growth factor receptor pathway

**Abstract**
***Background:*** Gastric cancer is the second most common cause of cancer-related deaths worldwide. There are large geographic variations in the incidence of these tumors, with 60% occurring in East Asia. For patients with resectable disease, surgery and perioperative treatment can be effective. For patients with advanced gastric cancer, chemotherapy regimens result in a median survival of 9–11 months. In general, the prognosis for advanced disease is poor and 5-year overall survival rates are around 15%. Combination therapies yield better survival rates, albeit with increased toxicity. Therefore, more effective and less toxic treatment regimens are needed. ***Summary:*** The molecular aberrations that characterize the different subgroups of gastric cancer have been used as therapeutic targets. However, the heterogeneity and complexity of gastric cancers is a major challenge for the development of effective targeted therapies. This review examines the main molecular targets in the treatment of gastric cancer, namely the vascular endothelial growth factor (VEGF), human epidermal growth factor receptor 2 (HER2), hepatocyte growth factor (HGF)/c-Met, epidermal growth factor receptor (EGFR) and phosphoinositide 3-kinase (PI3K)/Akt pathways. ***Key Message:*** The molecular aberrations characteristic of gastric cancer are being explored for the development of targeted therapies, including the VEGF, HER2, HGF/c-Met, EGFR and PI3K/Akt signaling pathways. ***Practical Implications:*** Trastuzumab, an antibody which targets HER2, is the first approved targeted therapy for the treatment of gastric cancer. However, trastuzumab is only effective in HER2-positive tumors (about 10–20% of all gastric cancers). Ramucirumab, which targets the VEGF receptor 2, has yielded benefits with respect to overall survival in a phase III trial and is an effective treatment for advanced gastric cancer with approval in second-line treatment. Apatinib and rilotumumab are another two promising new agents currently under development.


Dr. Nadine Schulte
Department of Medicine II, University Hospital Mannheim
Theodor-Kutzer-Ufer 1–3
DE–68167 Mannheim (Germany)
E-Mail nadine.schulte@umm.de

KARGER

Gastrointest Tumors 2014;1:180–194
DOI: 10.1159/000380786




## Introduction

Gastric cancer is the fourth most common cancer worldwide. About 600,000 men and 330,000 women were diagnosed with gastric cancer in the year 2002 worldwide [1]. Furthermore about 700,000 deaths occur due to gastric cancer, making it the second most common cause of cancer death worldwide. There are high geographic differences in prevalence, with 60% of gastric cancers arising in East Asia [2]. Apart from the decreasing incidence of gastric cancer in Western countries, the frequency of adenocarcinomas of the gastroesophageal junction (GEJ) is increasing [2, 3]. Surgery and perioperative treatment are potentially curative for patients with resectable cancer. The prognosis for advanced gastric cancer remains poor, the 5-year overall survival (OS) rates being approximate 15% [4, 5]. Chemotherapy remains the main treatment for patients with advanced disease and targeted therapy might be an option to improve the prognosis [6].

Targeted therapy depends on the presence of targets and is influenced by the heterogeneity of gastric cancer, which can be divided into several subgroups (histological, anatomical, epidemiological and molecular classifications) [7–10]. The overexpression of human epidermal growth factor receptor 2 (HER2) is more prevalent in proximal gastric cancer [11], similar to epidermal growth factor receptor (EGFR) expression (30–60% of proximal tumors) [12]. Also c-Met amplification occurs more frequently in gastroesophageal cancer [13]. Distal non-diffuse tumors are often related to chronic *Helicobacter pylori* infection [9]. This subtype expresses high vascular endothelial growth factor (VEGF) level [14]. Further molecular aberrations, including fibroblastic growth factor receptor 2 (FGFR2) signaling and phosphoinositide 3-kinase-Akt-mammalian target of rapamycin (PI3K/Akt/mTOR) pathway, have been described [15–17]. These multiple molecular alterations can therefore be considered as potential targets for specific biomolecular treatments. Recent data divided gastric carcinoma into five subgroups based on genomic amplifications: FGFR2 (9.3%), KRAS (8.8%), EGFR (7.7%), ERBB2 (7.2%) and c-Met (4%). These subgroups suggest that at least 37% of gastric cancer patients may be treatable by receptor tyrosine kinase/RAS-associated therapies [18].

Monoclonal antibodies as well as tyrosine kinase inhibitors and mTOR inhibitors have been administered to patients with gastric cancer in various clinical trials. However, molecular targeting therapy is actually less effective in gastric cancer compared to other cancers such as colorectal or breast cancer.

The ToGA (Trastuzumab for Gastric Cancer) trial confirmed that in HER2-positive inoperable gastric and GEJ cancers, trastuzumab plus cisplatin and either capecitabine or fluorouracil resulted in improved OS compared with chemotherapy alone [19]. This strategy has been approved as the standard regimen in HER2-positive patients. Ramucirumab was recently approved in gastric cancer based on these data in second-line setting. However, the approval of further targeted agents has been a challenge.

## Anti-VEGF/VEGFR Agents

Angiogenesis is an important aspect of tumorigenesis. Vascular endothelial growth factor A (VEGF-A) plays a central role in angiogenesis [20]. The activity of VEGF-A is mediated by two tyrosine kinase receptors, VEGFR-1 and VEGFR-2. VEGF enhances the permeability of tumor vessels [21], induces serine protease or metalloproteases [22, 23], inhibits apoptosis in endothelial cells [24, 25] and inhibits dendritic cell maturation [26].

### *Bevacizumab*

Bevacizumab is a monoclonal antibody targeting VEGF-A, which has shown activity in several solid tumors (i.e. colorectal cancer, breast cancer, non-small-cell lung cancer and glio-

blastoma). It binds to VEGF, preventing its interaction with VEGFR-1 and VEGFR-2. In patients with gastric cancer, VEGF expression has been linked to tumor aggressiveness [27] and poor prognosis [12].

In a multicenter phase II study, bevacizumab (15 mg/kg on day 1) plus platinum-containing chemotherapy had promising efficacy. The response rate was 65% (95% CI 46–80) and the median OS (mOS) was 12.3 months (95% CI 11.3–17.2) [28]. In a further phase II trial, bevacizumab (7.5 mg/kg) in addition to chemotherapy with docetaxel (70 mg/mq) and oxaliplatin (75 mg/mq) was administered in 38 patients. A disease control rate of 79% was reported, with a progression-free survival (PFS) of 6.6 months and an OS of 11.1 months [29].

Based on these data the AVAGAST study was initiated. 774 patients with previously untreated locally advanced or metastatic gastric cancer/GEJ cancer were included. Patients were treated with capecitabine (1,000 mg/mq twice daily for 14 days every 3 weeks) and cisplatin (80 mg/mq) in combination with either bevacizumab (7.5 mg/kg) or placebo. mOS was 12.1 months with bevacizumab and 10.1 months with placebo (hazard ratio [HR] = 0.87; 95% CI 0.73–1.03; p = 0.1002). Median PFS (mPFS) was 6.7 vs. 5.3 months, respectively (HR = 0.80; 95% CI 0.68–0.93; p = 0.0037) and overall response rate was 46.0 vs. 37.4% (p = 0.0315) [30]. Geographic differences in efficacy were observed in a subgroup analysis of AVAGAST. Patients enrolled in North and Latin America appeared to have a survival benefit with bevacizumab (median 11.5 vs. 6.8 months), whereas patients enrolled in Asia (90% from Japan and Korea) appeared to have no benefit. AVATAR, a study similar in design to AVAGAST, was initiated in Chinese patients with advanced gastric cancer. In total, 202 patients were included. The primary results did not show a difference in OS for bevacizumab compared to placebo (HR = 1.11; 95% CI 0.79–1.56; p = 0.5567). mPFS was also similar in both arms [31]. Further investigation with respect to benefit in subgroups is needed.

*Ramucirumab*

Ramucirumab is a fully humanized IgG1 monoclonal antibody specifically blocking the extracellular domain of VEGFR-2 [32]. A phase I/II trial showed promising clinical anti-tumor effects and tolerability. Four (15%) of 27 patients with measurable disease had partial response, and 11 (30%) of 37 patients had either partial response or stable disease lasting at least 6 months [33].

REGARD, an international randomized double-blind placebo-controlled phase III trial, showed benefit of monotherapy in patients with advanced gastric cancer. 355 patients were included with disease progression during first-line therapy. Patients were randomly assigned with a 2:1 ratio to receive best supportive care plus ramucirumab 8 mg/kg or placebo. mOS was 5.2 months (IQR 2.3–9.9) in patients in the ramucirumab group and 3.8 months (IQR 1.7–7.1) in those with placebo (HR = 0.776; 95% CI 0.603–0.998; p = 0.047). PFS was 2.1 months in the ramucirumab group vs. 1.3 months in the placebo group (HR = 0.483; p < 0.0001). The rates of serious adverse events were similar between arms [34].

The RAINBOW trial is a randomized multicenter double-blind placebo-controlled phase III study testing paclitaxel (80 mg/kg on days 1, 8, 15, every 4 weeks) with or without ramucirumab (8 mg/kg i.v. infusion on days 1 and 15 every 4 weeks) in patients with advanced gastric or GEJ adenocarcinoma after first-line chemotherapy. The study randomized 665 patients. mOS was 9.6 months for the combination vs. 7.4 months for paclitaxel alone (p = 0.0169). mPFS was 4.4 and 2.9 months, respectively (p < 0.0001) [35].

Ramucirumab was currently approved in gastric cancer based on these data in second-line setting.

**Table 1.** Clinical trials with anti-VEGF/VEGFR agents for advanced gastric cancer and GEJ cancer

| Trial | Phase | Setting | Regimen | Patients, n | OS, months | TTP/PFS, months |
|---|---|---|---|---|---|---|
| NCT00084604 | II | 1st | Iri/Cis/Bev | 47 | 12.3 | 8.3 |
| NCT00217581 | II | 1st | Doc/Ox/Bev | 38 | 11.1 | 6.6 |
| AVAGAST | III | 1st | Cis/Cap/Bev vs. Cis/Cap/placebo | 774 | 12.1 vs. 10.1 (p = 0.1002) | 6.7 vs. 5.3 (p = 0.0037) |
| AVATAR | III | 1st | Cis/Cap/Bev vs. Cis/Cap/placebo | 202 | 10.5 vs. 11.4 (p = 0.56) | 6.3 vs. 6.0 (p = 0.47) |
| REGARD | III | 2nd | Ram + BSC vs. BSC | 355 (2:1) | 5.2 vs. 3.8 (p = 0.047) | 2.1 vs. 1.3 (p < 0.0001) |
| RAINBOW | III | 2nd | Pac + Ram vs. Pac + placebo | 665 | 9.6 vs. 7.4 (p = 0.017) | 4.4 vs. 2.9 (p < 0.0001) |
| NCT00411151 | II | 2nd | Sun | 51 (ITT) | 5.8 | 1.3 |
| NCT00226811 | II | 2nd | Sun | 78 | 6.8 | 2.3 |
| NCT01238055 | II | 2nd | Doc + Sun vs. Doc | 107 | – | 3.9 vs. 2.6 (p = 0.206) |
| NCT00970138 | II | 3rd | Apa 850 mg daily vs. Apa 425 mg b.i.d. vs. placebo | 141 | 4.8 vs. 4.3 vs. 2.5 (p < 0.001/p = 0.0017) | 3.7 vs. 3.2 vs. 1.4 (p < 0.001) |

Apa = Apatinib; Bev = bevacizumab; BSC = best supportive care; Cap = capecitabine; Cis = cisplatin; Doc = docetaxel; Iri = irinotecan; ITT = intention to treat; Ox = oxaliplatin; Pac = paclitaxel; Ram = ramucirumab; Sun = sunitinib; TTP = time to progression.

*Sunitinib*

Sunitinib is an oral, multitargeted tyrosine kinase inhibitor of VEGF receptors, platelet-derived growth factor receptors, KIT and several other related receptor tyrosine kinases [36]. Sunitinib has received approval for the treatment of advanced imatinib-resistant/-intolerant gastrointestinal stromal tumors, advanced/metastatic renal cell carcinoma and unresectable or metastatic well-differentiated pancreatic neuroendocrine tumors.

A phase II study investigated the outcome of sunitinib monotherapy in pretreated patients with advanced gastric cancer (table 1). Sunitinib monotherapy appeared to be associated with a limited tumor response [37]. A second phase II study showed slightly better results for monotherapy of sunitinib (table 1) [38].

A randomized phase II trial investigated the effects of docetaxel and sunitinib in second-line treatment after pretreatment with fluoropyridine and platinum. Patients were assigned to either docetaxel monotherapy or a combination of docetaxel and sunitinib. The time to progression was not significantly prolonged in the docetaxel plus sunitinib arm (table 1). Patients in the docetaxel plus sunitinib arm had more frequently stomatitis, diarrhea and hand-foot syndrome [39].

In advanced gastric cancer sunitinib showed poor activity as a single drug as well as in combination in second-line setting.

*Apatinib*

Apatinib is a tyrosine kinase inhibitor selectively targeting VEGFR-2 [40]. A randomized phase II trial investigated apatinib in third-line setting in 144 patients with advanced gastric cancer. Patients were randomly assigned to receive placebo, apatinib 850 mg once daily or apatinib 425 mg twice daily. mOS was 2.5, 4.8 and 4.3 months, respectively. mPFS was 1.4, 3.7 and 3.2 months, respectively. There were statistically significant differences between the

apatinib and placebo groups for both PFS ($p < 0.001$) and OS ($p = 0.0017$). The most common adverse effects were hypertension and hand-foot syndrome. Patients who received apatinib once daily had fewer grade 3–4 adverse events. Therefore, the dosing regimen of 850 mg once daily was recommended for later studies [41].

A randomized phase III trial for third-line therapy is actually comparing apatinib (850 mg/daily) to placebo (NCT01512745). The estimated number of patients is 270. PFS and OS are the primary endpoints and recruitment is ongoing.

## HER2-Targeting Agents

The HER2 protein is a transmembrane tyrosine kinase and is composed of an extracellular ligand-binding domain, a transmembrane region and an intracellular domain with tyrosine kinase activity. The HER2 protein is a member of the EGFR family [42]. The activation of HER2 does not require ligand binding and induces a receptor dimerization that initiates phosphorylation cascades and activation of the PI3K/Akt/mTOR and Ras-Raf-ERK pathways [43].

Recent studies showed the role of HER2 in the development of several types of human cancer, including gastric cancer. HER2 is overexpressed in 10–38% of gastric cancer samples, with a higher prevalence in intestinal-type and proximal tumors than in diffuse-type and distal tumors [11].

Immunohistochemistry (IHC) should be used as the primary test for analyzing HER2 status; patients with scores of 3+ would be candidates for HER2-directed therapy, those with 2+ should be re-tested using fluorescence in situ hybridization (FISH), and FISH-positive patients would be eligible for trastuzumab with chemotherapy [44].

### *Trastuzumab*

Trastuzumab is a humanized monoclonal antibody that binds to the extracellular domain of the HER2 receptor, blocks dimerization, induces antibody-dependent cellular cytotoxicity and increased endocytosis of the receptor [45]. These therapeutic effects are enhanced in combination with chemotherapy (i.e. cisplatin, capecitabine, irinotecan, doxorubicin and taxanes). To pick out one of the several phase II trials, Grávalos et al. [46] included 228 patients under first-line therapy with HER2 overexpression. Treatment consisted of trastuzumab (8 mg/kg on cycle 1 day 1 as loading; 6 mg/kg in subsequent cycles) and cisplatin (75 mg/mq) both i.v. on day 1, every 21 days. Median time to progression was 5.1 months.

The ToGA trial established trastuzumab as the first biological therapy with longer OS in patients with gastric cancer. It was a randomized phase III study of trastuzumab in combination with chemotherapy for patients with HER2-positive advanced gastric cancer and cancer of the GEJ. 594 patients were randomized to receive 5-fluorouracil (5-FU) (800 mg/$m^2$/day on days 1–5 continuous infusion) or capecitabine (1,000 mg/$m^2$/day on days 1–14) and cisplatin (80 mg/$m^2$ on day 1) with trastuzumab (8 mg/kg loading dose on day 1 followed by 6 mg/kg) every 3 weeks for 6 cycles, or chemotherapy alone. Of all patients screened for HER2, 22.1% were HER2-positive. The addition of trastuzumab to chemotherapy showed a significantly longer PFS (6.7 vs. 5.5 months, $p = 0.0002$) and a significantly longer OS (13.8 vs. 11.1 months, $p = 0.0046$). Patients with higher levels of HER2 expression (IHC score of 3+ or 2+ and FISH-positive) had the greatest benefit with an OS reaching 16 months. The safety profiles in both groups were similar [19].

Currently the HELOISE study is recruiting. This randomized open-label multicenter phase IIIb study will compare the efficacy and safety of two different trastuzumab dose regimens in combination with cisplatin/capecitabine. Patients for first-line treatment will be

**Table 2.** Clinical phase III trials with anti-HER2 agents for advanced gastric cancer and GEJ cancer

| Trial | Phase | Setting | Regimen | Patients, n | OS, months | TTP/PFS, months |
|---|---|---|---|---|---|---|
| ToGA | III | 1st | 5-FU or Cap + Cis + Tras vs. 5-FU or Cap + Cis | 594 | 13.8 vs. 11.1 (p = 0.0046) | 6.7 vs. 5.5 (p = 0.0002) |
| LOGiC | III | 1st | Cap/Ox + Lap vs. Cap/Ox + placebo | 545 | 11.9 vs. 10.4 (p = 0.3244) | 6.0 vs. 5.4 |
| TYTAN | III | 2nd | Pac + Lap vs. Pac | 430 | 11.0 vs. 8.9 (p = 0.2088); subgroup: 14.0 vs. 7.6 (p = 0.0176) | 5.4 vs. 4.4 (p = 0.2441); subgroup: 5.6 vs. 4.2 (p = 0.0101) |
| JACOB | III | 1st | Tras + 5-FU/Cis + Per vs. Tras + 5-FU/Cis + placebo | 780 (est.) | ongoing | ongoing |

Cap = Capecitabine; Cis = cisplatin; Lap = lapatinib; Ox = oxaliplatin; Pac = paclitaxel; Per = pertuzumab; Tras = trastuzumab; TTP = time to progression.

randomized to receive trastuzumab either at 8 mg/kg loading dose followed by 6 mg/kg every 3 weeks or at 8 mg/kg loading dose followed by 10 mg/kg every 3 weeks. 400 patients will be enrolled in this trial [unpublished data, 47].

Trastuzumab emtansine (T-DM1) is an antibody-drug conjugate with trastuzumab and the cytotoxic antimicrotubule DM1 (derivative of maytansine). When T-DM1 binds to HER2, the receptors are internalized by endocytosis, with a consequent intracellular release of the active form of DM1, causing cell death. T-DM1 showed high efficiency in preclinical models of HER2-positive cancer [48]. Therefore a phase II/III trial is investigating the efficacy and safety of T-DM1 compared to standard taxane treatment in patients with HER2-positive advanced gastric cancer in second-line therapy. First-line therapy must include a combination of a platinum- and a fluoropyrimidine-based treatment. Patients will be randomized to one of three treatment arms (T-DM1 3.6 mg/kg every 3 weeks, T-DM1 2.4 mg/kg every week or standard taxane therapy with docetaxel or paclitaxel) [unpublished data, 49].

*Lapatinib*

Lapatinib is an oral tyrosine kinase inhibitor for both EGFR and HER2 which could be used in trastuzumab-resistant breast cancer. One of several phase II trials tested lapatinib as a first-line therapy in 47 patients with advanced gastric cancer showing moderate single-agent activity, with a median time to treatment failure of 1.9 months and an OS of 4.8 months. Only 7% of patients showed partial response and 20% stable disease [50].

Two different phase III studies analyzed the efficacy of lapatinib in combination with chemotherapy in patients with HER2-positive gastric cancer. The LOGiC study (Lapatinib Optimization Study in HER2 Positive Gastric Cancer) is a phase III study to evaluate the efficacy and safety of lapatinib in first-line treatment. Patients received chemotherapy (oxaliplatin 130 mg/mq day 1; capecitabine 850 mg/m$^2$ b.i.d. days 1–14, every 3 weeks) plus daily lapatinib (1,250 mg) or placebo. 545 patients were randomized and 487 had HER2 positivity centrally confirmed. The primary endpoint was not met. OS in all randomized patients was 11.9 months in the lapatinib group vs. 10.4 months in the placebo group (p = 0.3244). PFS was 6.0 vs. 5.4 months, respectively [unpublished data, 51].

The TYTAN trial combined lapatinib and paclitaxel in the second-line therapy of gastric cancer. The study included 430 HER2-positive patients (table 2). mOS was 11.0 months in the lapatinib group compared with 8.9 months with paclitaxel alone (p = 0.2088). In a subgroup analysis of the HER2 status, IHC 3+ patients had an OS of 14.0 and 7.6 months, respectively

(p = 0.0176). PFS was 5.6 vs. 4.2 months (p = 0.0101) [52]. Based on these results, lapatinib may be an effective treatment against HER2 (IHC 3+) gastric cancers; however, further investigation is needed.

*Pertuzumab*

Pertuzumab is a humanized anti-HER2 antibody which inhibits the dimerization of HER2 and prevents ligand-dependent HER2 signaling. Because of different mechanisms of HER2 inhibition, the combination of pertuzumab and trastuzumab may be more effective. As has already been shown, the combination of pertuzumab and trastuzumab increases antitumor activity in HER2-positive human gastric cancer xenograft models [53].

For dose finding of pertuzumab the JOSHUA phase II trial was developed, evaluating two different doses in first-line setting of gastric cancer. Patients were randomized to receive either pertuzumab 840 mg for cycle 1 and 420 mg for cycles 2–6 or pertuzumab 840 mg every 3 weeks in combination with trastuzumab and cisplatin and capecitabine or 5-FU. The mean concentration was higher in patients with 840 mg pertuzumab continuously. Based on these data, a pertuzumab dose of 840 mg every 3 weeks was recommended [54].

The ongoing phase III JACOB trial is a double-blind placebo controlled randomized multicenter study to evaluate the efficacy and safety of pertuzumab in gastric cancer. Patients will be randomized to receive either pertuzumab 840 mg or placebo in combination with trastuzumab (initial dose of 8 mg/kg i.v. followed by 6 mg/kg i.v. every 3 weeks) and cisplatin and capecitabine or 5-FU for the first 6 treatment cycles. Patients will continue to receive pertuzumab or placebo and trastuzumab until disease progression or increased toxicity [unpublished data, 55].

## Hepatocyte Growth Factor-c-Met Pathway

The receptor tyrosine kinase c-Met and its ligand, the hepatocyte growth factor (HGF), are involved in the regulation of multiple cellular processes including cell proliferation, invasion and angiogenesis [56]. High c-Met expression is associated with poor prognosis [57]. c-Met amplification was shown in approximately 4–10% of gastric tumors [58] and c-Met protein overexpression by IHC was shown in about 50% of advanced gastric cancers [59]. There are several drugs that inhibit c-Met activity, some of which we would like to highlight below.

*Foretinib*

Foretinib is an oral multikinase inhibitor targeting c-Met. A phase II study evaluated the safety, tolerability and response of two dosing regimens (240 mg/day for 5 days every 2 weeks or 80 mg/day) in 74 patients with gastric cancer. 93% of the patients had received prior therapy. Best response was stable disease in 10 (23%) patients receiving intermittent dosing and 5 (20%) receiving daily dosing. Stable disease duration was 1.9–7.2 months (median 3.2). Treatment-related adverse events occurred in 91% of patients. The rates of hypertension (35 vs. 15%) and elevated aspartate aminotransferase (23 vs. 8%) were higher with intermittent dosing. These results indicate that single-agent foretinib lacked efficacy in unselected patients with metastatic gastric cancer [60].

*Tivantinib*

Tivantinib is a selective small-molecule c-Met inhibitor. In a phase II trial the activity of tivantinib was tested in 30 gastric cancer patients in second- or third-line setting. No objective response was observed. mPFS was 43 days (95% CI 29.0–92.0). Grade 3 or 4 adverse events occurred in 13 patients (43.3%). Tivantinib as a single agent showed modest efficacy in previously treated gastric cancer [61].

**Table 3.** Phase II/III trials of c-Met inhibitors for advanced gastric cancer and GEJ cancer

| Trial | Phase | Setting | Regimen | Patients, n | OS, months | TTP/PFS, months |
|---|---|---|---|---|---|---|
| NCT00725712 | II | 1st, 2nd, 3rd | For | 74 | – | 3.2 |
| NCT01152645 | II | 2nd, 3rd | Tiv | 30 | – | 43 days |
| NCT00719550 | II | 1st | Ril 15 mg/kg + ECX vs. Ril 7.5 mg/kg + ECX vs. placebo + ECX | 121 | – | 5.1 (p = 0.164) vs. 6.8 (p = 0.009) vs. 4.2 |
| NCT01697072 | III | 1st | Ril + ECX vs. placebo + ECX | 600 (est.) | ongoing | ongoing |
| NCT02137343 | III | 1st | Ril + CX vs. placebo + CX | 450 (est.) | ongoing | ongoing |
| NCT01662869 | III | 1st | mFOLFOX6 + Ona vs. mFOLFOX6 + placebo | 800 (est.) | ongoing | ongoing |

CX = Cisplatin, capecitabine; ECX = epirubicin, cisplatin, capecitabine; For = foretinib; mFOLFOX6 = modified regimen with 5-FU, folinic acid and oxaliplatin; Ona = onartuzumab; Ril = rilotumumab; Tiv = tivantinib.

*Crizotinib*

Crizotinib is a potent c-Met inhibitor, targeting ATP binding sites of the c-Met kinase domain. Therefore, it might represent a potential drug for the treatment of gastric cancer with c-Met amplification. Crizotinib was recently approved for treatment of non-small-cell lung cancer. Treatment with crizotinib resulted in induction of apoptosis and inhibition of Akt in gastric cancer cells with c-Met amplification, but not in those without. Crizotinib exhibited a marked antitumor effect in gastric cancer xenografts positive for c-Met amplification, whereas it had little effect on those negative for this genetic change [62]. Clinical studies to evaluate in vivo efficacy should confirm these in vitro findings.

*Rilotumumab*

Rilotumumab is a human IgG2 antibody targeting human HGF/scatter factor that blocks the binding of HGF/scatter factor to its receptor c-Met. It results in inhibition of the c-Met signaling pathways as shown in animal model [63]. In a phase I/II study, patients with unresectable gastric cancer were treated with rilotumumab in first-line setting (initial dose 15 mg/kg i.v. on day 1) plus ECX (epirubicin, cisplatin, capecitabine). 121 patients were randomly assigned (40 to rilotumumab 15 mg/kg, 42 to rilotumumab 7.5 mg/kg, 39 to placebo). mPFS was 5.1 months (95% CI 2.9–7.0) in the rilotumumab 15 mg/kg group, 6.8 months (95% CI 4.5–7.5) in the rilotumumab 7.5 mg/kg group, 5.7 months (95% CI 4.5–7.0) in both rilotumumab groups combined, and 4.2 months (95% CI 2.9–4.9) in the placebo group. The HR for PFS compared with placebo was 0.69 (80% CI 0.49–0.97; p = 0.164) for rilotumumab 15 mg/kg, 0.53 (80% CI 0.38–0.73; p = 0.009) for rilotumumab 7.5 mg/kg, and 0.60 (80% CI 0.45–0.79; p = 0.016) for combined rilotumumab. Rilotumumab plus ECX had no unexpected adverse events and showed greater activity than placebo plus ECX [64]. According to these results, two phase III randomized double-blind placebo-controlled studies (RILOMET-1 and RILOMET-2) are currently active. RILOMET-1 (NCT01697072) is evaluating epirubicin, cisplatin and capecitabine with rilotumumab (15 mg/kg) or placebo for untreated advanced c-Met-positive gastric cancer. RILOMET-2 (NCT02137343) is evaluating cisplatin and capecitabine with rilotumumab or placebo (table 3).

*Onartuzumab*

Onartuzumab is a monovalent, humanized anti-c-Met antibody that targets the extracellular domain of c-Met. A randomized multicenter double-blind placebo-controlled phase III

study evaluating the efficacy and safety of onartuzumab in combination with mFOLFOX6 in patients with metastatic HER2-negative and c-Met positive adenocarcinoma of the stomach or GEJ is now ongoing. Patients are being randomized in a 1:1 ratio to receive onartuzumab or placebo in combination with mFOLFOX6 (NCT01662869).

## Anti-EGFR Therapies

EGFR-HER1 is one of four receptors involved in the pathway of epidermal growth factor-mediated signaling. It is a transmembrane receptor composed of an extracellular binding domain, a transmembrane portion and an intracellular cytoplasmic domain with tyrosine kinase activity [65]. It is activated by epidermal growth factor and several other factors. Ligand binding induces dimerization with consecutive tyrosine kinase autophosphorylation and activation [66]. This leads to several intracellular signal cascades, including the Ras/Raf/mitogen-activated protein kinase (MAPK) or the Akt/mTOR pathway, which regulate cell proliferation and growth, inhibition of apoptosis, tumor-induced angiogenesis as well as invasive and metastatic growth [67].

The expression of EGFR in gastric cancer was examined in a large study including 511 samples via IHC and FISH. As a result it was shown that the samples were positive for IHC (2+ and 3+) in 27.4% of all cases; EGFR overexpression was associated with older age ($p = 0.001$), moderately or poorly differentiated histology ($p = 0.001$) and higher-stage disease ($p = 0.046$). Sixteen cases (3.1%) showed high polysomy and 12 cases (2.3%) had gene amplification by FISH. The correlation between IHC and FISH was statistically significant ($p < 0.001$) [68].

In colorectal cancer, the presence of a KRAS mutation is usually associated with a downstream activation of the Ras/MAPK pathway, leading to cell proliferation, which in turn leads to resistance against anti-EGFR antibodies. The prevalence of KRAS mutations in 712 samples of gastric cancer was seen overall in 30 samples (4.2%) [69]. These results do not indicate that KRAS gene mutation is frequent in gastric cancer, and later studies were not restricted to patients with wild-type RAS.

## Monoclonal Antibodies

*Cetuximab*

Cetuximab is a chimeric (mouse/human) IgG1 antibody which binds to the extracellular EGFR domain, occluding the ligand-binding region. It results in receptor internalization and degradation. Furthermore, cetuximab initiates an immune-mediated antitumor response (antibody-dependent cell-mediated cytotoxicity) [65].

In a small phase II trial conducted by Lordick et al. [70], 52 patients received cetuximab (400 mg/m$^2$ at first infusion followed by weekly infusions of 250 mg/m$^2$) with chemotherapy (oxaliplatin 50 mg/m$^2$, 5-FU 2,000 mg/m$^2$ and folinic acid 200 mg/m$^2$ on days 1, 8, 15 and 22 every 5 weeks). Among 46 patients assessable for response, the median time to progression was 7.6 months and the mOS was 9.5 months. In another phase II study, the efficacy of cetuximab in addition to irinotecan (80 mg/m$^2$) and folinic acid (200 mg/m$^2$) and 5-FU (1,500 mg/m$^2$) was tested. mPFS and OS times were 9.0 and 16.5 months, respectively [71].

According to these promising data, the EXPAND trial was initiated [72]. In this phase III trial, 904 patients with gastric cancer were randomized to receive capecitabine (1,000 mg/m$^2$ twice daily on days 1–15) and cisplatin (80 mg/m$^2$) with or without cetuximab (400 mg/m$^2$ followed by 250 mg/m$^2$ per week) every 3 weeks. mPFS was 4.4 months (95% CI 4.2–5.5) in the cetuximab arm compared to 5.6 months (95% CI 5.1–5.7) in the chemotherapy arm

**Table 4.** Phase II/III trials of anti-EGFR agents for advanced gastric cancer and GEJ cancer

| Trial | Phase | Setting | Regimen | Patients, n | OS, months | TTP/PFS, months |
|---|---|---|---|---|---|---|
| AIO (Lordick) | II | 1st | FUFOX + Cet | 52 | 9.5 | 7.6 |
| NCT01123811 | II | 1st | FOLFIRI + Cet | 49 | 16.5 | 9.0 |
| EXPAND | III | 1st | Cis/Cap + Cet vs. Cis/Cap | 904 | 9.4 vs. 10.7 (p = 0.9547) | 4.4 vs. 5.6 (p = 0.3158) |
| REAL3 | III | 1st | EOC vs. mEOC + Pan | 553 | 11.3 vs. 8.8 | 7.4 vs. 6.0 |
| NCT01813253 | II | 2nd | Nim + Iri vs. Iri | 83 | 250.5 vs. 232.0 days (p = 0.9778) | 73.0 vs. 85.0 days (p = 0.5668) |
| MATRIX | II | 1st | ECX + Mat vs. ECX | 72 | 4.8 vs. 7.1 | 9.4 vs. 12.2 |

Cap = Capecitabine; Cet = cetuximab; Cis = cisplatin; ECX = epirubicin, cisplatin, capecitabine; EOC = epirubicin, oxaliplatin, capecitabine; FOLFIRI = 5-FU, folinic acid, irinotecan; FUFOX = 5-FU, folinic acid, oxaliplatin; Iri = irinotecan; Mat = matuzumab; mEOC = modified EOC; Nim = nimotuzumab; Pan = panitumumab; TTP = time to progression.

(HR = 1.091; 95% CI 0.920–1.292; p = 0.3158). mOS was 9.4 months (95% CI 8.3–10.6) in the cetuximab arm and 10.7 months (95% CI 9.4–11.3) in the chemotherapy arm (HR = 1.004; 95% CI 0.866–1.165; p = 0.9547). These results suggest that cetuximab in addition to standard chemotherapy is not effective in patients with advanced gastric cancer.

*Panitumumab*

Panitumumab is a fully human IgG2 monoclonal antibody targeting the EGFR. The efficacy of its addition to standard treatment in advanced esophagogastric cancer has been tested in a large phase III trial, the REAL3 study [73]. 553 patients were randomized to receive either EOC (epirubicin 50 mg/m$^2$, oxaliplatin 130 mg/m$^2$ and capecitabine 1,250 mg/m$^2$/day) or modified-dose EOC (epirubicin 50 mg/m$^2$, oxaliplatin 100 mg/m$^2$, capecitabine 1,000 mg/m$^2$/day) and panitumumab 9 mg/kg (table 4). The median survival time was 11.3 months in the EOC arm compared to 8.8 months in the modified-dose EOC plus panitumumab arm (HR = 1.37; 95% CI 1.07–1.76; p = 0.013). The mPFS was 7.4 and 6.0 months, respectively (HR = 1.22; 95% CI 0.98–1.52; p = 0.068). According to these results, the addition of a monoclonal antibody targeting EGFR is not effective in advanced gastric cancer.

*Nimotuzumab*

Nimotuzumab (h-R3) is a humanized IgG1 monoclonal antibody targeting human EGFR. Its activity in advanced gastric cancer has been studied in a randomized phase II trial. 83 patients received either nimotuzumab with irinotecan or irinotecan monotherapy in second-line setting. mPFS was 73.0 and 85.0 days, respectively (p = 0.5668), and mOS was 250.5 and 232.0 days, respectively (p = 0.9778). mPFS/mOS in the EGFR 2+/3+ subgroups were 118.5/59.0 days (HR = 0.341; 95% CI 0.080–1.457; p = 0.1293) and 358.5/229.5 days (HR = 0.369; 95% CI 0.110–1.242; p = 0.0944), respectively [74]. These results did not show a clear benefit when nimotuzumab was added to standard chemotherapy, but it might show some activity in EGFR 2+, 3+ patients.

*Matuzumab*

Matuzumab is a humanized IgG1 monoclonal antibody against human EGFR. It was tested in a phase II trial (MATRIX study) in gastric cancer. There was no benefit of adding matuzumab to chemotherapy with epirubicin, cisplatin and capecitabine [75].

## PI3K/Akt/mTOR-Targeted Therapy

The PI3K/Akt pathway is an intracellular signaling pathway transducing signals from cell membrane receptors (i.e. VEGF, HER2, IGF) with an important role in proliferation and apoptosis as well as in protein translation, synthesis and angiogenesis [76]. PI3K/Akt/mTOR activation was described in 30–60% of all tumors, including gastric cancer [77].

*Everolimus*

Everolimus (RAD001) is an inhibitor of the mTOR serine-threonine kinase inhibiting the PI3K/Akt/mTOR pathway. Everolimus showed efficacy in preclinical and phase I/II studies in gastric cancer [78]. The efficacy of the drug has been tested in a further phase II study. 53 patients with gastric cancer received everolimus (10 mg orally daily) in second-line setting. mPFS was 2.7 months (95% CI 1.6–3.0), mOS was 10.1 months (95% CI 6.5–12.1) [79]. Based on these results, a global phase III trial (GRANITE-1) was conducted to compare everolimus versus placebo in 656 patients with advanced gastric cancer with progressive disease after prior treatment with first- or second-line chemotherapy. mOS was 5.4 months with everolimus and 4.3 months with placebo (HR = 0.90; 95% CI 0.75–1.08; p = 0.124). mPFS was 1.7 and 1.4 months in the everolimus and placebo arms, respectively (HR = 0.66; 95% CI 0.56–0.78) [80]. Compared with best supportive care, everolimus did not significantly improve OS for advanced gastric cancer.

The ongoing randomized double-blind multicenter phase III study (RADPAC) evaluates paclitaxel with and without everolimus in patients with gastric carcinoma who have progressed after therapy with a fluoropyrimidine-containing regimens. A total of 480 patients will be enrolled in the study (NCT01248403, ClinicalTrials.gov).

## Fibroblastic Growth Factor Receptor-Targeting Therapy

Alterations in fibroblastic growth factor signaling (FGFR2 amplification) have been reported in gastric cancer in up to 9%, with a higher frequency in diffuse-type gastric cancer [18]. Various inhibitors of FGFR signaling are in development and showed efficacy in gastric cancer cell lines in vitro [81]. The ongoing SHINE study (NCT01457846) is a phase II study to compare the selective FGFR1, FGFR2 and FGFR3 inhibitor AZD4547 to paclitaxel in second-line setting of patients with FGFR polysomy or amplification in gastric cancer. Several further studies are being planned.

## Conclusion

There is growing knowledge of the different molecular alterations in gastric cancer. Treatment will probably move away from the current ‘one size fits all’ cytotoxic chemotherapy regimens to a more individualized approach. The first success to this strategy was the ToGA trial, as trastuzumab is now a standard of therapy in first-line setting of HER2-positive gastric cancer.

For evaluation of new targeted therapies, selection of patients is critical as only small subgroups show benefit. The AVAGAST study did not meet its endpoint of improved OS. However, the subgroup of patients from Pan-America might have shown a benefit whereas patients from Asia do not seem to benefit from addition of bevacizumab. Therefore failure in patient selection might lead to rejection of potentially efficacious drugs. Based on new data for ramucirumab it received approval in the second-line treatment in gastric cancer. Further

promising data exist for apatinib, a tyrosine kinase inhibitor selectively targeting VEGFR-2. Furthermore, there are promising data for c-Met inhibitors such as rilotumumab in phase II studies. Three phase III studies for either rilotumumab or another c-Met inhibitor, onartuzumab, are ongoing to evaluate efficacy in c-Met-positive gastric cancer. In summary, further investigation is needed to improve patient selection, predictive markers and targeted therapy in order to provide new treatment approaches to patients with advanced gastric cancer.

## Acknowledgements

This work was supported by the Zentrum für Geriatrische Onkologie und Biologie in der Metropolregion Rhein-Neckar (ZOBEL).

## References

1 Kamangar F, Dores GM, Anderson WF: Patterns of cancer incidence, mortality, and prevalence across five continents: defining priorities to reduce cancer disparities in different geographic regions of the world. J Clin Oncol 2006;24:2137–2150.
2 Parkin DM, Bray F, Ferlay J, Pisani P: Global cancer statistics, 2002. CA Cancer J Clin 2005;55:74–108.
3 Bosetti C, Bertuccio P, Levi F, Lucchini F, Negri E, La Vecchia C: Cancer mortality in the European Union, 1970–2003, with a joinpoint analysis. Ann Oncol 2008;19:631–640.
4 Ajani JA: Gastroesophageal cancers: progress and problems. J Natl Compr Canc Netw 2008;6:813–814.
5 Goscinski MA, Larsen SG, Warloe T, Stoldt S, Nesland JM, Suo ZH, Giercksky KE: Adenocarcinomas on the rise – does it influence survival from oesophageal cancer? Scand J Surg 2009;98:214–220.
6 Haghighat P, Bekaii-Saab T: An update on biochemotherapy of advanced gastric and gastroesophageal adenocarcinoma. J Natl Compr Canc Netw 2008;6:895–900.
7 Lauren P: The two histological main types of gastric carcinoma: diffuse and so-called intestinal-type carcinoma. An attempt at a histo-clinical classification. Acta Pathol Microbiol Scand 1965;64:31–49.
8 Correa P, Haenszel W, Cuello C, Zavala D, Fontham E, Zarama G, Tannenbaum S, Collazos T, Ruiz B: Gastric precancerous process in a high risk population: cross-sectional studies. Cancer Res 1990;50:4731–4736.
9 Peek RM, Blaser MJ: *Helicobacter pylori* and gastrointestinal tract adenocarcinomas. Nat Rev Cancer 2002;2:28–37.
10 Uemura N, Okamoto S, Yamamoto S, Matsumura N, Yamaguchi S, Yamakido M, Taniyama K, Sasaki N, Schlemper RJ: *Helicobacter pylori* infection and the development of gastric cancer. N Engl J Med 2001;345:784–789.
11 Grabsch H, Sivakumar S, Gray S, Gabbert HE, Müller W: HER2 expression in gastric cancer: rare, heterogeneous and of no prognostic value – conclusions from 924 cases of two independent series. Cell Oncol 2010;32:57–65.
12 Lieto E, Ferraraccio F, Orditura M, Castellano P, Mura AL, Pinto M, Zamboli A, De Vita F, Galizia G: Expression of vascular endothelial growth factor (VEGF) and epidermal growth factor receptor (EGFR) is an independent prognostic indicator of worse outcome in gastric cancer patients. Ann Surg Oncol 2008;15:69–79.
13 Janjigian YY, Tang LH, Coit DG, Kelsen DP, Francone TD, Weiser MR, Jhanwar SC, Shah MA: MET expression and amplification in patients with localized gastric cancer. Cancer Epidemiol Biomarkers Prev 2011;20:1021–1027.
14 Kitadai Y, Sasaki A, Ito M, Tanaka S, Oue N, Yasui W, Aihara M, Imagawa K, Haruma K, Chayama K: *Helicobacter pylori* infection influences expression of genes related to angiogenesis and invasion in human gastric carcinoma cells. Biochem Biophys Res Commun 2003;311:809–814.
15 Hattori Y, Itoh H, Uchino S, Hosokawa K, Ochiai A, Ino Y, Ishii H, Sakamoto H, Yamaguchi N, Yanagihara K, Hirohashi S, Sugimura T, Terada M: Immunohistochemical detection of K-sam protein in stomach cancer. Clin Cancer Res 1996;2:1373–1381.
16 Kobayashi M, Nagata S, Iwasaki T, Yanagihara K, Saitoh I, Karouji Y, Ihara S, Fukui Y: Dedifferentiation of adenocarcinomas by activation of phosphatidylinositol 3-kinase. Proc Natl Acad Sci USA 1999;96:4874–4879.
17 Murayama T, Inokuchi M, Takagi Y, Yamada H, Kojima K, Kumagai J, Kawano T, Sugihara K: Relation between outcomes and localisation of p-mTOR expression in gastric cancer. Br J Cancer 2009;100:782–788.
18 Deng N, Goh LK, Wang H, Das K, Tao J, Tan IB, Zhang S, Lee M, Wu J, Lim KH, Lei Z, Goh G, Lim QY, Tan AL, Sin Poh DY, Riahi S, Bell S, Shi MM, Linnartz R, Zhu F, Yeoh KG, Toh HC, Yong WP, Cheong HC, Rha SY, Boussioutas A, Grabsch H, Rozen S, Tan P: A comprehensive survey of genomic alterations in gastric cancer reveals systematic patterns of molecular exclusivity and co-occurrence among distinct therapeutic targets. Gut 2012;61:673–684.
19 Bang YJ, Van Cutsem E, Feyereislova A, Chung HC, Shen L, Sawaki A, Lordick F, Ohtsu A, Omuro Y, Satoh T, Aprile G, Kulikov E, Hill J, Lehle M, Rüschoff J, Kang YK; ToGA Trial Investigators: Trastuzumab in combination with chemotherapy versus chemotherapy alone for treatment of HER2-positive advanced gastric or gastro-oesophageal junction cancer (ToGA): a phase 3, open-label, randomised controlled trial. Lancet 2010;376:687–697.

20 Ferrara N, Gerber HP, LeCouter J: The biology of VEGF and its receptors. Nat Med 2003;9:669–676.
21 Dvorak HF, Brown LF, Detmar M, Dvorak AM: Vascular permeability factor/vascular endothelial growth factor, microvascular hyperpermeability, and angiogenesis. Am J Pathol 1995;146:1029–1039.
22 Pepper MS, Ferrara N, Orci L, Montesano R: Vascular endothelial growth factor (VEGF) induces plasminogen activators and plasminogen activator inhibitor-1 in microvascular endothelial cells. Biochem Biophys Res Commun 1991;181:902–906.
23 Unemori EN, Ferrara N, Bauer EA, Amento EP: Vascular endothelial growth factor induces interstitial collagenase expression in human endothelial cells. J Cell Physiol 1992;153:557–562.
24 Shaheen RM, Davis DW, Liu W, Zebrowski BK, Wilson MR, Bucana CD, McConkey DJ, McMahon G, Ellis LM: Antiangiogenic therapy targeting the tyrosine kinase receptor for vascular endothelial growth factor receptor inhibits the growth of colon cancer liver metastasis and induces tumor and endothelial cell apoptosis. Cancer Res 1999;59:5412–5416.
25 Gerber HP, McMurtrey A, Kowalski J, Yan M, Keyt BA, Dixit V, Ferrara N: Vascular endothelial growth factor regulates endothelial cell survival through the phosphatidylinositol 3′-kinase/Akt signal transduction pathway. Requirement for Flk-1/KDR activation. J Biol Chem 1998;273:30336–30343.
26 Lissoni P, Malugani F, Bonfanti A, Bucovec R, Secondino S, Brivio F, Ferrari-Bravo A, Ferrante R, Vigoré L, Rovelli F, Mandalà M, Viviani S, Fumagalli L, Gardani GS: Abnormally enhanced blood concentrations of vascular endothelial growth factor (VEGF) in metastatic cancer patients and their relation to circulating dendritic cells, IL-12 and endothelin-1. J Biol Regul Homeost Agents 2001;15:140–144.
27 Kim SE, Shim KN, Jung SA, Yoo K, Lee JH: The clinicopathological significance of tissue levels of hypoxia-inducible factor-1alpha and vascular endothelial growth factor in gastric cancer. Gut Liver 2009;3:88–94.
28 Shah MA, Ramanathan RK, Ilson DH, Levnor A, D'Adamo D, O'Reilly E, Tse A, Trocola R, Schwartz L, Capanu M, Schwartz GK, Kelsen DP: Multicenter phase II study of irinotecan, cisplatin, and bevacizumab in patients with metastatic gastric or gastroesophageal junction adenocarcinoma. J Clin Oncol 2006;24:5201–5206.
29 El-Rayes BF, Zalupski M, Bekai-Saab T, Heilbrun LK, Hammad N, Patel B, Urba S, Shields AF, Vaishampayan U, Dawson S, Almhanna K, Smith D, Philip PA: A phase II study of bevacizumab, oxaliplatin, and docetaxel in locally advanced and metastatic gastric and gastroesophageal junction cancers. Ann Oncol 2010;21:1999–2004.
30 Ohtsu A, Shah MA, Van Cutsem E, Rha SY, Sawaki A, Park SR, Lim HY, Yamada Y, Wu J, Langer B, Starnawski M, Kang YK: Bevacizumab in combination with chemotherapy as first-line therapy in advanced gastric cancer: a randomized, double-blind, placebo-controlled phase III study. J Clin Oncol 2011;29:3968–3976.
31 Shen L, Li J, Xu J, Pan H, Dai G, Qin S, Wang L, Wang J, Yang Z, Shu Y, Xu R, Chen L, Liu Y, Yu S, Bu L, Piao Y: Bevacizumab plus capecitabine and cisplatin in Chinese patients with inoperable locally advanced or metastatic gastric or gastroesophageal junction cancer: randomized, double-blind, phase III study (AVATAR study). Gastric Cancer 2015;18:168–176.
32 Lu D, Shen J, Vil MD, Zhang H, Jimenez X, Bohlen P, Witte L, Zhu Z: Tailoring in vitro selection for a picomolar affinity human antibody directed against vascular endothelial growth factor receptor 2 for enhanced neutralizing activity. J Biol Chem 2003;278:43496–43507.
33 Spratlin JL, Cohen RB, Eadens M, Gore L, Camidge DR, Diab S, Leong S, O'Bryant C, Chow LQ, Serkova NJ, Meropol NJ, Lewis NL, Chiorean EG, Fox F, Youssoufian H, Rowinsky EK, Eckhardt SG: Phase I pharmacologic and biologic study of ramucirumab (IMC-1121B), a fully human immunoglobulin G1 monoclonal antibody targeting the vascular endothelial growth factor receptor-2. J Clin Oncol 2010;28:780–787.
34 Fuchs CS, Tomasek J, Yong CJ, Dumitru F, Passalacqua R, Goswami C, Safran H, dos Santos LV, Aprile G, Ferry DR, Melichar B, Tehfe M, Topuzov E, Zalcberg JR, Chau I, Campbell W, Sivanandan C, Pikiel J, Koshiji M, Hsu Y, Liepa AM, Gao L, Schwartz JD, Tabernero J; REGARD Trial Investigators: Ramucirumab monotherapy for previously treated advanced gastric or gastro-oesophageal junction adenocarcinoma (REGARD): an international, randomised, multicentre, placebo-controlled, phase 3 trial. Lancet 2014;383:31–39.
35 Wilke H, Muro K, Van Cutsem E, Oh SC, Bodoky G, Shimada Y, Hironaka S, Sugimoto N, Lipatov O, Kim TY, Cunningham D, Rougier P, Komatsu Y, Ajani J, Emig M, Carlesi R, Ferry D, Chandrawansa K, Schwartz JD, Ohtsu A; RAINBOW Study Group: Ramucirumab plus paclitaxel versus placebo plus paclitaxel in patients with previously treated advanced gastric or gastro-oesophageal junction adenocarcinoma (RAINBOW): a double-blind, randomised phase 3 trial. Lancet Oncol 2014;15:1224–1235.
36 Mendel DB, Laird AD, Xin X, Louie SG, Christensen JG, Li G, Schreck RE, Abrams TJ, Ngai TJ, Lee LB, Murray LJ, Carver J, Chan E, Moss KG, Haznedar JO, Sukbuntherng J, Blake RA, Sun L, Tang C, Miller T, Shirazian S, McMahon G, Cherrington JM: In vivo antitumor activity of SU11248, a novel tyrosine kinase inhibitor targeting vascular endothelial growth factor and platelet-derived growth factor receptors: determination of a pharmacokinetic/pharmacodynamic relationship. Clin Cancer Res 2003;9:327–337.
37 Moehler M, Mueller A, Hartmann JT, Ebert MP, Al-Batran SE, Reimer P, Weihrauch M, Lordick F, Trarbach T, Biesterfeld S, Kabisch M, Wachtlin D, Galle PR: An open-label, multicentre biomarker-oriented AIO phase II trial of sunitinib for patients with chemo-refractory advanced gastric cancer. Eur J Cancer 2011;47:1511–1520.
38 Bang YJ, Kang YK, Kang WK, Boku N, Chung HC, Chen JS, Doi T, Sun Y, Shen L, Qin S, Ng WT, Tursi JM, Lechuga MJ, Lu DR, Ruiz-Garcia A, Sobrero A: Phase II study of sunitinib as second-line treatment for advanced gastric cancer. Invest New Drugs 2011;29:1449–1458.
39 Yi JH, Lee J, Lee J, Park SH, Park JO, Yim DS, Park YS, Lim HY, Kang WK: Randomised phase II trial of docetaxel and sunitinib in patients with metastatic gastric cancer who were previously treated with fluoropyrimidine and platinum. Br J Cancer 2012;106:1469–1474.

40 Tian S, Quan H, Xie C, Guo H, Lü F, Xu Y, Li J, Lou L: YN968D1 is a novel and selective inhibitor of vascular endothelial growth factor receptor-2 tyrosine kinase with potent activity in vitro and in vivo. Cancer Sci 2011;102:1374–1380.
41 Li J, Qin S, Xu J, Guo W, Xiong J, Bai Y, Sun G, Yang Y, Wang L, Xu N, Cheng Y, Wang Z, Zheng L, Tao M, Zhu X, Ji D, Liu X, Yu H: Apatinib for chemotherapy-refractory advanced metastatic gastric cancer: results from a randomized, placebo-controlled, parallel-arm, phase II trial. J Clin Oncol 2013;31:3219–3225.
42 Lin SY, Makino K, Xia W, Matin A, Wen Y, Kwong KY, Bourguignon L, Hung MC: Nuclear localization of EGF receptor and its potential new role as a transcription factor. Nat Cell Biol 2001;3:802–808.
43 De Vita F, Di Martino N, Fabozzi A, Laterza MM, Ventriglia J, Savastano B, Petrillo A, Gambardella V, Sforza V, Marano L, Auricchio A, Galizia G, Ciardiello F, Orditura M: Clinical management of advanced gastric cancer: the role of new molecular drugs. World J Gastroenterol 2014;20:14537–14558.
44 Albarello L, Pecciarini L, Doglioni C: HER2 testing in gastric cancer. Adv Anat Pathol 2011;18:53–59.
45 Sliwkowski MX, Lofgren JA, Lewis GD, Hotaling TE, Fendly BM, Fox JA: Nonclinical studies addressing the mechanism of action of trastuzumab (Herceptin). Semin Oncol 1999;26:60–70.
46 Grávalos C, Gómez-Martín C, Rivera F, Alés I, Queralt B, Márquez A, Jiménez U, Alonso V, García-Carbonero R, Sastre J, Colomer R, Cortés-Funes H, Jimeno A: Phase II study of trastuzumab and cisplatin as first-line therapy in patients with HER2-positive advanced gastric or gastroesophageal junction cancer. Clin Transl Oncol 2011;13:179–184.
47 ClinicalTrials.gov: HELOISE study: a study of Herceptin (trastuzumab) in combination with cisplatin/capecitabine chemotherapy in patients with HER2-positive metastatic gastric or gastro-esophageal junction cancer. http://clinicaltrials.gov/show/NCT01450696.
48 Lewis Phillips GD, Li G, Dugger DL, Crocker LM, Parsons KL, Mai E, Blättler WA, Lambert JM, Chari RV, Lutz RJ, Wong WL, Jacobson FS, Koeppen H, Schwall RH, Kenkare-Mitra SR, Spencer SD, Sliwkowski MX: Targeting HER2-positive breast cancer with trastuzumab-DM1, an antibody-cytotoxic drug conjugate. Cancer Res 2008;68:9280–9290.
49 ClinicalTrials.gov: A study of trastuzumab emtansine versus taxane in patients with advanced gastric cancer. http://clinicaltrials.gov/ct2/show/NCT01641939.
50 Iqbal S, Goldman B, Fenoglio-Preiser CM, Lenz HJ, Zhang W, Danenberg KD, Shibata SI, Blanke CD: Southwest Oncology Group study S0413: a phase II trial of lapatinib (GW572016) as first-line therapy in patients with advanced or metastatic gastric cancer. Ann Oncol 2011;22:2610–2615.
51 ClinicalTrials.gov: LOGiC – lapatinib optimization study in ErbB2 (HER2) positive gastric cancer: a phase III global, blinded study designed to evaluate clinical endpoints and safety of chemotherapy plus lapatinib. http://clinicaltrials.gov/ct2/show/results/NCT00680901.
52 Satoh T, Xu RH, Chung HC, Sun GP, Doi T, Xu JM, Tsuji A, Omuro Y, Li J, Wang JW, Miwa H, Qin SK, Chung IJ, Yeh KH, Feng JF, Mukaiyama A, Kobayashi M, Ohtsu A, Bang YJ: Lapatinib plus paclitaxel versus paclitaxel alone in the second-line treatment of HER2-amplified advanced gastric cancer in Asian populations: TyTAN – a randomized, phase III study. J Clin Oncol 2014;32:2039–2049.
53 Yamashita-Kashima Y, Iijima S, Yorozu K, Furugaki K, Kurasawa M, Ohta M, Fujimoto-Ouchi K: Pertuzumab in combination with trastuzumab shows significantly enhanced antitumor activity in HER2-positive human gastric cancer xenograft models. Clin Cancer Res 2011;17:5060–5070.
54 Kang YK, Rha SY, Tassone P, Barriuso J, Yu R, Szado T, Garg A, Bang Y: A phase IIa dose-finding and safety study of first-line pertuzumab in combination with trastuzumab, capecitabine and cisplatin in patients with HER2-positive advanced gastric cancer. Br J Cancer 2014;111:660–666.
55 ClinicalTrials.gov: A study of Perjeta (pertuzumab) in combination with Herceptin (trastuzumab) and chemotherapy in patients with HER2-positive metastatic gastroesophageal junction or gastric cancer. http://clinicaltrials.gov/show/NCT01774786.
56 Appleman LJ: MET signaling pathway: a rational target for cancer therapy. J Clin Oncol 2011;29:4837–4838.
57 Graziano F, Galluccio N, Lorenzini P, Ruzzo A, Canestrari E, D'Emidio S, Catalano V, Sisti V, Ligorio C, Andreoni F, Rulli E, Di Oto E, Fiorentini G, Zingaretti C, De Nictolis M, Cappuzzo F, Magnani M: Genetic activation of the MET pathway and prognosis of patients with high-risk, radically resected gastric cancer. J Clin Oncol 2011;29:4789–4795.
58 Hara T, Ooi A, Kobayashi M, Mai M, Yanagihara K, Nakanishi I: Amplification of c-myc, K-sam, and c-met in gastric cancers: detection by fluorescence in situ hybridization. Lab Invest 1998;78:1143–1153.
59 Amemiya H, Kono K, Itakura J, Tang RF, Takahashi A, An FQ, Kamei S, Iizuka H, Fujii H, Matsumoto Y: c-Met expression in gastric cancer with liver metastasis. Oncology 2002;63:286–296.
60 Shah MA, Wainberg ZA, Catenacci DV, Hochster HS, Ford J, Kunz P, Lee FC, Kallender H, Cecchi F, Rabe DC, Keer H, Martin AM, Liu Y, Gagnon R, Bonate P, Liu L, Gilmer T, Bottaro DP: Phase II study evaluating 2 dosing schedules of oral foretinib (GSK1363089), cMET/VEGFR2 inhibitor, in patients with metastatic gastric cancer. PLoS One 2013;8:e54014.
61 Kang YK, Muro K, Ryu MH, Yasui H, Nishina T, Ryoo BY, Kamiya Y, Akinaga S, Boku N: A phase II trial of a selective c-Met inhibitor tivantinib (ARQ 197) monotherapy as a second- or third-line therapy in the patients with metastatic gastric cancer. Invest New Drugs 2014;32:355–361.
62 Okamoto W, Okamoto I, Arao T, Kuwata K, Hatashita E, Yamaguchi H, Sakai K, Yanagihara K, Nishio K, Nakagawa K: Antitumor action of the MET tyrosine kinase inhibitor crizotinib (PF-02341066) in gastric cancer positive for MET amplification. Mol Cancer Ther 2012;11:1557–1564.

63 Cao B, Su Y, Oskarsson M, Zhao P, Kort EJ, Fisher RJ, Wang LM, Vande Woude GF: Neutralizing monoclonal antibodies to hepatocyte growth factor/scatter factor (HGF/SF) display antitumor activity in animal models. Proc Natl Acad Sci USA 2001;98:7443–7448.
64 Iveson T, Donehower RC, Davidenko I, Tjulandin S, Deptala A, Harrison M, Nirni S, Lakshmaiah K, Thomas A, Jiang Y, Zhu M, Tang R, Anderson A, Dubey S, Oliner KS, Loh E: Rilotumumab in combination with epirubicin, cisplatin, and capecitabine as first-line treatment for gastric or oesophagogastric junction adenocarcinoma: an open-label, dose de-escalation phase 1b study and a double-blind, randomised phase 2 study. Lancet Oncol 2014;15:1007–1018.
65 Ciardiello F, Tortora G: EGFR antagonists in cancer treatment. N Engl J Med 2008;358:1160–1174.
66 Hynes NE, Lane HA: ERBB receptors and cancer: the complexity of targeted inhibitors. Nat Rev Cancer 2005; 5:341–354.
67 Oda K, Matsuoka Y, Funahashi A, Kitano H: A comprehensive pathway map of epidermal growth factor receptor signaling. Mol Syst Biol 2005;1:2005.0010.
68 Kim MA, Lee HS, Lee HE, Jeon YK, Yang HK, Kim WH: EGFR in gastric carcinomas: prognostic significance of protein overexpression and high gene copy number. Histopathology 2008;52:738–746.
69 van Grieken NC, Aoyama T, Chambers PA, Bottomley D, Ward LC, Inam I, Buffart TE, Das K, Lim T, Pang B, Zhang SL, Tan IB, Carvalho B, Heideman DA, Miyagi Y, Kameda Y, Arai T, Meijer GA, Tsuburaya A, Tan P, Yoshikawa T, Grabsch HI: KRAS and BRAF mutations are rare and related to DNA mismatch repair deficiency in gastric cancer from the East and the West: results from a large international multicentre study. Br J Cancer 2013;108: 1495–1501.
70 Lordick F, Luber B, Lorenzen S, Hegewisch-Becker S, Folprecht G, Wöll E, Decker T, Endlicher E, Röthling N, Schuster T, Keller G, Fend F, Peschel C: Cetuximab plus oxaliplatin/leucovorin/5-fluorouracil in first-line metastatic gastric cancer: a phase II study of the Arbeitsgemeinschaft Internistische Onkologie (AIO). Br J Cancer 2010;102:500–505.
71 Moehler M, Mueller A, Trarbach T, Lordick F, Seufferlein T, Kubicka S, Geissler M, Schwarz S, Galle PR, Kanzler S; German Arbeitsgemeinschaft Internistische Onkologie: Cetuximab with irinotecan, folinic acid and 5-fluorouracil as first-line treatment in advanced gastroesophageal cancer: a prospective multi-center biomarker-oriented phase II study. Ann Oncol 2011;22:1358–1366.
72 Lordick F, Kang YK, Chung HC, Salman P, Oh SC, Bodoky G, Kurteva G, Volovat C, Moiseyenko VM, Gorbunova V, Park JO, Sawaki A, Celik I, Götte H, Melezínková H, Moehler M; Arbeitsgemeinschaft Internistische Onkologie and EXPAND Investigators: Capecitabine and cisplatin with or without cetuximab for patients with previously untreated advanced gastric cancer (EXPAND): a randomised, open-label phase 3 trial. Lancet Oncol 2013;14: 490–499.
73 Waddell T, Chau I, Cunningham D, Gonzalez D, Okines AF, Okines C, Wotherspoon A, Saffery C, Middleton G, Wadsley J, Ferry D, Mansoor W, Crosby T, Coxon F, Smith D, Waters J, Iveson T, Falk S, Slater S, Peckitt C, Barbachano Y: Epirubicin, oxaliplatin, and capecitabine with or without panitumumab for patients with previously untreated advanced oesophagogastric cancer (REAL3): a randomised, open-label phase 3 trial. Lancet Oncol 2013;14:481–489.
74 Satoh T, Lee KH, Rha SY, Sasaki Y, Park SH, Komatsu Y, Yasui H, Kim TY, Yamaguchi K, Fuse N, Yamada Y, Ura T, Kim SY, Munakata M, Saitoh S, Nishio K, Morita S, Yamamoto E, Zhang Q, Kim JM, Kim YH, Sakata Y: Randomized phase II trial of nimotuzumab plus irinotecan versus irinotecan alone as second-line therapy for patients with advanced gastric cancer. Gastric Cancer 2014, Epub ahead of print.
75 Rao S, Starling N, Cunningham D, Sumpter K, Gilligan D, Ruhstaller T, Valladares-Ayerbes M, Wilke H, Archer C, Kurek R, Beadman C, Oates J: Matuzumab plus epirubicin, cisplatin and capecitabine (ECX) compared with epirubicin, cisplatin and capecitabine alone as first-line treatment in patients with advanced oesophagogastric cancer: a randomised, multicentre open-label phase II study. Ann Oncol 2010;21:2213–2219.
76 Shitara K, Doi T, Ohtsu A: Molecular targeting therapy and biomarker for advanced gastric cancer. J Phys Chem Biophys 2012;2:107.
77 Markman B, Atzori F, Pérez-García J, Tabernero J, Baselga J: Status of PI3K inhibition and biomarker development in cancer therapeutics. Ann Oncol 2010;21:683–691.
78 Cidon EU, Ellis SG, Inam Y, Adeleke S, Zarif S, Geldart T: Molecular targeted agents for gastric cancer: a step forward towards personalized therapy. Cancers (Basel) 2013;5:64–91.
79 Doi T, Muro K, Boku N, Yamada Y, Nishina T, Takiuchi H, Komatsu Y, Hamamoto Y, Ohno N, Fujita Y, Robson M, Ohtsu A: Multicenter phase II study of everolimus in patients with previously treated metastatic gastric cancer. J Clin Oncol 2010;28:1904–1910.
80 Ohtsu A, Ajani JA, Bai YX, Bang YJ, Chung HC, Pan HM, Sahmoud T, Shen L, Yeh KH, Chin K, Muro K, Kim YH, Ferry D, Tebbutt NC, Al-Batran SE, Smith H, Costantini C, Rizvi S, Lebwohl D, Van Cutsem E: Everolimus for previously treated advanced gastric cancer: results of the randomized, double-blind, phase III GRANITE-1 study. J Clin Oncol 2013;31:3935–3943.
81 Gozgit JM, Wong MJ, Moran L, Wardwell S, Mohemmad QK, Narasimhan NI, Shakespeare WC, Wang F, Clackson T, Rivera VM: Ponatinib (AP24534), a multitargeted pan-FGFR inhibitor with activity in multiple FGFR-amplified or mutated cancer models. Mol Cancer Ther 2012;11:690–699.