

ADF Perspectives

Antiangiogenic cancer therapies get their act together: current developments and future prospects of growth factor- and growth factor receptor-targeted approaches

Jens Gille. Antiangiogenic cancer therapies get their act together: current developments and future prospects of growth factor- and growth factor receptor-targeted approaches.

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Abstract: Targeting the vascular endothelial growth factor (VEGF) in combination with standard chemotherapy has recently proved successful in the treatment of different types of advanced cancer. The achievements of combinatorial anti-VEGF monoclonal antibody bevacizumab (BEV) renewed the confidence in targeted antiangiogenic approaches to constitute a complementary therapeutic modality in addition to surgery, radiotherapy and chemotherapy. While several second-generation multitargeted tyrosine kinase inhibitors show promise in defined tumor entities, these novel antiangiogenic compounds have yet to meet or exceed the efficacy of combinatorial BEV therapy in ongoing clinical trials. Current developments of targeted antiangiogenic agents include their use in the adjuvant setting and the combination of different antiangiogenesis inhibitors to take a more comprehensive approach in blocking tumor angiogenesis. The identification of surrogate markers that can monitor the activity and efficacy of antiangiogenic drugs in patients belongs to the most critical challenges to exploit the full potential of antiangiogenic therapies. The opportunities and obstacles in further development of growth factor- and growth factor receptor-targeted antiangiogenic approaches for advanced cancer, including malignant melanoma, will be discussed herein with particular reference to selected ongoing clinical trials.

Jens Gille

Department of Dermatology, Dermato-Oncology Unit, Johann Wolfgang Goethe-University, Frankfurt am Main, Germany

Key words: angiogenesis inhibitors – anticancer drug combinations – clinical trials – investigational drugs – malignant neoplasms

Jens Gille, MD
Zentrum der Dermatologie und Venerologie
Klinikum der Johann Wolfgang Goethe-Universität
Theodor-Stern-Kai 7
D-60590 Frankfurt am Main
Germany
Tel.: +49 69 6301 5418
Fax: +49 69 6301 7948
e-mail: gille@em.uni-frankfurt.de

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Introduction

The assumption that tumor growth and metastasis are angiogenesis-dependent was proposed initially by Judah Folkman in 1971 (1). Its key implication is that inhibition of new vessel formation can serve as

a universal strategy to interfere with tumor growth and progression. Over the past three decades, the dependence of tumor growth on neovascularization has been firmly established by extensive experimental evidence, demonstrating that tumors as small as a few cubic millimeters in size are not able to continue to grow without vigorously inducing new blood vessel formation. As a result, tumor starvation through interference with tumor blood supply has become a well-recognized approach of cancer therapy (2,3). Conceptionally, antiangiogenic therapy pursues a strategy that is directed against the vascular constituent of the tumor stroma rather

Abbreviations: BEV, bevacizumab; CRC, colorectal cancer; EGF, epidermal growth factor; EGFR, epidermal growth factor receptor; GIST, gastrointestinal stromal tumor; PDGF, platelet-derived growth factor; PDGFR, platelet-derived growth factor receptor; NSCLC, non-small cell lung cancer; OS, overall survival; PFS, progression-free survival; RCC, renal cell cancer; TKI, tyrosine kinase inhibitor; TTP, time to progression; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

than the conventional target, which is the tumor cell itself (4). The tumor microenvironment as a primary target of such a stromal therapy approach carries the prospect of being less prone to resistance mechanisms due to genetic stability (5).

The angiogenic cascade is subject to very dynamic and complex regulation (6,7). A large number of proangiogenic and antiangiogenic factors have been demonstrated to control angiogenesis (8). Under the influence of endogenous angiogenesis inhibitors, the proliferation of tumor cell clusters may be kept in check by an equivalent rate of cell death (9). However, once the effect of proangiogenic molecules is no longer balanced by that of antiangiogenic factors, the angiogenic 'switch' is turned on and the vascular phase of tumor growth is being initiated (6).

Proangiogenic and antiangiogenic regulators not only emanate from cancer cells but also derive from stromal cells, circulating inflammatory cells, and the extracellular matrix. Among the several crucial angiogenic growth factor receptor pathways identified to date, the vascular endothelial growth factor (VEGF) family of proteins and receptors has been the major focus of targeted drug development in oncology (10,11). In February 2004, the humanized anti-VEGF antibody bevacizumab (BEV) was the first antiangiogenic compound that was approved by the US Food and Drug Administration for use in conjunction with standard chemotherapy in advanced colorectal cancer (CRC) patients (12). After suffering several setbacks during development of antiangiogenic cancer treatments in early clinical trials, combination of VEGF-targeted therapy in conjunction with traditional cytotoxic therapy finally proved successful.

This review will focus on the opportunities and challenges of growth factor- and growth factor receptor-targeted antiangiogenic drugs in the current clinical development of cancer therapy.

Targets of antiangiogenic therapy

The detailed understanding of the molecular changes underlying angiogenesis-dependent tumor growth has led to identification of various attractive targets for potential intervention (Fig.1). Next to the critical VEGF family of proteins and receptors, the platelet-derived growth factor (PDGF) and the epidermal growth factor (EGF) families are considered important complementary regulators of tumor angiogenesis and neovascularization (3).

The VEGF family of proteins and receptors plays a primary role in angiogenesis-dependent growth of most cancer types (11,13). Among these factors, VEGF-A (usually referred to as VEGF) is recognized as the most potent angiogenic molecule, which after binding to its high-affinity receptors VEGFR-1 and VEGFR-2 can trigger and promote essentially all segments of the angiogenic process. VEGFR-2 mediates the pivotal signaling effects of VEGF-A, including microvascular permeability, endothelial cell proliferation and survival. VEGF-C and VEGF-D are proteolytically cleaved from precursor proteins to give rise to forms with higher binding affinities to their receptors. While VEGF-C and VEGF-D largely act as lymphangiogenic growth factors via activation of VEGFR-3, they may also bind VEGFR-2 to elicit vascular angiogenic

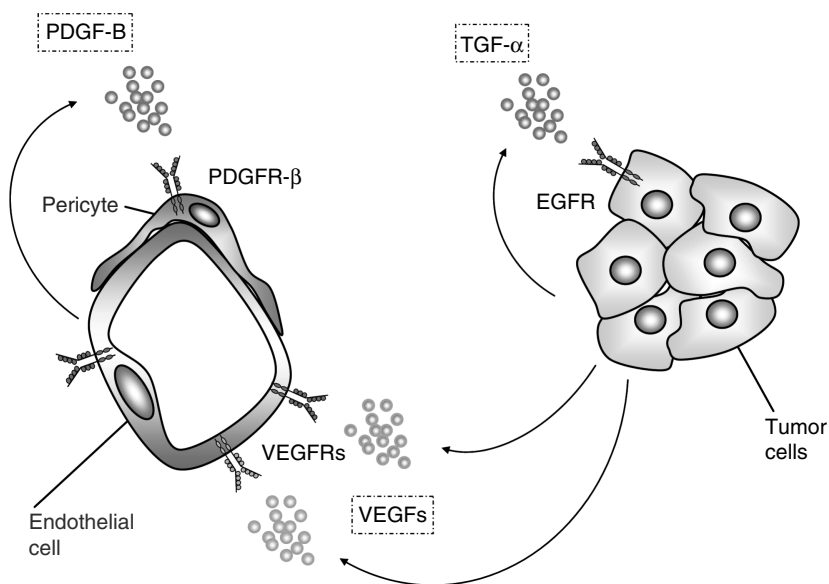


Figure 1. Schematic representation of potential targets of antiangiogenic therapy. EGFR, epidermal growth factor receptor; PDGF, platelet-derived growth factor; PDGFR, PDGFR receptor; TGF- α , transforming growth factor- α ; VEGFs, vascular endothelial growth factors; VEGFRs, vascular endothelial growth factor receptors.

responses (14). In contrast to VEGFR-2 that is expressed by most endothelial cells, VEGFR-3 expression in the adult is restricted largely to lymphatic endothelial cells and to proliferating angiogenic endothelial cells (15).

The PDGF family of growth factors is increasingly being recognized as a complementary target of antiangiogenic therapy. PDGF members not only increase tumor growth by autocrine stimulation of cancer cells via PDGF receptor (PDGFR) activation and overexpression but also by enhancing tumor angiogenesis (16). The PDGF family members mediate their effects through interaction with two individual receptors, namely PDGFR- α and PDGFR- β , the latter of which is the predominant form expressed by pericytes of tumor-related endothelial cells (17). Pericytes are smooth muscle-like mural cells that intimately associate with endothelial cells in capillaries and small blood vessels. These perivascular cells are viewed as important regulators of blood vessel stabilization and maturation, providing endothelial cells with essential survival signals. PDGFR inhibition has been shown to substantially impair pericyte function, leading to detachment of pericytes of established tumor vessels, and thereby rendering endothelial cells more susceptible to apoptotic cell death. It is therefore assumed that combined VEGFR and PDGFR inhibition constitutes a more effective antiangiogenic approach for cancer therapy (17,18).

The EGF receptor (EGFR) has been established as an important therapeutic target in a large number of epithelial tumors (19). Aberrant EGFR activation leads to cell cycle progression, reduced apoptotic capacity, and to enhanced angiogenesis. The latter observation may be attributed largely to an increased expression of critical angiogenesis factors because of EGFR activation (e.g. VEGF-A). In line with this assumption, compounds targeted against signaling via the EGFR have been previously linked to decreased secretion of proangiogenic factors by tumor cells and to an increased apoptotic rate in tumor-associated endothelial cells (20,21). Hence, targeted EGFR inhibition may not only be directed against the tumor cells themselves but may also interfere with angiogenesis via indirect inhibitory mechanisms.

Common misconceptions: Lessons from VEGF/VEGFR-directed therapies in clinical trials

Antiangiogenic cancer therapy is frequently perceived as 'magic bullet', capable to eventually

cure any type of cancer. This unrealistic perception is largely based on overwhelming preclinical experiences with first generation antiangiogenic agents. Realistically, however, the primary goal of antiangiogenic cancer therapy is to inhibit the tumor's capacity to grow beyond considerable size (22). On the basis of this assumption, tumor starvation through interference with tumor blood supply can be expected to primarily induce tumor stabilization in a short-term perspective and to potentially accomplish long-term tumor regression. As a great surprise, various preclinical studies have demonstrated that VEGF/VEGFR-targeted therapies alone are capable of not only suppressing the growth of established tumors but also of inducing remarkable tumor regressions or even eradication of metastatic disease (11).

The impressive results of these preclinical studies created expectations as to the potential of such targeted antiangiogenic approaches, which could not be met in the clinical setting (23). When VEGF/VEGFR-directed therapies were administered as single agents, only modest objective responses were seen without yielding long-term survival benefits (11). As a consequence, combinatorial strategies were subsequently pursued, simultaneously targeting the vascular compartment along with cancer cells. On the basis of preclinical evidence indicating that antiangiogenic agents can act synergistically with traditional chemo- and radiotherapy, angiogenesis inhibitors were increasingly studied in conjunction with standard cytotoxic regimens in clinical trials.

The mechanisms by which combined administration of antiangiogenic and cytotoxic therapies enhances anti-tumor activity are yet to be fully understood. A model has been recently proposed that antiangiogenic therapy may 'normalize' the structurally and functionally aberrant tumor vasculature, resulting in more efficient oxygen supply and delivery of cytotoxic drugs to the tumor cell compartment via lowering interstitial fluid pressure and improving blood flow (24). Accumulating evidence in support of this hypothesis indeed suggests that both VEGF/VEGFR- and PDGF/PDGFR-targeted therapies can alleviate interstitial fluid pressure and can increase transvascular transport of tracer substances or cytotoxic compounds (16,25). Improved anti-vascular effects of standard cytotoxic agents by addition of antiangiogenic compounds may also result from an increased capacity to exert collateral damaging effects on endothelial cell-cycling and additional key endothelial cell functions that are crucial for tumor-associated new vessel formation (26). To better interfere with the apparent

ability of slowly proliferating endothelial cells to repair and recover during the usual rest periods of traditional chemotherapy protocols, a strategy of more regular schedules with adapted lower doses of cytotoxic agents (referred to now as 'metronomic' chemotherapy) is currently evaluated in a number of clinical trials (26,27).

The strategy to target VEGF-A in combination with chemotherapy finally proved successful, as evidenced by the approval of the anti-VEGF antibody BEV in combination with standard chemotherapy in advanced CRC patients (28). Importantly, this was the first study to definitively show a benefit of an antiangiogenic compound when combined with chemotherapy, thereby reinstalling the confidence in antiangiogenic cancer therapy.

BEV in conjunction with chemotherapy shows promise beyond its role in CRC

The experiences with the anti-VEGF antibody BEV in clinical phase III trials suggest that tumor type, tumor stage and prior treatment status affect efficacy of antiangiogenic cancer therapy (Table 1). While BEV improves progression-free survival (PFS) and overall survival (OS) for both first-line and second-line chemotherapy of metastatic colorectal cancer (CRC) patients (28,29), the anti-VEGF antibody failed to prolong survival in pretreated advanced breast cancer patients (30). By comparison, when BEV was given in a randomized fashion with chemotherapy in women with previously untreated metastatic breast cancer, PFS and OS significantly increased in the BEV arm (31). Hence, BEV appears to be more effective in the setting of limited breast cancer disease and/or in women

with no or limited prior chemotherapy in the metastatic setting. Potentially, angiogenesis-dependent growth of breast cancer at later stages, in contrast to CRC, may not primarily depend on VEGF-A alone but may also rely on other critical proangiogenic factors, therefore allowing them to circumvent VEGF-A-targeted therapies. In addition to advanced breast and CR cancer patients, BEV as first-generation angiogenesis inhibitor shows also promise in phase III clinical data of untreated non-squamous non-small cell lung cancer (NSCLC) patients (32).

Multitargeted inhibitors for antiangiogenic cancer therapy in clinical trials

In the era of targeted cancer therapy, the development of orally available small-molecule kinase inhibitors has emerged as an attractive alternative to humanized monoclonal antibodies (33). First generation antiangiogenic tyrosine kinase inhibitors (TKIs) have been selected largely on their capacity to primarily target VEGFR2 tyrosine kinase activity as the major mediator of angiogenic signaling. However, the focus has shifted towards TKIs that target a broader set of receptor and non-receptor tyrosine kinases due to the enhanced understanding of the complexity of angiogenesis regulation. The newer compounds under investigation and development therefore inhibit multiple VEGFRs, mostly in addition to the tyrosine kinases of other important signaling pathways (e.g. PDGFR and EGFR) (Table 2). Orally available small-molecule kinase inhibitors may significantly increase toxicity of chemotherapy protocols (Table 3). Hypertension constitutes a commonly observed grade 3/4 adverse event among antiangiogenic

Table 1. Efficacy of combinatorial BEV therapies, phase III clinical trials

| Drug [Company] | Target | Tumor type | Dose | Regimen | Median PFS | Median OS | Additional information | Reference |
|--|--------------|---|-------------------------|-------------------------|---|---|--------------------------------------|-----------|
| BEV (Avastin [®]) [Genentech/Roche] | VEGF-A (MAB) | Previously untreated metastatic CRC | 5 mg/kg, biweekly | IFL ± BEV | 10.6 ¹ vs. 6.2 months (<i>P</i> < 0.001) | 20.3 ¹ vs. 15.6 months (<i>P</i> < 0.001) | Phase III, 813 patients | (28) |
| | | Previously treated CRC | 10 mg/kg, biweekly | FOLFOX ± BEV | 7.2 ¹ vs. 4.8 months (<i>P</i> < 0.0001) | 12.9 ¹ vs. 10.8 months (<i>P</i> = 0.0018) | Phase III, Trial E3200, 579 patients | (29) |
| | | Locally recurrent or metastatic breast cancer | 10 mg/kg, biweekly | Paclitaxel weekly ± BEV | 11.0 ¹ vs. 6.1 months (<i>P</i> < 0.001) | HR 0.67 (<i>P</i> = 0.01) | Phase III, Trial E2100, 715 patients | (31) |
| | | Previously treated breast cancer | 15 mg/kg, every 3 weeks | Capecitabine ± BEV | 4.9 vs. 4.2 months | 15.1 vs. 14.5 months | Phase III, 462 patients | (30) |

BEV, bevacizumab; CRC, colorectal cancer; FOLFOX, fluorouracil, leucovorin, oxaliplatin; HR, hazard ratio; IFL, irinotecan, fluorouracil, leucovorin; MAB, monoclonal antibody; NSCLC, non-small cell lung cancer; OS, overall survival; PFS, progression-free survival; VEGF, vascular endothelial growth factor.

¹Difference statistically significant.

Table 2. Synopsis of selected oral small molecule receptor tyrosine kinase inhibitors

| Agent | Company | Targets | | | | | | |
|--|-------------------|---------|---------|---------|-------|------|-------|-------|
| | | VEGFR-1 | VEGFR-2 | VEGFR-3 | PDGFR | EGFR | c-Kit | Other |
| PTK/ZK, Vatalanib | Novartis/Schering | + | + | + | (β) | | (+) | |
| BAY 43–9006, Sorafenib (Nexavar [®]) | Onyx/Bayer | | + | + | β | | + | RAF |
| SU 11248, Sunitinib (Sutent [®]) | Pfizer | + | + | + | α,β | | + | |
| ZD 6474 (Zactima [™]) | AstraZenica | + | + | + | | + | | ABL |
| AEE 788 | Novartis | + | + | | | + | | HER2 |
| Imatinib (Glivec [®]) | Novartis | | | | β | | + | ABL |
| Gefitinib (Iressa [™]) | AstraZenica | | | | | + | | |
| Erlotinib (Tarceva [™]) | OSI/Roche | | | | | + | | |

EGFR, epidermal growth factor receptor; PDGFR, platelet-derived growth factor receptor; VEGFR, vascular endothelial growth factor receptor

agents. Conversely, adverse effects frequently associated with administration of anti-VEGF antibody BEV (e.g. haemorrhage or arterial thromboembolic events) were not found to be generally increased in first clinical trials involving antiangiogenic TKIs (Table 3). Remarkably, dermatological toxicity (e.g. hand-foot syndrome or skin rash) appears to be a common severe adverse event of kinase inhibitors. A matter of concern is a potential increase in neurotoxicity by VEGF-targeted strategies, as reduced VEGF levels can promote motor neuron degeneration by limiting neural tissue perfusion and VEGF-

dependent neuroprotection (34). However, an increase in grade 3/4 sensory neuropathy has not yet been observed despite the fact that platinum analogues with their particular neurotoxic potential (e.g. oxaliplatin and carboplatin) are commonly used in chemotherapy combination regimens.

Depending on the tumor entity, oral multitargeted TKIs can exert both antiangiogenic and anti-tumor activities at the same time. As a consequence, multitargeted compounds may improve the outcome of cancer patients as single-agent treatment (Table 4). For instance,

Table 3. Increased drug-related grade 3/4 adverse events of selected antiangiogenic agents compared with placebo

| Drug [Company] | Target | Grade 3/4 | Reference |
|---|--|---|----------------|
| BEV (Avastin [®]) [Genentech/Roche] | VEGF-A (MAB) | Hypertension Proteinuria Bleeding Diarrhea Neutropenia Thromboembolic events Gastrointestinal perforations (in CRC patients) Congestive heart failure | (28–32,49) |
| SU11248, Sunitinib (Sutent [®]) [Pfizer] | VEGFR-1, -2, -3; PDGFR-α/-β, KIT (TKI) | Hypertension Fatigue Diarrhea Hand-foot skin reaction Neutropenia Thrombocytopenia Hyperlipasemia | (36,50,51) |
| PTK/ZK, Vatalanib [Novartis/Schering] | VEGFR-1, -2, -3 (TKI) | Hypertension Dizziness Pulmonary embolism Diarrhea Nausea | (39,52) |
| BAY 43-9006, Sorafenib (Nexavar [®]) [Onyx/Bayer] | VEGFR-2, -3; PDGFR-β;RAF; KIT (TKI) | Hypertension Diarrhea Hand-foot skin reaction Neutropenia Skin rash | (37,41,53, 54) |

BEV, bevacizumab; CRC, colorectal cancer; EGFR, epidermal growth factor receptor; MAB, monoclonal antibody; PDGFR, platelet-derived growth factor receptor; TKI, tyrosine kinase inhibitor; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

Table 4. Efficacy of multitargeted antiangiogenic agents, phase III clinical trials

| Drug [Company] | Target | Tumor type | Dose | Combination regimen | Median PFS | Median OS | Additional information | Reference |
|---|--|---|---|-----------------------|--|---------------------------------------|---|-----------|
| PTK/ZK Vatalanib [Novartis/ Schering] | VEGFR-1, -2, -3 (TKI) | Previously untreated metastatic CRC | 1250 mg, daily | FOLFOX ± PTK/ZK | HR 0.88 (P < 0.188) patients with high LDH only ¹ ; HR 0.68 (P < 0.012) | | Phase III, CONFIRM-1, 1168 patients enrolled | (39) |
| BAY 43-9006, Sorafenib (Nexavar [®]) [Onyx/Bayer] | VEGFR-2, -3; PDGFR- β ; RAF; KIT (TKI) | Metastatic clear cell RCC | 400 mg, bid | ± Sorafenib | 24 ¹ vs. 12 weeks (P < 0.00001) | | Phase III, 769 of planned 884 patients enrolled | (37) |
| SU11248, Sunitinib (Sutent [®]) [Pfizer] | VEGFR-1, -2, -3; PDGFR- α/β ; KIT (TKI) | Imatinib-resistant metastatic GIST | 50 mg, daily for 4 weeks, with a 2-week break | ± SU11248 | TTP; 6.3 ¹ vs. 1.5 months (P < 0.00001) | HR 0.49 ¹ (P < 0.00001) | Phase III, 312 patients enrolled | (36) |
| ZD6474, (Zactima [™]) [AstraZenica] | VEGFR-2, -3; EGFR (TKI) | Previously treated NSCLC | 100 mg, (300 mg), daily | Docetaxel ± ZD6474 | 18.7 weeks (100 mg) vs. 12.0 weeks (placebo) [HR 0.64; P = 0.074] | | Randomized phase II, 127 patients enrolled | (40) |

CRC, colorectal cancer; EGFR, epidermal growth factor receptor; FOLFOX, fluorouracil, leucovorin, oxaliplatin; GIST, gastrointestinal stromal tumor; HR, hazard ratio; IFL, irinotecan, fluorouracil, leucovorin; LDH, lactate dehydrogenase; NSCLC, non-small cell lung cancer; OS, overall survival; PDGFR, platelet-derived growth factor receptor; PFS, progression-free survival; RCC, renal cell cancer; TKI, tyrosine kinase inhibitor; TTP, time to progression; VEGFR, vascular endothelial growth factor receptor.

¹Difference statistically significant.

the biology of gastrointestinal stromal tumors (GISTs) has been demonstrated to critically involve constitutive activation of c-Kit and PDGFR- α (35). Thus, the compound SU11248 can be expected to target both tumor cells (via inhibition of c-Kit and PDGFR- α) and the endothelial cell compartment (via inhibition of VEGFRs and PDGFR- β). Remarkably, SU11248 has been recently reported to significantly prolong time to tumor progression (TTP) and OS in patients with metastatic GIST disease (36) that have progressed on imatinib (Gleevec[®]; TKI of c-Kit and PDGFR- β). Likewise, the oral multitargeted inhibitor sorafenib (BAY 43-9006) can improve the outcome of metastatic clear cell renal cell cancer (RCC) patients as single agent treatment (37), in all probability also based on the particular biology of the disease (38). In sporadic and familial clear cell RCC, the von Hippel-Lindau suppressor gene is inactivated or dysfunctional, leading to constitutive activation of the hypoxia inducible factor HIF-1 α . As a result, the expression of several hypoxia-induced genes, including VEGF and PDGF, is profoundly increased. Hence, VEGF- and VEGFR-targeted strategies have been rightfully anticipated to be specifically successful in this subset of RCC patients.

In most tumor entities, however, also second-generation antiangiogenic drugs have to be combined with standard chemotherapy protocols to yield demonstrable efficacy in advanced cancer patients. The results on the use of PTK/ZK in conjunction with cytotoxic therapy in chemo-naive CRC patients have been awaited with much anticipation (Table 5). In contrast to the encouraging findings with BEV in a comparable cohort of patients (Table 1), PTK/ZK failed to significantly impact PFS in a phase III clinical study, in which almost 1200 patients were enrolled (39).

ZD6474 represents a further, yet distinct second-generation antiangiogenic compound, as it exerts inhibitory capacity for both the VEGFRs and the EGFR (Table 2). It is therefore frequently referred to as 'dual-kinase' inhibitor. Given the importance of EGFR activation for the progression of NSCLC patients, the addition of ZD6474 to docetaxel chemotherapy was tested in a placebo-controlled fashion. The preliminary results indicate that adding ZD6474 may indeed prolong PFS, although the improvement did not attain statistical significance in this phase II clinical trial (40). Currently, ZD6474 is administered to previously untreated metastatic or recurrent NSCLC in a randomized phase II trial in

Table 5. Combinatorial targeted antiangiogenic therapies, randomized phase II/III clinical trials

| Drug [Company] | Target | Tumor type | Dose | Regimen | Endpoints | Additional information |
|---|--|--|---|---|---|---|
| BAY 43-9006, Sorafenib (Nexavar [®]) [Onyx/Bayer] | VEGFR-2, -3; PDGFR- β ; RAF; KIT (TKI) | Chemonaive unresectable stage III or metastatic melanoma | 400 mg bid | Carboplatin/ Pacitaxel \pm Sorafenib | I. OS II. PFS, response rate | Phase III, ECOG-E2603; projected accrual: 800 patients |
| BEV (Avastin [®]) [Genentech/Roche] | VEGF-A (MAB) | Locally advanced or metastatic pancreatic cancer | 10 mg/kg, biweekly | Gemcitabine \pm BEV | I. OS II. Response rate, duration of response, PFS | Phase III, CALGB-80303; projected accrual: 590 patients |
| PTK/ZK, Vatalanib [Novartis/Schering] | VEGFR-1, -2, -3 (TKI) | Hormone-refractory metastatic prostate cancer | 15 mg/kg, every 3 weeks | Docetaxel/Prednisone \pm BEV | I. OS II. PFS | Phase III, CALGB-90401; projected accrual: 1020 patients |
| SU11248, Sunitinib (Sutent [®]) [Pfizer] | VEGFR-2, -3; PDGFR- α/β , KIT (TKI) | Previously treated metastatic CRC | 1250 mg, daily | FOLFOX \pm PTK/ZK | I. OS II. PFS | Phase III, CONFIRM-2; accrual completed: 855 patients |
| ZD6474, Zactima [™] [Astrazenica] | VEGFR-2, -3; EGFR (TKI) | Previously untreated metastatic RCC | 50 mg, daily for 4 weeks, with a 2-week break | SU11248 vs. IFN- α | I. TTP II. Response rate | Phase III, UCLA-0406015-01; projected accrual: 690 patients |
| | | Previously untreated stage IIIB or metastatic or recurrent NSCLC | Daily | Arm I: ZD6474 Arm II: ZD6474 + Carboplatin/ Pacitaxel Arm III: Placebo + Carboplatin/ Pacitaxel | I. Tolerable dose of ZD6474 II. TTP | Phase II, UCLA-0405022-01; projected accrual: 200 patients |

BEV, bevacizumab; CRC, colorectal cancer; EGFR, epidermal growth factor receptor; FOLFOX, fluorouracil, leucovorin, oxaliplatin; IFN, interferon; MAB, monoclonal antibody; NSCLC, non-small cell lung cancer; OS, overall survival; PDGFR, platelet-derived growth factor receptor; PFS, progression-free survival; RCC, renal cell cancer; TKI, tyrosine kinase inhibitor; TTP, time to progression; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

combination with carboplatin and paclitaxel chemotherapy (Table 5), paralleling the clinical setting in which BEV could be demonstrated to significantly improve both PFS and OS (32). Next to BEV and ZD6474, also the above-mentioned oral inhibitors SU11248, PTK/ZK, and sorafenib are intensely trialed in distinct randomized protocols and different tumor entities. Selected randomized clinical studies that are currently ongoing, or of which results are expected to be reported shortly, are summarized in Table 5. On the basis of encouraging data from phase I/II trials, the addition of sorafenib to carboplatin and paclitaxel chemotherapy is hoped to yield survival benefits in advanced melanoma patients (41). Sorafenib as multitargeted TKI that also inhibits RAF kinases was initially expected to be particularly useful for melanoma therapy, as in more than 60% of all affected melanoma patients activating B-RAF mutations are detected. However, the remarkable responses that were seen with sorafenib were independent of the B-RAF mutational status. Regrettably, detection of RAF mutations can therefore not serve as a predictor of sorafenib activity in melanoma patients.

Current directions of targeted antiangiogenic cancer therapy

In particular, two trends of targeted antiangiogenic cancer therapy have become apparent. Firstly, antiangiogenic compounds with therapeutic activity in advanced disease are increasingly being tested in the adjuvant clinical setting

(Table 6). Hence, in particular, BEV is currently evaluated in large randomized phase III clinical trials, either alone or in addition with chemotherapy. In the adjuvant setting, however, tolerability and safety of therapy become an even more relevant issue of increased awareness and concern. BEV treatment in CRC patients has been shown to be associated with an increased severity and/or risk of hypertension, gastrointestinal bleeding, and thromboembolic events (32), the latter of which were not observed at a higher rate in more recent BEV-containing regimens, as patients with prior history for thrombotic events are no longer eligible for these trials (29). The issue of tolerability would also apply for second-generation multitargeted antiangiogenic compounds in the adjuvant setting, as quality of life could be significantly compromised due to related toxicities, such as hypertension, fatigue and asthenia, nausea and diarrhea, or rashes and hand/foot-skin reactions (Table 3). As an extension to adjuvant therapy, targeted antiangiogenic agents are currently also evaluated in 'neoadjuvant' or 'postjuvant' clinical settings (Table 6). As to particular instances, ZD6474 is currently tested in a randomized fashion with the intention to maintain the response after induction chemotherapy in small cell lung cancer patients.

Secondly, there is a clear tendency at present to combine different antiangiogenic agents to accomplish a more comprehensive approach in blocking tumor angiogenesis. Not only the anti-VEGF antibody BEV but also the multitargeted inhibitors SU11248 and sorafenib are currently tested in combination with EGFR inhibitors (Table 7) in phase I/II trials to complement their

Table 6. Targeted antiangiogenic agents in adjuvant or 'postjuvant' settings, phase II/III clinical trials

| Drug [Company] | Target | Tumor type | Dose | Regimen | Endpoints | Additional information |
|---|----------------------------|---|-------------------------|---|-----------------------------|--|
| BEV (Avastin [®]) [Genentech/Roche] | VEGF-A (MAB) | High-risk stage II and stage III CRC | 15 mg/kg, every 3 weeks | Arm I: FOLFOX Arm II: BEV + FOLFOX Arm III: BEV + Oxaliplatin/ Capecitabine | I. DFS II. OS | Phase III UCLA-0412086-01; Projected accrual: 3450 patients |
| | | Resected stage II or III colon cancer | 10 mg/kg, biweekly | FOLFOX ± BEV | I. DFS II. OS | Phase III, NSABP-C-08; projected accrual: 2632 patients |
| ZD6474, (Zactima [™]) [AstraZenica] | VEGFR-2, -3; EGFR (TKI) | Small cell lung cancer after response to induction chemotherapy | Daily | ±ZD6474 | I. PFS II. Response rate | 'Postjuvant', maintenance, phase II CAN-NCIC-BR20; projected accrual: 120 patients |

BEV, bevacizumab; CRC, colorectal cancer; DFS, disease-free survival; EGFR, epidermal growth factor receptor; FOLFOX, fluorouracil, leucovorin, oxaliplatin; MAB, monoclonal antibody; OS, overall survival; PFS, progression-free survival; TKI, tyrosine kinase inhibitor; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

Table 7. Combinations of different molecular targeted agents with the intention to enhance antiangiogenic efficacy, phase I/II clinical trials

| Drug [Company] | Target | Tumor type | Dose | Regimen | Endpoints | Additional information |
|---|---|--|--------------------|---|--|---|
| BEV (Avasitin [®]) [Genentech/Roche] | VEGF-A (MAB) | Advanced melanoma or other advanced cancer | 10 mg/kg, biweekly | +Imatinib (PDGFR- β , ABL-, KIT-TKI) | Response rate, TTP, OS (phase II) | Phase I/II, UPCC-03903; projected accrual: 23-40 patients (phase II) |
| | | Advanced pancreatic cancer | 10 mg/kg, biweekly | Arm I: BEV/Gemcitabine + Erlotinib (EGFR-TKI) Arm II: BEV/Gemcitabine + Cetuximab (EGFR-MAB) + Gefitinib (EGFR-TKI) | I. Response rate II. Toxicity III. PFS, OS | Phase II, UCRC-13200A; projected accrual: 54-126 patients |
| SU11248, Sunitinib (Sutent [®]) [Pfizer] | VEGFR-1, -2, -3; PDGFR- α / β , KIT (TKI) | Metastatic RCC | | | Safety, efficacy | Phase I/II, A6181038 |
| AEE788 [Novartis] | VEGFR-1, -2; EGFR (TKI) | Recurrent or relapsed glioblastoma multiforme | | +Everolimus (mTOR-Inhibitor) | Response rate, PFS, OS (phase II) | Phase I/II, UCLA-0409004-01; projected accrual: 65 patients (phase II) |

BEV, bevacizumab; DFS, disease-free survival; EGFR, epidermal growth factor receptor; FOLFOX, fluorouracil, leucovorin, oxaliplatin; MAB, monoclonal antibody; mTOR, mammalian target of rapamycin; OS, overall survival; PDGFR, platelet-derived growth factor receptor; PFS, progression-free survival; TKI, tyrosine kinase inhibitor; TTP, time to progression; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

modes of action (Table 2). In addition, different multitargeted receptor TKIs (e.g. AEE788) are evaluated in combination with non-receptor TKIs, such as inhibitors of the mammalian target of rapamycin (mTOR), which is engaged in the regulation of a wide range of growth-related cellular functions. Fittingly, mTOR inhibitors (e.g. rapamycin and everolimus) have been shown to interfere both with cellular processes involved in tumor cell proliferation and with signaling pathways that are essential for endothelial cell survival (42).

Frequently underestimated problems and challenges of targeted antiangiogenic cancer therapy

One of the major challenges in the development of antiangiogenic therapy is the appropriate selection of patients who are more likely to benefit from this particular modality of cancer therapy (2). At present, targeted antiangiogenic compounds have to be administered to cancer patients largely in an untargeted fashion. Ideally, however, efficacy of targeted antiangiogenic drugs could be predicted by prior verification of critical angiogenic pathways in readily obtained clinical samples, in a fashion comparable with that of pioneering molecular targeted drugs, such as imatinib in the treatment of BCR/Abl kinase-positive chronic myelogenous leukemia (43). In all probability, it will be most difficult, if not impossible, to achieve the goal of proper patient selection on the basis of target validation, as the vast majority of all tumors are highly heterogeneous with regard to their dependency on angiogenic pathways.

Consequently, the major focus in the development of antiangiogenic therapy is to identify and to validate surrogate markers that can monitor activity and efficacy of targeted antiangiogenic drugs in patients (44). Obviously, circulating expression levels of VEGFs as well as other angiogenic growth factors have been initially regarded as potential surrogate markers of antiangiogenic drug efficacy (2). Although most of the angiogenic growth factors are found at higher levels in local and metastatic disease, the evidence has been rather elusive as to their reliability to predict drug efficacy and disease progression. Similarly, diverse results have been reported from different studies as to the potential of the number of circulating endothelial progenitors and/or mature circulating endothelial cells to be used as surrogate predictors (45,46). Hence, reliable biomarkers are yet to be determined to more precisely and correctly monitor the activity and

efficacy of antiangiogenic drugs in patients. Potentially, genomic- and/or proteomic-based analyses of clinical samples prior and during the course of antiangiogenic therapy will ultimately identify more appropriate sets of surrogate markers that can guide patient selection and can monitor early therapeutic efficacy. Currently, improved high-resolution imaging techniques are increasingly being implemented and tested in clinical trials to provide serial measures of tumor blood perfusion, vascular volume, and vascular permeability as methods to better understand the modes of action of antiangiogenic cancer therapy (47,48). These pieces of information will additionally help to optimize sequencing and combining antiangiogenic and standard cytotoxic therapy to further improve therapeutic efficacy in cancer patients.

Conclusions and future prospects

The successes of combinatorial BEV therapy in different randomized phase III studies with distinct tumor entities have made antiangiogenic therapy a complementary therapeutic modality in addition to surgery, radiotherapy, and chemotherapy. While further advances in the development of antiangiogenic cancer therapy are expected to result from ongoing clinical trials, the identification of surrogate biomarkers will be both instrumental to be able to select patients likely to benefit from antiangiogenic strategies, and to optimize sequencing, dosing and choices of drugs of combination therapies.

It can be anticipated that interfering with angiogenic signaling in conjunction with tumor type-adapted targeting of cell-survival and cell-division pathways will constitute a very powerful way to render advanced cancer patients more sensitive to universal chemotherapy regimens (Fig. 2). Hopefully, the integration of both antiangiogenic stromal and targeted cancer therapies into cytotoxic treatment cycles will even allow for lower doses of chemotherapy to be effective, thereby reducing side effects and enabling long-term treatment approaches. Turning cancer into a chronic disease at relatively symptom-free conditions would represent a major advance that may well be achieved by integration of distinct therapeutic modalities in numerous very heterogeneous cancer entities, including malignant melanoma. In this respect, targeted antiangiogenic therapy is a very welcome complementary therapeutic approach for future cancer therapy regimens.

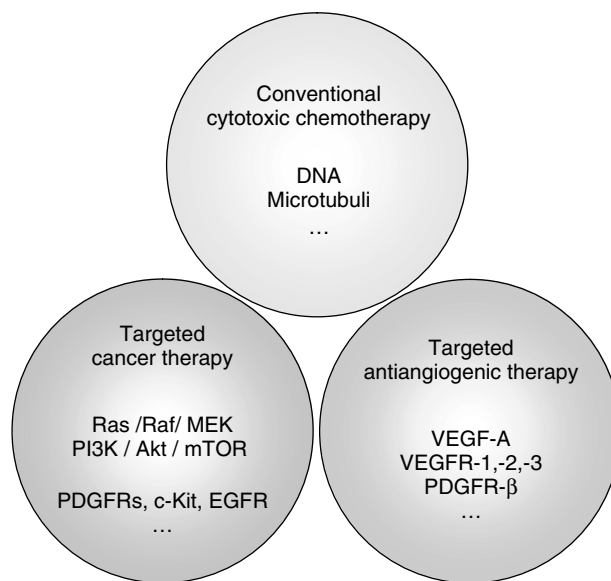


Figure 2. Integration of distinct therapeutic modalities. Yellow circle, conventional cytotoxic therapy with typical targets (e.g. DNA and microtubule); blue circle, cancer-directed targeted therapies against cell-division pathways (e.g. Ras/Raf/MEK), cell-survival pathways (e.g. PI3K/Akt/mTOR), and activated receptor tyrosine kinases (e.g. EGFR, c-Kit, and PDGFR); auburn circle, antiangiogenic therapy targeted against angiogenic growth factors or growth factor receptors (e.g. VEGF-A and VEGFR2). Akt, protein kinase B; EGFR, epidermal growth factor receptor; MAPK, mitogen-activated protein kinase; MEK, MAPK kinase; mTOR, mammalian target of rapamycin; PDGF, platelet-derived growth factor; PDGFR, platelet-derived growth factor receptor; PI3K, phosphatidylinositol-3-kinase; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor.

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