Published in final edited form as:

Curr Oncol Rep. 2014 February ; 16(2): 368. doi:10.1007/s11912-013-0368-7.

Aflibercept—a Decoy VEGF Receptor

Kristen K. Ciombor and

Division of Medical Oncology, Department of Internal Medicine, The Ohio State University Comprehensive Cancer Center, Arthur G. James Cancer Hospital and Richard J. Solove Research Institute, A445A Starling Loving Hall, 320 West 10th Avenue, Columbus, OH 43210, USA

Jordan Berlin

Division of Hematology/Oncology, Department of Medicine, Vanderbilt-Ingram Cancer Center, 777 Preston Research Building, 2220 Pierce Avenue, Nashville, TN 37232, USA

Kristen K. Ciombor: kristen.ciombor@osumc.edu; Jordan Berlin: jordan.berlin@vanderbilt.edu

Abstract

Aflibercept (known as ziv-aflibercept in the USA and sold under the trade name Zaltrap®) is a human recombinant fusion protein with antiangiogenic effects that functions as a decoy receptor to bind vascular endothelial growth factors A and B and placental growth factor. Its unique mechanism of action with respect to other agents targeting angiogenesis led investigators to speculate that it may be more ubiquitously efficacious in tumors highly dependent on pathologic angiogenesis for their growth. Despite encouraging preclinical studies in various tumor types, aflibercept has not been proven efficacious in most later-phase clinical studies. In fact, its only currently held US Food and Drug Administration indication is in metastatic colorectal cancer in combination with 5-fluorouracil, leucovorin, and irinotecan for those patients previously treated with an oxaliplatin-containing chemotherapy regimen. Given aflibercept's toxicity profile and cost, further investigation is needed to better understand its mechanism of action and to discover predictive biomarkers for optimization of its appropriate use in treatment of cancer patients.

Keywords

Aflibercept; Metastatic colorectal cancer; Vascular endothelial growth factor; Angiogenesis; Evolving therapies

Correspondence to: Jordan Berlin, jordan.berlin@vanderbilt.edu.

This article is part of the Topical Collection on *Evolving Therapies*

Compliance with Ethics Guidelines

Conflict of Interest Kristen K. Ciombor has served as a consultant for Bayer, has received travel/accommodation expenses reimbursed by Bayer, and has received honoraria from *Clinical Advances in Hematology and Oncology*.

Jordan Berlin has served on the Scientific Advisory Board of Amgen and on the Advisory Board of Genentech/Roche.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

Introduction

As oncologic treatments are increasingly moving toward targeted therapies as single agents or in combination with cytotoxic chemotherapeutics, the development of agents that target processes such as angiogenesis is of critical importance. Aflibercept, one of the more recent agents with antiangiogenic properties to be developed, has a unique biological mechanism as a decoy vascular endothelial growth factor receptor (VEGFR). This review will place the process of angiogenesis in context with important oncologic targets, discuss the mechanism of action, toxicity, efficacy, and cost of aflibercept as compared with other agents with antiangiogenic properties, and discuss its approved use in metastatic colorectal cancer (CRC), as well as its potential future opportunities.

Tumor Angiogenesis and Key Players

Angiogenesis, or new blood vessel formation, has long been known to be a critical component of tumorigenesis [1]. Physiologic angiogenesis is characterized by various well-organized steps, including proteolysis of the extracellular matrix, proliferation, migration, and assembly of the endothelial cells, mural cell recruitment and differentiation, and extracellular matrix production [2]. Pathologic angiogenesis is more heterogeneous and chaotic, often demonstrating tortuous vessel organization, hypoxic voids of various sizes, uneven and imperfect vessel walls and linings, and ineffective perfusion [3]. These unique characteristics of new blood vessel formation in tumors have made therapeutic targeting of angiogenesis a formidable challenge. Nonetheless, given the ubiquitous nature of tumor neovascularization and our increasing knowledge of the angiogenic process, antiangiogenic drug development remains an area of great interest.

Key players in the neovascularization process, and therefore common targets of antiangiogenic drugs, include the various vascular endothelial growth factors [VEGFs; VEGF-A, VEGF-B, VEGF-C, VEGF-D, and placental growth factor (PlGF)], VEGFRs (VEGFR-1/FLT1, VEGFR-2/KDR/FLK1, and VEGFR-3/FLT4), neuropilins 1 and 2, or co-receptors for the VEGFRs, and the TIE2 receptor and its angiopoietin ligands [4–6]. Although these ligands often have distinct roles, their overall effects overlap, and elevated levels are commonly associated with a poor prognosis in patients [7–10].

Targeting Angiogenesis: Mechanisms of Action

Targeted antiangiogenic therapies have had some success in controlling tumor growth, with several approved by the US Federal Drug Administration (FDA), but they have not been the magic bullet the cancer community has sought. These drugs ultimately fail to control disease through either primary or acquired resistance. However, in the following examples, they have proven to be beneficial to particular subsets of patients, and their differing mechanisms of action are in part responsible for their success.

Bevacizumab (Avastin®) is a recombinant humanized monoclonal antibody currently approved for the treatment of patients with metastatic CRC, nonsquamous non-small-cell lung cancer, glioblastoma multiforme, and metastatic renal cell carcinoma (RCC). It binds VEGF-A and prevents interaction with VEGFR-1 and VEGFR-2.

In addition to bevacizumab, small-molecule tyrosine kinase inhibitors of angiogenesis have also been developed and FDA-approved. Given the functional redundancy of angiogenic pathway components, these drugs target multiple kinases, such as VEGFRs. They include pazopanib (Votrient®), an inhibitor of VEGF, platelet-derived growth factor receptor (PDGFR), and KIT for the treatment of patients with advanced RCC; sorafenib (Nexavar®), an inhibitor of VEGFR-1, VEGFR-2, VEGFR-3, PDGFR- β , and RAF1 for patients with advanced RCC or hepatocellular carcinoma; sunitinib (Sutent®), an inhibitor of VEGFR-1, VEGFR-2, VEGFR-3, PDGFR- β , and RET for patients with advanced RCC, gastrointestinal stromal tumor, or pancreatic neuroendocrine tumor; vandetanib (Caprelsa®), an inhibitor of VEGFR and epidermal growth factor receptor for patients with medullary thyroid cancer; cabozantinib (Cometriq®), an inhibitor of MET and VEGFR2 for patients with medullary thyroid cancer; axitinib (Inlyta®), an inhibitor of VEGFR-1, VEGFR-2, and VEGFR-3 for patients with advanced RCC; and regorafenib (Stivarga®), an inhibitor of VEGFR-1, VEGFR-2, VEGFR-3, TIE2, PDGFR, fibroblast growth factor receptor, KIT, RET, and RAF for patients with metastatic CRC or gastrointestinal stromal tumor.

In contrast to bevacizumab and the small-molecule multi-tyrosine kinase inhibitors, aflibercept is a decoy receptor that inhibits angiogenesis by targeting VEGF-A, VEGF-B, and PlGF [11]. It is a recombinant fusion protein of the second immunoglobulin (Ig) domain of VEGFR-1 and the third Ig domain of VEGFR-2, fused to the constant region (Fc) of human IgG1 [11]. Although its potential advantages over bevacizumab in light of its mechanism beyond VEGF-A are clear, it also has a higher affinity than either VEGFR or bevacizumab for VEGF-A. In addition, its unique PlGF targeting at least conceptually lends the possibility of increased efficacy over the other antiangiogenic drugs, particularly given the increase in PlGF levels seen when patients were exposed to other anti-VEGF therapies [12, 13].

Preclinical Activity of Aflibercept

Preclinical studies of aflibercept first demonstrated inhibition of VEGF-induced phosphorylation of VEGFR-2 in human umbilical vein endothelial cells and VEGF-induced fibroblast proliferation [11], which led to studies of single-agent aflibercept in various animal models. In vivo studies in mice revealed capillary regression and endothelial cell apoptosis, among other effects [14]. In various tumor xenograft models, aflibercept reduced blood vessel density, tumor growth, and metastasis formation, while increasing survival [15, 16]. When combined with administration of other chemotherapeutic agents such as paclitaxel, docetaxel, gemcitabine, or 5-fluorouracil and irinotecan, administration of aflibercept led to synergistic reductions in tumor burden, vasculature, and tumor proliferation [17–19]. These favorable antitumor results were the impetus to take aflibercept into early-phase clinical trials in humans.

Safety and Early Clinical Activity of Aflibercept

Phase I studies were undertaken with aflibercept administered either subcutaneously or intravenously, and the toxicities seen were as expected for an agent with antiangiogenic effects [20, 21•]. In a phase I study by Tew et al. [20], 38 patients with advanced solid

tumors were treated with aflibercept administered subcutaneously at seven dosages ranging from 25 µg/kg per week to 800 µg/kg twice weekly, with the maximum tolerated dose not reached, and dosages not increased above 800 µg/kg twice weekly owing to solubility and administration difficulties. Common toxicities included proteinuria (37 %), fatigue (32 %), injection-site reactions (18 %), nausea (17 %), myalgia (16 %), anorexia (16 %), hypertension (13 %), and voice hoarseness (11 %). Half-lives of 3 days for free aflibercept and 18 days for bound aflibercept were seen in this study, and no anti-aflibercept antibodies were detected. VEGF–aflibercept complex formation was maximized at approximately 800 µg/kg weekly. Stable disease was maintained for at least 10 weeks in 18 patients (47 %).

In the phase I study of intravenously administered aflibercept, 47 patients received doses of 0.3 to 7.0 mg/kg intravenously every 2 weeks, with dose-limiting toxicities of rectal ulceration and proteinuria with the 7 mg/kg dose [21•]. On the basis of significantly increasing toxicities for doses exceeding 4 mg/kg, this was considered the recommended phase II dose of aflibercept. The half-life of aflibercept at 0.3 mg/kg was 1.7 days and was 5.1 days at 7.0 mg/kg, yet the phase II dose of aflibercept was recommended to be administered every 2 weeks. In this study, Response Evaluation Criteria in Solid Tumors defined partial responses were observed, one at 3 mg/kg and two at 7.0 mg/kg, and maximal VEGF-bound aflibercept complex levels were reached at doses of 2.0 mg/kg and above.

Phase I studies of aflibercept therapy in combination with cytotoxic chemotherapy were also performed, with acceptable safety and tolerability being seen [22–24]. Aflibercept was relatively well tolerated in 38 patients treated concomitantly with irinotecan, 5-fluorouracil, and leucovorin [25]. In this study, two grade 3 dose-limiting toxicities with a dose of 4 mg/kg, one of proteinuria and one of acute nephrotic syndrome with thrombotic microangiopathy, were observed. However, nine patients had partial responses, five with the 4 mg/kg dose, and 22 patients had stable disease. When aflibercept therapy was combined with treatment with docetaxel at 75 mg/m² or pemetrexed at 500 mg/m² and cisplatin at 75 mg/m² every 3 weeks, the recommended phase II dose of aflibercept was 6 mg/kg on the basis of similar dose-limiting toxicities and pharmacokinetics [23, 24].

Clinical Efficacy of Aflibercept: Phase II Studies

Once aflibercept's safety, toxicities, and preliminary efficacy were known, phase II studies in various tumor types were undertaken to more closely examine possible efficacy of the drug, as reviewed by Gaya and Tse [26•]. Several phase II trials failed to show significant efficacy of aflibercept as a single agent. For example, in one phase II trial single-agent aflibercept was administered at 4 mg/kg intravenously every 2 weeks to 75 patients with metastatic CRC who had previously progressed following treatment with at least one line of therapy, of which 68 % had received prior treatment with bevacizumab [27]. In the bevacizumab-naïve cohort, five of 24 patients had stable disease for 16 weeks or more. In the cohort that had received prior treatment with bevacizumab, one patient had a partial response that was sustained for 20 weeks, and six of 51 patients had stable disease for 16 weeks or more. Median progression-free survival for the bevacizumab-naïve group and the group that had received prior treatment with bevacizumab was 2.0 and 2.4 months, respectively. Median overall survival for the same groups was 10.4 and 8.5 months,

respectively. Typical anti-VEGF toxicities and other toxicities were seen, including grade 3 or higher hypertension, proteinuria, fatigue, and headache, and dosing was reduced, delayed, or discontinued in a significant number of patients. The pharmacokinetic data suggested that free aflibercept was present in an amount to bind endogenous VEGF, but no association was found between the free to VEGF-bound aflibercept ratio and clinical benefit, or between hypertension and clinical benefit [27].

In the hope that aflibercept therapy might show efficacy when combined with cytotoxic chemotherapy in first-line metastatic CRC, a phase II noncomparative, randomized trial was undertaken [28]. In this study, 236 patients were randomized to receive infusional fluorouracil, leucovorin, and oxaliplatin (modified FOLFOX6) with or without aflibercept, 4 mg/kg, every 2 weeks. The primary end point of progression-free survival rate at 12 weeks was 25.8 % [95 % confidence interval (CI), 17.2–34.4] for the modified FOLFOX6 plus aflibercept group and 21.2 % (95 % CI, 12.2–30.3) for the modified FOLFOX6 alone group. The response rates were 49.1 % (95 % CI, 39.7–58.6) for the combined arm and 45.9 % (95 % CI, 36.4–55.7) for the modified FOLFOX6 alone arm, with median progression-free survival of 8.48 months (95 % CI, 7.89–9.92) and 8.77 months (95 % CI, 7.62–9.27), respectively. The study was not powered to compare the two arms, but it did not appear that the addition of aflibercept significantly improved the response rate or progression-free survival in this trial. As expected, toxicities were greater in the combined treatment arm. Several other phase II trials in combination with various cytotoxic chemotherapy regimens in other tumor types have now been completed or are ongoing.

Clinical Efficacy of Aflibercept: Phase III Studies

Despite some promising safety and efficacy data for aflibercept in various tumor types in early-phase clinical trials, it failed to improve survival in combination with chemotherapy in prostate cancer, non-small-cell lung cancer, or pancreatic cancer in randomized, phase III clinical trials (Table 1) [29–31]. In the VENICE study, 1,224 men with metastatic castration-resistant prostate cancer were randomized to receive either aflibercept at 6 mg/kg or placebo in combination with docetaxel at 75 mg/m² every 3 weeks and 5 mg orally administered prednisone twice daily as first-line therapy in the metastatic setting [31]. Median overall survival was 22.1 months for the aflibercept group and 21.2 months for the placebo group [stratified hazard ratio (HR), 0.94, 95.6 % CI, 0.82–1.08, $p=0.38$], with higher incidence of grade 3–4 toxicities and treatment-related fatal adverse events seen in the aflibercept group.

Similarly, treatment with aflibercept in the phase III VANILLA clinical trial failed to improve overall survival for patients with metastatic pancreatic cancer. In this study, patients were treated with first-line aflibercept at 4 mg/kg every 2 weeks or placebo in combination with gemcitabine at 1,000 mg/m² weekly for 7 weeks of 8 weeks, then weekly for 3 weeks of 4 weeks [30]. The VANILLA trial was stopped for futility after a planned interim analysis of overall survival in 427 randomized patients. Median overall survival was 7.8 months in the gemcitabine plus placebo group versus 6.5 months in the gemcitabine plus aflibercept group (HR, 1.165, 95 % CI, 0.921–1.473, $p=0.2034$), with treatment discontinuations due to adverse events occurring more frequently in the aflibercept-containing-treatment arm.

The phase III VITAL trial, in contrast to the other phase III trials with negative findings, studied aflibercept in combination with second-line chemotherapy, but aflibercept failed to improve survival in this setting as well. The VITAL trial randomized 913 platinum-pretreated patients with advanced or meta-static non-small-cell lung cancer to aflibercept at 6 mg/kg or placebo every 3 weeks in combination with docetaxel at 75 mg/m² [29]. Median overall survival was 10.1 months for patients in the aflibercept arm versus 10.4 months for patient in the placebo arm (HR, 1.01, 95 % CI, 0.87–1.17, stratified log-rank $p=0.90$). In addition, grade 3 or higher toxicities occurred more frequently in the aflibercept arm.

In contrast, one phase III trial of aflibercept in patients with pretreated CRC demonstrated positive results, leading to FDA approval of the drug in 2012 (Table 1). In the randomized phase III VELOUR trial, 1,226 patients with metastatic CRC previously treated with oxaliplatin were randomized to receive aflibercept at 4 mg/kg intravenously or placebo every 2 weeks in combination with infusional fluorouracil, leucovorin, and irinotecan (FOLFIRI) [32•]. Significantly, 30.4 % of the overall trial population had previously received bevacizumab therapy, and this was well balanced among the two groups. Median overall survival was 13.50 months for patients in the aflibercept arm versus 12.06 months for patients in the placebo arm (HR, 0.817, 95.34 % CI, 0.713–0.937, $p=0.0032$). Progression-free survival was also significantly improved in the aflibercept arm compared with the placebo arm (6.9 vs 4.67 months; HR, 0.758, 95 % CI, 0.661–0.869, $p<0.0001$). The response rate was improved as well, with 19.8 % of patients treated with FOLFIRI plus aflibercept responding versus 11.1 % of the FOLFIRI plus placebo group ($p=0.0001$). As expected, the frequency of grade 3 and grade 4 toxicities was increased in the aflibercept arm (grade 3, 62.0 % vs 45.1 %; grade 4, 21.4 % vs 17.4 %), including adverse events typically associated with anti-VEGF therapy. On the basis of these data, on August 3, 2012, the FDA approved aflibercept for use in combination with FOLFIRI for the treatment of patients with metastatic CRC that is resistant to or has progressed following treatment with an oxaliplatin-containing regimen.

Aflibercept: Unanswered Questions and Future Directions

Although aflibercept has proven to be of benefit in combination with chemotherapy in a certain subset of patients, many unanswered questions about its use remain. Ongoing trials of aflibercept in tumor types other than CRC hope to find effective chemotherapy regimens or dosing that may expand its use to other patients. Even within the setting of CRC, it remains to be seen whether aflibercept has a role in other treatment settings such as in the neoadjuvant, adjuvant, or first-line metastatic settings, as well as in combination with radiation therapy. Predictive biomarkers for aflibercept have been investigated, but no effective ones have yet been found. Of critical importance, ongoing and future trials need to incorporate correlative science and tissue biopsies to best study the underlying mechanisms and efficacy of aflibercept, as some are doing.

Even in the metastatic CRC setting in which aflibercept's role is more closely defined, its efficacy when compared with that of other agents with antiangiogenic activity is unclear. Although bevacizumab has clear efficacy when combined with chemotherapy in the first-line setting for metastatic CRC [33, 34], it was not definitively known until recently whether it

had value with chemotherapy beyond disease progression. A phase III multinational study, ML18147, randomized 820 patients with metastatic CRC who had received bevacizumab in the first line to chemotherapy with or without bevacizumab therapy in the second line [35•]. Median overall survival was 11.2 months (95 % CI, 10.4–12.2) for the bevacizumab-containing-treatment arm and 9.8 months (95 % CI, 8.9–10.7) for the chemotherapy alone arm (HR, 0.81, 95 % CI, 0.69–0.94; $p=0.0062$). In light of these data, it has become a standard to continue bevacizumab therapy beyond the first line in metastatic CRC patients with no contra-indication to bevacizumab. However, it is unclear how bevacizumab therapy in combination with chemotherapy in the second-line setting compares with the combination of aflibercept therapy and chemotherapy, both in efficacy as well as in toxicity. As one third of patients in the VELOUR trial had received bevacizumab prior to aflibercept, perhaps additional benefit from aflibercept is present, but the possible mechanism of this additional benefit warrants further investigation.

Shortly after aflibercept gained approval from the FDA, another agent with antiangiogenic properties, regorafenib (Stivarga®), was approved for treatment of metastatic CRC. In the CORRECT trial, 760 patients with metastatic CRC who had been previously treated with fluoropyrimidine-, oxaliplatin-, and irinotecan-based chemotherapy were randomized to receive single-agent regorafenib, a multi-tyrosine kinase inhibitor targeting, for example, VEGFR1, VEGFR2, VEGFR3, PDGFR- α , PDGFR- β , and TIE2, or placebo [36•]. A preplanned interim analysis demonstrated that patients treated with regorafenib had a statistically significant improvement in overall survival. Median overall survival was 6.4 months in the regorafenib arm versus 5.0 months in the placebo arm (HR, 0.77, 95 % CI, 0.64–0.94). Adverse events were significant for those treated with regorafenib, including hand–foot skin reaction, fatigue, diarrhea, hypertension, and rash. Despite its similar efficacy in survival time, regorafenib appears to have a clearer niche in the metastatic CRC treatment landscape than aflibercept. Unlike aflibercept, it can be given as a single agent and is approved for refractory meta-static CRC patients. However, it remains to be seen how its further development and use will affect aflibercept, both biologically as well as in practice.

In addition to efficacy and toxicity comparisons between aflibercept and other agents with antiangiogenic activity, cost comparisons are significant as well. Although aflibercept was originally priced to compare with bevacizumab at a dosage of 10 mg/kg every 2 weeks, it was strenuously argued by the oncology community that this was a bevacizumab dose that many physicians do not use. When compared with the 5 mg/kg dose of bevacizumab every 2 weeks, aflibercept was much more expensive for what appeared to be similar benefit. As a result of market pressures, as illustrated by one academic institution's formulary policy [37], Sanofi-Aventis ultimately offered a 50 % discount on the purchase price of aflibercept. Aflibercept now competes financially more favorably with other antiangiogenic agents on the market than it previously did, although it remains unclear whether the original pricing had durable effects on the market share of the drug.

Conclusions

Agents with antiangiogenic properties such as aflibercept have long been heralded as potentially promising weapons in the treatment of cancer. In aflibercept's case, however, its

efficacy has only been proven in a subset of metastatic CRC patients, despite extensive testing in many tumor types thus far; namely, those who are resistant to or have progressed following treatment with an oxaliplatin-containing regimen, and even then only in combination with FOLFIRI. It remains unclear whether aflibercept has a role in other settings as well, and further investigation into the underlying mechanisms of the action of and resistance to this class of drug is needed to answer these questions. Furthermore, given the limited efficacy, toxicity, and cost of this agent, predictive biomarkers to select patients most likely to benefit from treatment with aflibercept are greatly needed.

References

Papers of particular interest, published recently, have been highlighted as:

- Of importance

1. Folkman J. Tumor angiogenesis: therapeutic implications. *N Engl J Med.* 1971; 285(21):1182–6. [PubMed: 4938153]
2. Carmeliet P, Jain RK. Molecular mechanisms and clinical applications of angiogenesis. *Nature.* 2011; 473(7347):298–307. [PubMed: 21593862]
3. Jain RK. Molecular regulation of vessel maturation. *Nat Med.* 2003; 9(6):685–93. [PubMed: 12778167]
4. Ellis LM, Hicklin DJ. VEGF-targeted therapy: mechanisms of anti-tumour activity. *Nat Rev Cancer.* 2008; 8(8):579–91. [PubMed: 18596824]
5. Holash J, et al. Vessel cooption, regression, and growth in tumors mediated by angiopoietins and VEGF. *Science.* 1999; 284(5422):1994–8. [PubMed: 10373119]
6. Tsigkos S, Koutsilieris M, Papapetropoulos A. Angiopoietins in angiogenesis and beyond. *Expert Opin Investig Drugs.* 2003; 12(6):933–41.
7. Lohela M, et al. VEGFs and receptors involved in angiogenesis versus lymphangiogenesis. *Curr Opin Cell Biol.* 2009; 21(2):154–65. [PubMed: 19230644]
8. Fischer C, et al. FLT1 and its ligands VEGFB and PlGF: drug targets for anti-angiogenic therapy? *Nat Rev Cancer.* 2008; 8(12):942–56. [PubMed: 19029957]
9. Jussila L, Alitalo K. Vascular growth factors and lymphangiogenesis. *Physiol Rev.* 2002; 82(3):673–700. [PubMed: 12087132]
10. Park JE, et al. Placenta growth factor. Potentiation of vascular endothelial growth factor bioactivity, in vitro and in vivo, and high affinity binding to Flt-1 but not to Flk-1/KDR. *J Biol Chem.* 1994; 269(41):25646–54. [PubMed: 7929268]
11. Holash J, et al. VEGF-Trap: a VEGF blocker with potent antitumor effects. *Proc Natl Acad Sci U S A.* 2002; 99(17):11393–8. [PubMed: 12177445]
12. Willett CG, et al. Efficacy, safety, and biomarkers of neoadjuvant bevacizumab, radiation therapy, and fluorouracil in rectal cancer: a multidisciplinary phase II study. *J Clin Oncol.* 2009; 27(18):3020–6. [PubMed: 19470921]
13. Rini BI, et al. Antitumor activity and biomarker analysis of sunitinib in patients with bevacizumab-refractory metastatic renal cell carcinoma. *J Clin Oncol.* 2008; 26(22):3743–8. [PubMed: 18669461]
14. Baffert F, et al. Cellular changes in normal blood capillaries undergoing regression after inhibition of VEGF signaling. *Am J Physiol Heart Circ Physiol.* 2006; 290(2):H547–59. [PubMed: 16172161]
15. Fukasawa M, Korc M. Vascular endothelial growth factor-trap suppresses tumorigenicity of multiple pancreatic cancer cell lines. *Clin Cancer Res.* 2004; 10(10):3327–32. [PubMed: 15161686]

16. Verheul HM, et al. Vascular endothelial growth factor trap blocks tumor growth, metastasis formation, and vascular leakage in an orthotopic murine renal cell cancer model. *Clin Cancer Res.* 2007; 13(14):4201–8. [PubMed: 17634549]
17. Le XF, et al. Specific blockade of VEGF and HER2 pathways results in greater growth inhibition of breast cancer xenografts that overexpress HER2. *Cell Cycle.* 2008; 7(23):3747–58. [PubMed: 19029832]
18. Hu L, et al. Vascular endothelial growth factor trap combined with paclitaxel strikingly inhibits tumor and ascites, prolonging survival in a human ovarian cancer model. *Clin Cancer Res.* 2005; 11(19 Pt 1):6966–71. [PubMed: 16203789]
19. Cheron MVPLP, Demers B, Leopold D, Bissery MC. Synergistic activity of aflibercept (VEGF Trap) in combination with 5-fluorouracil and irinotecan in preclinical tumor models: abstract A13. Proceedings from the AACR-NCI-EORTC: molecular targets and cancer therapeutics. 2007
20. Tew WP, et al. Phase I study of aflibercept administered subcutaneously to patients with advanced solid tumors. *Clin Cancer Res.* 2010; 16(1):358–66. [PubMed: 20028764]
21. Lockhart AC, et al. Phase I study of intravenous vascular endothelial growth factor trap, aflibercept, in patients with advanced solid tumors. *J Clin Oncol.* 2010; 28(2):207–14. This phase I trial of intravenously administered aflibercept established its safety and toxicity profile, as well as its recommended phase II dose. [PubMed: 19949018]
22. Khayat D, et al. Intravenous aflibercept administered in combination with irinotecan, 5-fluorouracil and leucovorin in patients with advanced solid tumours: results from the expansion cohort of a phase I study. *Eur J Cancer.* 2013; 49(4):790–7. [PubMed: 23312881]
23. Isambert N, et al. Phase I dose-escalation study of intravenous aflibercept in combination with docetaxel in patients with advanced solid tumors. *Clin Cancer Res.* 2012; 18(6):1743–50. [PubMed: 22261804]
24. Diaz-Padilla I, et al. A phase I dose-escalation study of aflibercept administered in combination with pemetrexed and cisplatin in patients with advanced solid tumours. *Br J Cancer.* 2012; 107(4):604–11. [PubMed: 22805331]
25. Van Cutsem E, et al. Phase I dose-escalation study of intravenous aflibercept administered in combination with irinotecan, 5-fluorouracil and leucovorin in patients with advanced solid tumours. *Eur J Cancer.* 2013; 49(1):17–24. [PubMed: 22921183]
26. Gaya A, Tse V. A preclinical and clinical review of aflibercept for the management of cancer. *Cancer Treat Rev.* 2012; 38(5):484–93. This excellent review comprehensively discusses the preclinical and clinical development of aflibercept. [PubMed: 22264850]
27. Tang PA, et al. Phase II clinical and pharmacokinetic study of aflibercept in patients with previously treated metastatic colorectal cancer. *Clin Cancer Res.* 2012; 18(21):6023–31. [PubMed: 22977191]
28. Pericay CFG, Saunders M, Thomas A, Roh JK, Lopez R, et al. Phase 2 randomized, noncomparative open-label study of aflibercept and modified FOLFOX6 in the first line treatment of metastatic colorectal cancer (AFFIRM): abstract O = 0024. *Ann Oncol.* 2012; 23(Suppl 4):iv5–18. [PubMed: 22774231]
29. Ramlau R, et al. Aflibercept and docetaxel versus docetaxel alone after platinum failure in patients with advanced or metastatic non-small-cell lung cancer: a randomized, controlled phase III trial. *J Clin Oncol.* 2012; 30(29):3640–7. [PubMed: 22965962]
30. Rougier P, et al. Randomised, placebo-controlled, double-blind, parallel-group phase III study evaluating aflibercept in patients receiving first-line treatment with gemcitabine for metastatic pancreatic cancer. *Eur J Cancer.* 2013; 49(12):2633–42. [PubMed: 23642329]
31. Tannock IF, et al. Aflibercept versus placebo in combination with docetaxel and prednisone for treatment of men with metastatic castration-resistant prostate cancer (VENICE): a phase 3, double-blind randomised trial. *Lancet Oncol.* 2013; 14(8):760–8. [PubMed: 23742877]
32. Van Cutsem E, et al. Addition of aflibercept to fluorouracil, leucovorin, and irinotecan improves survival in a phase III randomized trial in patients with metastatic colorectal cancer previously treated with an oxaliplatin-based regimen. *J Clin Oncol.* 2012; 30(28):3499–506. This pivotal phase III trial demonstrated a survival benefit of aflibercept in combination with FOLFIRI for

patients with oxaliplatin-pretreated metastatic CRC and led to its FDA approval for this indication. [PubMed: 22949147]

33. Hurwitz H, et al. Bevacizumab plus irinotecan, fluorouracil, and leucovorin for metastatic colorectal cancer. *N Engl J Med*. 2004; 350(23):2335–42. [PubMed: 15175435]
34. Macedo LT, da Costa Lima AB, Sasse AD. Addition of bevacizumab to first-line chemotherapy in advanced colorectal cancer: a systematic review and meta-analysis, with emphasis on chemotherapy subgroups. *BMC Cancer*. 2012; 12:89. [PubMed: 22414244]
- 35•. Bennouna J, et al. Continuation of bevacizumab after first progression in metastatic colorectal cancer (ML18147): a randomised phase 3 trial. *Lancet Oncol*. 2013; 14(1):29–37. This phase III trial demonstrated the survival benefit of continuing bevacizumab therapy in the second -line setting beyond progression with bevacizumab therapy in the first-line setting for metastatic CRC. [PubMed: 23168366]
- 36•. Grothey A, et al. Regorafenib monotherapy for previously treated metastatic colorectal cancer (CORRECT): an international, multicentre, randomised, placebo-controlled, phase 3 trial. *Lancet*. 2013; 381(9863):303–12. This phase III trial showed a survival benefit of single-agent regorafenib for patients with refractory meta-static CRC and led to its FDA approval for this indication. [PubMed: 23177514]
37. Bach PB, Saltz LB, Wittes RE. In cancer care, cost matters. *New York Times*. 2012 Oct 15.:A25.

Table 1

Completed phase III clinical trials of aflibercept

Phase III Clinical Trial	Patient Population	Treatment Regimens	Efficacy Outcomes (Aflibercept vs. Placebo)	Reference
VENICE	Metastatic castration-resistant prostate cancer	Aflibercept 6 mg/kg IV or placebo plus docetaxel IV 75 mg/m ² every 3 weeks and oral prednisone 5 mg twice daily	PFS: 6.9 vs. 6.2 mo; $p=0.31$	Tannock, et al. [31]
	$N=1224$		OS: 22.1 vs. 21.2 mo; $p=0.38$	
	First-line		RR: 38.4 vs. 28.1 %; $p=0.0043$	
VANILLA	Metastatic pancreatic cancer	Aflibercept 4 mg/kg IV or placebo every 2 weeks plus gemcitabine 1,000 mg/m ² IV weekly for 7 weeks out of 8, then weekly for 3 weeks out of 4	PFS: 3.7 vs. 3.7 mo; $p=0.8645$	Rougier, et al. [30]
	$N=427$ (stopped for futility at interim analysis)		OS: 6.5 vs. 7.8 mo; $p=0.2034$	
	First-line			
VITAL	Platinum-retreated advanced or metastatic non-small cell lung cancer (NSCLC)	Aflibercept 6 mg/kg IV or placebo plus docetaxel 75 mg/m ² IV every 3 weeks	PFS: 5.2 vs. 4.1 mo; $p=0.0035$	Ramlau, et al. [29]
	$N=913$		OS: 10.1 vs. 10.4 mo; $p=0.90$	
	Second-line		RR: 23.3 vs. 8.9 %; $p<0.001$	
VELOUR	Oxaliplatinpretreated metastatic colorectal cancer	Aflibercept 4 mg/kg IV or placebo plus FOLFIRI (irinotecan 180 mg/m ² , leucovorin 400 mg/m ² , bolus 5-FU 400 mg/m ² , infusional 5-FU 2,400 mg/m ²) IV every 2 weeks	PFS: 6.90 vs. 4.67 mo; $p<0.0001$	Van Cutsem, et al. [32•]
	$N=1226$		OS: 13.50 vs. 12.06 mo; $p=0.0032$	
	Second-line		RR: 19.8 vs. 11.1 %; $p=0.0001$	