Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

CCR Drug Updates

Clinical Cancer Research

Crizotinib for *ALK*-Rearranged Non–Small Cell Lung Cancer: A New Targeted Therapy for a New Target

Leena Gandhi[1,2,4] and Pasi A. Jänne[1–4]

Abstract

Crizotinib (PF02341066, Xalkori; Pfizer) was recently approved by the U.S. Food and Drug Administration for treatment of *ALK*-positive non–small cell lung cancer (NSCLC) as defined by a jointly approved diagnostic test using a break-apart fluorescence *in situ* hybridization assay. The approval was based on dramatic response rates in *ALK*-positive NSCLC patients of 54% to 61% in phase I and II trials. To date, the overall disease control rates in these trials are close to 90%. Progression-free survival approaches 10 months. This review focuses on the ALK-inhibitory activity of crizotinib in preclinical and clinical trials that led to approval, as well as the diagnostic methods to classify patients with *ALK*-positive NSCLC. Although these patients represent a small subset of all patients with NSCLC, the rapid time course from identification of this unique target to an approved targeted therapy with striking benefit serves as a paradigm for the development of targeted therapeutics in an era of personalized medicine. *Clin Cancer Res; 18(14); 3737–42. ©2012 AACR.*

Introduction

Non–small cell lung cancer (NSCLC) is the most common cause of cancer-related deaths in both men and women in the United States. Most patients present with advanced disease, and standard chemotherapeutic agents have marginal effects on survival. However, select populations of patients with NSCLC carry somatic genomic activating mutations that, when present, seem to singularly drive the growth of the cancer. The most frequent examples of this are mutations in the epidermal growth factor receptor (EGFR), which are present in ∼15% of all patients with NSCLC in the United States. EGFR-targeted tyrosine kinase inhibitors (TKI) show markedly greater response rates and progression-free survival than first-line chemotherapy in *EGFR*-mutant patients (1–3).

Anaplastic Lymphoma Kinase Rearrangement in NSCLC

Anaplastic lymphoma kinase (ALK) rearrangements were first identified as a fusion to a portion of the nucleophosmin (NPM) gene in 60% of anaplastic large cell lymphoma, a small subset of non-Hodgkin lymphomas.

ALK rearrangements in NSCLC were identified in late 2007, primarily as fusions to echinoderm microtubule-like protein 4 [EML4 (4, 5)]. These fusion proteins are found in ∼3% to 7% of patients with NSCLC overall but have been identified primarily in lung adenocarcinomas and more frequently in younger patients and in never or light smokers (4, 6, 7). Like *EGFR*, *KRAS*, *ERBB2*, and *BRAF*, they are mostly found exclusive of other known activating mutations (8, 9).

Although the most frequent fusion partner is *EML4*, several different breakpoints of *EML4* have been described (4, 10, 11). In addition, other, rare fusion partners have also been described, such as *TFG* and *KIF5B* (12). Most fusions link to *ALK* in frame with exon 20, and the resulting protein carries a coiled-coil basic domain from the upstream fusion partner, which may facilitate activation of the ALK tyrosine kinase by promoting dimerization (4).

Most of the identified EML4-ALK fusion proteins have been shown to be oncogenic both *in vitro* and *in vivo* (4, 10). EML4-ALK variant 1 expressed in mouse fibroblasts generates tumors in nude mice and results in multiple adenocarcinomas when expressed in the lungs of transgenic mice (4, 13, 14). Although the detailed mechanism of oncogenic activation by ALK is not fully characterized, overexpression in cell lines results in activation of both STAT3 and extracellular signal-regulated kinase (ERK) (15). Studies of crizotinib and TAE684, another small-molecule ALK inhibitor, have shown downregulation of both AKT and ERK phosphorylation (16, 17). One study showed that TAE684 results in apoptosis mediated by BIM upregulation that is ERK dependent, and survivin downregulation that is STAT3 dependent (15).

Crizotinib

Crizotinib is a potent and specific small-molecule inhibitor of both ALK and c-MET tyrosine kinases. It is a 3-benzyloxy-2-aminopyridine series inhibitor that was derived from a first-generation c-MET inhibitor,

Authors' Affiliations: [1]Lowe Center for Thoracic Oncology, [2]Department of Medical Oncology, and [3]Belfer Institute for Applied Cancer Science, Dana-Farber Cancer Institute; [4]Department of Medicine, Brigham and Women's Hospital, Boston, Massachusetts

Corresponding Author: Pasi A. Jänne, Lowe Center for Thoracic Oncology, Dana-Farber Cancer Institute, 450 Brookline Avenue, HIM223, Boston, MA 02215. Phone: 617-632-6076; Fax: 617-582-7683; E-mail: pjanne@partners.org

doi: 10.1158/1078-0432.CCR-11-2393

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

PHA-66752, using the cocrystal structure of this inhibitor with c-MET to further optimize active-site binding (18). In a screen for kinase selectivity, crizotinib inhibited autophosphorylation of both c-MET and ALK with high potency and specificity across a panel of >120 kinases (18). Crizotinib binds in a bidentate manner at the hinge region of c-MET and competes with ATP binding in both kinases [a crystal structure of crizotinib binding in the inactive conformation of ALK has been reported as well (PDB ID: 2XP2)].

Pharmacokinetics and pharmacodynamics

Crizotinib blocks c-MET phosphorylation and c-MET–dependent cell growth and invasion in a variety of cell lines at an IC_{50} of 5 to 20 nmol/L (18, 19). The IC_{50} of crizotinib in blocking cell proliferation and apoptosis in ALK-rearranged lymphoma cell lines is 25 to 50 nmol/L (18, 20). The IC_{50} of crizotinib in NSCLC lines carrying an *EML4-ALK* fusion is significantly smaller, ranging from 250 to 340 nmol/L (16, 21). This is notably less potent than other ALK inhibitors in development (17).

In a pharmacokinetic/pharmacodynamic modeling study from Pfizer, crizotinib was projected to reach adequate plasma levels to inhibit ALK phosphorylation and achieve an antitumor effect. Specifically, in xenograft models using either H3122 cells (NSCLC line with EML4-ALK fusion) or Karpas299 cells (ALCL line with NPM-ALK fusion), ALK inhibition was seen at an EC_{50} of 233 and 660 nmol/L, respectively, and tumor growth inhibition was observed at 255 and 875 nmol/L, suggesting that $\geq$50% ALK inhibition is required for antitumor efficacy (22). On the basis of clinical pharmacokinetic data, >70% ALK inhibition was projected at the now U.S. Food and Drug Administration (FDA)–approved dose of 250 mg twice daily.

In a phase I clinical study of crizotinib, patients treated at the recommended phase II dose were noted to reach a C_{max} of crizotinib at a median T_{max} of 4 hours, with a terminal half-life of 42 hours after a single dose (23). Steady-state levels were reached within 15 days, with nonlinear pharmacokinetics (decreasing mean value of clearance with multiple dosing). This is believed to be due to autoinhibition of CYP3A after reaching steady state. Because of these observations, the phase I study has been amended to evaluate once-daily dosing, and to date, safety has been observed at doses up to 650 mg daily (K. Wilner, Pfizer, unpublished data). Of note, the C_{max} and area under the curve (AUC) for crizotinib in a population of Asian patients were ~1.5-fold greater than those in non-Asian patients, but without increased toxicities.

Safety

Crizotinib was well tolerated overall, with rare grade 3/4 toxicities that included pneumonitis (1.6% across phase I and II studies) that usually occurred within 2 months of initiating the drug (24). Two deaths attributed to treatment-related pneumonitis were reported in the phase II study (25). Grade 3/4 alanine aminotransferase elevation (4% in the phase I study and 7% in stage II) was also noted, resulting in 1 treatment discontinuation. Overall, elevated transaminases were attributable to the drug in 19% of the patients.

More common treatment-related toxicities included visual disturbances, which occurred in 62% of the patients, but all were grade 1 or 2 effects. These were primarily described as light trails (following a moving lighted object) or light flashes or brief image persistence (i.e., following camera flashbulb use). Flipped dark–light registration in high-contrast images was also reported. No ophthalmologic abnormalities were noted in the limited examinations that were performed.

Other frequent treatment-related toxicities that were primarily grade 1 or 2 included nausea (53%), diarrhea (43%), vomiting (40%), constipation (27%), edema (28%), fatigue (20%), anorexia (19%), dizziness (16%), neuropathy (13%), and dysgeusia (12%). Rash was noted in 10% of the patients, and reflux or other esophageal symptoms were observed in 11%. After a food-effect evaluation was done, yielding findings of only a 14% reduction in C_{max} and AUC after a high-fat meal, patients were allowed to take the drug in a nonfasting state, which resulted in a substantial reduction of nausea and vomiting.

Clinical efficacy

Preclinical data suggested the potential efficacy of crizotinib against both MET and ALK; however, it was the dramatic results seen in *ALK*-positive patients with NSCLC that served as the impetus for accelerated FDA approval. Following responses seen in 2 *ALK*-positive patients during dose escalation, the phase I study of crizotinib in solid tumors was amended to include an expanded cohort of *ALK*-positive patients with lung cancer, into which 119 patients have been enrolled. The overall response rate among these patients is 61% (26), with an overall clinical benefit rate (complete response + partial response + stable disease) of 88%. In patients who were chemotherapy naïve ($n = 14$), the response rate was 68%. Preliminary median survival was 10 months. These striking data served as the basis for rapid initiation of both a phase II trial of crizotinib for patients who have relapsed following first-line chemotherapy and a phase III randomized trial of crizotinib versus second-line therapy. As reported to date, in the phase II trial, the response rate in the population of 76 patients evaluable for response was 54%, with a 91% disease control rate [partial response + stable disease (25)].

These results led to the accelerated FDA approval of crizotinib for *ALK*-positive patients with NSCLC in record time—the timeframe from discovery of the target in late 2007 to FDA approval of a targeted therapy in August 2011 was <4 years (Fig. 1). The approval was granted while both the phase I and phase II trials were ongoing. In addition, a phase III trial comparing crizotinib with standard second-line therapy is still accruing, and a more recent phase III trial comparing crizotinib with first-line platinum doublet chemotherapy has been launched as well. Of importance, the FDA approval was not linked to any line of therapy, and given the commercial availability of crizotinib, accrual to

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

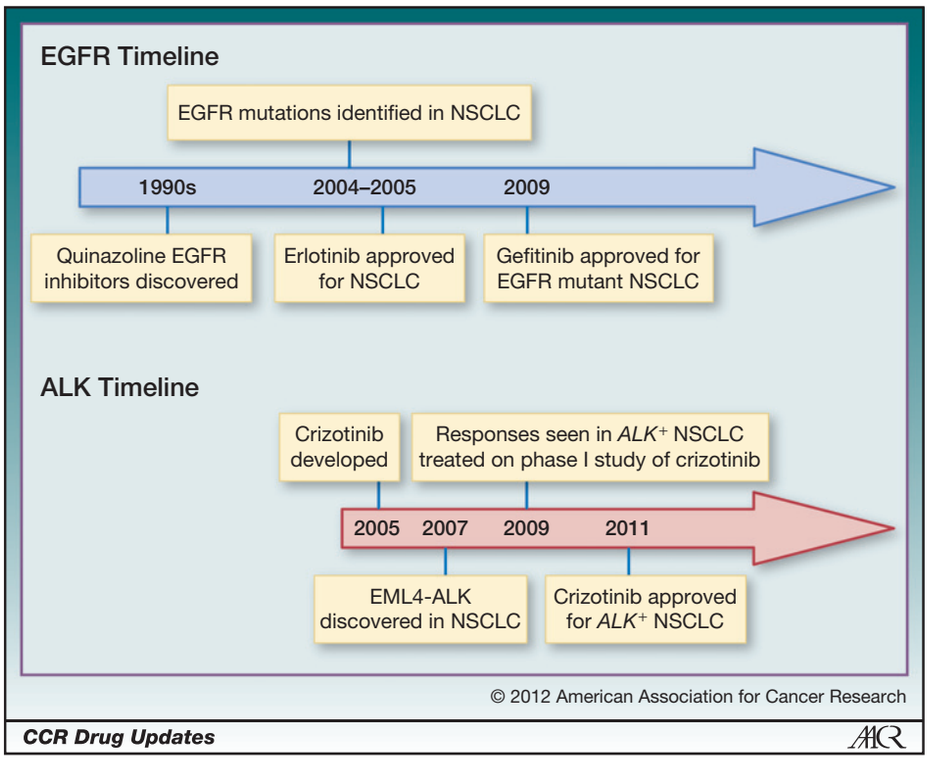

Figure 1. Timeline for approval of kinase inhibitors for molecular subsets of NSCLC. EGFR inhibitors were approved initially for a broader population and subsequently for *EGFR*-mutant patients with NSCLC. In contrast, clinical development and approval of crizotinib have been limited to *ALK*-positive patients with NSCLC.

these large phase III trials, especially the first-line trial, may be slowed.

The approval of the drug also makes a prospective evaluation of the overall survival benefit of crizotinib challenging. Although EGFR-directed TKIs produce markedly better results than first-line chemotherapy in terms of progression-free survival for *EGFR*-mutated patients, the overall survival is not statistically different because most patients are likely to receive an EGFR TKI during some point of their therapy (1–3), and the same is likely to be true of crizotinib. A retrospective study examining the outcomes of 36 *ALK*-positive patients who were not treated with crizotinib compared with 56 patients who were treated in the phase I expanded cohort of crizotinib showed that patients treated in the second or third line showed improved overall survival compared with those treated with other therapies [HR 0.36; 95% confidence interval (CI), 0.17–0.75; $P = 0.004$ (27)].

Future clinical directions

Although much of the attention on crizotinib has focused on its activity against *ALK*-positive NSCLC, equally dramatic responses have been seen against *ALK*-positive lymphoma (28) as well as *MET*-amplified gastroesophageal cancers and NSCLC (29, 30). An independent study of crizotinib in *ALK*-positive lymphoma is now ongoing (ClinicalTrials.gov, identifier NCT00939770). In addition, *in vitro* studies identified a *ROS1*-rearranged cell line as being sensitive to TAE684, another ALK inhibitor (16), and this alteration has been identified in clinical specimens as well (5). An effort to look for *ROS1* translocations (using a break-apart FISH probe similar to that employed for *ALK*) showed *ROS1* rearrangements in 18 of 1,073 of NSCLC patients screened (31). One of these patients was treated in the expanded cohort of the phase I study of crizotinib and achieved a dramatic and sustained response similar to those observed in *ALK*-positive patients. These observations suggest a broader role for crizotinib as an effective therapy against multiple targets in multiple tumor types.

ALK Diagnostics

Although the phase I trial of crizotinib allowed several different local laboratory determinations of *ALK* rearrangement for approval of eligibility, the phase II trial mandated central testing for *ALK* rearrangement using the now FDA-approved Vysis ALK Break Apart FISH Probe Kit (Abbott Molecular). The test, which uses a red and a green probe flanking the *ALK* translocation breakpoint, is considered positive if ≥15% of at minimum 50 nuclei evaluated have a split or isolated red signal that results from the intervening fusion of a translocated protein fragment such as *EML4*. Although this approach was clinically validated in the sense that it was the test used in crizotinib trials, in comparison with other methods, it is associated with several practical challenges. FISH requires specialized equipment and expertise, and it can cost more than $1,000 per test. In addition, the interpretation is sometimes not clear-cut depending on how close the signals are to each other. Multiple alternative methods of evaluation have been and are being studied, but each has potential drawbacks (Table 1).

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

Table 1. Methods to diagnose ALK rearrangements

Test	Advantages	Disadvantages
Break-apart FISH	Can detect any rearrangement	Cost
	Performed on FFPE tissue	Expertise required
	Used clinically in crizotinib trials	Often difficult to interpret
CISH	Cost	Expertise required
	Performed on FFPE tissue	Often difficult to interpret
	Simultaneous histology evaluation	
Reverse transcriptase PCR	Needs minimal material	Will not identify new fusions
	Can identify fusion partners	RNA degradation
		Possibility of contamination
IHC	Cost	Low-level ALK expression
	Can be performed in any laboratory	No preferred antibody as yet
	Performed on FFPE tissues	

Abbreviations: FFPE, formalin-fixed paraffin-embedded; IHC, immunohistochemistry.

Chromogenic *in situ* hybridization (CISH) offers a lower-cost alternative to FISH and allows simultaneous histologic evaluation. A study that used CISH for detection of ALK rearrangement showed a high concordance with FISH [95% (32)]. However, CISH does not get around the problem of a sometimes subtle or difficult-to-interpret hybridization pattern.

Reverse transcriptase PCR (RT-PCR) is an alternative diagnostic method that uses multiplexed primer pairs to amplify known possible ALK fusions. This test has been shown to be sensitive and specific for detecting and identifying particular ALK fusions but will by definition miss as-yet-uncharacterized fusion partners (10). In addition, the success of the test depends on the quality of the sample and whether the RNA is degraded. The test has been optimized on formalin-fixed paraffin-embedded (FFPE) specimens in addition to fresh tissue, but the quality of the former is more variable.

Immunohistochemistry (IHC) for ALK protein is another alternative test, because there is essentially no ALK expression in normal lung epithelia. However, even within ALK-positive lung tumors, the expression of ALK is relatively low (compared with ALK expression in *ALK*-rearranged lymphoma), and antibodies that are used routinely for detection of ALK-positive lymphoma, such as the DAKO clone ALK1, showed specificity but relatively low sensitivity for detecting ALK in NSCLC (33). However, increased concentrations of antibody and the use of novel methods to amplify the secondary signal did result in improved sensitivity and higher concordance with FISH (34).

Other antibodies also have improved sensitivity and higher concordance with FISH, including DF3 (developed by Cell Signaling Technology), which showed a sensitivity of 100% and specificity of 99%, and much improved staining compared with the ALK1 antibody (35). Another promising antibody for routine use is the 5A4 antibody from Novocastra, which showed a sensitivity of 100% and specificity of 96% (36). However, moderate staining (+2) is more variable in determining *ALK* rearrangement as detected by other methods (36, 37).

Studies comparing all 3 methods (excluding CISH) have not shown a clear superiority for all aspects of any one method (reviewed in ref. 38). Rather, they suggest that ALK testing may one day mirror HER2 testing in breast cancer, with an initial screen by IHC (using an as-yet-to-be-agreed-upon antibody and IHC procedure) with grading of intensity on a scale of +1 to +3, followed by FISH confirmation of moderate staining. RT-PCR may be used to identify specific fusion partners when indicated.

Crizotinib Resistance

Additional methods for ALK evaluation will also be required to streamline the detection of resistance mutations. Several resistance mutations have already been identified in patients treated with crizotinib (12, 17, 39). The first report of 2 acquired point mutations, C1156Y and L1196M, was published concurrently with the first report of activity of crizotinib (12). Although some of the identified resistance mutations, such as L1196M and G1269A, are found within the active site and likely interfere with crizotinib binding (L1196M is analogous to resistance mutations involving a gatekeeper residue found in *EGFR* and *ABL*), other mutations (i.e., L1152 and C1156Y) do not have a clear mechanism for causing resistance to crizotinib (40). Studies of acquired resistance *in vitro* (41, 42) have identified these clinically identified mutations as well as other resistance mutations, all of which have variable resistance to crizotinib *in vitro*. In the 7 mutations that have been clinically identified to date, the degree of resistance to crizotinib is variable. Some are still sensitive to structurally different ALK inhibitors with greater potency than crizotinib, as well as to Hsp90 inhibitors, which are clinically active against *ALK*-positive NSCLCs (17).

Unlike *EGFR* resistance to erlotinib, which occurs by a common mechanism in >50% of cases (i.e., the T790M

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

gatekeeper mutation), *ALK* resistance seems to be more variable. Not only have more varied mutations been identified but also in many cases no identified *ALK* secondary mutation has been identified. EGFR activation and *ALK* copy number gain have been identified as mechanisms of acquired resistance in both patients and *in vitro* models (17, 31, 39, 40). Clinically, *KIT* amplification (with concomitant increased expression of the KIT ligand, SCF) and the emergence of independent *KRAS*-mutant clones have also been identified as mechanisms of resistance to crizotinib (17, 39). The largest series published to date suggests that <25% of resistance occurs via acquisition of a secondary point mutation and >50% of resistance may occur via alternate tyrosine kinase activation. With the use of more potent ALK inhibitors, alternate pathway activation may become an even more important issue with which investigators will have to grapple.

Conclusions

ALK-targeted therapy, as well as the identification of *ALK*-positive NSCLC tumors, is a rapidly evolving field that has been put on the map by crizotinib.

Although future refinements in diagnostic tests are likely and more potent inhibitors are currently in clinical development, crizotinib and the break-apart FISH test have clearly distinguished a group of NSCLC patients with dramatically improved outcomes compared with the majority of patients. This example of a molecularly defined target and an effective therapeutic strategy mirrors the experience with *EGFR*-mutant NSCLC and EGFR-directed TKIs, and, it is hoped, will serve as a model for more examples to come.

Disclosure of Potential Conflicts of Interest

L. Gandhi is a consultant/advisory board member of Chugai Corporation. P.A. Janne is a consultant/advisory board member of Pfizer, Chugai Pharmaceuticals, and Abbott Molecular.

Authors' Contributions

Conception and design: L. Gandhi, P.A. Janne
Writing, review, and/or revision of the manuscript: L. Gandhi, P.A. Janne

Grant Support

National Institutes of Health (R01CA136851 to P.A. Jänne); National Cancer Institute Lung SPORE (P50CA090578 to P.A. Jänne).

Received February 23, 2012; revised April 9, 2012; accepted April 17, 2012; published OnlineFirst April 30, 2012.

References

1. Mok TS, Wu YL, Thongprasert S, Yang CH, Chu DT, Saijo N, et al. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med 2009;361:947–57.
2. Maemondo M, Inoue A, Kobayashi K, Sugawara S, Oizumi S, Isobe H, et al.North-East Japan Study Group. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N Engl J Med 2010;362:2380–8.
3. Zhou C, Wu YL, Chen G, Feng J, Liu XQ, Wang C, et al. Erlotinib versus chemotherapy as first-line treatment for patients with advanced EGFR mutation-positive non-small-cell lung cancer (OPTIMAL, CTONG-0802): a multicentre, open-label, randomised, phase 3 study. Lancet Oncol 2011;12:735–42.
4. Soda M, Choi YL, Enomoto M, Takada S, Yamashita Y, Ishikawa S, et al. Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer. Nature 2007;448:561–6.
5. Rikova K, Guo A, Zeng Q, Possemato A, Yu J, Haack H, et al. Global survey of phosphotyrosine signaling identifies oncogenic kinases in lung cancer. Cell 2007;131:1190–203.
6. Koivunen JP, Mermel C, Zejnullahu K, Murphy C, Lifshits E, Holmes AJ, et al. EML4-ALK fusion gene and efficacy of an ALK kinase inhibitor in lung cancer. Clin Cancer Res 2008;14:4275–83.
7. Shaw AT, Yeap BY, Mino-Kenudson M, Digumarthy SR, Costa DB, Heist RS, et al. Clinical features and outcome of patients with non-small-cell lung cancer who harbor EML4-ALK. J Clin Oncol 2009;27:4247–53.
8. Inamura K, Takeuchi K, Togashi Y, Hatano S, Ninomiya H, Motoi N, et al. EML4-ALK lung cancers are characterized by rare other mutations, a TTF-1 cell lineage, an acinar histology, and young onset. Modern Pathol 2009;22:508–15.
9. Kris MG, Johnson BE, Kwiatkowski DJ, Iafrate AJ, Wistuba II, Aronson SL, et al. Identification of driver mutations in tumor specimens from 1000 patients with lung adenocarcinoma: the NCI's Lung Cancer Mutation Consortium (LCMC). J Clin Oncol 29: 2011 (suppl; abstr CRA7506).
10. Takeuchi K, Choi YL, Soda M, Inamura K, Togashi Y, Hatano S, et al. Multiplex reverse transcription-PCR screening for EML4-ALK fusion transcripts. Clin Cancer Res 2008;14:6618–24.
11. Sasaki T, Rodig SJ, Chirieac LR, Jänne PA. The biology and treatment of EML4-ALK non-small cell lung cancer. Eur J Cancer 2010;46:1773–80.
12. Choi YL, Soda M, Yamashita Y, Ueno T, Takashima J, Nakajima T, et al. ALK Lung Cancer Study Group. EML4-ALK mutations in lung cancer that confer resistance to ALK inhibitors. N Engl J Med 2010;363:1734–9.
13. Soda M, Takada S, Takeuchi K, Choi YL, Enomoto M, Ueno T, et al. A mouse model for EML4-ALK-positive lung cancer. Proc Natl Acad Sci U S A 2008;105:19893–7.
14. Chen Z, Sasaki T, Tan X, Carretero J, Shimamura T, Li D, et al. Inhibition of ALK, PI3K/MEK, and HSP90 in murine lung adenocarcinoma induced by EML4-ALK fusion oncogene. Cancer Res 2010;70:9827–36.
15. Takezawa K, Okamoto I, Nishio K, Jänne PA, Nakagawa K. Role of ERK-BIM and STAT3-survivin signaling pathways in ALK inhibitor-induced apoptosis in EML4-ALK-positive lung cancer. Clin Cancer Res 2011;17:2140–8.
16. McDermott U, Iafrate AJ, Gray NS, Shioda T, Classon M, Maheswaran S, et al. Genomic alterations of anaplastic lymphoma kinase may sensitize tumors to anaplastic lymphoma kinase inhibitors. Cancer Res 2008;68:3389–95.
17. Katayama R, Shaw AT, Khan TM, Mino-Kenudson M, Solomon BJ, Halmos B, et al. Mechanisms of acquired crizotinib resistance in ALK-rearranged lung cancers. Sci Transl Med 2012;4:120ra17.
18. Cui JJ, Tran-Dubé M, Shen H, Nambu M, Kung PP, Pairish M, et al. Structure based drug design of crizotinib (PF-02341066), a potent and selective dual inhibitor of mesenchymal-epithelial transition factor (c-MET) kinase and anaplastic lymphoma kinase (ALK). J Med Chem 2011;54:6342–63.
19. Zou HY, Li Q, Lee JH, Arango ME, McDonnell SR, Yamazaki S, et al. An orally available small-molecule inhibitor of c-Met, PF-2341066, exhibits cytoreductive antitumor efficacy through antiproliferative and antiangiogenic mechanisms. Cancer Res 2007;67:4408–17.
20. Christensen JG, Zou HY, Arango ME, Li Q, Lee JH, McDonnell SR, et al. Cytoreductive antitumor activity of PF-2341066, a novel inhibitor of anaplastic lymphoma kinase and c-Met, in experimental models of anaplastic large-cell lymphoma. Mol Cancer Ther 2007;6:3314–22.

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

21. Costa DB, Kobayashi S, Pandya SS, Yeo WL, Shen Z, Tan W, et al. CSF concentration of the anaplastic lymphoma kinase inhibitor crizotinib. J Clin Oncol 2011;29:e443–5.
22. Yamazaki S, Vicini P, Shen Z, Zou HY, Lee J, Li Q, et al. Pharmacokinetic-pharmacodynamic modeling of crizotinib for anaplastic lymphoma kinase inhibition and anti-tumor efficacy in human tumor xenograft mouse models. J Pharmacol Exp Ther 2012;340: 549–57.
23. Li C, Alvey C, Bello KD, Wilner KD, Tan WPfizer Inc. Pharmacokinetics (PK) of crizotinib (PF-02341066) in patients with advanced non-small cell lung cancer (NSCLC) and other solid tumors. J Clin Oncol 29: 2011 (suppl; abstr e13065).
24. U.S. Food and Drug Administration [homepage on the Internet]. Xalkori (crizotinib) capsules—safety [updated 2011; cited 2012 Jan 5]. Available from: http://www.accessdata.fda.gov/drugsatfda_docs/nda/2011/202570Orig1s000MedR.pdf
25. Crinò L, Kim D, Riely GJ, Janne PA, Blackhall FH, Camidge DR, et al. Initial phase II results with crizotinib in advanced ALK-positive non-small cell lung cancer (NSCLC): PROFILE 1005. J Clin Oncol 29: 2011 (suppl; abstr 7514).
26. Camidge DR, Bang Y, Kwak EL, Shaw AT, Iafrate AJ, Maki RG, et al. Progression-free survival (PFS) from a phase I study of crizotinib (PF-02341066) in patients with ALK-positive non-small cell lung cancer. J Clin Oncol 29: 2011 (suppl; abstr 2501).
27. Shaw AT, Yeap BY, Solomon BJ, Riely GJ, Gainor J, Engelman JA, et al. Effect of crizotinib on overall survival in patients with advanced non-small-cell lung cancer harbouring ALK gene rearrangement: a retrospective analysis. Lancet Oncol 2011;12: 1004–12.
28. Gambacorti-Passerini C, Messa C, Pogliani EM. Crizotinib in anaplastic large-cell lymphoma. N Engl J Med 2011;364:775–6.
29. Ou SH, Kwak EL, Siwak-Tapp C, Dy J, Bergethon K, Clark JW, et al. Activity of crizotinib (PF02341066), a dual mesenchymal-epithelial transition (MET) and anaplastic lymphoma kinase (ALK) inhibitor, in a non-small cell lung cancer patient with de novo MET amplification. J Thorac Oncol 2011;6:942–6.
30. Lennerz JK, Kwak EL, Ackerman A, Michael M, Fox SB, Bergethon K, et al. MET amplification identifies a small and aggressive subgroup of esophagogastric adenocarcinoma with evidence of responsiveness to crizotinib. J Clin Oncol 2011;29:4803–10.
31. Bergethon K, Shaw AT, Ou SH, Katayama R, Lovly CM, McDonald NT, et al. ROS1 rearrangements define a unique molecular class of lung cancers. J Clin Oncol 2012;30:863–70.
32. Kim H, Yoo SB, Choe JY, Paik JH, Xu X, Nitta H, et al. Detection of ALK gene rearrangement in non-small cell lung cancer: a comparison of fluorescence in situ hybridization and chromogenic in situ hybridization with correlation of ALK protein expression. J Thorac Oncol 2011;6:1359–66.
33. Rodig SJ, Mino-Kenudson M, Dacic S, Yeap BY, Shaw A, Barletta JA, et al. Unique clinicopathologic features characterize ALK-rearranged lung adenocarcinoma in the western population. Clin Cancer Res 2009;15:5216–23.
34. Yi ES, Boland JM, Maleszewski JJ, Roden AC, Oliveira AM, Aubry MC, et al. Correlation of IHC and FISH for ALK gene rearrangement in non-small cell lung carcinoma: IHC score algorithm for FISH. J Thorac Oncol 2011;6:459–65.
35. Mino-Kenudson M, Chirieac LR, Law K, Hornick JL, Lindeman N, Mark EJ, et al. A novel, highly sensitive antibody allows for the routine detection of ALK-rearranged lung adenocarcinomas by standard immunohistochemistry. Clin Cancer Res 2010;16:1561–71.
36. Paik JH, Choe G, Kim H, Choe JY, Lee HJ, Lee CT, et al. Screening of anaplastic lymphoma kinase rearrangement by immunohistochemistry in non-small cell lung cancer: correlation with fluorescence in situ hybridization. J Thorac Oncol 2011;6:466–72.
37. Just PA, Cazes A, Audebourg A, Cessot A, Pallier K, Danel C, et al. Histologic subtypes, immunohistochemistry, FISH or molecular screening for the accurate diagnosis of ALK-rearrangement in lung cancer: a comprehensive study of Caucasian non-smokers. Lung Cancer 2011;76:309–15.
38. Shaw AT, Solomon B, Kenudson MM. Crizotinib and testing for ALK. J Natl Compr Canc Netw 2012;9:1335–41.
39. Doebele RC, Pilling AB, Aisner DL, Kutateladze TG, Le AT, Weickhardt AJ, et al. Mechanisms of resistance to crizotinib in patients with ALK gene rearranged non-small cell lung cancer. Clin Cancer Res 2012;18: 1472–82.
40. Sasaki T, Koivunen J, Ogino A, Yanagita M, Nikiforow S, Zheng W, et al. A novel ALK secondary mutation and EGFR signaling cause resistance to ALK kinase inhibitors. Cancer Res 2011;71: 6051–60.
41. Katayama R, Khan TM, Benes C, Lifshits E, Ebi H, Rivera VM, et al. Therapeutic strategies to overcome crizotinib resistance in non-small cell lung cancers harboring the fusion oncogene EML4-ALK. Proc Natl Acad Sci U S A 2011;108:7535–40.
42. Zhang S, Wang F, Keats J, Zhu X, Ning Y, Wardwell SD, et al. Crizotinib-resistant mutants of EML4-ALK identified through an accelerated mutagenesis screen. Chem Biol Drug Des 2011;78:999–1005.

Published OnlineFirst April 30, 2012; DOI: 10.1158/1078-0432.CCR-11-2393

Clinical Cancer Research

Crizotinib for *ALK*-Rearranged Non–Small Cell Lung Cancer: A New Targeted Therapy for a New Target

Leena Gandhi and Pasi A. Jänne

Clin Cancer Res 2012;18:3737-3742. Published OnlineFirst April 30, 2012.