Kasr El-Aini J. Clin. Oncol. Nucl. Med.
Vol. 10, No. 3-4, Jul-Oct. 2014:47-55

Original article

EGFR Inhibitors in Treatment of Lung Adenocarcinoma

Ehab Ibrahim

Kasr El-Aini Centre of Clinical Oncology and Nuclear Medicine, Faculty of Medicine, Cairo University, Egypt

The aim of this review is to provide an overview of current molecular targeted therapies for epidermal growth factor receptor (EGFR) mutation-positive lung adenocarcinoma. Current strategies aim to identify patients with driver mutations such as epidermal growth factor receptor gene mutations and anaplastic lymphoma kinase rearrangements. Previous treatment strategies have been less than satisfactory regarding response rates and significant systemic toxicities. EGFR has been shown to be deregulated by various mechanisms in lung adenocarcinoma (AC), including overexpression, amplification and mutation. Reversible EGFR tyrosine kinase inhibitors (TKIs) such as Erlotinib and Gefitinib offered a therapeutic alternative that has proven its superiority over standard platinum-based chemotherapy for patients with metastatic EGFR mutation-positive lung cancer. Irreversible EGFR tyrosine kinase inhibitors (TKIs) such as Afatinib and Dacomitinib came into practice to provide sustained disease control in adenocarcinoma of the lung with primary or acquired resistance to first generation EGFR TKIs.

Key words: Targeted therapy, NSCLC, Mutation, TKIs, Resistance.
Corresponding Author: Ehab Ibrahim, MD **E-mail:** ehabhas@hotmail.com

INTRODUCTION

Lung cancer is the leading cause of cancer death worldwide. Approximately 80% of lung cancers are grouped as non-small cell lung carcinoma (NSCLC), which are clinically and pathologically different from small cell lung carcinoma (SCLC). Adenocarcinoma (AC) of lung constitutes about 40% to 50% of all lung cancers. NSCLC also includes squamous cell carcinoma (SQCC) and large cell carcinoma (LCLC)[1, 2].

The epidermal growth factor receptor (EGFR) family of receptor tyrosine kinases (RTKs), referred to as the HER or ErbB family, consists of four members namely: (HER1/ErbB1), (HER2/ErbB2), (HER3/ErbB3) and (HER4/ErbB4). They regulate many developmental, metabolic and physiological processes. The intracellular TK activity of EGFR is increased as a consequence of the binding of various cognate ligands, which include EGF, transforming growth factor-α, amphiregulin and others leading to the homodimerization of two EGFRs or the heterodimerization of EGFR with other family members, most commonly HER2.[3] Heterodimerization with HER2, which is over-expressed in some tumors, is a more potent activator of EGFR TK than is EGFR homodimerization. The activation of receptor TK leads to the autophosphorylation of the intracellular domain of EGFR, and the phosphotyrosine residues that are formed act as a docking site for various adapter molecules, resulting in the activation of the Ras/mitogen-activated protein kinase pathway, the PI3K/Akt pathway and signal transducers and activators of transcription signaling pathways[4].

EGFR mutations have been observed in NSCLC especially in adenocarcinoma which results in dysregulation of cellular growth and proliferation.[5] Based on these findings EGFR tyrosine kinase inhibitors (TKIs) have been utilized as part of treatment strategies for selected patients with NSCLC[6]. The presence of an *EGFR* mutation strongly predicts likelihood of response to TKI therapy, with an observed response rate of about 80% among individuals whose tumors express the mutation and only 10% among those whose tumors do not[7].

Several types of activating mutations are known to occur in EGFR in NSCLC: Class I - exon 19 in-frame deletions (44% of all EGFR mutations), Class II - single amino acid changes (L858R 41%, G719 4%, other missense mutations 6%) and Class III - exon 20 in-frame duplication/insertions (5%). These mutations occur in the tyrosine kinase domain of EGFR. Eighty-five percent of all EGFR-activating mutations are exon 19 in-frame deletions or L858R, and they tend to be sensitive to currently available EGFR inhibitors. Class III mutations (exon 20) are generally insensitive to EGFR inhibitors[8].

Higher EGFR mutation frequency is observed in non-smokers, women, and in non-mucinous tumors. EGFR has been shown to be dysregulated by various mechanisms in adenocarcinoma, including overexpression, amplification, and mutation[9].

In unselected patients, clinicopathological features like adenocarcinoma histology (40% vs 3% in other histologies), East Asian descent (30% vs 8% in non-Asians), female sex (42% vs 14% in male patients), and particularly never-smoking status (51% vs 10% in current or ever-smokers) are associated with an increased *EGFR* mutation frequency within the tumor[5].

Nearly 50%–60% of patients with NSCLC have at least one identifiable driver mutation, with the most common mutations being in the Kirsten ras (*KRAS*) gene (24%) and the epidermal growth factor receptor (*EGFR*) gene (13%–22%), with translocations involving anaplastic lymphoma kinase (*ALK*) in another 5%–6[11, 10]%.

EGFR INHIBITORS

Patients with adenocarcinoma and large cell histology NSCLC should be tested at diagnosis for EGFR mutations as those who harbor such mutations benefit from upfront EGFR inhibitors. Many clinical trials have shown that patients with EGFR mutations have improved progression-free survival (PFS) and overall survival (OS) when treated with EGFR inhibitors compared to patients with the same mutations who received the standard chemotherapy regimens[12-14].

In a pivotal trial of Erlotinib versus chemotherapy, PFS was improved from 4.6 months to 13.1 months with Erlotinib compared to those who received standard doublet chemotherapy[15].

Early phase II trials of Erlotinib and Gefitinib in the first line setting demonstrated modest efficacy in unselected patients with rather low and disappointing response rates (4–23%), PFS (1.6–3.5 months) and OS (5–13 months)[16–22]. This concurs with results of the large second- and third-line trials BR.21 and ISEL which showed again minor clinical benefit of EGFR TKI therapy compared to placebo in unselected patient population[23, 24]. Subgroup analyses and the discovery of activating *EGFR* gene mutations as predictors for TKI effectiveness since then yielded a conceptual change in selecting patients for EGFR-targeted therapies[16, 18, 19, 25, 26]. The following trials have used clinical criteria (such as smoking status, sex, histology, and ethnicity) and some have used the *EGFR* mutational status, whereas others have selected patients with potential intolerance to chemotherapy based on poor performance status and higher age[20, 22, 27–29]. In contrast, several phase II trials, and in particular the phase III study IPASS, selected patients for clinical surrogate markers (i.e. female patients, Asians, never- or former light-smokers). All of these trials reported an increase in ORR (13–43%) and PFS (4–5.9 months). Additional *EGFR*-mutational analyses revealed that the benefit was consistently highest in patients with tumors harboring the activating *EGFR* gene mutations[14, 19, 30–32]. These findings eventually stimulated 4 randomized phase III trials comparing first-line Erlotinib or Gefitinib with cytotoxic chemotherapy in patients with proven *EGFR*-mutant NSCLC. In such molecularly selected populations, the OPTIMAL, WJTOG3405, and NEJSG002 trials in Asian patients and the EURTAC trial in Caucasian patients clearly demonstrated the superiority of Erlotinib or Gefitinib over platinum doublet chemotherapy – the standard of care – in terms of PFS (OPTIMAL: 13.1 vs. 4.6 months, hazard ratio (HR) 0.16; WJTOG3405: 9.2 vs. 6.3 months, HR 0.48; NEJSG002: 10.8 vs. 5.4 months, HR 0.30; EURTAC: 9.7 vs. 5.2 months, HR 0.37). Also the objective response rate (ORR) was more than doubled in the EGFR TKI arms (OPTIMAL: 83% vs. 36%; WJTOG3405: 62% vs. 32%; NEJSG002: 74% vs. 31%; EURTAC: 58% vs. 15%)[33–36]. Recently published meta-analyses consistently confirmed that *EGFR* mutations predict response to EGFR TKI with much higher sensitivity (0.78; 95% confidence interval (CI) (0.74–0.82) than do *EGFR* gene copy numbers or EGFR expression levels[37]. First-line treatment with Erlotinib or Gefitinib in selected patients with EGFR mutations increases the chance of obtaining an ORR more than 2-fold (70% vs. 33%) when compared to chemotherapy and the same time the hazard of progression is reduced by 65[38]%. In *EGFR* mutation- positive patients treated with Erlotinib or Gefitinib, those with exon 19-deleted tumors appear to have a longer PFS (14.6 vs. 9.7 months) and OS (30.8 vs. 14.8 months) as compared to patients with *EGFR* L858R-mutant tumors,[39, 40] although this was not consistently reproducible throughout all trials[35, 36, 41, 42]. The European Medicines Agency (EMA) approved Gefitinib in 2009 due to results from the IPASS and INTEREST trials, and Erlotinib in 2011 due to EURTAC trial results for the first-line treatment of patients with *EGFR*-mutant NSCLC, but both drugs have yet to be licensed for this indication by the U.S. Food and Drug Administration (FDA). This is in part due to the fact that none of these trials nor the meta-analysis (HR 0.96, 2p = 0.71) has actually demonstrated a significant improvement in OS for recipients of first-line EGFR TKI therapy[38]. There is evidence, however, that the response rates are higher when TKIs are given upfront to chemotherapy- naive patients compared to chemotherapy treated patients[43]. It is highly unlikely that such an overall survival advantage will ever be demonstrated, as nearly all patients with known *EGFR* mutations who receive first-line chemotherapy cross over to TKI treatment.

Furthermore, EGFR TKIs are also active in the second-line and maintenance treatment (SATURN, INFORM trial) – especially in *EGFR*-mutant tumors[44, 45] . In many trials the HR for OS was slightly in favor of first-line EGFR TKI[38] and that the quality of life was maintained for much longer in TKI-treated patients[46] . Patients who are older or have a poor performance status will have a greater benefit from first-line EGFR TKIs[32, 47] . These trials also clearly show that proper selection of patients is absolutely crucial for treatment with EGFR-targeted agents. First-line EGFR TKIs in patients with unknown or wild-type *EGFR* status were detrimental in terms of PFS and OS as compared to chemotherapy[14, 48, 49] . In line with this, early trials combining EGFR TKIs with first-line chemotherapy in unselected patients (INTACT-1 and -2 trials) did not lead to an increased treatment efficacy,[50, 51] but recent trials in selected patients suggest that a combination may work (CALGB 30406 trial)[52] .

Afatinib is an irreversible ErbB family blocker that covalently binds to the cysteine residue of EGFR (as well as HER2), providing longer inhibition of EGFR[53] . In a decisive large randomized trial, Afatinib in a preselected EGFR-mutant population was compared to the state-of-the-art chemotherapy with pemetrexed and cisplatin, and was shown to be superior[41] . Afatinib is undergoing clinical testing as single therapy or in combination treatments in many clinical trials. In July 2013 Afatinib was approved as the first-line treatment for advanced stage NSCLC with class I and class II EGFR mutations.

Icotinib is another EGFR inhibitor clinically developed in China and reported to be active against mutant and wild-type forms of the receptor. A phase 3 randomized trial of Icotinib versus Gefitinib showed the non-inferiority of Icotinib and a much better profile in terms of drug-related side effects[54].

Dacomitinib, a pan-EGFR-family irreversible inhibitor is currently in several trials for NSCLC. In a recent trial, treatment with Dacomitinib was associated with a median PFS of 12.4 weeks compared with 8.3 weeks for Erlotinib in patients with adenocarcinoma. However, a superior PFS with Dacomitinib compared to Erlotinib was not observed in other histological subtypes of NSCLC[55] .

There are a number of other trials evaluating the efficacy of combining first- or second-generation EGFR inhibitors with other molecularly targeted agents.

Many clinical trials for recurrent or advanced NSCLC involve Erlotinib in combination with other therapies. Results of a trial using combination of Erlotinib and Tivantinib/ARQ 197, a non-ATP-competitive inhibitor of MET, showed clinical activity in patients with NSCLC, with 6 of 8 patients achieving stable disease[56].

A phase 3 randomized trial explored addition of Erlotinib to the doublet chemotherapy with cisplatin/ gemcitabine as a - treatment in patients with advanced NSCLC. Addition of Erlotinib significantly prolonged PFS in patients with mutant EGFR[57].

The use of RTK inhibitors in wild-type EGFR NSCLC patients remains a subject of controversy, with some investigators advocating it,[58, 59] while others provide data analysis indicating no benefit in this large heterogeneous population of patients[60].

The First-Line Erbitux in Lung Cancer (FLEX) phase 3 worldwide study demonstrated that Cetuximab, a monoclonal antibody directed against EGFR when to a platinum-based doublet (cisplatin and vinorelbine) can extend median OS in patients with advanced EGFR-expressing NSCLC (stage wet IIIB or stage IV). As a result, the use of Cetuximab combined with cisplatin-vinorelbine has been endorsed by National Comprehensive Cancer Network (NCCN) guidelines as a first-line option for the treatment of advanced NSCLC[61].

RESISTANCE TO EGFR INHIBITORS

Resistance to EGFR inhibitors arises because of cellular heterogeneity within an oncogene-addicted tumor. Tumor shrinkage indicates a response to a molecularly targeted therapy, but residual tumor may be a source of slow growing drug-tolerant “persistor” cells that promote tumor regrowth, regeneration, and heterogeneity[62].

Nearly all patients with EGFR-mutant- AC develop resistance to the first-generation inhibitors Erlotinib and Gefitinib, with a median PFS of 14 months[63].

Although EGFR kinase mutations are associated with an enhanced sensitivity to Gefitinib and Erlotinib, not all tumors that have activating mutations are associated with an enhanced response. Tumors that fail to respond to EGFR TKIs despite the presence of an activating mutation might have an additional genetic lesion that relieves the tumor of its dependence on the EGFR signaling pathway. One mechanism that has been linked to insensitivity of NSCLC to EGFR TKIs is the occurrence of insertion point mutations in exon 20 of the *EGFR* gene[64, 65].

The most common and first identified secondary mutation is the threonine-790 to methionine (T790M) point mutation in exon 20 which represents

approximately 50% of all acquired resistance in NSCLC58. The development of such genetic alteration restores the EGFR TK affinity to ATP, rendering first-generation TKIs inactive[66, 67]. T790M mutation has been identified in pre-treatment tumors and is responsible for both a lower sensitivity and duration of response to the first generation TKIs[68].

Other critical resistance mechanisms have been identified to be linked to the inhibition of EGFR and include MET amplification[69], PTEN loss[70], HER2 kinase domain[71] or PIK3CA and transformation to small cell lung cancer[72].

Concerns have been raised about resistance to TKIs due to mutations in the KRAS gene (which encodes for a protein downstream to the EGFR in the pathway initiated by the activation of the tyrosine kinase) or due to mutations that occur in other proteins of the same cascade such as BRAF[73].

Other resistance point mutations, such as aspartic acid-761 to tyrosine (D761Y), have been reported, some of which may weaken the interaction of EGFR TKI with its target[74].

In common with resistance mechanisms identified for EGFR, ALK-rearranged cancers have been reported to develop secondary mutations[75].

OVERCOMING RESISTANCE TO EGFR INHIBITORS

Management of EGFR tumor resistance has become the next challenge in order to extend these patients' overall survival; identification of the molecular resistance mechanisms will allow for the treatment of TKI-resistant tumors.

A new class of drugs, the so-called second-generation TKIs, may be able to overcome the T790M mutation resistant cell. Compared to first-generation TKIs, these molecules show higher affinity for the ATP-binding domain, form an irreversible covalent bond to the ATP-binding site and are able to stimulate other receptors (e.g. HER2).

The second-generation TKIs Afatinib irreversibly inhibit RTKs of EGFR family, as well as the T790M variant of EGFR[53].

The second-generation TKIs Neratinib hasn't shown good RR when tested on patients with known T790M mutation[76].

Dacomitinib is another irreversible TKI able to target the activity of all HER TKs and has shown activity in NSCLC cell lines harboring the T790M mutation. This molecule has been evaluated in two phase II trials: The first one was after failure of one or two chemotherapy regimens and failure on Erlotinib; the second one compared it with Erlotinib in the second and third-line in patients with advanced NSCLC with promising results[55, 77 -79].

Another approach for overcoming resistance to reversible EGFR TKIs involves targeting parallel- or convergent signaling pathways. The mammalian target of the rapamycin (mTOR) signaling pathway integrates nutrient and mitogen signals to regulate cell proliferation, survival and angiogenic pathways, and has been implicated in resistance to EGFR inhibitors. In both sensitive and resistant tumor cell lines, the mTOR inhibitor, Everolimus, reduces the expression of EGFR signaling effectors and cooperates with Gefitinib to overcome resistance[80].

Various drugs or antibodies capable of inhibiting MET (e.g., Crizotinib, Foretinib, ARQ 197, MetMAb) could, in principle, be combined with the first (Erlotinib) or second (Dacomitinib, Afatinib) generation EGFR-TKIs. Concurrent inhibition of both may improve patient outcomes. Small-1399 molecule inhibitors of MET and MetMAb/Onartuzumab are currently being tested in NSCLC. However, the phase III trial of Onartuzumab combined with Erlotinib in MET positive EGFR mutant NSCLC failed to improve PFS or OS in spite of the positive results from a phase II trial[81].

Many cancers have increased levels of active Hsp90, which is involved in protein folding. HSP90 is a molecular chaperone that is critical for tumor growth and proliferation. Client proteins of HSP90 include many signaling kinases such as RTKs and intracellular kinases essential for cancer cell survival, since lack of HSP90 triggers protein degradation. Hsp90 inhibitors may thus block multiple signaling pathways that are functioning aberrantly in cancer cells. Hsp90 inhibitors such as AUY922 and STA9090 are in many clinical trials for lung cancer. Both inhibitors showed good efficacy in preclinical models of NSCLC[82- 84].

Ongoing trials for recurrent or advanced NSCLC will test the efficacy of various combination therapies including EGFR inhibitors, second-generation tyrosine kinase inhibitors, dual MET/ VEGFR2 inhibitor, and targeted drugs to different proteins disregulated in lung cancer.

SIDE EFFECTS ASSOCIATED WITH EGFR TKIS THERAPY

EGFR-targeting agents are generally associated with less serious toxicities than traditional chemotherapeutic agents. Chemotherapeutic agents carry the risk of organ

toxicities, severe myelosuppression and neutropenic sepsis, with treatment-related fatal events in up to 8% of patients, especially in patients with poor performance status[85]. Fatal events during EGFR TKI treatment in the form of lung or liver toxicity have been rarely reported[86, 87]. The most common adverse events leading to TKI dose reductions are cutaneous reactions (acneiform 'rash'), paronychia and diarrhea, because of the abundance of EGFR in skin and mucosa. Meta-analysis confirmed that the appearance of skin rash was an independent predictive factor for survival (HR 0.30; $p< 0.00001$) and tumor progression (HR 0.50; $p< 0.00001$) during Erlotinib and Gefitinib treatment[38]. In first-line NSCLC trials of Erlotinib and Gefitinib, most patients developed rash of grades 1 or 2. Only few patients experienced grade 3 rash (Erlotinib: 67%, 2–13% grade ≥ 3; Gefitinib: 66%, 3% grade ≥ 3). Skin reactions normally appear after 1 week and reach maximum severity following 2–3 weeks of TKI treatment, after which they gradually and spontaneously disappear[88].

Diarrhea also affects a substantial part of EGFR TKI-treated patients. It is thought to result from excess chloride secretion ('secretory diarrhea'), which can lead to dehydration, electrolyte imbalances, renal insufficiency, and malnutrition[88]. In first-line NSCLC trials, the diarrhea incidence was reported as high as 52% (5% grade ≥ 3) for Erlotinib and 47% (4% grade ≥ 3) for Gefitinib[33, 34].

A much less frequent but potentially lethal (30–50% lethality) side effect of EGFR TKIs therapy is interstitial lung disease (ILD), which in general occurs during the first 3 months of treatment (median 24–42 days) in < 1% of all patients, with higher risk in Japanese populations (1.6–3.5%)[86]. Pre-existing pulmonary fibrosis, prior thoracic irradiation and smoking history have been identified as further risk factors for developing ILD and should be taken into account when considering a patient for first-line EGFR TKI treatment[89].

Other side effects may occur and mostly reversible include fatigue, nausea/vomiting, and increased liver enzyme levels[35, 36].

CONCLUSIONS

Lung cancer remains the number-one worldwide cause of cancer-related mortality. Adenocarcinoma (AC) of lung comprises about 40% to 50% of all lung cancers. Histology is now an important consideration for treatment selection in NSCLC. The majority of individuals with NSCLC are considered inoperable at initial evaluation owing to the presence of locally advanced or metastatic disease. For those patients who present with locally advanced or metastatic NSCLC, palliative chemotherapy is associated with only modest survival prolongation and indeterminate impact on quality of life. Personalized therapy for NSCLC patients should include a genetic testing of the EGFR mutational status for individual patients. Genomic testing for personalized treatment of lung cancer is now associated with improved survival, likely due to getting targeted kinase inhibitors to the indicated patients. All patients with *EGFR* mutation-positive lung adenocarcinoma must be offered an EGFR TKI therapy. The main approach to block the EGFR pathway is by competing with ATP for binding to the tyrosine kinase domain. The EGFR TKIs Erlotinib and Gefitinib are reversible inhibitors of the EGFR kinase and are also called "first-generation" small molecular inhibitors. The second-generation TKIs Afatinib may overcome resistance to the treatment of Erlotinib or Gefitinib through the T790M gatekeeper mutation. Several irreversible EGFR inhibitors blocked multiple EGFR family members, interrupting the cooperative signal pathway among EGFR members and resulted in a more complete blockage. EGFR inhibitors are better tolerated than chemotherapy. Side effects like rash and diarrhea are manageable in the outpatient setting under close surveillance, in particular during the first weeks of treatment. The problem with targeted therapies is that most of the time clinical responses to them are short-lived. The mechanisms of resistance are multifaceted, and might involve new resistance-conferring mutations in the target protein itself (such as T790M in EGFR) or activate lateral signal-transduction pathways via new mutations or changes in the expression level of the key proteins. Close cooperation between clinicians, surgeons, molecular biologists and pathologists is crucial for a continuous improvement in the field of NSCLC target therapy.

Acknowledgment: This work was not supported by any grants.

Conflict of interest: The author has declared that no competing interest exists.

REFERENCES

1. Clinical Lung Cancer Genome Project (CLCGP); Network Genomic Medicine (NGM). A genomics-based classification of human lung tumors. Sci Transl Med. 2013;5:209ra153.
2. Travis W, Brambilla E, Muller-Hermelink H, et al (eds): Pathology and Genetics of Tumors of the Lung, Pleura, Thymus and Heart. Lyon, France, IARC Press, 2004.
3. Bazley LA and Gullick WJ. The epidermal growth factor receptor family. Endocr Relat Cancer. 2005;12 (Suppl 1) :S17–S27.

4. Kumar A, Petri ET, Halmos B, et al. Structure and clinical relevance of the epidermal growth factor receptor in human cancer. J Clin Oncol. 2008;26:1742–1751.
5. Shigematsu H, Lin L, Takahashi T, et al. Clinical and biological features associated with epidermal growth factor receptor gene mutations in lung cancers. J Natl Cancer Inst. 2005;97:339–346.
6. Wigle DA. Personalized therapy for non-small cell lung cancer: hype or clinical reality? Semin Thorac Cardiovasc Surg. 2011;23:30–35.
7. Carr LL, Finigan JH and Kern JA. Evaluation and treatment of patients with non-small cell lung cancer. Med Clin North Am. 2011;95:1041–1054.
8. Yasuda H, Park E, Yun CH, et al. Structural, biochemical, and clinical characterization of epidermal growth factor receptor (EGFR) exon 20 insertion mutations in lung cancer. Sci Transl Med. 2013;5:216ra177.
9. Scagliotti GV, Selvaggi G, Novello S, et al. The biology of epidermal growth factor receptor in lung cancer. Clin Cancer Res. 2004;10:4227s-4232s.
10. Sequist LV, Heist RS, Shaw AT, et al. Implementing multiplexed genotyping of non-small-cell lung cancers into routine clinical practice. Ann Oncol. 2011; 22:2616–2624.
11. Kris MG, Johnson BE, Berry LD, et al. Using multiplexed assays of oncogenic drivers in lung cancers to select targeted drugs. JAMA. 2014;311:1998-2006.
12. Inoue A, Kobayashi K, Usui K, et al. First-line Gefitinib for patients with advanced non-small-cell lung cancer harboring epidermal growth factor receptor mutations without indication for chemotherapy. J Clin Oncol. 2009;27:1394-1400.
13. Lee CK, Brown C, Gralla RJ, et al. Impact of EGFR Inhibitor in Non-Small Cell Lung Cancer on Progression-Free and Overall Survival: A Meta- Analysis. J Natl Cancer Inst. 2013;105:595-605.
14. Mok TS, Wu YL, Thongprasert S, et al. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med. 2009;361:947-957.
15. Somaiah N, Simon NG and Simon GR. A tabulated summary of targeted and biologic therapies for non-small-cell lung cancer. J Thorac Oncol. 2012; 7:S342-368.
16. Giaccone G, Gallegos Ruiz M, Le Chevalier T, et al. Erlotinib for frontline treatment of advanced non-small cell lung cancer: a phase II study. Clin Cancer Res. 2006;12:6049–6055.
17. Reck M, Buchholz E, Romer KS, et al. Gefitinib monotherapy in chemotherapy-naive patients with inoperable stage III/IV non-small-cell lung cancer. Clin Lung Cancer. 2006;7:406–411.
18. Akerley W, Boucher KM, Bentz JS, et al. A phase II study of Erlotinib as initial treatment for patients with stage IIIB–IV nonsmall cell lung cancer. J Thorac Oncol. 2009;4:214–219.
19. Jackman DM, Yeap BY, Lindeman NI, et al. Phase II clinical trial of chemotherapy-naive patients > or = 70 years of age treated with Erlotinib for advanced non-small-cell lung cancer. J Clin Oncol. 2007;25:760–766.
20. Hesketh PJ, Chansky K, Wozniak AJ, et al. Southwest Oncology Group phase II trial (S0341) of erlotinib (OSI-774) in patients with advanced nonsmall cell lung cancer and a performance status of 2. J Thorac Oncol. 2008;3:1026–1031.
21. Crino L, Cappuzzo F, Zatloukal P, et al. Gefitinib versus vinorelbine in chemotherapy-naive elderly patients with advanced non-small-cell lung cancer (INVITE): a randomized, phase II study. J Clin Oncol. 2008;26:4253–4260.
22. Lilenbaum R, Axelrod R, Thomas S, et al. Randomized phase II trial of Erlotinib or standard chemotherapy in patients with advanced non-small-cell lung cancer and a performance status of 2. J Clin Oncol. 2008;26:863–869.
23. Shepherd FA, Rodrigues Pereira J, Ciuleanu T, et al. Erlotinib in previously treated non-small-cell lung cancer. N Engl J Med. 2005;353:123–132.
24. Thatcher N, Chang A, Parikh P, et al. Gefitinib plus best supportive care in previously treated patients with refractory advanced non-small-cell lung cancer: results from a randomised, placebo-controlled, multicentre study (Iressa Survival Evaluation in Lung Cancer). Lancet 2005;366:1527–1537.
25. Paez JG, Janne PA, Lee JC, et al. EGFR mutations in lung cancer: correlation with clinical response to Gefitinib therapy. Science. 2004;304:1497–1500.
26. Lynch TJ, Bell DW, Sordella R, et al. Activating mutations in the epidermal growth factor receptor underlying responsiveness of non-small-cell lung cancer to Gefitinib. N Engl J Med. 2004;350:2129–2139.
27. Lee SM, Khan I, Upadhyay S, et al. First-line Erlotinib in patients with advanced non-small-cell lung cancer unsuitable for chemotherapy (TOPICAL): a double-blind, placebo-controlled, phase 3 trial. Lancet Oncol. 2012;13:1161–1170.
28. Morere JF, Brechot JM, Westeel V, et al. Randomized phase II trial of Gefitinib or gemcitabine or docetaxel chemotherapy in patients with advanced non-small-cell lung cancer and a performance status of 2 or 3 (IFCT-0301 study). Lung Cancer. 2010;70:301–307.
29. Goss G, Ferry D, Wierzbicki R, et al. Randomized phase II study of Gefitinib compared with placebo in chemotherapy-naive patients with advanced non-small-cell lung cancer and poor performance status. J Clin Oncol. 2009;27:2253–2260.
30. Yang CH, Yu CJ, Shih JY, et al. Specific EGFR mutations predict treatment outcome of stage IIIB/IV patients with chemotherapy-naive nonsmall- cell lung cancer receiving first-line Gefitinib monotherapy. J Clin Oncol. 2008;26:2745–2753.
31. Niho S, Kubota K, Goto K, et al. First-line single agent treatment with Gefitinib in patients with advanced non-small-cell lung cancer: a phase II study. J Clin Oncol. 2006;24:64–69.

32. Ebi N, Semba H, Tokunaga SJ, et al. A phase II trial of Gefitinib monotherapy in chemotherapynaive patients of 75 years or older with advanced non-small cell lung cancer. J Thorac Oncol. 2008; 3:1166–1171.
33. Zhou C, Wu YL, Chen G, et al. Erlotinib versus chemotherapy as first-line treatment for patients with advanced EGFR mutation-positive nonsmall- cell lung cancer (OPTIMAL, CTONG-0802): a multicentre, open-label, randomised, phase 3 study. Lancet Oncol. 2011;12:735–742.
34. Maemondo M, Inoue A, Kobayashi K, et al. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N Engl J Med. 2010; 362:2380–2388.
35. Rosell R, Carcereny E, Gervais R, et al. Erlotinib versus standard chemotherapy as first-line treatment for European patients with advanced EGFR mutation-positive non-small-cell lung cancer (EURTAC): a multicentre, open-label, randomized phase 3 trial. Lancet Oncol. 2012;13:239–246.
36. Mitsudomi T, Morita S, Yatabe Y, et al. Gefitinib versus cisplatin plus docetaxel in patients with non-small-cell lung cancer harbouring mutations of the epidermal growth factor receptor (WJTOG3405): an open label, randomised phase 3 trial. Lancet Oncol. 2010;11:121–128.
37. Dahabreh IJ, Linardou H, Siannis F, et al. Somatic EGFR mutation and gene copy gain as predictive biomarkers for response to tyrosine kinase inhibitors in non-small cell lung cancer. Clin Cancer Res. 2010;16:291–303.
38. Petrelli F, Borgonovo K, Cabiddu M, et al. Efficacy of EGFR tyrosine kinase inhibitors in patients with EGFR-mutated non-small-cell lung cancer: a meta-analysis of 13 randomized trials. Clin Lung Cancer. 2012;13:107–114.
39. Jackman DM, Miller VA, Cioffredi LA, et al. Impact of epidermal growth factor receptor and KRAS mutations on clinical outcomes in previously untreated non-small cell lung cancer patients: results of an online tumor registry of clinical trials. Clin Cancer Res. 2009;15:5267–5273.
40. Fukuoka M, Wu YL, Thongprasert S, et al. Biomarker analyses and final overall survival results from a phase III, randomized, open-label, first-line study of Gefitinib versus carboplatin/paclitaxel in clinically selected patients with advanced nonsmall- cell lung cancer in Asia (IPASS). J Clin Oncol. 2011;29:2866–2874.
41. Sequist LV, Yang JC, Yamamoto N, et al. Phase III study of Afatinib or cisplatin plus pemetrexed in patiens with metastatic lung adenocarinoma with EGFR mutations. J Clin Oncol. 2013;31:3327-3334.
42. Yang JC, Hirsh V, Schuler M, et al. Symptom control and quality of life in LUX-Lung 3: a phase III study of Afatinib or cisplatin/pemetrexed in patients with advanced lung adenocarcinoma with EGFR mutations. J Clin Oncol. 2013;31:3342-3350.
43. Wu JY, Yu CJ, Yang CH, et al. First- or second-line therapy with Gefitinib produces equal survival in non-small cell lung cancer. Am J Respir Crit Care Med. 2008;178:847–853.
44. Zhang L, Ma S, Song X, et al. Gefitinib versus placebo as maintenance therapy in patients with locally advanced or metastatic non-small-cell lung cancer (INFORM; C-TONG 0804): a multicentre, double-blind randomised phase 3 trial. Lancet Oncol. 2012;13:466–475.
45. Cappuzzo F, Ciuleanu T, Stelmakh L, et al. Erlotinib as maintenance treatment in advanced non-small-cell lung cancer: a multicentre, randomised, placebo-controlled phase 3 study. Lancet Oncol. 2010;11:521–529.
46. Oizumi S, Kobayashi K, Inoue A, et al. Quality of life with Gefitinib in patients with EGFR-mutated non-small cell lung cancer: quality of life analysis of North East Japan Study Group 002 Trial. Oncologist. 2012;17:863–870.
47. Asami K, Koizumi T, Hirai K, et al. Gefitinib as first-line treatment in elderly epidermal growth factor receptor-mutated patients with advanced lung adenocarcinoma: results of a Nagano Lung Cancer Research Group study. Clin Lung Cancer. 2011;12:387–392.
48. Gridelli C, Butts C, Ciardiello F, et al. An international, multicenter, randomized phase III study of first-line Erlotinib followed by second-line cisplatin/ gemcitabine versus first-line cisplatin/gemcitabine followed by second-line Erlotinib in advanced non-small-cell lung cancer: treatment rationale and protocol dynamics of the TORCH trial. Clin Lung Cancer. 2008;9:235–238.
49. Thomas M, Reuss A, Fischer JR, et al. Innovations: Randomized phase II trial of Erlotinib (E)/bevacizumab (B) compared with cisplatin (P)/gemcitabine (G) plus B in first-line treatment of advanced nonsquamous (NS) non-small cell lung cancer (NSCLC). J Clin Oncol. 2011;29(suppl; abstr 7504).
50. Giaccone G, Herbst RS, Manegold C, et al. Gefitinib in combination with gemcitabine and cisplatin in advanced non-small-cell lung cancer: a phase III trial – INTACT 1. J Clin Oncol. 2004; 22:777–784.
51. Herbst RS, Giaccone G, Schiller JH, et al. Gefitinib in combination with paclitaxel and carboplatin in advanced non-small-cell lung cancer: a phase III trial – INTACT 2. J Clin Oncol. 2004;22:785–794.
52. Janne PA, Wang X, Socinski MA, et al. Randomized phase II trial of Erlotinib alone or with carboplatin and paclitaxel in patients who were never or light former smokers with advanced lung adenocarcinoma: CALGB 30406 trial. J Clin Oncol. 2012;30:2063–2069.
53. Nelson V, Ziehr J, Agulnik M, et al. Afatinib: emerging next-generation tyrosine kinase inhibitor for NSCLC. Onco Targets Ther. 2013;6:135-143.
54. Shi Y, Zhang L, Liu X, et al. Icotinib versus Gefitinib in previously treated advanced non-small-cell lung cancer (ICOGEN): a randomised, double-blind phase 3 non-inferiority trial. Lancet Oncol. 2013;14:953-961.
55. Ramalingam SS, Blackhall F, Krzakowski M, et al. Randomized phase II study of dacomitinib (PF-00299804), an irreversible pan-human epidermal growth

factor receptor inhibitor, versus Erlotinib in patients with advanced non-small-cell lung cancer. J Clin Oncol. 2012; 30:3337- 3344.
56. Goldman JW, Laux I, Chai F, et al. Phase 1 dose-escalation trial evaluating the combination of the selective MET (mesenchymal-epithelial transition factor) inhibitor tivantinib (ARQ 197) plus Erlotinib. Cancer. 2012; 118:5903-5911.
57. Wu YL, Lee JS, Thongprasert S, et al. Intercalated combination of chemotherapy and Erlotinib for patients with advanced stage non-small-cell lung cancer (FASTACT-2): a randomised, double-blind trial. Lancet Oncol. 2013;14:777-786.
58. Laurie SA and Goss GD. Role of epidermal growth factor receptor inhibitors in epidermal growth factor receptor wild-type non-small-cell lung cancer. J Clin Oncol. 2013;31:1061-1069.
59. Laurie SA and Goss GD. Reply to F. Gelsomino et al. J Clin Oncol. 2013;31:3293-2394.
60. Gelsomino F, Agustoni F, Niger M, et al. Epidermal growth factor receptor tyrosine kinase inhibitor treatment in patients with EGFR wild- type non-small-cell lung cancer: the never-ending story. J Clin Oncol. 2013;31:3291-3293.
61. Pirker R, Pereira JR, Szczesna A, et al. Cetuximab plus chemotherapy in patients with advanced non-small-cell lung cancer (FLEX): an open-label randomised phase III trial. Lancet. 2009;373:1525-31.
62. Sharma SV and Settleman J. Oncogene addiction: setting the stage for molecularly targeted cancer therapy. Genes Dev. 2007; 21:3214–3231.
63. Jackman D, Pao W, Riely GJ, et al. Clinical definition of acquired resistance to epidermal growth factor receptor tyrosine kinase inhibitors in non-small-cell lung cancer. J Clin Oncol. 2010;28:357-360.
64. Greulich H, Chen TH, Feng W, et al. Oncogenic transformation by inhibitor-sensitive and-resistant EGFR mutants. PLoS Med. 2005;2:e313.
65. Sharma SV, Bell DW, Settleman J, et al. Epidermal growth factor receptor mutations in lung cancer. Nat Rev Cancer. 2007;7:169–181.
66. Pao W, Miller VA, Politi KA, et al. Acquired resistance of lung adenocarcinomas to Gefitinib or Erlotinib is associated with a second mutation in the EGFR kinase domain. PLoS Med. 2005; 2: e73.
67. Yun CH, Mengwasser KE, Toms AV, et al. The T790M mutation in EGFR kinase causes drug resistance by increasing the affinity for ATP. Proc Natl Acad Sci USA. 2008;105:2070-2075.
68. Bean J, Riely GJ, Balak M, et al. Acquired resistance to epidermal growth factor receptor kinase inhibitors associated with a novel T854A mutation in a patient with EGFR-mutant lung adenocarcinoma. Clin Cancer Res. 2008;14:7519-7525.
69. Benedettini E, Sholl LM, Peyton M, et al: Met activation in non-small cell lung cancer is associated with de novo resistance to EGFR inhibitors and the development of brain metastasis. Am J Pathol. 2010, 177:415–423.
70. Sos ML, Koker M, Weir BA, et al: PTEN loss contributes to Erlotinib resistance in EGFR-mutant lung cancer by activation of Akt and EGFR. Cancer Res. 2009; 69:3256–3261.
71. Wang SE, Narasanna A, Perez-Torres M, et al. HER2 kinase domain mutation results in constitutive phosphorylation and activation of HER2 and EGFR and resistance to EGFR tyrosine kinase inhibitors. Cancer Cell. 2006;10:25–38.
72. Sequist LV, Waltman BA, Dias-Santagata D, et al: Genotypic and histological evolution of lung cancers acquiring resistance to EGFR inhibitors. Sci Transl Med. 2011;3:75–26.
73. Ohashi K, Sequist LV, Arcila ME, et al. Lung cancers with acquired resistance to EGFR inhibitors occasionally harbor BRAF gene mutations but lack mutations in KRAS, NRAS, or MEK1. Proc Natl Acad Sci U S A. 2012; 109: E2127-2133.
74. Balak MN, Gong Y, Riely GJ, et al. Novel D761Y and common secondary T790M mutations in epidermal growth factor receptor–mutant lung adenocarcinomas with acquired resistance to kinase inhibitors. Clin Cancer Res. 2006;12:6494–6501.
75. Sasaki T, Okuda K, Zheng W, et al. The neuroblastoma-associated F1174L ALK mutation causes resistance to an ALK kinase inhibitor in ALK-translocated cancers. Cancer Res. 2010;70:10038–10043.
76. Sequist LV, Besse B, Lynch TJ, et al. Neratinib, an irreversible pan-ErbB receptor tyrosine kinase inhibitor: results of phase II trial in patients with advanced non small-cell lung cancer. J Clin Oncol. 2010;28:3076-3083.
77. Engelman JA, Zejnullahu K, Gale CM, et al. PF00299804, an irreversible pan-ERBB inhibitor, is effective in lung cancers models with EGFR and ERBB2 mutations that are resistant to Gefitinib. Cancer Res. 2007;67:11924-11932.
78. Boyer MJ, Blackhall FH, Park K, et al. Efficacy and safety of PF00299805 versus Erlotinib: a global, randomised phase 2 trial in patients with advanced non small cell lung cancer after failure of chemotherapy. J Clin Oncol. 2010;28:18s (suppl; abstr LBA7523).
79. Takahashi T, Boku N, Murakami H, et al. Phase I and pharmacokinetic study of dacomitinib (PF-00299804), an oral irreversible, small molecule inhibitor of human epidermal growth factor receptor-1, -2, and -4 tyrosine kinases, in Japanese patients with advanced solid tumors. Invest New Drugs. 2012;30:2352-2363.
80. Bianco R, Gaofalo S, Rosa R, et al. Inhibition of mTOR pathway by everolimus cooperates with EGFR inhibitors in human tumours sensitive and resistant to anti-EGFR drugs. Br J Cancer. 2008;98:923–930.
81. Spigel DR, Ervin TJ, Ramlau RA, et al. Randomized phase II trial of Onartuzumab in combination with Erlotinib in patients with advanced non-small-cell lung cancer. J Clin Oncol. 2013;31:4105-4114.

82. Garon EB, Finn RS, Hamidi H, et al. The HSP90 inhibitor NVP-AUY922 potently inhibits non-small cell lung cancer growth. Mol Cancer Ther. 2013;12:890-900.
83. Shimamura T, Perera SA, Foley KP, et al. Ganetespib (STA-9090), a nongeldanamycin HSP90inhibitor, has potent antitumor activity in in vitro and in vivo models of non-small cell lung cancer. Clin Cancer Res. 2012;18:4973-4985.
84. Ueno T, Tsukuda K, Toyooka S, et al. Strong anti-tumor effect of NVP-AUY922, a novel Hsp90 inhibitor, on non-small cell lung cancer. Lung Cancer. 2012;76:26-31.
85. O'Brien ME, Borthwick A, Rigg A, et al. Mortality within 30 days of chemotherapy: a clinical governance benchmarking issue for oncology patients. Br J Cancer 2006;95:1632–1636.
86. Ren S, Li Y, Li W, et al. Fatal asymmetric interstitial lung disease after Erlotinib for lung cancer. Respiration. 2012;84:431–435.
87. Schacher-Kaufmann S and Pless M. Acute fatal liver toxicity under Erlotinib. Case Rep Oncol. 2010;3:182–188.
88. Hirsh V. Managing treatment-related adverse events associated with EGFR tyrosine kinase inhibitors in advanced non-small-cell lung cancer. Curr Oncol. 2011;18:126–138.
89. Nakagawa M, Nishimura T, Teramukai S, et al. Interstitial lung disease in Gefitinib-treated Japanese patients with non-small cell lung cancer – a retrospective analysis: JMTO LC03–02. BMC Res Notes. 2009;2:157.