Cancer Medicine

ORIGINAL RESEARCH

Advances in molecular-based personalized non-small-cell lung cancer therapy: targeting epidermal growth factor receptor and mechanisms of resistance

Robert M. Jotte[1] & David R. Spigel[2]

[1]Rocky Mountain Cancer Centers, Lone Tree, Colorado
[2]Sarah Cannon Research Institute, Nashville, Tennessee

Abstract

Molecularly targeted therapies, directed against the features of a given tumor, have allowed for a personalized approach to the treatment of advanced non-small-cell lung cancer (NSCLC). The reversible epidermal growth factor receptor (EGFR) tyrosine kinase inhibitors (TKIs) gefitinib and erlotinib had undergone turbulent clinical development until it was discovered that these agents have preferential activity in patients with NSCLC harboring activating EGFR mutations. Since then, a number of phase 3 clinical trials have collectively shown that EGFR-TKI monotherapy is more effective than combination chemotherapy as first-line therapy for EGFR mutation-positive advanced NSCLC. The next generation of EGFR-directed agents for EGFR mutation-positive advanced NSCLC is irreversible TKIs against EGFR and other ErbB family members, including afatinib, which was recently approved, and dacomitinib, which is currently being tested in phase 3 trials. As research efforts continue to explore the various proposed mechanisms of acquired resistance to EGFR-TKI therapy, agents that target signaling pathways downstream of EGFR are being studied in combination with EGFR TKIs in molecularly selected advanced NSCLC. Overall, the results of numerous ongoing phase 3 trials involving the EGFR TKIs will be instrumental in determining whether further gains in personalized therapy for advanced NSCLC are attainable with newer agents and combinations. This article reviews key clinical trial data for personalized NSCLC therapy with agents that target the EGFR and related pathways, specifically based on molecular characteristics of individual tumors, and mechanisms of resistance.

Introduction

At least 85% of lung cancers are histologically classified as non-small-cell lung cancer (NSCLC), often requiring systemic therapy for advanced disease [1]. As conventional chemotherapy with platinum-based doublets is associated with improved clinical outcomes, but also potentially higher toxicity [1], the evaluation of molecularly targeted therapies has led to extensive investigation and several U.S. Food and Drug Administration (FDA) approvals for advanced NSCLC [2–5].

This review article describes the framework upon which epidermal growth factor receptor tyrosine kinase inhibitor (EGFR-TKI) therapy has been built and summarizes the ongoing work in the development of personalized medicine in the EGFR-TKI field.

EGFR and ErbB Family Pathways

Overview

EGFR, or human epidermal growth factor receptor 1 (HER1)/ErbB1, is the first of the four receptor tyrosine kinases (TKs) within the ErbB family [6]. Autophosphorylation of EGFR and other family members is a key step toward activation of several pathways involved in cellular proliferation, including the retrovirus-associated DNA sequences (Ras)/v-raf 1

murine leukemia viral oncogene homolog 1 (Raf)/mitogen-activated protein kinase (MAPK) pathway and phospho-inositide-3 kinase (PI3K)/protein kinase B (Akt) pathway [7]. Additional downstream effects of ErbB family signaling include signal transducers and activation of transcription (STAT) recruitment and phosphorylation (Fig. 1) [7, 8].

Role of EGFR and the ErbB family in NSCLC therapy

Increased EGFR expression and its association with advanced disease in NSCLC [9] prompted early efforts toward clinical development of agents targeting the EGFR pathway. The first-generation reversible EGFR TKIs gefitinib (Iressa®, AstraZeneca, Wilmington, DE) [2] and erlotinib (Tarceva®, Genentech, South San Francisco, CA) [4] were the first EGFR-directed therapies to be approved by the FDA in NSCLC.

Of note, gefitinib use has been discontinued in the United States. Based on data establishing that EGFR TKIs confer the most benefit when used in molecularly selected patients, erlotinib was granted an additional indication by the FDA in May 2013 for first-line treatment of patients with metastatic NSCLC whose tumors have EGFR exon 19 deletions or exon 21 (L858R) mutations as detected by an FDA-approved test [4]. In July 2013, the irreversible ErbB family TKI afatinib (Gilotrif®, Boehringer Ingelheim, Ingelheim, Germany) was approved by the FDA in the same setting and also accompanied by an FDA-approved mutational test [10].

Discovery and implications of activating EGFR mutations

A key discovery toward personalized therapy for NSCLC was the association between activating somatic EGFR
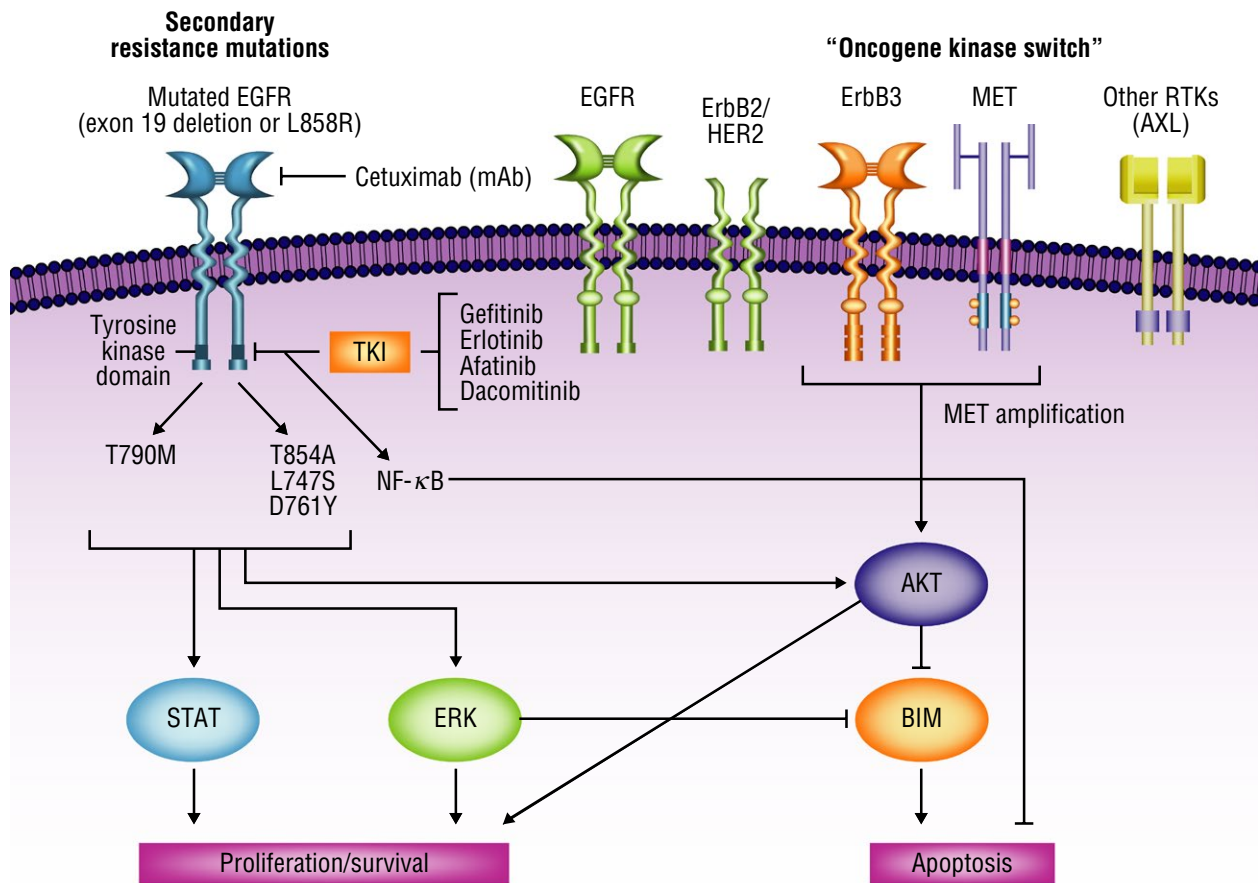

Figure 1. Mechanisms of acquired resistance to gefitinib/erlotinib in EGFR-mutated NSCLC. EGFR, epidermal growth factor receptor; ErbB3, v-erb-b2 avian erythroblastic leukemia viral oncogene homolog 3; NSCLC, non-small-cell lung cancer; RTK, receptor tyrosine kinase; MET, met proto-oncogene; AXL, AXL receptor tyrosine kinase; mAb, monoclonal antibody; TKI, tyrosine kinase inhibitor; NF-κB, nuclear factor kappa-light-chain-enhancer of activated B cells; AKT, v-akt murine thymoma viral oncogene homolog 1; STAT, signal transducers and activation of transcription; ERK, extracellular signal-regulated kinase; BIM, BCL2-like 11 (apoptosis facilitator). Adapted from Nguyen et al. [43], with permission from Elsevier.

mutations and response to gefitinib and erlotinib [11], observed at a higher rate in Asian compared with Western populations [12]. Known EGFR-activating mutations are located on exons 18–21 within the TK domain [13], with ~90% estimated to involve deletions in exon 19 and point mutations in exon 21 [12]. This molecular characterization served to explain the clustering of responses in early clinical trials among patients with certain characteristics, including East Asian ethnicity, adenocarcinoma histology, female gender, and nonsmoking history [13, 14]. While EGFR mutations are more commonly observed in patients with these clinical characteristics (i.e., Asian ethnicity, adenocarcinoma histology, etc.), they can occur in patients who do not fit these characteristics as well. In related findings, it is now known that KRAS mutations are often mutually exclusive with EGFR-activating mutations and may be associated with EGFR-TKI resistance [1, 15].

The criticality of EGFR mutations in predicting response to EGFR TKIs makes molecular testing important in both clinical trials and clinical practice [1]. Per the 2015 National Comprehensive Cancer Network (NCCN) guidelines [1], routine EGFR mutation testing is recommended in NSCLC of adenocarcinoma, large cell, or unknown histology, but not in squamous cell carcinoma (except in never smokers and mixed histology or small biopsy specimens) given its rarity in this subtype. The 2013 guidelines from the College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology recommend EGFR mutation testing for adenocarcinomas and mixed lung cancers with an adenocarcinoma component regardless of clinical characteristics or risk factors [16]. Guidelines recommend laboratories use validated molecular testing methods with sufficient performance characteristics [16]; options include direct sequencing [17], immunohistochemistry (IHC) [17], and polymerase chain reaction-based analysis (e.g., Scorpion Amplification Refractory Mutation System technology [DxS]) [18]. From a clinical practice standpoint, EGFR reflex testing of resected pulmonary adenocarcinoma has demonstrated feasibility [19].

Phase 3 clinical trials in molecularly selected NSCLC populations

Available data from completed phase 3 trials of EGFR or ErbB family TKIs in EGFR mutation-positive NSCLC or clinically selected populations are summarized in Tables 1 and 2 and discussed below, along with recent phase 2 data for the newer generation of irreversible agents.

Reversible EGFR TKIs (gefitinib and erlotinib)

In the phase 3 NEJ002 trial of gefitinib versus carboplatin/paclitaxel in 230 patients with EGFR-activating mutations [20],

gefitinib significantly improved median progression-free survival (PFS, 10.8 vs. 5.4 months; $P < 0.001$) and response rate (RR, 73.7% vs. 30.7%; $P < 0.001$). Based on updated overall survival (OS) results, median OS was 27.7 months with gefitinib and 26.6 months with chemotherapy ($P = 0.483$), with this lack of difference potentially attributable to a high rate of crossover to gefitinib in the control arm [21]. WJTOG3405 was a phase 3 trial of gefitinib versus cisplatin/docetaxel in 177 Japanese patients with advanced or recurrent NSCLC with EGFR-activating mutations [22]. As in NEJ002, gefitinib significantly prolonged median PFS (9.2 vs. 6.3 months; $P < 0.0001$) and increased RR (62.1% vs. 32.2%; $P < 0.0001$) [22]; median OS was 34.8 months with gefitinib versus 37.3 months with cisplatin/docetaxel [23].

Results of the IPASS phase 3 trial, conducted in a population of 1217 patients with clinical characteristics predictive of EGFR mutations (namely East Asian nonsmokers with adenocarcinoma), provide additional support for the activity of gefitinib in this setting [24, 25]. Gefitinib was associated with a median PFS of 5.7 months that was noninferior to carboplatin/paclitaxel (5.8 months; $P < 0.001$ for noninferiority) and a significantly higher 12-month PFS (24.9% vs. 6.7%, respectively; $P < 0.001$) and RR (43.0% vs. 32.2%; $P < 0.001$) [24]. Median OS was 18.8 months for gefitinib and 17.4 months for chemotherapy ($P = 0.109$) [25]. In the subgroup of patients with EGFR-activating mutations ($n = 261$), PFS was significantly longer with gefitinib versus chemotherapy ($P < 0.001$) and the RR was significantly higher (71.2% vs. 47.3%; $P < 0.001$); conversely, in the EGFR mutation-negative subgroup, PFS was significantly shorter ($P < 0.001$) and the RR was significantly lower with gefitinib versus chemotherapy (1.1% vs. 23.5%; $P = 0.001$) [24]. OS was similar regardless of treatment in EGFR mutation-positive patients (21.6 vs. 21.9 months; $P = 0.990$), but was likely confounded by a high rate of crossover to EGFR-TKI therapy [25]. Exploration of biomarkers in IPASS found that gefitinib significantly prolonged PFS in cases where tumors had high EGFR gene copy number and an EGFR mutation, but not when high EGFR gene copy number was unaccompanied by an EGFR mutation; in the latter subset, PFS was significantly shorter with gefitinib versus carboplatin/paclitaxel.

The most recently published phase 3 data for first-line gefitinib versus chemotherapy for advanced NSCLC are from the Korean First-SIGNAL phase 3 trial, which included never smokers with lung adenocarcinoma [26]. In the overall study population ($N = 309$), there were no significant benefits for gefitinib versus cisplatin/gemcitabine with respect to RR (55.4% vs. 46.0%; $P = 0.101$), PFS (5.8 vs. 6.4 months; $P = 0.138$), or the primary endpoint of OS (22.3 vs. 22.9 months; $P = 0.604$). Among

Table 1. Phase 3 clinical trial results for EGFR or ErbB family TKIs as first-line therapy in molecularly selected NSCLC.

Trial	Study	Treatment arms	RR, %	Median PFS	Median OS	Most common grade ≥3 AEs (TKI vs. chemotherapy)
Gefitinib						
WJTOG3405 [22, 23, 71]	Japanese study of 177 patients with EGFR mutations	Gefitinib vs. cisplatin/ docetaxel	62.1 vs. 32.2 ($P < 0.0001$)	9.2 vs. 6.3 months (HR, 0.489; 95% CI, 0.336–0.710; $P < 0.0001$)	34.8 vs. 37.3 months (HR, 1.252; 95% CI, 0.883–1.775)	ALT elevation (24% vs. 2%), AST elevation (14% vs. 1%)
NEJ002 [20, 21]	Japanese study of 230 patients with EGFR mutations	Gefitinib vs. carboplatin/ paclitaxel	73.7 vs. 30.7 ($P < 0.001$)	10.8 vs. 5.4 months (HR, 0.322; 95% CI, 0.236–0.438; $P < 0.001$)	27.7 vs. 26.6 months (HR, 0.887; 95% CI, 0.634–1.241; $P = 0.483$)	Aminotransferase elevation (26% vs. 1%), appetite loss (5% vs. 6%)
IPASS [24]	East Asian study of 261 patients with EGFR mutations	Gefitinib vs. carboplatin/ paclitaxel	71.2 vs. 47.3 ($P < 0.001$)	HR, 0.48; 95% CI, 0.36–0.64; $P < 0.001$	HR, 0.91; 95% CI, 0.76–1.10	Diarrhea (3.8% vs. 1.4%), neutropenia (3.7% vs. 67.1%), rash/acne (3.1% vs. 0.8%)
First-SIGNAL [26]	Korean study of 42 patients with EGFR mutations	Gefitinib vs. cisplatin/ gemcitabine	84.6 vs. 37.5 ($P = 0.002$)	8.0 vs. 6.3 months (HR, 0.544; 95% CI, 0.269–1.100; $P = 0.086$)	27.2 vs. 25.6 months (HR, 1.043; 95% CI, 0.498–2.182)	Rash (29.3% vs. 2.0%), anorexia (13.8% vs. 57.3%), AST (11.3% vs. 2.0%)
Erlotinib						
OPTIMAL [27]	Chinese study of 165 patients with EGFR mutations	Erlotinib vs. carboplatin/ gemcitabine (up to four cycles)	83 vs. 36 ($P < 0.0001$)	13.1 vs. 4.6 months (HR, 0.16; 95% CI, 0.10–0.26; $P < 0.0001$)	NR	ALT elevation (4% vs. 1%), skin rash (2% vs. 0%)
EURTAC [28][1]	European study of 173 patients with EGFR mutations	Erlotinib vs. platinum-based chemotherapy (up to four cycles)	58 vs. 15 ($P < 0.0001$)	9.7 vs. 5.2 months (HR, 0.37; 95% CI, 0.25–0.54; $P < 0.0001$)	19.3 vs. 19.5 months (HR, 1.04; 95% CI, 0.65–1.68; $P = 0.87$)	Rash (13% vs. 0%), fatigue (6% vs. 20%)
Afatinib						
LUX-Lung 6 [33, 34][2]	Asian study of 364 patients with EGFR mutations	Afatinib vs. cisplatin/ gemcitabine (up to six cycles)	66.9 vs. 23.0 ($P < 0.0001$)	11.0 vs. 5.6 months (HR, 0.28; 95% CI, 0.20–0.39; $P < 0.0001$)	23.1 vs. 23.5 months (HR, 0.93; $P = 0.6137$)	Rash/acne (14.6% vs. 0%), diarrhea (5.4% vs. 0%), stomatitis/mucositis (5.4% vs. 0%)
LUX-Lung 3 [34, 35][2]	Global study of 345 patients with EGFR mutations	Afatinib vs. cisplatin/ pemetrexed (up to six cycles)	56 vs. 23 ($P = 0.001$)	11.1 vs. 6.9 months (HR, 0.58; 95% CI, 0.43–0.78; $P = 0.001$)	28.2 vs. 28.2 months (HR, 0.88; $P = 0.3850$)	Rash (16.2% vs. 0%), diarrhea (14.4% vs. 0%), paronychia (11.4% vs. 0%), stomatitis/mucositis (8.7% vs. 0.9%)

EGFR, epidermal growth factor receptor; TKIs, tyrosine kinase inhibitors; NSCLC, non-small-cell lung cancer; RR, response rate; PFS, progression-free survival; OS, overall survival; AEs, adverse events; HR, hazard ratio; CI, confidence interval; ALT, alanine aminotransferase; AST, aspartate aminotransferase; NR, not reported.

[1]Investigator assessed.

[2]Independent review.

42 patients with EGFR mutation-positive disease, the RR was significantly higher with gefitinib versus cisplatin/ gemcitabine (84.6% vs. 37.5%; $P = 0.002$), but PFS was not significantly different ($P = 0.086$).

As with gefitinib, phase 3 data are available to support the activity of erlotinib versus doublet chemotherapy as first-line therapy for EGFR mutation-positive NSCLC. The OPTIMAL trial compared erlotinib against carboplatin/ gemcitabine as first-line therapy in 165 Chinese patients with EGFR-activating mutations, with significant benefits for erlotinib with respect to PFS (13.1 vs. 4.6 months; $P < 0.0001$) and RR (83% vs. 36%; $P < 0.0001$) observed [27].

Table 2. PFS and OS from phase 3 clinical trials for EGFR or ErbB family TKIs for NSCLC by Del19 and L858R EGFR mutation subtypes.

Trial	Treatment arms	Median PFS, Del19	Median PFS, L858R	Median OS, Del19	Median OS, L858R
Gefitinib					
WJTOG3405 [22]	Gefitinib vs. cisplatin/docetaxel (up to six cycles)	9.0 vs. 6.0 months (HR, 0.45; 95% CI, 0.27–0.77)	9.6 vs. 6.7 months (HR, 0.51; 95% CI, 0.29–0.90)	NR	NR
NEJ002 [20, 21]	Gefitinib vs. carboplatin/paclitaxel (up to six cycles)	11.5 months for gefitinib (HR, 0.35; 95% CI, 0.23–0.52)	10.8 months for gefitinib (HR, 0.32; 95% CI, 0.20–0.50)	NR	NR
IPASS [24, 25, 72]	Gefitinib vs. carboplatin/paclitaxel (up to six cycles)	HR, 0.38; 95% CI, 0.26–0.56	HR, 0.55; 95% CI, 0.35–0.87	HR, 0.79; 95% CI, 0.54–1.15	HR, 1.44; 95% CI, 0.90–2.30
Erlotinib					
OPTIMAL [27]	Erlotinib vs. carboplatin/gemcitabine (up to four cycles)	HR, 0.13; 95% CI, 0.07–0.25	HR, 0.26; 95% CI, 0.14–0.49	NR	NR
EURTAC [4, 28]	Erlotinib vs. platinum-based chemotherapy (up to four cycles)	11.0 vs. 4.6 months (HR, 0.30; 95% CI, 0.18–0.50; $P < 0.0001$)	8.4 vs. 6.0 months (HR, 0.55; 95% CI, 0.29–1.02; $P = 0.0539$)	HR, 0.94; 95% CI, 0.57–1.54	HR, 0.99; 95% CI, 0.56–1.76
Afatinib					
LUX-Lung 6 [33, 34]	Afatinib vs. cisplatin/gemcitabine (up to six cycles)	HR, 0.20[1]; 95% CI, 0.13–0.33	HR, 0.32[1]; 95% CI, 0.19–0.52	31.4 vs. 18.4 months (HR, 0.64; 95% CI, 0.44–0.94; $P = 0.0229$)	HR, 1.22; 95% CI, 0.81–1.83
LUX-Lung 3 [34, 35]	Afatinib vs. cisplatin/pemetrexed (up to six cycles)	HR, 0.28[1]; 95% CI, 0.18–0.44	HR, 0.73[1]; 95% CI, 0.46–1.17	33.3 vs. 21.1 months (HR, 0.54; 95% CI, 0.36–0.79; $P = 0.0015$)	HR, 1.30; 95% CI, 0.80–2.11

PFS, progression-free survival; OS, overall survival; EGFR, epidermal growth factor receptor; TKIs, tyrosine kinase inhibitors; NSCLC, non-small-cell lung cancer; HR, hazard ratio; CI, confidence interval; NR, not reported.
[1]Independent review.

Similarly, the EURTAC phase 3 trial assessed erlotinib versus first-line platinum-based chemotherapy in 174 patients with EGFR mutation-positive advanced NSCLC [28]. At preplanned interim analysis, erlotinib significantly improved the investigator-assessed primary endpoint of PFS (9.4 vs. 5.2 months; $P < 0.0001$), prompting early closure of the study. The final results of EURTAC (Table 1) were consistent with those in the interim analysis.

Both gefitinib and erlotinib continue to be studied in EGFR mutation-positive advanced NSCLC in ongoing phase 3 and 4 trials (Table 3).

Irreversible ErbB family TKIs (afatinib and dacomitinib)

Afatinib is an irreversible ErbB family inhibitor of EGFR/Erb1, ErbB2/HER2, and ErbB4/HER4 [29, 30]. It has also been shown to inhibit phosphorylation of ErB3/HER3 in vitro [31]. In the LUX-Lung 2 phase 2 trial of first-line or second-line afatinib in EGFR mutation-positive advanced or recurrent NSCLC ($N = 129$), median PFS was 10.1 months, median OS was 24.8 months, and the independent confirmed RR was 61% [32]. In subgroups based on specific mutation type, the RR was 66% among 106 patients with common mutations (exon deletion 19 or exon 21 L858R) and 39% among 23 patients with other mutations. Results are also available from phase 3 trials (LUX-Lung 3 and LUX-Lung 6) of afatinib in patients with EGFR mutation-positive lung adenocarcinoma (Table 1) [33, 34]. Data from LUX-Lung 6, which evaluated afatinib versus cisplatin plus gemcitabine in Asian patients, showed significantly prolonged PFS with afatinib versus gemcitabine/cisplatin (11.0 vs. 5.6 months, respectively; $P < 0.0001$) and a significantly higher RR (66.9% vs. 23.0%; $P < 0.0001$) by independent review [33]. The global LUX-Lung 3 study evaluated afatinib versus

Table 3. Ongoing phase 3/4 clinical trials of EGFR or ErbB family TKIs as first-line therapy in EGFR mutation-positive or clinically selected advanced NSCLC.

Trial	Estimated enrollment	Key eligibility	Treatment arm(s)	Primary outcome	Status[1]
Gefitinib					
Phase 4 (Europe)—NCT01203917	1060	Caucasian race EGFR mutation-positive disease	Gefitinib	RR	Active, not recruiting
Erlotinib					
Phase 3—NCT01667562	30	EGFR mutation-positive disease	Erlotinib	PFS	Active, not recruiting
Afatinib					
Phase 3—NCT01121393 (LUX-Lung 6)	364	Adenocarcinoma EGFR mutation-positive disease	Afatinib vs. cisplatin/ gemcitabine	PFS	Active, not recruiting
Phase 3—NCT00949650 (LUX-Lung 3)	345	Adenocarcinoma EGFR mutation-positive disease	Afatinib vs. cisplatin/ pemetrexed	PFS	Active, not recruiting
Dacomitinib					
Phase 3—NCT01774721 (ARCHER 1050)	440	Known histology EGFR mutation-positive disease, specifically exon 19 deletion or L858R mutation in exon 21 (may occur with an exon 20 T790M mutation)	Dacomitinib vs. gefitinib	PFS	Active, not recruiting

EGFR, epidermal growth factor receptor; TKIs, tyrosine kinase inhibitors; NSCLC, non-small-cell lung cancer; RR, response rate; PFS, progression-free survival.
[1]Per the U.S. National Institutes of Health ClinicalTrials.gov database, accessed July 2015.

pemetrexed/cisplatin and also showed significantly prolonged PFS with afatinib both overall (11.1 vs. 6.9 months; $P = 0.001$) and in patients with common (exon 19 deletions or L858R) EGFR mutations (13.6 vs. 6.9 months; $P < 0.0001$) by independent review; results also showed a significantly higher RR (56% vs. 23%; $P = 0.001$) and delayed worsening of lung cancer-related symptoms with afatinib [34–36]. Of note, randomization in both the LUX-Lung 6 and LUX-Lung 3 trials was stratified by type of EGFR mutation (L858R, Del19, or other); efficacy analyses by EGFR mutation type were prespecified [33, 35]. Analysis of OS by EGFR mutation type in LUX-Lung 6 and LUX-Lung 3 revealed an OS benefit in afatinib-treated patients with Del19 mutations (Table 2) [34]. In LUX-Lung 6, the median OS in patients with Del19 EGFR mutation treated with afatinib versus chemotherapy was 31.4 versus 18.4 months ($P = 0.0229$). However, no significant differences in OS were observed between treatment groups among patients with L858R mutation. Similarly, in LUX-Lung 3, afatinib-treated patients in the Del19 subgroup demonstrated prolonged OS compared with chemotherapy-treated patients (33.3 vs. 21.1 months; $P = 0.0015$), but no significant OS differences were observed between the two treatment arms in patients with L858R mutation.

Dacomitinib (Pfizer, New London, CT) is an irreversible pan-HER inhibitor of EGFR/ErbB1, ErbB2/HER2, and ErbB4/HER4 [37]. A phase 3 trial, ARCHER 1050 (NCT01774721), will evaluate dacomitinib versus gefitinib in the first-line treatment of EGFR mutation-positive advanced NSCLC (Table 3). An ongoing phase 3 trial of dacomitinib in advanced NSCLC unresponsive to standard therapy, including one to three lines of chemotherapy and an EGFR TKI (BR26; NCT01000025), is not focused on molecularly selected patients; however, its secondary endpoints include OS in EGFR mutation-positive or wild-type *KRAS* subsets. Preliminary results were recently presented and showed that dacomitinib did not improve OS versus placebo (6.8 vs. 6.3 months; $P = 0.99$), but did significantly improve PFS (2.7 vs. 1.4 months; $P < 0.0001$) and RR (7% vs. 1%; $P = 0.001$) [38]. Effect of dacomitinib on OS was similar regardless of EGFR mutation status; however, OS results appeared to differ by *KRAS* mutation status, with dacomitinib improving OS in patients with *KRAS* wild-type tumors (7.0 vs. 5.2 months; hazard ratio [HR], 0.79; 95% confidence interval [CI], 0.61–1.03), but worsening OS in patients with *KRAS* mutation-positive NSCLC (5.8 vs. 8.3 months; HR, 2.1; 95% CI, 1.05–4.22; interaction $P = 0.08$). In a randomized phase 2 trial of dacomitinib versus erlotinib after one or two lines of chemotherapy

for advanced NSCLC, PFS was prolonged with dacomitinib in the overall population (2.86 vs. 1.91 months for erlotinib; $P = 0.012$) and most clinically or molecularly defined subgroups, including patients with EGFR mutation-positive disease (3.71 vs. 1.91 months; $P = 0.006$) [39]. Preliminary results are available from a phase 2 trial of first-line dacomitinib in 74 patients with adenocarcinoma who were either nonsmokers/former light smokers or had documented EGFR mutations; median PFS was 9.30 months in all patients, but had not been reached in the 27-patient subset with confirmed EGFR mutations (all of whom had experienced tumor shrinkage) [40]. In the EGFR mutation-positive subgroup, 4-, 6-, and 9-month PFS rates were 95.7%, 84.7%, and 84.7%, respectively (higher than those in the overall population, which were 73.3%, 67.0%, and 57.1%, respectively). In an updated analysis of 47 patients with EGFR mutations involving exons 19 or 21, the partial RR was 74% and 1-year and median PFS were 77% and 17 months, respectively [41]. Preliminary results from an ongoing phase 3 trial (ARCHER 1009) in unselected patients with advanced NSCLC suggest similar PFS with dacomitinib versus erlotinib as second- or third-line therapy in the coprimary populations, all patients (2.6 vs. 2.6 months; $P = 0.229$) and KRAS wild-type patients (2.6 vs. 2.6 months; $P = 0.587$); OS and outcomes for patients with EGFR mutation are not mature [42].

Compensatory ErbB Family Signaling

Just as there are several sensitizing EGFR mutations that predict response to EGFR TKIs, other mutations have been linked to acquired resistance. The T790M mutation in exon 20 was the first identified mechanism of acquired resistance to EGFR TKIs and is thought to influence receptor affinity toward ATP and occur in at least 50% of cases of acquired resistance to EGFR-TKI therapy [43]. Interestingly, patients with T790M mutations have been shown to have a more favorable disease course in the postprogression period relative to patients with acquired resistance without T790M mutation [44]. Few secondary mutations other than T790M have been identified to date and include D761Y, L747S, and T854A; these non-T790M mutations are thought to occur in <5% of EGFR-mutated TKI-resistant patients [43]. To specifically evaluate mechanisms of acquired resistance to EGFR-TKI therapy for NSCLC, investigators at the Memorial Sloan Kettering Cancer Center implemented a prospective registry (NCT00579683) to compare EGFR gene sequence at relapse versus prior to EGFR-TKI therapy. Secondary outcomes include identification of novel EGFR mutations and resistance mechanisms and a more precise characterization of the frequency and clinical implications of T790M mutations.

A number of signaling pathways share downstream targets with EGFR and have been implicated in resistance to EGFR TKIs (Fig. 1) [43], including hepatocyte growth factor receptor (MET) [45], AXL receptor TK [46], and nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) [47, 48]. Other resistance mechanisms include ERBB2/HER2 amplification [49], epithelial-to-mesenchymal transition (EMT) [50], and BIM polymorphism [51]. In addition, PI3K/Akt/mTOR (downstream mediator of EGFR signaling) may function as a compensatory EGFR signaling pathway; mutations in the main catalytic subunit of PI3K (PIK3CA) have been associated with primary and acquired EGFR-TKI resistance, coexisting with EGFR mutations in some cases [52, 53]. Systematic genetic and histologic analyses of tumor biopsy specimens from 37 patients with drug-resistant EGFR mutation-positive NSCLC were performed to determine mechanisms of acquired resistance and found that all tumors retained their pretreatment activating EGFR mutations and many acquired other resistance mechanisms, including T790M or MET gene amplification (Fig. 2) [53]. The authors also reported histology transformation in tumors with acquired resistance; five patients with lung adenocarcinoma before EGFR-TKI treatment were found to have small-cell lung cancer (SCLC) in drug-resistant
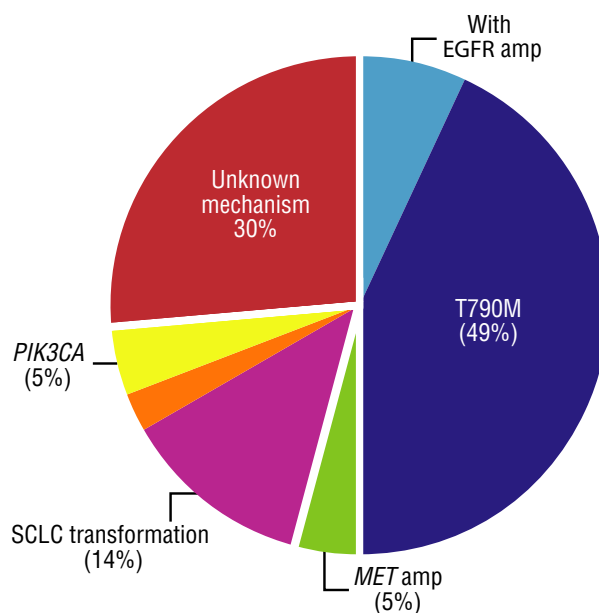

Figure 2. Frequency of observed drug resistance mechanisms in 37 patients with NSCLC biopsied at the time of acquired resistance. Note that orange wedge represents one patient who had both SCLC transformation and acquisition of a PIK3CA mutation. EGFR, epidermal growth factor receptor; MET, met proto-oncogene; PIK3CA, phosphatidylinositol-4,5-bisphosphate 3-kinase, catalytic subunit alpha; NSCLC, non-small-cell lung cancer; SCLC, small-cell lung cancer. From Sequist et al. [53]. Adapted with permission from AAA.

tumor biopsies, while retaining the original EGFR mutation [53]. This transition from NSCLC to SCLC appears to be specific to EGFR-TKI resistance and supports the importance of repeat biopsies at the time of resistance.

Agents that target each of these various resistance pathways are in clinical development, with several evaluated in completed or ongoing phase 3 trials in molecularly selected NSCLC populations.

As *MET* oncogene amplification has been implicated in both primary and acquired resistance to EGFR inhibition [54], MET inhibitors are being studied in combination with erlotinib for molecularly selected, advanced NSCLC. The combination of onartuzumab (MetMAb; Genentech, South San Francisco, CA), a monoclonal antibody (mAb) targeting the MET receptor [55], plus erlotinib is being evaluated in a phase 3 trial in previously treated, MET-expressing (by IHC), advanced NSCLC (NCT01456325). Based on preliminary data suggesting that the addition of onartuzumab to erlotinib did not improve OS (6.8 vs. 9.1 months; $P = 0.068$), PFS (2.7 vs. 2.6 months; $P = 0.92$), or RR (8.4% vs. 9.6%; $P = 0.63$) versus erlotinib/placebo, an independent data review committee recommended stopping the trial for futility; however, exploratory analyses by molecular subgroups are pending [56]. In a placebo-controlled phase 2 trial of onartuzumab plus erlotinib in 137 unselected, previously treated patients with advanced NSCLC, onartuzumab recipients with MET-expressing (by IHC) tumors had significantly prolonged PFS ($P = 0.04$) and OS ($P = 0.002$) [57]. Conversely, onartuzumab was associated with shortened PFS in the MET expression-negative setting ($P = 0.05$). Tivantinib (ARQ 197; ArQule, Inc., Woburn, MA), a MET TKI [58], is being evaluated in combination with erlotinib in a phase 3 trial in Asian patients with wild-type EGFR advanced NSCLC (ATTENTION; NCT01377376). Preliminary data from the trial suggest some benefit from adding tivantinib to erlotinib in this patient population (OS: 12.9 vs. 11.2 months with placebo/erlotinib, $P = 0.427$; PFS: 2.9 vs. 2.0 months; $P = 0.019$); however, the trial lacked statistical power due to premature termination for toxicity concern (imbalance in interstitial lung disease between groups) [59]. In a placebo-controlled phase 2 trial of tivantinib plus erlotinib in 167 unselected, previously treated patients with advanced NSCLC, tivantinib/erlotinib recipients with wild-type EGFR had numerically improved PFS ($P = 0.25$) and OS ($P = 0.25$) [60]. Tivantinib/erlotinib-treated patients with *KRAS* mutations had significantly improved PFS ($P < 0.01$) and numerically improved OS ($P = 0.17$).

Another approach to attenuating acquired resistance to EGFR TKIs is to prevent the associated epigenetic changes and EMT that have been observed during EGFR-TKI therapy [61]. The histone deacetylase inhibitor entinostat, which has these capabilities, was evaluated in a placebo-controlled phase 2 trial with erlotinib in chemotherapy-pretreated,

molecularly unselected, advanced NSCLC [61]. The combination of erlotinib plus entinostat did not improve 4-month PFS (18% vs. 20% with erlotinib/placebo; $P = 0.7$) or other efficacy outcomes. However, a planned biomarker analysis found that the 26-patient subset with high baseline expression of E-cadherin had significantly longer OS with entinostat/erlotinib versus erlotinib alone (9.4 vs. 5.4 months; $P = 0.03$) and numerically prolonged PFS (3.7 vs. 1.9 months; $P = 0.19$), supporting further study of the combination in patients with high E-cadherin expression.

Conclusions and Future Directions

Personalized therapy is now a clinical reality in NSCLC—an era that began with the reversible EGFR TKIs gefitinib and erlotinib for EGFR mutation-positive disease. Interim data for several phase 3 trials of gefitinib or erlotinib monotherapy versus doublet chemotherapy were favorable enough to warrant early study closures, with subsequent treatment crossover as a confounding factor in the analysis of OS. In contrast, data show less favorable outcomes with EGFR TKIs versus conventional platinum-based chemotherapy in patients with EGFR wild-type NSCLC and thus EGFR TKIs cannot be recommended in the first-line metastatic setting without evidence of a sensitizing EGFR mutation [16]. Results are awaited from ongoing phase 3 trials of investigational treatments and EGFR-TKI-containing combination regimens in molecularly selected NSCLC. We recommend that EGFR mutation testing be done both at the time of diagnosis for patients who are suitable for therapy and also considered at the time of recurrence or progression in an effort to determine the mechanism(s) of resistance and to more effectively direct future therapies [16]. Recent data suggest clinical benefits with the continuation of EGFR TKIs beyond progression in patients developing acquired resistance [1, 16, 62]. In fact, discontinuation of EGFR-TKI therapy in patients who were once sensitive to EGFR inhibition may lead to more rapid cancer progression [63, 64]. In terms of treatment options when genomic data are unavailable, erlotinib has been shown to significantly prolong PFS and OS compared with placebo irrespective of EGFR mutation status both as switch maintenance therapy following conventional chemotherapy and in patients with NSCLC after failure of first- or second-line chemotherapy [14, 65]. Similarly, continuing treatment with afatinib in patients with metastatic NSCLC who had progressed following treatment with reversible EGFR TKIs and afatinib revealed that afatinib treatment beyond progression significantly improves PFS and objective response rate versus chemotherapy alone [62].

To date, clinical investigations of EGFR-directed therapy for EGFR mutation-positive NSCLC have focused on the use of TKI monotherapy for advanced disease. Molecularly

focused evaluations of other types of therapies, such as anti-EGFR therapeutic vaccines [66] or TKI/mAb combinations that more broadly target the ErbB family (e.g., erlotinib plus the HER2-targeted mAb pertuzumab [67], and afatinib plus the EGFR-targeted mAb cetuximab [68]), may be worthwhile. Outstanding questions include whether EGFR TKIs confer clinical benefit when used in less advanced disease, which will be addressed by ongoing and recently completed phase 3 trials of gefitinib or erlotinib versus combination chemotherapy in the adjuvant or neoadjuvant setting for patients with EGFR mutation-positive NSCLC (NCT01405079 [ADJUVANT trial of gefitinib vs. vinorelbine/platinum stage II–IIIA(N1–N2) disease]; NCT01407822 [EMERGING trial of erlotinib vs. gemcitabine/cisplatin as neoadjuvant therapy for stage IIIA(N2) disease], and NCT00373425 [RADIANT trial of erlotinib vs. placebo added to adjuvant chemotherapy for stage IB–IIIA disease]). Preliminary results from the RADIANT trial were recently presented and suggest that adjuvant erlotinib may prolong disease-free survival (DFS) in patients with EGFR mutation-positive NSCLC, but adjuvant erlotinib did not significantly improve DFS over placebo in the overall patient population [69, 70]. There is also a need for pooled testing of targets with limited quantities of tissue for testing and the centralization of data repositories for this information.

Acknowledgments

Conflict of Interest

Robert Jotte serves on the Speaker's Bureau for Bristol-Myers Squibb. David Spigel has served in an uncompensated advisory role for Genentech and Boehringer Ingelheim.

References

1. National Comprehensive Cancer Network. 2015. NCCN clinical practice guideline in oncology. Non-Small Cell Lung Cancer. Version 5.2015.

2. Iressa. 2005. Iressa® (gefitinib tablets) [package insert]. AstraZeneca Pharmaceuticals LP, Wilmington, DE.

3. Avastin. 2014. AVASTIN® (bevacizumab). Solution for intravenous infusion [package insert]. Genentech, Inc., South San Francisco, CA.

4. Tarceva. 2014. TARCEVA® (erlotinib) tablets, for oral use [package insert]. Genentech, Inc., South San Francisco, CA.

5. XALKORI. 2014. XALKORI® (crizotinib) capsules, oral [package insert]. Pfizer Labs, New York, NY.

6. Herbst, R. S., and D. M. Shin. 2002. Monoclonal antibodies to target epidermal growth factor receptor-positive tumors: a new paradigm for cancer therapy. Cancer 94:1593–1611.

7. Hirsch, F. R., G. V. Scagliotti, C. J. Langer, M. Varella-Garcia, and W. A. Franklin. 2003. Epidermal growth factor family of receptors in preneoplasia and lung cancer: perspectives for targeted therapies. Lung Cancer 41(Suppl. 1):S29–S42.

8. Buettner, R., L. B. Mora, and R. Jove. 2002. Activated STAT signaling in human tumors provides novel molecular targets for therapeutic intervention. Clin. Cancer Res. 8:945–954.

9. Ohsaki, Y., S. Tanno, Y. Fujita, E. Toyoshima, S. Fujiuchi, Y. Nishigaki, et al. 2000. Epidermal growth factor receptor expression correlates with poor prognosis in non-small cell lung cancer patients with p53 overexpression. Oncol. Rep. 7:603–607.

10. GILOTRIF. 2014. GILOTRIF® (afatinib) tablets, for oral use [package insert]. Boehringer Ingelheim Pharmaceuticals, Inc., Ridgefield, CT.

11. Pao, W., V. Miller, M. Zakowski, J. Doherty, K. Politi, I. Sarkaria, et al. 2004. EGF receptor gene mutations are common in lung cancers from "never smokers" and are associated with sensitivity of tumors to gefitinib and erlotinib. Proc. Natl. Acad. Sci. USA 101:13306–13311.

12. Sharma, S. V., D. W. Bell, J. Settleman, and D. A. Haber. 2007. Epidermal growth factor receptor mutations in lung cancer. Nat. Rev. Cancer 7:169–181.

13. Paez, J. G., P. A. Janne, J. C. Lee, S. Tracy, H. Greulich, S. Gabriel, et al. 2004. EGFR mutations in lung cancer: correlation with clinical response to gefitinib therapy. Science 304:1497–1500.

14. Shepherd, F. A., J. Rodrigues Pereira, T. Ciuleanu, E. H. Tan, V. Hirsch, S. Thongprasert, et al; National Cancer Institute of Canada Clinical Trials Group. 2005. Erlotinib in previously treated non-small-cell lung cancer. N. Engl. J. Med. 353:123–132.

15. Eberhard, D. A., B. E. Johnson, L. C. Amler, A. D. Goddard, S. L. Heldens, R. S. Herbst, et al. 2005. Mutations in the epidermal growth factor receptor and in KRAS are predictive and prognostic indicators in patients with non-small-cell lung cancer treated with chemotherapy alone and in combination with erlotinib. J. Clin. Oncol. 23:5900–5909.

16. Lindeman, N. I., P. T. Cagle, M. B. Beasley, D. A. Chitale, S. Dacic, G. Giaccone, et al. 2013. Molecular testing guideline for selection of lung cancer patients for EGFR

and ALK tyrosine kinase inhibitors: guideline from the College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology. J. Thorac. Oncol. 8:823–859.

17. Yu, J., S. Kane, J. Wu, E. Benedettini, D. Li, C. Reeves, et al. 2009. Mutation-specific antibodies for the detection of EGFR mutations in non-small-cell lung cancer. Clin. Cancer Res. 15:3023–3028.

18. DxS Diagnostic Innovations cQI. DxS EGFR mutation test kit: for the detection of 29 mutations in the epidermal growth factor receptor (EGFR) gene. Instructions For Use. Product Codes: EG-03 and EG-04. Instructions Version: RU001b. Date of Revision: July 2009. Available at: http://www.dxsdiagnostics.com/Site/PDF/CMP/RUO-EGFR29/IFU-EGFR29-RUO-US.pdf (accessed 28 December 2009).

19. D'Angelo, S. P., B. Park, C. G. Azzoli, M. G. Kris, V. Rusch, M. Ladanyi, et al. 2011. Reflex testing of resected stage I through III lung adenocarcinomas for EGFR and KRAS mutation: report on initial experience and clinical utility at a single center. J. Thorac. Cardiovasc. Surg. 141:476–480.

20. Maemondo, M., A. Inoue, K. Kobayashi, S. Sugawara, S. Oisumi, H. Isobe, et al; North-East Japan Study Group. 2010. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N. Engl. J. Med. 362:2380–2388.

21. Inoue, A., K. Kobayashi, M. Maemondo, S. Sugawara, S. Oisumi, H. Isobe, et al; North-East Japan Study Group. 2013. Updated overall survival results from a randomized phase III trial comparing gefitinib with carboplatin-paclitaxel for chemo-naive non-small cell lung cancer with sensitive EGFR gene mutations (NEJ002). Ann. Oncol. 24:54–59.

22. Mitsudomi, T., S. Morita, Y. Yatabe, S. Negoro, I. Okamoto, J. Tsuratani, et al; West Japan Study Group. 2010. 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. 11:121–128.

23. Yoshioka, H., T. Mitsudomi, S. Morita, Y. Yatabe, S. Negoro, I. Okamoto, et al; West Japan Oncology Group. 2014. Final overall survival results of WJTOG 3405, a randomized phase 3 trial comparing gefitinib (G) with cisplatin plus docetaxel (CD) as the first-line treatment for patients with non-small cell lung cancer (NSCLC) harboring mutations of the epidermal growth factor receptor (EGFR). J. Clin. Oncol. 32: Abstract 8117.

24. Mok, T. S., Y. L. Wu, S. Thongprasert, C. H. Yang, D. T. Chu, N. Saijo, et al. 2009. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N. Engl. J. Med. 361:947–957.

25. Fukuoka, M., Y. L. Wu, S. Thongprasert, P. Sunpaweravong, S. S. Leong, V. Sriuranpong, et al. 2011. 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 non-small-cell lung cancer in Asia (IPASS). J. Clin. Oncol. 29:2866–2874.

26. Han, J. Y., K. Park, S. W. Kim, D. H. Lee, H. Y. Kim, M. J. Ahn, et al. 2012. First-SIGNAL: first-line single-agent Iressa versus gemcitabine and cisplatin trial in never-smokers with adenocarcinoma of the lung. J. Clin. Oncol. 30:1122–1128.

27. Zhou, C., Y. L. Wu, G. Chen, J. Feng, X. Q. Liu, C. Wang, et al. 2011. 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. 12:735–742.

28. Rosell, R., E. Carcereny, R. Gervais, A. Vergnenegre, B. Massuti, E. Felip, et al; Spanish Lung Cancer Group in collaboration with Groupe Francais de Pneumo-Cancerology and Associazione Italiana Oncologia Toracica. 2012. 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, randomised phase 3 trial. Lancet Oncol. 13:239–246.

29. Li, D., L. Ambrogio, T. Shimamura, S. Kubo, M. Takahashi, L. R. Chirieac, et al. 2008. BIBW2992, an irreversible EGFR/HER2 inhibitor highly effective in preclinical lung cancer models. Oncogene 27:4702–4711.

30. Solca, F., G. Dahl, A. Zoephel, G. Bader, M. Sanderson, C. Klein, et al. 2012. Target binding properties and cellular activity of afatinib (BIBW 2992), an irreversible ErbB family blocker. J. Pharmacol. Exp. Ther. 343:342–350.

31. Ioannou, N., A. M. Seddon, A. Dalgleish, D. Mackintosh, and H. Modjtahedi. 2013. Treatment with a combination of the ErbB (HER) family blocker afatinib and the IGF-IR inhibitor, NVP-AEW541 induces synergistic growth inhibition of human pancreatic cancer cells. BMC Cancer 13:41.

32. Yang, J. C., J. Y. Shih, W. C. Su, T. C. Hsia, C. M. Tsia, S. H. Ou, et al. 2012. Afatinib for patients with lung adenocarcinoma and epidermal growth factor receptor mutations (LUX-Lung 2): a phase 2 trial. Lancet Oncol. 13:539–548.

33. Wu, Y. L., C. Zhou, C. P. Hu, J. Feng, S. Lu, Y. Huang, et al. 2014. Afatinib versus cisplatin plus gemcitabine for first-line treatment of Asian patients with advanced non-small-cell lung cancer harbouring EGFR mutations (LUX-Lung 6): an open-label, randomised phase 3 trial. Lancet Oncol. 15:213–222.

34. Yang, J. C. H., Y. L. Wu, M. Schuler, M. Sebastiam, S. Popat, N. Yamamoto, et al. 2015. Afatinib versus cisplatin-based chemotherapy for EGFR mutation-positive lung adenocarcinoma (LUX-Lung 3 and LUX-Lung 6): analysis of overall survival data from two randomised, phase 3 trials. Lancet Oncol. 16:141–151.

35. Sequist, L. V., J. C. Yang, N. Yamamoto, K. O'Byrne, V. Hirsch, T. Mok, et al. 2013. Phase III study of afatinib or cisplatin plus pemetrexed in patients with metastatic lung adenocarcinoma with EGFR mutations. J. Clin. Oncol. 31:3327–3334.

36. Yang, J. C., V. Hirsh, M. Schuler, M. Yamamoto, K. J. O'Byrne, T. S. Mok, et al. 2013. 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. 31:3342–3350.

37. Engelman, J. A., K. Zejnullahu, C. M. Gale, E. Lifshits, A. J. Gonzales, T. Shimamura, et al. 2007. PF00299804, an irreversible pan-ERBB inhibitor, is effective in lung cancer models with EGFR and ERBB2 mutations that are resistant to gefitinib. Cancer Res. 67:11924–11932.

38. Ellis, P. M., G. Liu, M. Millward, F. Perrone, F. A. Shepherd, S. Sun, et al. 2014. NCIC CTG BR.26: a phase III randomized, double blind, placebo controlled trial of dacomitinib versus placebo in patients with advanced/metastatic non-small cell lung cancer (NSCLC) who received prior chemotherapy and an EGFR TKI. J. Clin. Oncol. 32: Abstract 8036.

39. Ramalingam, S. S., F. Blackhall, M. Krzakowski, C. H. Barrios, K. Park, I. Bover, et al. 2012. 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. 30:3337–3344.

40. Mok, T., D. R. Spigel, K. Park, M. A. Socinski, S. Y. Tung, D.-W. Kim, et al. 2010. Efficacy and safety of PF299804 as first-line treatment (TX) of patients (pts) with advanced (adv) NSCLC selected for activating mutation (mu) of epidermal growth factor receptor (EGFR). Ann. Oncol. 21:viii8. Abstract LBA18.

41. Kris, M. G., T. Mok, S.-H. I. Ou, R. Martins, D.-W. Kim, Z. Goldberg, et al. 2012. First-line dacomitinib (PF-00299804), an irreversible pan-HER tyrosine kinase inhibitor, for patients with EGFR-mutant lung cancers. J. Clin. Oncol. 30: Abstract 7530.

42. Ramalingam, S. S., P. A. Janne, T. Mok, K. O'Byrne, M. J. Boyer, H. Zhang, et al. 2014. Randomized, double-blinded study of dacomitinib, an irreversible pan-human epidermal growth factor receptor (HER) inhibitor, versus erlotinib for second-line/third-line therapy of locally advanced/metastatic non-small cell lung cancer (ARCHER 1009). J. Clin. Oncol. 32: Abstract 8018.

43. Nguyen, K. S., S. Kobayashi, and D. B. Costa. 2009. Acquired resistance to epidermal growth factor receptor tyrosine kinase inhibitors in non-small-cell lung cancers dependent on the epidermal growth factor receptor pathway. Clin. Lung Cancer 10:281–289.

44. Oxnard, G. R., M. E. Arcila, C. S. Sima, G. J. Riely, J. Chmielecki, M. G. Kris, et al. 2011. Acquired resistance to EGFR tyrosine kinase inhibitors in EGFR-mutant lung cancer: distinct natural history of patients with tumors harboring the T790M mutation. Clin. Cancer Res. 17:1616–1622.

45. Mazzone, M., and P. M. Comoglio. 2006. The Met pathway: master switch and drug target in cancer progression. FASEB J. 20:1611–1621.

46. Zhang, Z., J. C. Lee, L. Lin, V. Olivas, V. Au, T. LaFramboise, et al. 2012. Activation of the AXL kinase causes resistance to EGFR-targeted therapy in lung cancer. Nat. Genet. 44:852–860.

47. Bivona, T. G., H. Hieronymus, J. Parker, K. Chang, M. Taron, R. Rosell, et al. 2011. FAS and NF-kappaB signalling modulate dependence of lung cancers on mutant EGFR. Nature 471:523–526.

48. Blakely, C. M., E. Pazarentzos, V. Olivas, S. Athana, J. J. Yan, I. Tan, et al. 2015. NF-kappaB-activating complex engaged in response to EGFR oncogene inhibition drives tumor cell survival and residual disease in lung cancer. Cell Rep. 11:98–110.

49. Takezawa, K., V. Pirazzoli, M. E. Arcila, C. A. Nebhan, X. Song, E. de Stanchina, et al. 2012. HER2 amplification: a potential mechanism of acquired resistance to EGFR inhibition in EGFR-mutant lung cancers that lack the second-site EGFRT790M mutation. Cancer Discov. 2:922–933.

50. Byers, L. A., L. Diao, J. Wang, P. Saintigny, L. Girard, M. Peyton, et al. 2013. An epithelial-mesenchymal transition gene signature predicts resistance to EGFR and PI3K inhibitors and identifies Axl as a therapeutic target for overcoming EGFR inhibitor resistance. Clin. Cancer Res. 19:279–290.

51. Ng, K. P., A. M. Hillmer, C. T. Chuah, W. C. Juan, T. K. Ko, A. S. Teo, et al. 2012. A common BIM deletion polymorphism mediates intrinsic resistance and inferior responses to tyrosine kinase inhibitors in cancer. Nat. Med. 18:521–528.

52. Engelman, J. A., T. Mukohara, K. Zejnullahu, E. Lifshits, A. M. Borras, C. M. Gale, et al. 2006. Allelic dilution obscures detection of a biologically significant resistance mutation in EGFR-amplified lung cancer. J. Clin. Invest. 116:2695–2706.

53. Sequist, L. V., B. A. Waltman, D. Dias-Santagata, S. Digumarthy, A. B. Turke, P. Fidias, et al. 2011. Genotypic and histological evolution of lung cancers acquiring resistance to EGFR inhibitors. Sci. Transl. Med. 3:75ra26.

54. Bean, J., C. Brennan, J. Y. Shih, G. Riely, A. Viale, L. Wang, et al. 2007. MET amplification occurs with or without T790M mutations in EGFR mutant lung tumors with acquired resistance to gefitinib or erlotinib. Proc. Natl. Acad. Sci. USA 104:20932–20937.

55. Jin, H., R. Yang, Z. Zheng, M. Romero, J. Ross, H. Bou-Reslan, et al. 2008. MetMAb, the one-armed 5D5 anti-c-Met antibody, inhibits orthotopic pancreatic tumor growth and improves survival. Cancer Res. 68:4360–4368.

56. Spigel, D. R., M. J. Edelman, K. O'Byrne, L. Paz-Ares, D. S. Shames, W. Yu, et al. 2014. Onartuzumab plus erlotinib versus erlotinib in previously treated stage IIIb or IV NSCLC: results from the pivotal phase III randomized, multicenter, placebo-controlled METLung (OAM4971g) global trial. J. Clin. Oncol. 32: Abstract 8000.

57. Spigel, D. R., T. J. Ervin, R. A. Ramlau, D. B. Daniel, J. H. Jr Goldschmidt, G. R. Jr Blumenschein, et al. 2013. Randomized phase II trial of onartuzumab in combination with erlotinib in patients with advanced non-small-cell lung cancer. J. Clin. Oncol. 31:4105–4114.

58. Goldman, J. W., I. Laux, F. Chai, R. E. Savage, D. Ferrari, E. G. Garmey, et al. 2012. Phase 1 dose-escalation trial evaluating the combination of the selective MET (mesenchymal-epithelial transition factor) inhibitor tivantinib (ARQ 197) plus erlotinib. Cancer 118:5903–5911.

59. Azuma, K., H. Yoshioka, N. Yamamoto, T. Takahashi, M. Nishio, N. Katakami, et al. 2014. Tivantinib plus erlotinib versus placebo plus erlotinib in Asian patients with previously treated nonsquamous NSCLC with wild-type EGFR: first report of a phase III ATTENTION trial. J. Clin. Oncol. 32: Abstract 8044.

60. Sequist, L. V., J. von Pawel, E. G. Garmey, W. L. Akerley, W. Brugger, D. Ferrari, et al. 2011. Randomized phase II study of erlotinib plus tivantinib versus erlotinib plus placebo in previously treated non-small-cell lung cancer. J. Clin. Oncol. 29:3307–3315.

61. Witta, S. E., R. M. Jotte, K. Konduri, M. A. Neubauer, A. L. Spira, R. L. Ruxer, et al. 2012. Randomized phase II trial of erlotinib with and without entinostat in patients with advanced non-small-cell lung cancer who progressed on prior chemotherapy. J. Clin. Oncol. 30:2248–2255.

62. Schuler, M. H., C. H. Yang, K. Park, J. Bennouna, Y. M. Chen, C. Chouaid, et al. 2014. Continuation of afatinib beyond progression: results of a randomized, open-label, phase III trial of afatinib plus paclitaxel (P) versus investigator's choice chemotherapy (CT) in patients (pts) with metastatic non-small cell lung cancer (NSCLC) progressed on erlotinib/gefitinib (E/G) and afatinib-LUX-Lung 5 (LL5). J. Clin. Oncol. 32: Abstract 8019.

63. Riely, G. J., M. G. Kris, B. Zhao, T. Akhurst, D. T. Milton, E. Moore, et al. 2007. Prospective assessment of discontinuation and reinitiation of erlotinib or gefitinib in patients with acquired resistance to erlotinib or gefitinib followed by the addition of everolimus. Clin. Cancer Res. 13:5150–5155.

64. Chaft, J. E., G. R. Oxnard, C. S. Sima, M. G. Kris, V. A. Miller, and G. J. Riely. 2011. Disease flare after tyrosine kinase inhibitor discontinuation in patients with EGFR-mutant lung cancer and acquired resistance to erlotinib or gefitinib: implications for clinical trial design. Clin. Cancer Res. 17:6298–6303.

65. Cappuzzo, F., T. Ciuleanu, L. Stelmakh, S. Cicenas, A. Szczesna, E. Juhesz, et al; SATURN Investigators. 2010. Erlotinib as maintenance treatment in advanced non-small-cell lung cancer: a multicentre, randomised, placebo-controlled phase 3 study. Lancet Oncol. 11:521–529.

66. Wu, Y. L., K. Park, R. A. Soo, Y. Sun, K. Tyroller, D. Wages, et al. 2011. INSPIRE: a phase III study of the BLP25 liposome vaccine (L-BLP25) in Asian patients with unresectable stage III non-small cell lung cancer. BMC Cancer 11:430.

67. Felip, E., M. Ranson, S. Cedres, E. Dean, M. Brewster, P. Martinez, et al. 2012. A phase Ib, dose-finding study of erlotinib in combination with a fixed dose of pertuzumab in patients with advanced non-small-cell lung cancer. Clin. Lung Cancer 13:432–441.

68. Janjigian, Y. Y., E. F. Smit, H. J. Groen, L. Horn, S. Gettinger, D. R. Camidge, et al. 2014. Dual inhibition of EGFR with afatinib and cetuximab in kinase inhibitor-resistant EGFR-mutant lung cancer with and without T790M mutations. Cancer Discov. 4:1036–1045.

69. Kelly, K., N. Altorki, W. E. E. Eberhardt, M. E. R. O'Brien, D. R. Spigel, L. Crino, et al. 2014. A randomized, double-blind phase 3 trial of adjuvant erlotinib (E) versus placebo (P) following complete tumor resection with or without adjuvant chemotherapy in patients (pts) with stage IB-IIIA EGFR positive (IHC/FISH) non-small cell lung cancer (NSCLC): RADIANT results. J. Clin. Oncol. 32: Abstract 7501.

70. Shepherd, F. A., N. K. Altorki, W. E. E. Eberhardt, M. E. R. O'Brien, D. R. Spigel, L. Crino, et al. 2014. Adjuvant erlotinib (E) versus placebo (P) in non-small cell lung cancer (NSCLC) patients (pts) with tumors carrying EGFR-sensitizing mutations from the RADIANT trial. J. Clin. Oncol. 32: Abstract 7513.

71. Mitsudomi, T., S. Morita, Y. Yantabe, S. Negoro, I. Okamoto, T. Seto, et al. 2012. Updated overall survival results of WJTOG 3405, a randomized phase III trial comparing gefitinib (G) with cisplatin plus docetaxel (CD) as the first-line treatment for patients with non-small cell lung cancer harboring mutations of the epidermal growth factor receptor (EGFR). J. Clin. Oncol. 30: Abstract 7521.

72. Yang, J., Y. L. Wu, N. Saijo, S. Thongprasert, D. T. Chu, Y. M. Chen, et al. 2011. Efficacy outcomes in first-line treatment of advanced NSCLC with gefitinib (G) vs carboplatin/paclitaxel (C/P) by epidermal growth factor receptor (EGFR) gene-copy number score and by most common EGFR mutation subtypes—exploratory data from IPASS. Eur. J. Cancer 47: Poster 9132.