Identification of Recurrent FGFR3–TACC3 Fusion Oncogenes from Lung Adenocarcinoma

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Abstract

Purpose: Targetable oncogenic alterations are detected more commonly in patients with non–small cell lung cancer (NSCLC) who never smoked cigarettes. For such patients, specific kinase inhibitors have emerged as effective clinical treatments. However, the currently known oncogenic alterations do not account for all never smokers who develop NSCLC. We sought to identify additional oncogenic alterations from patients with NSCLC to define additional treatment options.

Experimental Design: We analyzed 576 lung adenocarcinomas from patients of Asian and Caucasian ethnicity. We identified a subset of cancers that did not harbor any known oncogenic alteration. We performed targeted next-generation sequencing (NGS) assay on 24 patients from this set with >75% tumor cell content.

Results: EGFR mutations were the most common oncogenic alteration from both Asian (33%) and Caucasian (41.6%) patients. No known oncogenic alterations were present in 25.7% of Asian and 31% of Caucasian tumor specimens. We identified a FGFR3–TACC3 fusion event in one of 24 patients from this subset using targeted NGS. Two additional patients harboring FGFR3–TACC3 were identified by screening our entire cohort (overall prevalence, 0.5%). Expression of FGFR3–TACC3 led to IL3 independent growth in Ba/F3 cells. These cells were sensitive to pan-fibroblast growth factor receptor (pan-FGFR) inhibitors but not the epidermal growth factor (EGFR) inhibitor gefitinib.

Conclusions: FGFR3–TACC3 rearrangements occur in a subset of patients with lung adenocarcinoma. Such patients should be considered for clinical trials featuring FGFR inhibitors. Clin Cancer Res; 20(24); 6551–8. ©2014 AACR.

Introduction

The identification of somatic oncogenic alterations has translated into the development of effective targeted ther-

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Note: Supplementary data for this article are available at Clinical Cancer Research Online (http://clincancerres.aacrjournals.org/).

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Translational Relevance
The identification of oncogenic alterations in subsets of patients with non–small cell lung cancer (NSCLC) has accelerated the development of targeted therapies including epidermal growth factor receptor (EGFR) and anaplastic lymphoma kinase (ALK) inhibitors. Although oncogenic alterations are more prevalent in never smokers than smokers with NSCLC, not all cancers from never smokers contain a known alteration. By studying a cohort of lung adenocarcinomas from never smokers without a known oncogenic alteration, we identified recurrent FGFR3–TACC3 rearrangements. This fusion gene is oncogenic in vitro and the transformed cells are sensitive to fibroblast growth factor receptor (FGFR) kinase inhibitors. These findings identify a population of patients with lung cancer who could benefit from treatment with FGFR kinase inhibitors.

Patients and Methods

Patients
Tumor specimens from patients with NSCLC were obtained at the time of surgical resection and flash-frozen in liquid nitrogen. The initial discovery cohort of patients were limited to never (<100 cigarettes/lifetime) or former light (<10 pack years) smokers, but subsequently included 61 samples from former or current smokers whose cancers did not harbor a known genomic alteration. These studies identify a fusion product involving fibroblast growth factor receptor 3 (FGFR3) and transforming acidic coiled-coil 3 (TACC3) in a subset of patients suggesting that patients with NSCLC harboring this alteration should be enrolled in clinical trials evaluating FGFR kinase inhibitors.

Point mutations or small insertions and deletions were investigated by RT-PCR carried out using the QuantiTec Reverse Transcription Kit (Qiagen). Primers were specifically designed for the tyrosine kinase domain of EGFR and HER2 and the serine/threonine-protein kinase domain of BRAF. A DNA-based PCR was used to analyze KRAS exons 2 and 3. All primer sequences are available upon request. The amplicons were analyzed by direct sequencing and/or run on the Agilent 2100 Bioanalyzer (Agilent Technologies) using the DNA 1000 Kit. The DNA 1000 Kit allows an easy separation of fragments with sizes ranging from 25 to 1,000 bp aiding in the detection of small insertions/deletions and point mutations. Point mutations were identified following digestion with the endonuclease Surveyor (Surveyor Mutation Detection Kit; Transgenic), which specifically cuts portions of mismatched DNA as previously described.

Next-generation DNA sequencing
Library construction was performed as previously described using 50 to 200 ng of DNA sheared by sonication to approximately 100 to 400 bp prior to end-repair, dA

RNA and DNA extraction
Each frozen tissue sample was sectioned using a microtome to obtain between 10 and 15 mg of tissue for RNA and DNA extraction. RNA was extracted using Trizol (Ambion) according to the manufacturer’s instructions and subsequently purified using the RNasy Mini Kit (Qiagen). DNA was extracted from frozen tissue using the DNeasy Blood and Tissue Kit (Qiagen) and quantified using a standardized PicoGreen fluorescence assay (Invitrogen). RNA concentrations were measured using a NanoDrop ND-2000 Spectrophotometer (Thermo Scientific) or alternatively RiboGreen fluorescent assay (Invitrogen) and the RNA integrity was assessed using an Agilent 2100 Bioanalyzer (Agilent Technologies).

Genonomic profiling
All tumors were first genotyped to determine whether they contained a previously known oncogenic alteration. Point mutations or small insertions and deletions were investigated by RT-PCR and sequencing. DNA was sheared by sonication to approximately 100 to 400 bp prior to end-repair, dAT.
addition and ligation of indexed, Illumina sequencing adaptors (20). Enrichment of target sequences (3,320 exons of 182 cancer-related genes and 37 introns from 14 genes recurrently rearranged in cancer representing approximately 1.1 Mb of the human genome) was achieved by solution-based hybrid capture with a custom Agilent SureSelect biotinylated RNA baitset (20). The selected libraries were sequenced on an Illumina HiSeq 2000 platform using 49 × 49 paired-end reads. Sequence data from genomic DNA were mapped to the reference human genome (hg19) using the Burrows-Wheeler Aligner and were processed using the publicly available SAMtools, Picard, and Genome Analysis Tool kit (21, 22). Genomic rearrangements were detected by clustering chimeric reads mapped to targeted introns.

Ba/F3 cell construction and drug treatments

The full-length cDNA of FGFR3–TACC3 was obtained by RT-PCR amplification directly from tumor RNA. The primers used were 5′-TTGTGAGCCCGCACTATGGGCGCCC-3′ (forward) and 5′-TTCTTAGGAGATCTGCTCCATCTG-3′ (reverse). The fusion transcript was then inserted into the pDNRDual vector (BD Biosciences) using SalI and XbaI and recombined into a retroviral flag epitope tag containing expression vector PJ1540 (in-house). Retrovirus containing FGFR3–TACC3 was produced using 293T cells transfected with 3.0 μg of plasmid and p-Ampho packaging vector using FuGene6 (Roche) according to the manufacturer’s instructions. Retroviral infection and culture of Ba/F3 cells were performed using previously described methods (8, 15, 23). Ba/F3 cells were treated with increasing concentrations of BGI398, ponatinib, and gefitinib and their viability was tested in an MTS assay according to previously established methods (8, 23, 24). Results were plotted in logarithmic scale using GraphPad Prism Version 6.0 (GraphPad Software Inc.) and results were repeated in triplicate.

Western blotting and antibodies

Cells were lysed in NP-40 buffer. Thirty micrograms of total protein was loaded and separated by gel electrophoresis on 4% to 12% SDS Bis-Tris polyacrylamide gels (Invitrogen) and transferred to polyvinylidene difluoride (PVDF)-P membranes (Millipore). Membranes were probed overnight with anti-phosphotyrosine clone 4G10 antibody (Millipore), FGFR3 mAb Y724 (Abcam), anti-α-tubulin Ab and Flag antibody (Sigma-Aldrich). Detection by immunoblotting was according to the antibody manufacturer’s recommendations.

Results

Patients

Frozen tumor specimens were obtained from 664 patients. We excluded 88 tumors from the analyses because they were of non-adenocarcinoma histology (n = 56), had no specified ethnicity data (n = 6), had low (<20%) tumor cell content (n = 1), or lacked any clinical data (n = 25). A total of 576 patients were analyzed including 463 patients from Asia (Korea (n = 325), Japan (n = 123); collectively herein referred to as Asian patients) and the United States (n = 128). Fifteen of the 128 samples from the United States group were from patients of Asian ethnicity, and therefore included in the Asian patient group. The remaining 113 samples from the United States were from Caucasian patients and are herein referred to as Caucasian patients. The characteristics of the patients are listed in Table 1.

Genomic profiling

All tumors were initially genotyped and divided into two categories based on the findings: those with a known oncogenic alteration and those in which a known oncogenic alteration could not be detected [referred to as panwild-type (WT)]. A known oncogenic alteration was detected in 74.3% (n = 344) of Asian and 69% (n = 78) of Caucasian patients (Table 2 and Fig. 1). All the alterations identified were mutually exclusive with the exception of two cases of concurrent BRAF and KRAS mutations (Table 2). A subset of this cohort has

Table 1. Clinical characteristics of the patients cohort by ethnicity

<table>
<thead>
<tr>
<th>Histology</th>
<th>Caucasian (%)</th>
<th>Asian (%)</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADC</td>
<td>67.2</td>
<td>324</td>
<td>455</td>
</tr>
<tr>
<td>ADC NOS</td>
<td>20 (17.7)</td>
<td>0</td>
<td>20 (3.5)</td>
</tr>
<tr>
<td>AIS</td>
<td>14 (12.4)</td>
<td>3 (0.7)</td>
<td>17 (3.0)</td>
</tr>
<tr>
<td>MIA</td>
<td>2 (1.8)</td>
<td>4 (0.9)</td>
<td>6 (1.0)</td>
</tr>
<tr>
<td>ADC-SCC</td>
<td>1 (0.9)</td>
<td>1 (0.2)</td>
<td>2 (0.3)</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>88 (77.9)</td>
<td>324 (70.0)</td>
<td>412 (71.5)</td>
</tr>
<tr>
<td>Male</td>
<td>25 (22.1)</td>
<td>139 (30.0)</td>
<td>164 (28.5)</td>
</tr>
<tr>
<td>Smoking</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never Smoker</td>
<td>71 (62.8)</td>
<td>374 (80.8)</td>
<td>445 (77.3)</td>
</tr>
<tr>
<td>Light Smoker</td>
<td>39 (34.5)</td>
<td>31 (6.7)</td>
<td>70 (12.2)</td>
</tr>
<tr>
<td>Former/current smoker</td>
<td>3 (2.7)</td>
<td>58 (12.5)</td>
<td>61 (10.6)</td>
</tr>
<tr>
<td>Stage</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Early (I–IIIA)</td>
<td>20 (17.7)</td>
<td>362 (78.2)</td>
<td>382 (66.3)</td>
</tr>
<tr>
<td>Advanced (IIIB–IV)</td>
<td>1 (0.9)</td>
<td>47 (10.1)</td>
<td>48 (8.3)</td>
</tr>
<tr>
<td>N/A</td>
<td>92 (81.4)</td>
<td>54 (11.7)</td>
<td>146 (25.4)</td>
</tr>
</tbody>
</table>

NOTE: Never Smoker, people who smoked <100 cigarettes/ lifetime; Light Smoker, people who smoked 10–100 pack-years; Former/Current Smoker, people who smoked >10 pack-years.

Abbreviations: ADC, adenocarcinoma; ADC-NOS, adenocarcinoma non-otherwise specified; ADC-SCC, adenocarcinoma in situ; AIS, minimally invasive adenocarcinoma.
Clinical Cancer Research

Point mutations in exon 18 or 21 (not L858R), deletions resistance to available tyrosine kinase inhibitors (28).

EGFR mutations in EGFR 86.3% of all the EGFR spread alterations found in and point mutation in exon 21 (L858R) were the wide-

limited smoking histories (26, 27). Deletions in exon 19 consistent with prior studies in patients with NSCLC with Asian patients). These EGFR (detected in 41.6% of patients; most frequent genomic alteration in Caucasian patients (53%) and in 5.0% of patients). Similarly a t i o n A s i a np a t i e n t s w a sa n EGFR mutation (detected in Asian Caucasian

Table 2. Genomic alterations across 576 lung adenocarcinomas grouped by ethnicity

<table>
<thead>
<tr>
<th>Alteration</th>
<th>Caucasian (%)</th>
<th>Asian (%)</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>EGFR</td>
<td>47 (41.6)</td>
<td>245 (52.9)</td>
<td>292 (50.7)</td>
</tr>
<tr>
<td>ALK</td>
<td>7 (6.2)</td>
<td>36 (7.8)</td>
<td>43 (7.5)</td>
</tr>
<tr>
<td>KRASa</td>
<td>15 (13.3)</td>
<td>23 (5.0)</td>
<td>38 (6.8)</td>
</tr>
<tr>
<td>ROS1</td>
<td>3 (2.7)</td>
<td>13 (2.8)</td>
<td>16 (2.8)</td>
</tr>
<tr>
<td>HER2</td>
<td>2 (1.8)</td>
<td>10 (2.2)</td>
<td>12 (2.1)</td>
</tr>
<tr>
<td>RET</td>
<td>1 (0.9)</td>
<td>10 (2.2)</td>
<td>11 (1.9)</td>
</tr>
<tr>
<td>BRAFa</td>
<td>3 (2.7)</td>
<td>6 (1.3)</td>
<td>9 (1.6)</td>
</tr>
<tr>
<td>RIT1</td>
<td>1 (0.9)</td>
<td>1 (0.2)</td>
<td>1 (0.3)</td>
</tr>
<tr>
<td>UNKNOWNb</td>
<td>35 (31.0)</td>
<td>119 (25.7)</td>
<td>154 (26.7)</td>
</tr>
</tbody>
</table>

*aTwo sample showed a concomitant KRAS and BRAF mutation. An Asian sample has a KRAS G12V mutation together with a G469Del in BRAF. A Caucasian sample harbors a G12A mutation in the KRAS gene and a G469E mutation in BRAF. bThe "Unknown" population is the same of the pan-WT reported in the text.

been previously used to identify RET and NTRK1 rearrangements (8, 25). The most common oncogenic alteration in Asian patients was an EGFR mutation (detected in 53% of patients). Similarly EGFR mutations were the most frequent genomic alteration in Caucasian patients (detected in 41.6% of patients; P = 0.036 compared with Asian patients). These EGFR mutation frequencies were consistent with prior studies in patients with NSCLC with limited smoking histories (26, 27). Deletions in exon 19 and point mutation in exon 21 (L858R) were the widespread alterations found in EGFR, accounting for the 86.3% of all the EGFR mutations. Notably, insertion mutations in EGFR exon 20 accounted for 5.3% of all the EGFR mutations and are recognized to confer primary resistance to available tyrosine kinase inhibitors (28).

Point mutations in exon 18 or 21 (not L858R), deletions in exon 18, and insertions in exon 19 together with double point mutations were collectively referred as “Others” in Supplementary Table S2. KRAS mutations were found in approximately 7% of tumors from never/light former smokers and more frequently in Caucasian than Asian patients (Caucasian 13.3% vs. Asian 5.0%; P = 0.003; Table 2). Moreover, KRAS mutational analysis showed variants G12D and G12V to be prevalent among the population in study in agreement with what previous studies of KRAS mutations in never/limited former smokers (29). Twelve insertion mutations were identified in HER2 exon 20 (~2%) and nine BRAF mutations, mainly V600E were detected. Two cases harbored alterations in BRAF codon G469 and had concomitant KRAS mutations. Mutations identified in HER2, KRAS, and BRAF genes are shown in Supplementary Table S3. The incidence of ALK rearrangements was similar in the two patient populations and the most frequent EML4–ALK variants were variants 1 (E20:A13) and 3 (E20:A6). One rearrangement between KIF5B and ALK was identified but none involving TFG–ALK (30) or the newly described KLC1–ALK (31). RET fusions were identified in 11 cases (1.9%) of which 10 were KIF5B–RET and one was a CCDC6–RET rearrangement. RET fusions with different partners have been previously identified in papillary thyroid carcinomas (PTC; refs. 32, 33) and some of them have been reported at lower frequencies in lung cancer, such as those involving CCDC6 (PTC1), NCOA4 (PTC3), and TRIM33 (PTC7; ref. 16, 32). Chromosomal translocations involving ROS1 receptor tyrosine kinase were found in 2.8% of samples involving primarily CD74 and EZR as fusion partners. No fusions with SLC34A2, LRIG3-, and FIG-ROS1 were detected. CD74 was additionally found fused to the high-affinity nerve growth factor receptor (NTRK1) in a patient of Asian ethnicity (15). The specific ALK, ROS1, RET, and NTRK1 rearrange-

Figure 1. Pie charts demonstrating the distribution of oncogenic alterations in lung adenocarcinoma divided by ethnicity.
M90I alteration in lung and myeloid malignancies (34, 35). The second one, D51Y, occurs in the switch I domain of RIT1 and is the second alteration reported in this region of the gene other than one described in subjects with Noonan Syndrome (Supplementary Fig. S1; ref. 36). No oncogenic alteration was detected in 25.7% \( (n = 119) \) of Asian and 31% \( (n = 35) \) of Caucasian tumor specimens.

**NGS of pan-WT tumor specimens**

We next performed targeted NGS on a subset \( (n = 24; 12 \text{ Asian and } 12 \text{ Caucasian patients}) \) of tumors that did not contain any known oncogenic alteration based on our initial analyses (Supplementary Table S5). This subset was chosen because the average tumor content was \( >75\% \). In one tumor from a Caucasian patient, a rearrangement fusing exon 17 of the gene FGFR3 to exon 10 of TACC3 was detected, similar to that previously described in other tumor types (19, 24, 25, 37, 38). A schematic illustration of the identified FGFR3–TACC3 fusion is shown in Fig. 2A.

**Incidence of FGFR3–TACC3 rearrangements in lung adenocarcinoma**

We developed specific PCR primers flanking the breakpoint of FGFR3 (exon 17) and TACC3 (exon 10) and screened the remaining 130 pan-WT tumor specimens using RT-PCR. We detected FGFR3–TACC3 fusion transcripts in two additional tumor specimens from the Asian patient cohort (J2-109 and K2-49). Both samples harbored two different fusion variants (Supplementary Fig. S3A): sample J2-109 showed two variants linking exon 17 of FGFR3 to exon 11 or/and exon 8 of TACC3 and sample K2-49 showed an amplicon connecting exon 17 of FGFR3 and exon 11 of TACC3 and a second one joining together exon 17 of FGFR3 to exon 4 of TACC3 (Supplementary Fig. S3B). We further validate these findings by RNA-Seq and exome sequencing for sample J2-109 (Fig. 2B). The hematoxylin and eosin staining of samples J2-109 and CG-42 is shown in Supplementary Fig. S2. We were not able to further

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**Figure 2.** A, schematic representation of the genomic rearrangement from the tumor sample CG-42 harboring the FGFR3–TACC3 fusion oncogene using the FoundationOne Next-Generation Sequencing Assay. B, FGFR3–TACC3 fusion in sample J2-109 confirmed by RNA sequencing.
analyze sample K2-49 because the tissue sample was exhausted.

**FGFR3–TACC3 is oncogenic in vitro and sensitive to FGFR inhibitors**

The full-length FGFR3–TACC3 transcript from the index patient (CG-42) was cloned using RT-PCR into the pDNR-dual vector and confirmed by direct sequencing to be correct. The expression vector was then introduced into Ba/F3 cells using retroviral infection. The cells were grown in the presence and absence of IL3 and the FGFR3–TACC3 containing Ba/F3 cells were able to proliferate in the absence of IL3 consistent with oncogenic transformation (data not shown). We further evaluated whether pan-FGFR kinase inhibitors, BGJ398 and ponatinib, could represent a potential therapeutic strategy for patients with FGFR3–TACC3 NSCLC. In particular, BGJ398 is a selective FGFR inhibitor while ponatinib is a multitargeted agent with FGFR activity. Both BGJ398 and ponatinib, but not the EGFR kinase inhibitor gefitinib, could represent a potential therapeutic strategy for patients with FGFR3–TACC3 NSCLC. In particular, BGJ398 is a selective FGFR inhibitor while ponatinib is a multitargeted agent with FGFR activity. Both BGJ398 and ponatinib, but not the EGFR kinase inhibitor gefitinib, effectively inhibited the growth of the FGFR3–TACC3 Ba/F3 cells (Fig. 3A). The growth inhibition was mirrored by inhibition of FGFR3 phosphorylation as shown by p-Tyr and pFGFR3 antibodies (Fig. 3B). The decrease in the fusion protein (as measured by the anti-FLAG antibody) is likely due to degradation as only detected in the FGFR inhibitor but not gefitinib-treated cells (Fig. 3B).

**Discussion**

Recent advances in sequencing technologies have led to an increase in the discovery of novel and therapeutically actionable genomic alterations in a broad range of cancers, including those arising in the lung (8, 38, 39). In lung cancer, many of these alterations, including ALK, ROS1, RET, and NTRK1, are chromosomal inversions or translocations that would not have been detected using more conventional methods such as Sanger sequencing. In this study, using NGS, we identify recurrent FGFR3–TACC3 fusions in lung adenocarcinomas from patients who do not harbor any previously identified oncogenic alterations. We used an enrichment strategy and focused our studies on tumors that did not harbor any known oncogenic alterations.

Genomic alterations in FGFR family members have been identified in several cancers including glioblastoma multiforme (GBM), breast cancer, bladder cancer, cholangiocarcinoma, prostate cancer, thyroid cancer, oral cancer, and in lung cancer (19, 24, 25, 37, 38, 40). In lung cancer, FGFR alterations include FGFR1 amplifications in squamous cell lung cancer, FGFR2 and FGFR3 somatic mutations in both lung adenocarcinoma and lung squamous cell carcinoma, and recently identified FGFR3–TACC3 fusion genes in lung squamous cell carcinomas (38). This study adds to this growing list by identifying FGFR3–TACC3 fusions also in lung adenocarcinomas and predominantly from never/limited smokers. Unlike other fusion oncogenes found in lung cancer, the fusion here occurs after the FGFR3 kinase domain. Nevertheless, the function of the partner gene, TACC3, is still likely the same as in other fusion proteins, to force dimerization and consequently activation of FGFR3 kinase activity. In our study, two fusions

![Figure 3. Inhibition of FGFR fusion kinase activity following pan-FGFR kinase inhibitors. A, FGFR3–TACC3 containing Ba/F3 cells were treated at the indicated concentration of BGJ398, ponatinib, and gefitinib and viable cells were measured after 72 hours of treatment and plotted relative to the off-target gefitinib treatment. B, FGFR3–TACC3 containing Ba/F3 cells were treated with indicated concentrations of BGJ398, ponatinib, and gefitinib for 6 hours. Cells extracts were immunoblotted to detect the indicated proteins.](image-url)
were found in never smokers and a third in a smoker of Asian ethnicity (50-pack-year smoker). Interestingly, the fusion found in the smoker subject is the same one most commonly identified in lung squamous cell carcinoma, GBM and bladder cancer (19, 24, 25, 37). The reported fusions occur primarily in two exons of FGFR3 (17 and 18), preserving almost entirely the tyrosine kinase domain that ends after the first three codons of exons 18, but involving many different portions of the TACC3 gene (exons 4, 7, 8, 9, 10, and 11). The same variants have been identified in diverse cancers, suggesting that they are not tumor specific. However, it could be possible to detect additional breakpoints by increasing the number of cases tested. We further demonstrate that pan-FGFR inhibitors may be therapeutically effective in such cancers. Several clinical trials of FGFR inhibitors, including with BGI398 (NCT 01928459, NCT 01975701, NCT01697605, and NCT01004224), are currently under clinical development. Preliminary data of the phase I study of BGI398 in genetic preselected solid tumors showed a tolerable safety profile and tumor regression in patients with urothelial cell carcinomas, squamous cell carcinoma of the lung, cholangiocarcinomas, and breast cancers (41).

Disclosure of Potential Conflicts of Interest
P.A. Janne is a consultant/advisory board member for Abbots, AstraZeneca, Boehringer Ingelheim, Chugai, Clovis Oncology, Genentech, Merrimack, Pfizer, and Sanofi; has ownership interest (including patents) in Azekekeeper Pharmaceuticals, and reports receiving post-marketing royalties from a Dana-Farber Cancer Institute-owned patent on EGFR mutations, which is licensed to Lab Corp. D. Lipson, L. Young, P.J. Stephens, and V.A. Miller have ownership interest in and are employees of Foundation Medicine. D. Jablons is a consultant/advisory board member for Genentech and Eli Lilly. No potential conflicts of interest were disclosed by the other authors.

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Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): M. Capelletti, D. Ercan, P.S. Hammerman, D. Lipson, J. Young, P.J. Stephens, N.I. Lindeman, P.A. Janne
Writing, review, and/or revision of the manuscript: M. Capelletti, P.S. Hammerman, V.A. Miller, N.I. Lindeman, P.A. Janne
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Study supervision: J. Kim
Other (sample collection and acquisition and management of patients): H. Sasaki
Other (provided/processed tissue samples for analyses): K.J. Munir

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Study supervision: J. Kim
Other (sample collection and acquisition and management of patients): H. Sasaki
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