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## Activating Mutations in the Epidermal Growth Factor Receptor Underlying Responsiveness of Non–Small-Cell Lung Cancer to Gefitinib

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### ABSTRACT

#### BACKGROUND

Most patients with non–small-cell lung cancer have no response to the tyrosine kinase inhibitor gefitinib, which targets the epidermal growth factor receptor (EGFR). However, about 10 percent of patients have a rapid and often dramatic clinical response. The molecular mechanisms underlying sensitivity to gefitinib are unknown.

#### METHODS

We searched for mutations in the *EGFR* gene in primary tumors from patients with non–small-cell lung cancer who had a response to gefitinib, those who did not have a response, and those who had not been exposed to gefitinib. The functional consequences of identified mutations were evaluated after the mutant proteins were expressed in cultured cells.

#### RESULTS

Somatic mutations were identified in the tyrosine kinase domain of the *EGFR* gene in eight of nine patients with gefitinib-responsive lung cancer, as compared with none of the seven patients with no response ( $P < 0.001$ ). Mutations were either small, in-frame deletions or amino acid substitutions clustered around the ATP-binding pocket of the tyrosine kinase domain. Similar mutations were detected in tumors from 2 of 25 patients with primary non–small-cell lung cancer who had not been exposed to gefitinib (8 percent). All mutations were heterozygous, and identical mutations were observed in multiple patients, suggesting an additive specific gain of function. In vitro, EGFR mutants demonstrated enhanced tyrosine kinase activity in response to epidermal growth factor and increased sensitivity to inhibition by gefitinib.

#### CONCLUSIONS

A subgroup of patients with non–small-cell lung cancer have specific mutations in the *EGFR* gene, which correlate with clinical responsiveness to the tyrosine kinase inhibitor gefitinib. These mutations lead to increased growth factor signaling and confer susceptibility to the inhibitor. Screening for such mutations in lung cancers may identify patients who will have a response to gefitinib.

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**N**ON-SMALL-CELL LUNG CANCER IS THE leading cause of death from cancer in the United States. Chemotherapy slightly prolongs survival among patients with advanced disease, but at the cost of clinically significant adverse effects.<sup>1</sup> The success of the ABL tyrosine kinase inhibitor imatinib in the treatment of chronic myeloid leukemia (CML) has demonstrated the effectiveness of targeting the critical genetic lesion that promotes proliferative signals in cancer cells.<sup>2</sup> Gefitinib targets the ATP cleft within the tyrosine kinase epidermal growth factor receptor (EGFR),<sup>3</sup> which is overexpressed in 40 to 80 percent of non-small-cell lung cancers and many other epithelial cancers.<sup>4</sup> EGFR signaling is triggered by the binding of growth factors, such as epidermal growth factor (EGF), resulting in the dimerization of EGFR molecules or heterodimerization with other closely related receptors, such as HER2/neu. Autophosphorylation and transphosphorylation of the receptors through their tyrosine kinase domains leads to the recruitment of downstream effectors and the activation of proliferative and cell-survival signals.<sup>5</sup> Despite its ubiquitous expression, inactivation of the *EGFR* gene in the mouse causes minimal defects,<sup>6,7</sup> suggesting that pharmacologic inhibition of EGFR by gefitinib should have few adverse effects.

Gefitinib inhibits the growth of some cancer-derived cell lines and tumor xenografts, although this effect is not well correlated with the level of expression of EGFR or related members of the ErbB family of receptors.<sup>3</sup> In initial clinical studies, gefitinib had minimal adverse effects,<sup>8-10</sup> but tumor responses were observed in only 10 to 19 percent of patients with chemotherapy-refractory advanced non-small-cell lung cancer.<sup>11,12</sup> The addition of gefitinib to traditional chemotherapy provided no benefit.<sup>13,14</sup> Even in gliomas, in which the finding of frequent amplification and rearrangements of the *EGFR* gene suggests that EGFR plays an important role, gefitinib failed to induce clinically significant responses.<sup>15,16</sup> Despite these discouraging results, the remarkably rapid and often profound response to gefitinib in a subgroup of patients with non-small-cell lung cancer led to its approval as single-drug therapy for refractory lung cancer.<sup>17</sup> We evaluated tumors from patients with these dramatic responses to determine the underlying mechanisms.

## METHODS

### NUCLEOTIDE-SEQUENCE ANALYSIS OF TUMOR SPECIMENS

Tumor specimens were obtained during diagnostic or surgical procedures from patients with non-small-cell lung cancer who were subsequently treated with gefitinib according to a protocol approved by the institutional review board of Massachusetts General Hospital in Boston. Frozen tumor specimens, along with matched normal tissue, were available from four patients, and paraffin-embedded material was used from the other patients. In addition, specimens from 25 patients with primary non-small-cell lung cancer who had not been exposed to gefitinib (15 with bronchoalveolar cancer, 7 with adenocarcinoma, and 3 with large-cell lung cancer), with matched normal tissues, were obtained from the Massachusetts General Hospital tumor bank. For mutational analysis of the entire *EGFR* coding sequence, DNA was extracted from specimens, all 28 exons were amplified, and uncloned polymerase-chain-reaction (PCR) fragments were sequenced and analyzed in both sense and antisense directions for the presence of heterozygous mutations. All sequence variants were confirmed by multiple independent PCR amplifications. Primer sequences and amplification conditions are explained in the Supplementary Appendix, available with the full text of this article at [www.nejm.org](http://www.nejm.org). *EGFR* mutations in exons 19 and 21 were also sought in primary tumors of the breast (15 specimens), colon (20 specimens), kidney (16 specimens), pancreas (40 specimens), and brain (4 specimens), along with a panel of 108 cancer-derived cell lines representing diverse histologic types (listed in the Supplementary Appendix).

### FUNCTIONAL ANALYSIS OF MUTANT *EGFR* CONSTRUCTS

The L858R and delL747-P753insS mutations were introduced into the full-length *EGFR* coding sequence with the use of site-directed mutagenesis and inserted into a cytomegalovirus promoter-driven expression construct (pUSE, Upstate). Cos-7 cells were transfected (Lipofectamine 2000, Invitrogen) with 1  $\mu$ g of the expression constructs and then replated 18 hours later at a concentration of  $5 \times 10^4$  cells per well in 12-well plates (Costar) with Dulbec-

co's minimal essential medium without fetal-calf serum. After 16 hours of serum starvation, cells were stimulated with 10 ng of EGF per milliliter (Sigma). To determine whether the mutant receptors were inhibited by gefitinib, the drug was added to the culture medium three hours before the addition of 100 ng of EGF per milliliter. Cells were exposed to EGF for 30 minutes. Cell lysates were prepared in 100  $\mu$ l of Laemmli lysis buffer, followed by the resolution of proteins on 10 percent sodium dodecyl sulfate–polyacrylamide-gel electrophoresis, transfer to membranes, and Western blot analysis with the use of an enhanced chemiluminescence reagent (Amersham). Autophosphorylation of EGFR was measured with antibody against phosphotyrosine at position 1068, and standardized to total protein expression, shown with the use of antibody against EGFR (working concentration, 1:1000; Cell Signaling Technology).

## RESULTS

## CLINICAL CHARACTERISTICS OF PATIENTS WITH A RESPONSE TO GEFITINIB

Patients with advanced, chemotherapy-refractory non-small-cell lung cancer have been treated with gefitinib as a single agent since 2000 at Massachusetts General Hospital. A total of 275 patients were treated, both before its approval on May 2003 by the Food and Drug Administration (FDA), as part of a compassionate-use expanded-access program, and subsequently, with the use of a commercial supply. During this period, 25 patients were identified by physicians as having clinically significant responses to the drug. A clinically significant response was defined as a partial response according to the response evaluation criteria in solid tumors<sup>18</sup> for patients with measurable disease; for patients whose tumor burden could not be quantified with the use

**Table 1.** Characteristics of Nine Patients with Non-Small-Cell Lung Cancer and a Response to Gefitinib.

Patient No.	Sex	Age at Beginning of Gefitinib Therapy yr	Pathological Type*	No. of Prior Regimens	Smoking-Status†	Duration of Therapy mo	Overall Survival‡	EGFR Mutation§	Response¶
1	F	70	BAC	3	Never	15.6	18.8	Yes	Major; improved lung lesions
2	M	66	BAC	0	Never	>14.0	>14.0	Yes	Major; improved bilateral lung lesions
3	M	64	Adeno	2	Never	9.6	12.9	Yes	Partial; improved lung lesions and soft-tissue mass
4	F	81	Adeno	1	Former	>13.3	>21.4	Yes	Minor; improved pleural disease
5	F	45	Adeno	2	Never	>14.7	>14.7	Yes	Partial; improved liver lesions
6	M	32	BAC	3	Never	>7.8	>7.8	Yes	Major; improved lung lesions
7	F	62	Adeno	1	Former	>4.3	>4.3	Yes	Partial; improved liver and lung lesions
8	F	58	Adeno	1	Former	11.7	17.9	Yes	Partial; improved liver lesions
9	F	42	BAC	2	Never	>33.5	>33.5	No	Partial; improved lung nodules

\* Adenocarcinoma (Adeno) with any element of bronchoalveolar carcinoma (BAC) is listed as BAC.

† Smoking status was defined as former if the patient had not smoked any cigarettes within 12 months before entry and never if the patient had smoked less than 100 cigarettes in his or her lifetime.

‡ Overall survival was measured from the beginning of gefitinib treatment to death.

§ EGFR denotes the epidermal growth factor receptor gene.

¶ A partial response was evaluated with the use of response evaluation criteria in solid tumors; major and minor responses were evaluated by two physicians in patients in whom the response could not be measured with the use of these criteria.

of these criteria, the response was assessed by two physicians.

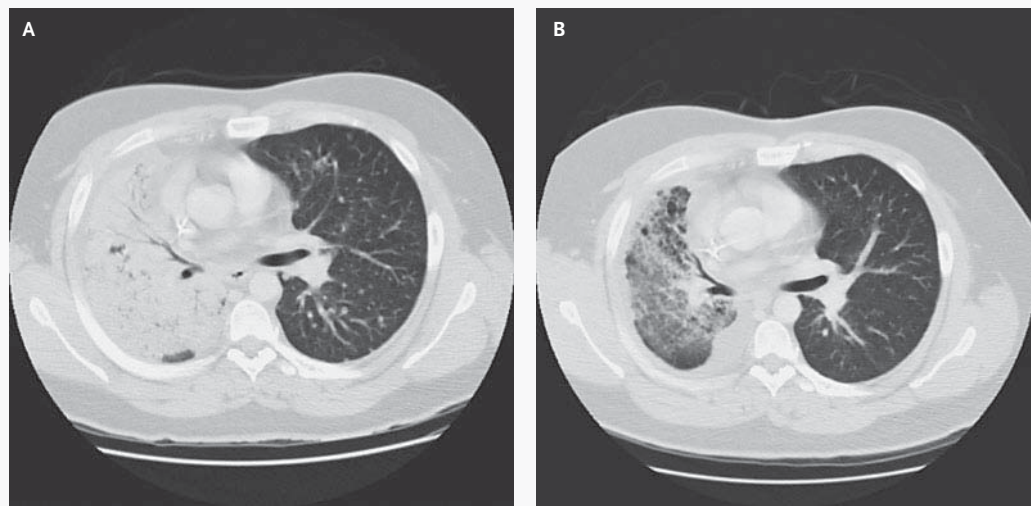
Table 1 shows the clinical characteristics of nine patients for whom tumor specimens obtained at the time of diagnosis were available. Tissue was not available from the other patients with a response to gefitinib, most commonly because diagnostic specimens were limited to needle aspirates. As a group, the nine patients derived a substantial benefit from gefitinib therapy. The median duration of survival from the start of drug treatment exceeded 18 months, and the median duration of therapy was greater than 16 months. Consistent with previous reports, we found that most patients with a response to gefitinib were women, had never smoked, and had bronchoalveolar tumors.<sup>11,12</sup> Patient 6 was representative of the cohort. This patient, a 32-year-old man with no history of smoking, presented with multiple brain lesions and bronchoalveolar carcinoma in the right lung. He was treated with whole-brain radiotherapy, followed by a series of chemotherapy regimens (carboplatin and gemcitabine, docetaxel, and vinorelbine) to which his tumor did not respond. With a declining functional status and progressive lung-tumor burden, he started therapy with 250 mg of gefitinib per day. His dyspnea promptly improved, and computed tomog-

raphy of the lung six weeks after the initiation of treatment revealed a dramatic improvement (Fig. 1).

#### EGFR MUTATIONS IN PATIENTS WITH A RESPONSE TO GEFITINIB

We hypothesized that patients with non-small-cell lung cancer who had striking responses to gefitinib had somatic mutations in the *EGFR* gene that would indicate the essential role of the EGFR signaling pathway in the tumor. To search for such mutations, we first looked for rearrangements within the extracellular domain of EGFR that are characteristic of gliomas<sup>15</sup>; none were detected. We therefore sequenced the entire coding region of the gene using PCR amplification of individual exons.

Heterozygous mutations were observed in eight of nine patients, all of which were clustered within the tyrosine kinase domain of EGFR (Table 2 and Fig. 2). Four tumors had in-frame deletions, removing amino acids 746 through 750 (delE746–A750) in Patient 1, 747 through 751 (delL747–T751insS) in Patient 2, and 747 through 753 (delL747–P753insS) in Patients 3 and 4. The second and third deletions were associated with the insertion of a serine residue, resulting from the generation of a novel codon at the deletion breakpoint. Remarkably, all these deletions overlapped, sharing the deletion of four ami-



**Figure 1.** Example of the Response to Gefitinib in a Patient with Refractory Non-Small-Cell Lung Cancer.

A computed tomographic scan of the chest in Patient 6 shows a large mass in the right lung before treatment with gefitinib was begun (Panel A) and marked improvement six weeks after gefitinib was initiated (Panel B).

no acids (leucine, arginine, glutamic acid, and alanine at codons 747 through 750) within exon 19.

Another three tumors had amino acid substitutions within exon 21: leucine to arginine at codon 858 (L858R) in Patients 5 and 6 and leucine to glutamine at codon 861 (L861Q) in Patient 7. The L861Q mutation is of particular interest, since the same amino acid change in the mouse *egfr* gene is responsible for the Dark Skin (*dsk5*) trait, associated with altered EGFR signaling.<sup>19</sup> A fourth missense mutation in the tyrosine kinase domain resulted in the substitution of cysteine for glycine at codon 719 within exon 18 (G719C) in Patient 8.

Matched normal tissue was available for Patients 1, 4, 5, and 6 and showed only the wild-type sequence, indicating that the mutations had arisen somatically during tumor formation. By compari-

son, no mutations were observed in seven patients with non-small-cell lung cancer who had had no response to gefitinib ( $P < 0.001$  by a two-sided Fisher's exact test).

#### PREVALENCE OF SPECIFIC EGFR MUTATIONS IN NON-SMALL-CELL LUNG CANCER AND OTHER TYPES OF CANCER

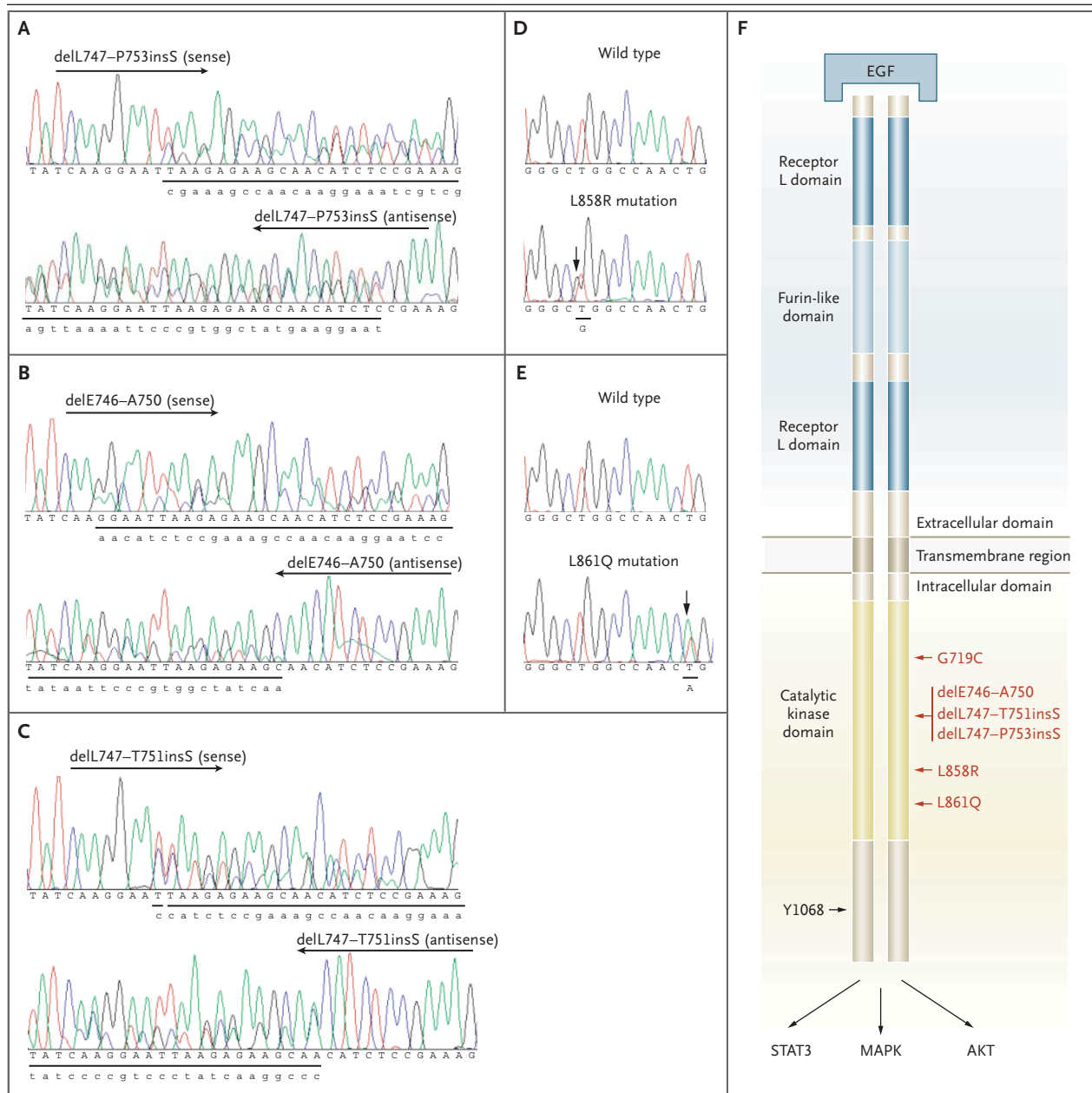
Unlike gliomas, in which rearrangements affecting the EGFR extracellular domain have been extensively studied,<sup>15</sup> the frequency of EGFR mutations in non-small-cell lung cancer has not been defined. We therefore sequenced the entire coding region of the gene in tumors from 25 patients with primary non-small-cell lung cancer who were not involved in the gefitinib study, including 15 with bronchoalveolar lung cancer, which has been associated with

**Table 2. Somatic Mutations in the Tyrosine Kinase Domain of EGFR in Patients with Non-Small-Cell Lung Cancer.**

Patient	Mutation	Effect of Mutation
<b>Patients with a response to gefitinib</b>		
Patient 1	Deletion of 15 nucleotides (2235–2249)	In-frame deletion (746–750)
Patient 2	Deletion of 12 nucleotides (2240–2251)	In-frame deletion (747–751) and insertion of a serine residue
Patient 3	Deletion of 18 nucleotides (2240–2257)	In-frame deletion (747–753) and insertion of a serine residue
Patient 4	Deletion of 18 nucleotides (2240–2257)	In-frame deletion (747–753) and insertion of a serine residue
Patient 5	Substitution of G for T at nucleotide 2573	Amino acid substitution (L858R)
Patient 6	Substitution of G for T at nucleotide 2573	Amino acid substitution (L858R)
Patient 7	Substitution of A for T at nucleotide 2582	Amino acid substitution (L861Q)
Patient 8	Substitution of T for G at nucleotide 2155	Amino acid substitution (G719C)
<b>Patients with no exposure to gefitinib*</b>		
Patient A	Deletion of 18 nucleotides (2240–2257)	In-frame deletion (747–753) and insertion of a serine residue
Patient B	Deletion of 15 nucleotides (2235–2249)	In-frame deletion (746–750)

\* Among the 25 patients with no exposure to gefitinib (15 with bronchoalveolar cancer, 7 with adenocarcinoma, and 3 with large-cell carcinoma), 2 (Patients A and B) — both of whom had bronchoalveolar cancer — had EGFR mutations. No mutations were found in 14 lung-cancer cell lines representing diverse histologic types: non-small-cell lung cancer (6 specimens), small-cell lung cancer (6 specimens), bronchus carcinoid (1 specimen), and an unknown type (1 specimen). Polymorphic variants identified within EGFR included the following: the substitution of A for G at nucleotide 1562, the substitution of A for T at nucleotide 1887, and a germ-line variant of unknown functional significance, the substitution of A for G at nucleotide 2885, within the tyrosine kinase domain.





**Figure 2.** Mutations in the *EGFR* Gene in Gefitinib-Responsive Tumors.

Panels A, B, and C show the nucleotide sequence of the *EGFR* gene in tumor specimens with heterozygous in-frame deletions within the tyrosine kinase domain (double peaks). Tracings in both sense and antisense directions are shown to demonstrate the two breakpoints of the deletion; the wild-type nucleotide sequence is shown in capital letters, and the mutant sequence is in lowercase letters. The 5' breakpoint of the delL747–T751insS mutation is preceded by a T-to-C substitution that does not alter the encoded amino acid. Panels D and E show heterozygous missense mutations (arrows) resulting in amino acid substitutions within the tyrosine kinase domain. The double peaks represent two nucleotides at the site of heterozygous mutations. For comparison, the corresponding wild-type sequence is also shown. Panel F shows dimerized EGFR molecules bound by the EGF ligand. The extracellular domain (containing two receptor ligand [L] domains and a furin-like domain), the transmembrane region, and the cytoplasmic domain (containing the catalytic kinase domain) are highlighted. The position of tyrosine<sup>1068</sup> (Y1068), a site of autophosphorylation used as a marker of receptor activation, is indicated, along with downstream effectors activated by EGFR autophosphorylation — STAT3, MAP kinase (MAPK), and AKT. The locations of tumor-associated mutations, all within the tyrosine kinase domain, are shown in red.

responsiveness to gefitinib in previous clinical trials.<sup>11,12</sup> Heterozygous mutations were detected in two patients with bronchoalveolar cancers. Both had in-frame deletions in the kinase domain that were identical to those found in the patients with a response to gefitinib — namely, delL747–P753insS and delE746–A750 (Table 2). Given the apparent clustering of *EGFR* mutations, we sequenced exons 19 and 21 in a total of 95 primary tumors and 108 cancer-derived cell lines, representing diverse tumor types (see the Supplementary Appendix). No mutations were detected, suggesting that only a subgroup of cancers, in which EGFR signaling may play a critical role in tumorigenesis, harbor *EGFR* mutations.

#### INCREASE IN EGF-INDUCED ACTIVATION AND GEFITINIB-INDUCED INHIBITION OF MUTANT EGFR PROTEINS

To study the functional properties encoded by these mutations, we expressed the receptor with the L747–P753insS deletion and the receptor with the L858R missense mutation in cultured cells. Transient transfection of wild-type and mutant constructs into Cos-7 cells demonstrated equivalent expression levels, indicating that the mutations do not affect the stability of the protein. EGFR activation was quantified by measuring phosphorylation of the tyrosine<sup>1068</sup> residue, commonly used as a marker of the autophosphorylation of EGFR.<sup>20</sup> In the absence of serum and associated growth factors, neither wild-type nor mutant EGFR demonstrated autophosphorylation (Fig. 3A and 3B). However, the addition of EGF doubled or tripled the activation of both mutant EGFRs, as compared with the activation of the wild-type receptor. Moreover, whereas the activation of normal EGFR was down-regulated after 15 minutes, consistent with the internalization of the receptor, the two mutant receptors demonstrated continued activation for up to three hours (Fig. 3A). Similar results were obtained with the use of antibodies to measure the total phosphorylation of EGFR after the addition of EGF (data not shown).

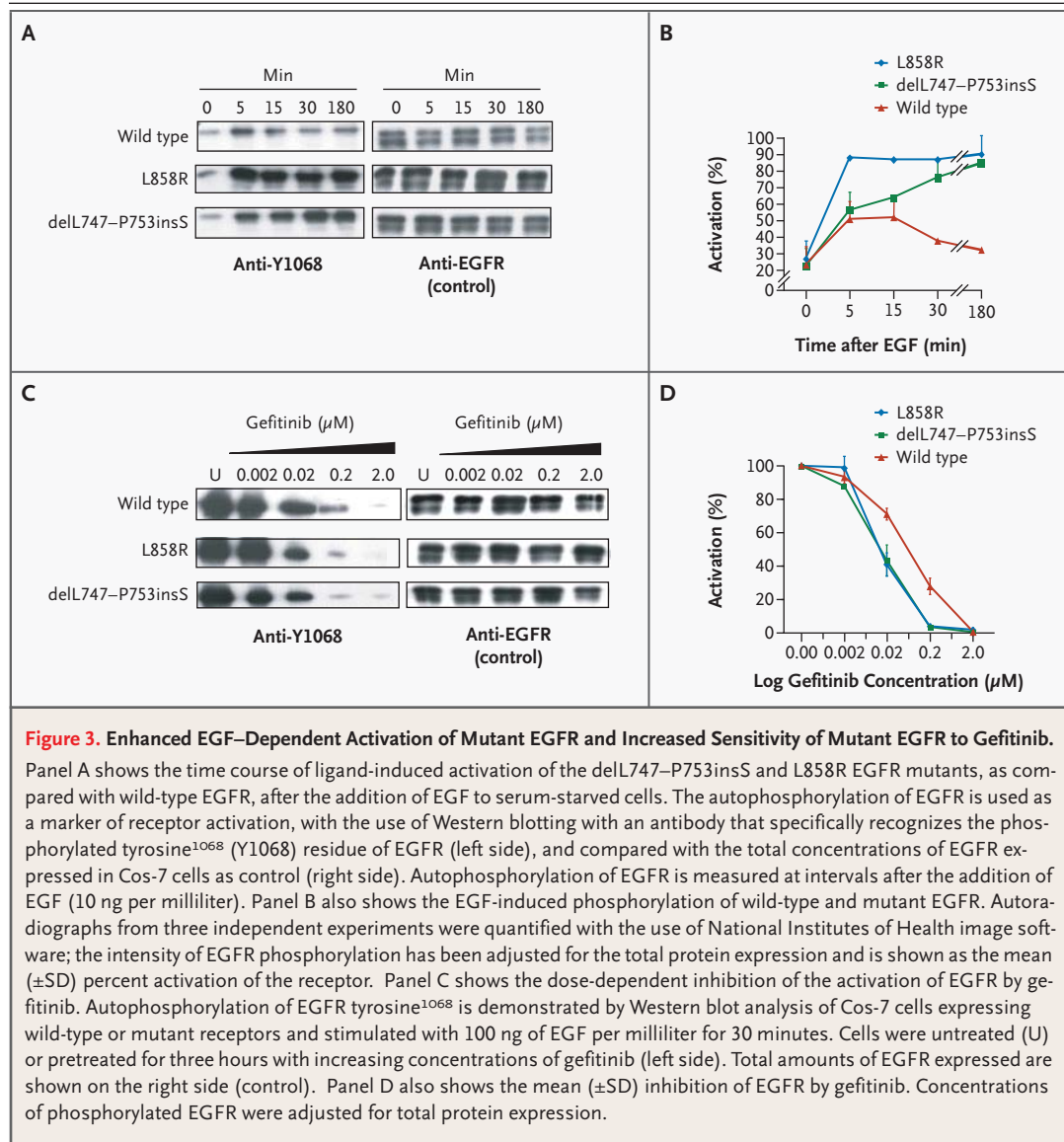
Since seven of the eight EGFR tyrosine kinase mutations reside near the ATP cleft, which is targeted by gefitinib, we assessed whether the mutant proteins have altered sensitivity to the inhibitor. EGF-induced autophosphorylation of EGFR was measured in cells pretreated with various concentrations of gefitinib. Remarkably, both mutant receptors were more sensitive than the wild-type re-

ceptor to inhibition by gefitinib. Wild-type EGFR was inhibited by 50 percent at a gefitinib concentration of 0.1  $\mu$ M and was completely inhibited by a concentration of 2.0  $\mu$ M, whereas the respective values for the two mutant proteins were 0.015  $\mu$ M and 0.2  $\mu$ M (Fig. 3C and 3D). This difference in drug sensitivity may be clinically relevant, since pharmacokinetic studies indicate that daily oral administration of 400 to 600 mg of gefitinib results in a mean steady-state trough plasma concentration of 1.1 to 1.4  $\mu$ M, whereas the currently recommended daily dose of 250 mg leads to a mean trough concentration of 0.4  $\mu$ M.<sup>21</sup>

#### DISCUSSION

Gefitinib is the first agent designed with a known molecular target to receive FDA approval for the treatment of lung cancer, yet its activity is limited to a subgroup of patients with non-small-cell lung cancer. We have identified specific activating mutations within the tyrosine kinase domain of *EGFR* as the molecular correlate of the dramatic responses to gefitinib in this subgroup. These somatic mutations were identified in eight of nine patients with a response to gefitinib; the ninth patient may have had an undetected mutation or a mutation in a heterodimerization partner of EGFR. These results, together with the finding of *EGFR* mutations in tumors from 2 of 25 patients with non-small-cell lung cancer who had not received gefitinib (8 percent), suggest that such mutations account for the majority of responses to gefitinib reported in clinical studies.<sup>11,12</sup>

The heterozygous nature of *EGFR* mutations suggests that they exert a dominant oncogenic effect, which is evident despite the presence of the second wild-type allele. The presence of an additive specific gain of function is further supported by the observation of identical somatic mutations in different tumors. These mutations are clustered near the ATP cleft of the tyrosine kinase domain, where they flank amino acids shown in crystallographic studies to mediate binding of 4-anilinoquinazoline compounds, such as gefitinib<sup>22</sup> (Fig. 4). We postulate that the mutations result in repositioning of these critical residues, stabilizing their interaction with both ATP and its competitive inhibitor gefitinib. Such a mechanism would explain both the increased receptor activation after ligand binding and the enhanced inhibition induced by gefitinib. Structural analysis of the mutant receptors will therefore

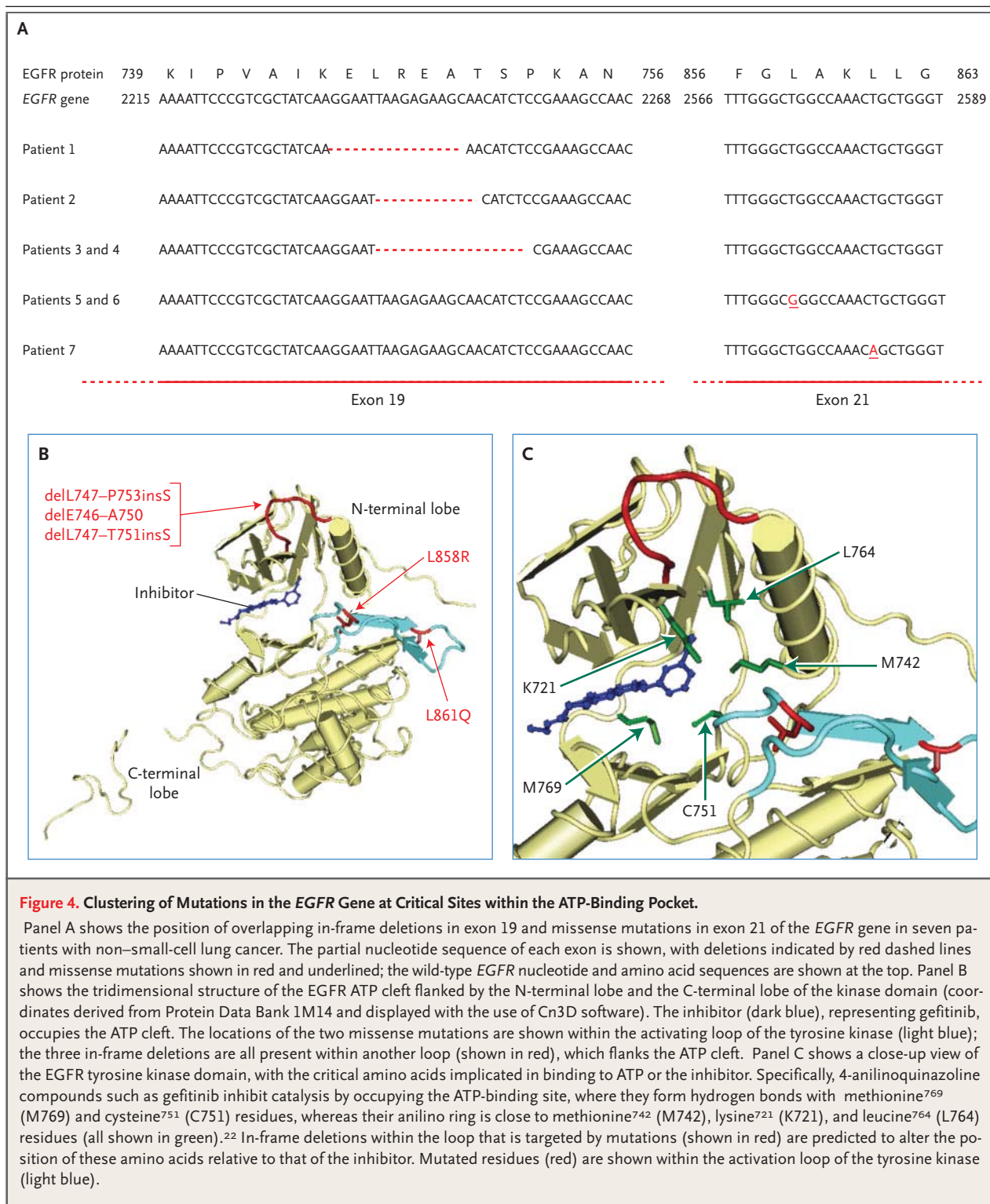


provide important insight into the mechanisms that regulate the activation of EGFR and the design of more potent inhibitors targeting the mutant receptors.

Our observations have implications for the identification of molecular targets for cancer therapy using small-molecule kinase inhibitors. The effectiveness of imatinib in CML is based on its ability to target the ABL tyrosine kinase, which is activated by the BCR-ABL translocation (i.e., the Philadelphia chromosome) in all patients with this disease and can transform hematopoietic cells.<sup>2,23</sup> Similar evidence designating a protein as an optimal thera-

peutic target is not available for most epithelial cancers. Our data suggest that EGFR tyrosine kinase mutations can be used to identify the subgroup of patients with non-small-cell lung cancer in whom this growth factor receptor may be essential to tumor growth, whereas the overexpression of EGFR in the absence of mutations may reflect the less critical role played by this factor in the majority of cases. This emphasis on genetic alterations is consistent with the observation that the amplification of the *HER2/neu* gene is a more reliable predictor than protein overexpression of the responsiveness of breast cancer to the targeting antibody trastu-





zumab and that *C-KIT* mutations can be used to determine the response of gastrointestinal stromal tumors to imatinib.<sup>24,25</sup> Ongoing, large-scale sequencing efforts may reveal additional mutations in other kinases, linking different cancers to potential therapeutic targets.<sup>26,27</sup>

Gefitinib has not elicited clinical responses in patients with gliomas, despite the high frequency of amplification and rearrangements of the *EGFR* gene in such patients.<sup>15,16</sup> However, the *EGFR* tyrosine kinase mutations in patients with non-small-cell lung cancer are fundamentally different from the glioma-associated deletions within the extracellular domain of *EGFR*. These truncated *EGFR* proteins resemble the avian erythroblastosis viral oncogene *v-erbB* in mediating constitutive, ligand-independent activation of the receptor, but they do not alter the ATP cleft of the tyrosine kinase that is bound by gefitinib. The enhanced sensitivity to gefitinib associated with tyrosine kinase mutations may therefore contribute substantially to the clinical responses of certain patients with non-small-cell lung cancer.

The plasma concentrations of gefitinib that can be achieved with the use of current dosage recommendations<sup>21</sup> exceed the drug concentration that suppressed autophosphorylation of the mutant *EGFR* tyrosine kinase in our assays but are below those required to suppress the wild-type receptor. In vitro analysis of wild-type *EGFR* has also suggested that low concentrations of gefitinib may be sufficient to suppress autophosphorylation at some tyrosine residues, but that abrogation of downstream signaling requires a higher dose.<sup>28</sup> Thus, in patients with gliomas, in whom biologic dependence on *EGFR* signaling is identified by the presence of gene amplification or deletions within the extracellular domain, a clinical response may

require plasma concentrations of an *EGFR* tyrosine kinase inhibitor that are sufficient to abrogate downstream signaling.

Understanding the molecular basis of responsiveness to gefitinib has immediate clinical implications with respect to patients with non-small-cell lung cancer. The clustering of mutations within specific regions of the *EGFR* tyrosine kinase domain makes possible the potential development of rapid and reliable diagnostic testing to guide the clinical use of gefitinib. For patients whose tumors have activating mutations of *EGFR*, the dramatic responses to gefitinib of patients whose disease has been refractory to all other therapies suggest that this agent may be more effective if used earlier in the course of treatment. Prospective validation of *EGFR* tyrosine kinase mutations as predictors of the responsiveness to gefitinib is warranted, and genotype-directed clinical trials of this tyrosine kinase inhibitor in the initial treatment of advanced non-small-cell lung cancer—and even in the adjuvant setting after surgical resection—should now be considered.

Similar results are being reported by other investigators.<sup>29</sup>

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