original report

Epidermal Growth Factor Receptor Mutational Status and Brain Metastases in Non–Small-Cell Lung Cancer

abstract

Introduction Epidermal growth factor receptor (*EGFR*) mutations in non–small-cell lung cancers (NSCLC) may be more common in patients with brain metastases. Previous studies, however, did not adjust for effects of confounding variables.

Methods This retrospective study included 1,522 consecutive patients with NSCLC, whose tumors were diagnosed and tested for *EGFR* mutations at the University of Nebraska Medical Center (Omaha, NE) and Tata Memorial Hospital (Mumbai, India). Multivariate logistic regression was used to identify any association between *EGFR* status and clinical factors.

Results EGFR mutations were more common in females than males (38.7% *v* 24.8%), Asians than whites (31.3% *v* 13.4%), nonsmokers than smokers (40.2% *v* 14.6%), alcohol nonconsumers than users (32.4% *v* 15.8%), adenocarcinoma than other histology types (32.7% *v* 10.3%), and patients with brain metastases than extracranial or no metastases (39.4% *v* 29.8% *v* 15.1%; $P < .001$ for all comparisons). There was a higher likelihood of an *EGFR* mutation among patients with brain metastases (odds ratio, 1.8; $P < .001$). The median overall survival (OS) was 19.8 months. Patients with brain metastases had a shorter median OS (15 *v* 20.6 months; $P = .02$). However, in the cohort of *EGFR* mutation–positive patients, there was no difference in median OS between patients with and without brain metastases (20.8 *v* 25.1 months; $P = .11$).

Conclusion There is a nearly two-fold higher incidence of *EGFR* mutations in NSCLC among patients with brain metastases at diagnosis. *EGFR* mutations did not predict for outcomes from brain metastases.

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INTRODUCTION

Approximately 40% of patients with lung cancer develop brain metastases; lung cancer accounts for up to half of all secondary brain tumors.[1] Conventional chemotherapy regimens are associated with significant toxicities and modest response rates in metastatic non–small-cell lung cancer (NSCLC), particularly among patients with brain metastases.[1,2] Consequently, some patients forego therapy because of anticipated intolerance.[3] In particular, older patients or those unfit for treatment may discontinue therapy as a result of toxicities. In recent years, epidermal growth factor receptor (EGFR) tyrosine kinase inhibitors (TKIs) have emerged as powerful therapeutic agents with significant improvements in response rates and progression-free survival in *EGFR*-mutant NSCLC.[2] Identifying patients with a higher probability of having tumors that harbor *EGFR* mutations has therefore become important. Although *EGFR* testing has been advocated in all advanced-stage adenocarcinoma regardless of clinical features,[4] this approach may not be cost effective, especially in low-resource settings. The ability to identify additional subgroups of patients, whose tumors are more likely to harbor an *EGFR* mutation and benefit from EGFR TKI therapy compared with conventional chemotherapy, may improve the cost effectiveness of *EGFR* testing.

The most common *EGFR* mutations in lung cancer include in-frame deletion within exon 19 (45%) and point mutation within exon 21 (p.L858R; 40%); exon 18 point mutations and exon 20 insertions or point mutations account for most of the remainder of mutations. The presence of activating *EGFR* mutations (exon 19 deletion, p.L858R mutation in exon 21, and codon 719 mutations in exon 18) predicts a response to EGFR TKIs (erlotinib, afatinib, and gefitinib).[5,6]

The incidence of *EGFR*-mutated lung cancer is approximately 16% among whites,[7] but there seems to be differences on the basis of histology type, sex, and smoking status. *EGFR* mutations were more frequent in adenocarcinoma compared with other NSCLC (21% *v* 2%), women compared with men (20% *v* 9%), and Asians compared with North Americans (26% *v* 2%).[8] A few studies assessing the role of targeted therapy as a single agent or in combination with radiation among patients with NSCLC with brain metastases have shown that the prevalence of *EGFR* mutation is higher among patients with NSCLC with brain metastases.[9] Given the poor survival of patients with brain metastases, such findings have therapeutic implications and may also help elucidate a possible role of *EGFR* mutation in the development of brain metastases. However, these studies were largely descriptive. A study with a sufficient sample size to adjust for sex, smoking status, ethnicity, and histology[10] could provide a closer estimate of the incidence of *EGFR* mutations in patients with NSCLC presenting with different stages, including those with and without brain metastases.

METHODS

A retrospective study of consecutive patients with NSCLC, who were examined for the presence of *EGFR* mutations, diagnosed between 2007 and 2014 at the University of Nebraska Medical Center (UNMC; Omaha, NE) and Tata Memorial Hospital (TMH; Mumbai, India) was conducted. A total of 1,609 patients were identified; however, 87 patients were excluded as a result of missing data.

Patient Selection

The diagnoses were based on microscopic examination of the biopsy of lung mass or metastases, pleural fluid cytology, or bronchoscopic brushing. Information obtained at diagnosis included age, sex, ethnicity, smoking status, history of alcohol consumption, and histology type. Ethnicity was divided into Asian, white, and others because the other categories included only a small number of patients. On the basis of patients' self-reports, smoking status was categorized as smoker versus nonsmoker, and alcohol consumption was divided into consumer versus nonconsumer. The categorization of patients into smoker and alcohol consumer required current or prior use of tobacco and alcohol consumption, regardless of the quantity.

Staging evaluation included computed tomography, bone scan and/or positron emission tomography, as well as imaging of brain (magnetic resonance imaging or contrasted computed tomography). The American Joint Committee on Cancer, 7th edition of tumor-node-metastasis classification system was used for tumor staging. Patients were divided into those without metastasis and with extracranial and/or brain metastases. Overall survival was defined as the time between diagnosis and date of death or last contact. Patients who were alive at last contact were censored for survival analysis. The study was conducted in accordance with institutional guidelines at both participating institutions.

EGFR Mutation Analysis at UNMC

Genomic DNA was extracted from formalin-fixed paraffin-embedded tissue containing at least 20% tumor tissue. When necessary, macrodissection was performed to obtain the threshold level of tumor. Sections were deparaffinized and then digested from a minimum of 4 hours to overnight. DNA was extracted and simultaneous real-time polymerase chain reaction amplification and mutation detection was performed using Amplification Refractory Mutation System technology and Scorpions dual primer-probes available in the *EGFR* RGQ PCR Kit (QIAGEN, Valencia, CA). Seven separate mutation amplification reactions and one wild-type control reaction were performed, allowing for the detection of 29 somatic mutations in exons 18 to 21. The limit of detection of this assay is 2.5% to 10%, depending on the specific mutation (point mutation or deletion/insertion).

EGFR Mutation Analysis at TMH

Formalin-fixed paraffin-embedded tissue sections containing at least 75% tumor were deparaffinized in xylene and the DNA was extracted (QIAamp FFPE Tissue Kit, QIAGEN). Mutation analysis was performed using a clinically validated real-time TaqMan assay described previously.[11] Exon 20 point mutations were performed additionally on specimens from August 2012 onward. The analytical sensitivity of this assay has a limit of 1% detection, and each run consisted of a positive and a negative control to check the integrity of the assay.

Statistical Analysis

χ^2 tests or Fisher's exact test were used to determine univariate associations with patient location (TMH or UNMC) and patient characteristics, and *EGFR* mutation status and patient characteristics. Age distributions were compared between groups with *t* tests. Multivariate logistic regression was used to determine associations between

Table 1. Patient Characteristics on Basis of Country of Origin

Variable	Total (N = 1,522)	TMH (n = 1,385)	UNMC (n = 137)	*P*
Age in years, median (range)	57 (19-89)	57 (19-83)	64 (42-89)	< .001
Sex				
Female	532 (35%)	466 (33.7%)	66 (48.2%)	< .001
Male	987 (65%)	916 (66.3%)	71 (51.8%)	
Ethnicity				
Asian	1,388 (91%)	1,385 (100%)	3 (2.2%)	< .001
Other	6 (0.4%)	0 (0%)	6 (4.4%)	
White	128 (8%)	0 (0%)	128 (93.4%)	
Smoking status				
No	894 (59%)	864 (62.4%)	30 (22.4%)	< .001
Yes	624 (41%)	520 (37.6%)	104 (77.6%)	
Alcohol intake				
No	1,272 (84%)	1,178 (85.1%)	94 (70.1%)	< .001
Yes	246 (16%)	206 (14.9%)	40 (29.9%)	
Stage				
I	20 (1%)	0 (0%)	20 (14.6%)	< .001
II	10 (1%)	3 (0.2%)	7 (5.1%)	
III	124 (8%)	106 (7.7%)	18 (13.1%)	
IV	1,361 (90%)	1,269 (92.1%)	92 (67.2%)	
Adenocarcinoma				
No	204 (13%)	184 (13.3%)	20 (14.6%)	.67
Yes	1,318 (87%)	1,201 (86.7%)	117 (85.4%)	
EGFR mutation				
No	1,070 (70%)	951 (68.7%)	119 (86.9%)	< .001
Yes	452 (30%)	434 (31.3%)	18 (13.1%)	
Metastatic disease				
No metastasis	165 (11%)	115 (8.3%)	50 (36.5%)	< .001
Metastasis other than brain	1,121 (74%)	1,074 (77.6%)	47 (34.3%)	
Brain metastasis	236 (16%)	196 (14.1%)	40 (29.2%)	
Metastatic disease				
No brain metastasis	1,286 (84%)	1,189 (85.9%)	97 (70.8%)	< .001
Brain metastasis	236 (16%)	196 (14.1%)	40 (29.2%)	

Abbreviations: *EGFR*, epidermal growth factor receptor; TMH, Tata Memorial Hospital; UNMC, University of Nebraska Medical Center.

EGFR status and demographic/clinical factors including sex, ethnicity, smoking status, alcohol use, stage, adenocarcinoma histology, metastatic location, and overall survival (OS). *P* values < .05 were considered to be statistically significant. SAS Software version 9.3 (SAS Institute Inc., Cary, NC) was used for statistical analysis.

RESULTS

This analysis included 1,522 patients. Patients' characteristics and differences on the basis of country of diagnosis and *EGFR* mutational status are depicted in Tables 1 and 2. Most patients (87%) had adenocarcinoma histology, which included pure adenocarcinoma as well as predominant adenocarcinoma with areas of neuroendocrine differentiation and adenosquamous histology. Patients diagnosed at UNMC rather than TMH were more likely to be older (median age 64 *v* 57 years), female (48.2% *v* 33.7%), white (93.4% *v* 0%), smokers (77.6% *v* 37.6%), consumers of alcohol (29.9% *v* 14.9%), and nonmetastatic (36% *v* 8%). Conversely, patients diagnosed at UNMC were less likely than those

Table 2. Patient Characteristics on Basis of Epidermal Growth Factor Receptor Mutational Status

Variable	*EGFR* Mutation		*P*
	Negative (n = 1,070)	Positive (n = 452)	
Location			
TMH	951 (68.7%)	434 (31.3%)	< .001
UNMC	119 (86.9%)	18 (13.1%)	
Age in years, median (range)	57 (19-88)	57 (26-89)	.12
Sex			
Female	326 (61.3%)	206 (38.7%)	< .001
Male	742 (75.2%)	245 (24.8%)	
Ethnicity			
White/other	116 (86.6%)	18 (13.4%)	< .001
Asian	954 (68.7%)	434 (31.3%)	
Smoking status			
No	535 (59.8%)	359 (40.2%)	< .001
Yes	533 (85.4%)	91 (14.6%)	
Alcohol intake			
No	860 (67.6%)	412 (32.4%)	< .001
Yes	207 (84.2%)	39 (15.8%)	
Stage			
I/II	25 (83.3%)	5 (16.7%)	< .001
III	109 (87.9%)	15 (12.1%)	
IV	932 (68.5%)	429 (31.5%)	
Adenocarcinoma			
No	183 (89.7%)	21 (10.3%)	< .001
Yes	887 (67.3%)	431 (32.7%)	
Metastatic disease			
No metastasis	140 (84.9%)	25 (15.1%)	< .001
Metastasis other than brain	787 (70.2%)	334 (29.8%)	
Brain metastasis	143 (60.6%)	93 (39.4%)	
Metastatic disease			
No brain metastasis	927 (72.1%)	359 (27.9%)	< .001
Brain metastasis	143 (60.6%)	93 (39.4%)	

NOTE. Totals may not add up as a result of missing values.

Abbreviations: *EGFR*, epidermal growth factor receptor; TMH, Tata Memorial Hospital; UNMC, University of Nebraska Medical Center.

at TMH to have an EGFR mutation (13.1% *v* 31.3%). For the entire cohort, tumor *EGFR* mutations were more commonly seen in females than males (38.7% *v* 24.8%), Asians than whites (31.3% *v* 13.4%), nonsmokers than smokers (40.2% *v* 14.6%), alcohol nonconsumers than consumers (32.4% *v* 15.8%), adenocarcinoma than other histology types (32.7% *v* 10.3%), and patients with brain metastases than extracranial metastases or no metastasis (39.4% *v* 29.8% *v* 15.1%). Eight patients with *EGFR*-mutant tumors from the UNMC cohort received erlotinib as part of their treatment regimen. In the TMH cohort, 42 patients did not receive a TKI during treatment. The type of mutation (exon 19 *v* 21) did not correlate with the presence of metastases (Appendix Table A1); the frequency of other mutations was too small to conduct an analysis.

In a multivariate analysis of all patients (presented in Table 3), ethnicity, smoking status, alcohol intake, adenocarcinoma, and metastatic disease were all significant predictors of having a tumor with an *EGFR* mutation. Pairwise comparisons revealed that patients with brain metastases were 1.9 times more likely to have *EGFR* mutations than patients with extracranial metastases ($P < .001$) or those with none and extracranial metastases (odds ratio [OR], 1.8; $P < .001$). The outcomes were largely similar when the data from the institutions were analyzed separately (Appendix Tables A2 and A3). For patients at UNMC, the incidence of *EGFR* mutation was significantly higher for nonsmokers than smokers.

The median OS of the entire cohort was 19.8 months (95% CI, 17.9 to 21.7 months). The OS was similar at the two sites (TMH, 19.7 months; UNMC, 21.6 months; $P = .2$). Patients with an *EGFR* mutation had a longer median OS compared with those without (25.1 *v* 16.8 months; $P < .001$; Appendix Table A4). Patients with tumors harboring an exon 19 deletion had a numerically better median OS compared with those with tumors containing an L858R mutation (27.1 *v* 21.1 months), but this difference was not statistically significant (odds ratio, 0.73; 95% CI, 0.53 to 1.01; $P = .056$; Table 4). Patients with brain metastases had a shorter median OS compared with those without brain metastases (15 months; 95% CI, 11.7 to 18.4 *v* 20.6 months; 95% CI, 18.3 to 22.8 months; $P = .02$). However, in the cohort of *EGFR* mutation–positive patients, there was no difference in median OS between patients with and without brain metastases (20.8 *v* 25.1 months; $P = .11$).

DISCUSSION

To our knowledge, the current study is the largest to date to demonstrate that *EGFR* mutations are significantly more frequent among tumors of patients with NSCLC with brain metastases at diagnosis. In multivariate analysis, the presence of brain metastases independently predicted a twofold increased likelihood of an *EGFR* mutation compared with tumors in patients without brain metastases. Several published studies have indicated the possibility of a higher rate of *EGFR* mutations in NSCLC metastatic to the brain.[10]

Table 3. Multivariate Analysis of Positive Epidermal Growth Factor Receptor Mutational Status for the Entire Cohort of Patients

Effect	Variable	OR (95% CI)	*P*
Sex	Female *v* male	1.13 (0.87 to 1.48)	NS
Ethnicity	Asian *v* white/other	2.14 (1.16 to 3.95)	.02
Smoking	No *v* yes	2.75 (2.04 to 3.72)	< .001
Alcohol intake	No *v* yes	1.61 (1.07 to 2.41)	.02
Stage	IV *v* I/II	1.76 (0.33 to 9.35)	NS
	III *v* I/II	0.51 (0.14 to 1.90)	
Adenocarcinoma	Yes *v* no	3.07 (1.87 to 5.03)	< .001
Metastatic disease	Brain metastasis *v* extracranial metastasis	1.85 (1.34 to 2.54)	< .001
	No metastasis *v* extracranial metastasis	1.64 (0.46 to 5.83)	NS

Abbreviations: NS, not significant; OR, odds ratio.

These studies with small sample sizes did not adjust for effects of potential confounding variables. A more recent Korean study demonstrated similar findings, with a strong association between *EGFR* mutation status and brain metastases (OR, 3.8; $P = .001$), but not between *EGFR* mutation status and extracranial metastases (OR, 1.73; $P = .079$). The authors also reported a higher number of brain metastases in patients with *EGFR* mutations and a higher risk of brain metastases after resection of lung primary tumors.[12] In another study, from Slovenia, patients with *EGFR* mutation–positive disease had a nonsignificant trend toward higher risk of brain metastases at diagnosis.[13]

No differences in the distribution of type of *EGFR* mutation (exon 19 *v* 21) on the basis of the presence or absence of brain metastases were identified in the current study. A Japanese study (N = 57) suggested a correlation between miliary brain metastases and exon 19 deletion, but not exon 21 point mutation.[14]

The development of an *EGFR* mutation seems to be an early event in the development of NSCLC,[15] whereas *EGFR* amplification is a relatively late event more prevalent in metastatic lesions[15-17] and may contribute to the development of brain metastases.[18] Other mechanisms for increased brain metastases in *EGFR*-mutant NSCLC may include Met activation[19] and epithelial-mesenchymal transition.[20,21] Studies performed in patients resistant to EGFR TKIs have demonstrated epithelial-mesenchymal transition in a subset of *EGFR*-mutant NSCLC.[20,21] *CRIPTO1* is an oncofetal gene related to the EGF–*Cripto-1/FRL-1/Cryptic* family, which has been noted in *EGFR*-mutant NSCLC to activate SRC and ZEB1 to promote epithelial-mesenchymal transition.[21] Loss of epithelial markers such as E-cadherin and cell-cell adhesion during epithelial-mesenchymal transition may result in increased motility and invasiveness of lung cancer cells.[20] A subset of patients with *EGFR*-mutant NSCLC may undergo epithelial-mesenchymal transition; therefore, their cancer may be more likely to metastasize to the brain. These findings would encourage increased surveillance for brain metastases even in asymptomatic patients, to treat them early, possibly with a local therapy (such as

Table 4. Multivariate Analysis of Survival in Patients With Epidermal Growth Factor Receptor Mutations

Effect	Variable	OR (95% CI)	*P*
Age		1.00 (0.48 to 1.03)	.93
Sex	Female *v* male	0.85 (0.6 to 1.19)	.34
Center	TMH *v* UNMC	0.76 (0.37 to 1.57)	.46
Smoking	No *v* yes	0.97 (0.62 to 1.53)	.9
Brain metastases	No *v* yes	0.71 (0.48 to 1.03)	.07
Extracranial metastases	No *v* yes	0.21 (0.03 to 1.51)	.12
Type of mutation	Exon 19 *v* exon 21	0.73 (0.53 to 1.01)	.06

Abbreviations: OR, odds ratio; TMH, Tata Memorial Hospital; UNMC, University of Nebraska Medical Center.

stereotactic radiosurgery rather than whole-brain radiation therapy), thereby avoiding significant morbidity.

The presence of brain metastases decreases OS, with a reported median survival in patients of only 3 to 5 months.[22] The results of the current study suggest that patients with tumors harboring an activating *EGFR* mutation have a better OS and that the presence of brain metastases does not necessarily worsen prognosis. Previous studies of prophylactic cranial irradiation (PCI) in NSCLC have shown that it decreases the risk of brain metastases without survival improvement.[23,24] Identification of patients with NSCLC at high risk of brain metastases may improve the beneficial effects of PCI. Prior studies have demonstrated clinical features (including increased size of primary tumor,[25] higher nodal stage,[25-27] and histology [adenocarcinoma or nonsquamous *v* squamous])[25-27] that are associated with an increased incidence of brain metastases in patients with NSCLC. One study found that PCI improved disease-free survival among patients with NSCLC with a higher risk of brain metastases; OS was similar, at least partly related to early discontinuation of therapy and relatively small sample size.[28]

Similar to previous reports, the presence of an *EGFR* mutation in this study predicted better OS.[29,30] However, for patients whose tumors had an *EGFR* mutation, none of the common factors considered (including the presence of brain metastases) predicted for survival. These results differ from those recently published by Li et al,[31] who found that the presence of *EGFR* mutations, especially exon 19 deletions, predicted for outcomes in patients with brain metastases. These differences may be a result of the sample size in each study. Li et al included 66 patients with *EGFR* mutations (33 patients each with exon 21 point mutation and exon 19 deletion), whereas the current study includes 452 patients with tumors harboring an *EGFR* mutation (exon 21 point mutation, n = 190; exon 19 deletion, n = 229; uncommon mutations, n = 32). In addition, the ethnicity of the patients could also contribute to the disparity. The study by Li et al included East Asian patients, whereas the current study consisted of South Asian and white patients.

This study did not find a significant difference in outcomes between patients whose tumors had exon 19 deletions and those with exon 21 mutations, although the former group had a 27% decreased risk of death compared with those with an exon 21 mutation. Older studies have not found a difference in OS,[32] but recent studies found a better prognosis with the presence of an exon 19 deletion. Survival analyses of the LUX-Lung 3 and the LUX-Lung 6 trials (afatinib *v* chemotherapy [cisplatin and pemetrexed, LUX-Lung 3; cisplatin and gemcitabine, LUX-Lung 6]) demonstrated a significant improvement in OS with afatinib compared with cisplatin-based chemotherapy in patients with tumors that harbored exon 19 deletions, but not L858R mutations.[33] A recent meta-analysis of 4,835 patients enrolled in clinical trials of EGFR TKIs suggested that patients with an exon 19 deletion have a better prognosis (hazard ratio, 0.61; 95% CI, 0.43 to 0.86).[34] Again, this discrepancy may reflect the sample size differences.

Integration of *EGFR* mutation status with other clinical predictors may identify patients with a higher risk of developing brain metastases, who may benefit from close monitoring and serve as candidates for enrollment in future trials of PCI. Further investigation into the role of *EGFR* mutations in the development of brain metastases in patients with NSCLC may also provide insights into the molecular biology of brain metastases for lung cancer and other malignancies that commonly metastasize to the brain.

A retrospective study design has inherent limitations. Patients more likely to have a tumor with an *EGFR* mutation on the basis of clinical features are more likely to be tested. A multivariate analysis was performed to adjust for known confounding variables, but unknown variables may exist. The incidences of *EGFR* mutations in each cohort are similar to those reported in the literature for the two ethnicities studied in this analysis.[35,36] A few studies have shown a difference in the incidence of *EGFR* mutation in never-smokers and former or current smokers (51% *v* 19% *v* 4%, respectively) as well as with the history of smoking pack-year numbers and smoke-free years.[37] The retrospective nature of this study did not allow such categorization of smoking status. Conversely, to our knowledge, this is the largest study to have assessed the incidence of *EGFR* mutation in NSCLC with brain metastases and effects of *EGFR* mutation status on OS. In conclusion, patients whose tumors harbored an *EGFR* mutation had a nearly twofold increased risk of brain metastases, but *EGFR* status did not predict for survival in patients who had brain metastases.

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AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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APPENDIX

Table A1. Impact of Epidermal Growth Factor Receptor Mutation Type on Metastasis for the Entire Cohort of Patients

	Mutation Type, No. (%)		
Variable	**Exon 19**	**Exon 21**	***P***
Metastatic disease			
No metastasis	9 (3.9)	12 (6.3)	.44
Metastasis other than brain	175 (76.4)	137 (72.1)	
Brain metastasis	45 (19.7)	41 (21.6)	
Metastatic disease			
No brain metastasis	184 (80.3)	149 (78.4)	.63
Brain metastasis	45 (19.7)	41 (21.6)	

Table A2. Multivariate Analysis of Positive Epidermal Growth Factor Receptor Mutational Status (Tata Memorial Hospital cohort)

Effect	**Variable**	**OR (95% CI)**	**P**
Sex	Female *v* male	1.19 (0.90 to 1.57)	.23
Smoking	No *v* yes	2.31 (1.69 to 3.15)	< .001
Alcohol intake	No *v* yes	1.78 (1.16 to 2.73)	.0089
Stage	IV *v* III	2.13 (0.43 to 10.60)	.36
Adenocarcinoma	Yes *v* no	3.18 (1.92 to 5.28)	< .001
Metastatic disease	Brain metastasis *v* metastasis other than brain	1.81 (1.30 to 2.51)	.0018
	No metastasis *v* metastasis other than brain	1.06 (0.21 to 5.37)	

Abbreviation: OR, odds ratio.

Table A3. Multivariate Analysis of Positive Epidermal Growth Factor Receptor Mutational Status (University of Nebraska Medical Center cohort)

Effect	Variable	OR (95% CI)	*P*
Sex	Female *v* male	1.58 (0.38 to 6.61)	.53
Smoking	No *v* yes	77.12 (12.57 to 473.03)	< .001
Alcohol intake	No *v* yes	0.48 (0.09 to 2.56)	.39
Stage	III/IV *v* I/II	0.26 (0.02 to 2.82)	.27
Adenocarcinoma	Yes *v* no	0.94 (0.09 to 10.46)	.96
Metastatic disease	Brain metastasis *v* metastasis other than brain	8.42 (1.09 to 65.14)	.095
	No metastasis *v* metastasis other than brain	1.04 (0.13 to 8.38)	

NOTE. As a result of the small sample sizes, ethnicity had to be excluded, and stage was reclassified. Alcohol use and adenocarcinoma were no longer significant. Nonsmoking status was highly significant. Again, pairwise comparisons were conducted between the categories of metastatic disease. The trend for brain metastasis is similar, and the pairwise comparison of brain metastasis versus other metastasis was significant ($P = .033$).
Abbreviation: OR, odds ratio.

Table A4. Multivariate Analysis of Factors Affecting Overall Survival

Effect	Variable	OR (95% CI)	*P*
Age		1.00 (0.99 to 1.01)	.08
Sex	Female *v* male	0.79* (0.65 to 0.95)	.01
Center	TMH *v* UNMC	1.3 (0.97 to 1.75)	.09
Smoking	No *v* yes	0.85 (0.70 to 1.02)	.09
Brain metastases	No *v* yes	0.75* (0.59 to 0.94)	.02
Extracranial metastases	No *v* yes	1.00 (0.59 to 1.69)	1.00
Activating *EGFR* mutation	No *v* yes	1.3* (1.08 to 1.57)	.006

Abbreviations: *EGFR*, epidermal growth factor receptor; OR, odds ratio; TMH, Tata Memorial Hospital; UNMC, University of Nebraska Medical Center.
*Statistically significant.