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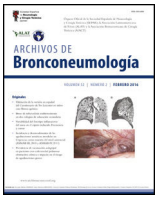

Original Article

Overall Survival Analysis and Characterization of an EGFR Mutated Non-Small Cell Lung Cancer (NSCLC) Population

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ABSTRACT

Background: Patients with activating somatic mutations in the Epidermal Growth Factor Receptor (EGFR) have better clinical outcomes when treated with Tyrosine Kinase Inhibitors (TKI) over chemotherapy. However, the impact of the use of TKIs on overall survival outside clinical trials is not well established.
Objective: To characterize and analyze the overall survival of a Caucasian population with NSCLC and EGFR mutations.
Methods: A retrospective cohort analysis of patients with NSCLC screened for EGFR mutations (exons 18–21) between October 2009 and July 2013 was conducted. Clinical and pathological characteristics, mutational EGFR status, treatment and overall survival were evaluated.
Results: From the 285 patients which performed screening for EGFR mutations, 54 (18.9%) had mutations, 25 (46.3%) of which in exon 19 and 20 of which (37.0%) in exon 21. The occurrence of mutations was associated with female sex and non-smoking habits (both, $P < .001$). The median survival of the global population was 12.0 months, with a better overall survival in mutated than non-mutated patients (20.0 vs 11.0 months, respectively; $P = .007$).
Conclusion: These data contribute for a better knowledge of our lung cancer population concerning the mutational status and clinical outcomes, confirming a better overall survival for the patients with EGFR TKI sensible mutations.

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Análisis de la supervivencia global y caracterización del perfil mutacional del gen EGFR en una población con cáncer de pulmón no microcítico

Palabras clave:
Receptor del factor de crecimiento epidérmico
Inhibidores de tirosina cinasa
Supervivencia global en cáncer de pulmón
Cáncer de pulmón no microcítico
Pulmón

RESUMEN

Antecedentes: Los pacientes con mutaciones somáticas activantes en el receptor del factor de crecimiento epidérmico (EGFR) obtienen mejor resultado clínico cuando se tratan con inhibidores de la tirosina cinasa (TQ) frente a quimioterapia. Sin embargo, el impacto de la terapia en inhibidores de TQ en la supervivencia global de los pacientes no está del todo establecido en la práctica clínica habitual.
Objetivo: Caracterizar y analizar la supervivencia global de una población caucásica con cáncer de pulmón no microcítico y mutaciones en el gen EGFR.
Métodos: Se realizó un análisis retrospectivo de una cohorte de pacientes con cáncer de pulmón no microcítico con mutaciones en el gen EGFR (exones 18-21) entre octubre de 2009 y julio de 2013. Se evaluaron las características clínicas y patológicas, el estatus mutacional del gen EGFR, el tratamiento y la supervivencia global.

Abbreviations: NSCLC, Non-Small Cell Lung Cancer; EGFR, Epidermal Growth Factor Receptor; TKI, Tyrosine Kinase Inhibitors.

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Resultados: De los 285 pacientes que se cribaron para caracterización de mutaciones en el gen EGFR, 54 (18,9%) presentaron mutaciones, de los cuales 25 (46,3%) tenían mutaciones en el exón 19 y 20 (37,0%) en el exón 21. Se observó que la ocurrencia de mutaciones estaba asociada al género femenino y al no consumo de tabaco (p < 0,001 en ambos casos). La supervivencia media de la población global fue de 12 meses, con una mejor supervivencia global en pacientes que presentaron mutaciones que en los que no las presentaron (20 vs. 11 meses, respectivamente, p = 0,007).
Conclusión: Estos datos contribuyen a mejorar el conocimiento de nuestra población con cáncer de pulmón con relación a su estatus mutacional y el resultado clínico, confirmando una mayor tasa de supervivencia global en los pacientes con mutaciones en el gen EGFR sensibles a inhibidores de TQ.
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Introduction

Lung Cancer is the most frequent cause of cancer death.[1] NSCLC is more often diagnosed in advanced stages,[2] which reduces the therapeutic options to cytotoxic chemotherapy, with modest outcomes.[3] Its poor prognosis turns this disease into an emergent area of investigation.

The Epidermal Growth Factor Receptor (EGFR) belongs to the ErbB family, composed by several transmembrane tyrosine kinase receptors.[4] These receptors mediate extracellular growth factors such as the epidermic growth factor.[4] Dysregulation of these receptors leads to uncontrolled proliferation and increased resistance to apoptosis[5] as well as modified cell adhesion and increased migration capacity, facilitating neoplastic invasion and metastization.[6]

The EGFR tyrosine kinase domain is found in the exons 18–24 and the more relevant mutations are located in the exons 18–21.[7] The most frequent EGFR mutations consist in in-frame deletions in exon 19, by the modification of the LREA amino-acid motif (delE746–750) and in missense mutations in exon 21 (L858R codon).[8]

The presence of EGFR mutations in NSCLC is associated to certain clinical characteristics as female sex, Asian ancestry, absence of smoking habits and histology of adenocarcinoma.[8]

EGFR tyrosine kinase inhibitors (TKI) were first used in a non-selective way in NSCLC treatment resulting in disappointing outcomes, being effective only in a small proportion of patients.[9]

The first evidence that EGFR mutations could be used as a target therapy emerged thirteen years ago.[8] Since then, several clinical trials concluded that TKI were more effective than chemotherapy in the treatment of NSCLC patients with EGFR mutation,[8,10] except when the EGFR mutation is in exon 20 (insertion mutations and T790m).[11] Current recommendations support EGFR TKI as the first-line treatment in patients with advanced NSCLC and EGFR TKI sensible mutations.[12,13]

The knowledge of EGFR mutations and its relation with treatment outcomes was one of the most recent important steps in lung cancer management. Tumor-free progression and quality of life parameters are better with TKI when compared to chemotherapy,[14,15] but until now, only studies with Afatinib proved survival advantage for the exon 19 deletion over chemotherapy.[16] Real life data, outside clinical trials information, is poor. In our population neither the correlation between specific clinical features and EGFR mutations nor the effect on survival are well established.

In the present study, a *cohort* of NSCLC patients tested for EGFR mutation was characterized and a survival analysis was conducted.

Patients and Methods

Study Design and Population Selection

Retrospective study of a cohort of lung cancer patients (n = 285) followed in Centro Hospitalar São João with stage IIIB/IV NSCLC or with recurrence or progression at the time of the inclusion period, who were submitted to EGFR mutation screening, between October 2009 and June 2013. Criteria for screening were adenocarcinoma histology, specific characteristics as female sex, younger age or absence of smoking habits. Follow up for survival was censured in January 2016.

Study ethical approval was obtained by the Ethical Committee from the present Hospital.

Data

Age at diagnosis, sex, smoking habits, tumor stage, histopathology, EGFR mutational status, treatments, overall survival and tumor progression were evaluated.

Tumor staging was based on the 6th TNM system in patients diagnosed until the end of 2010 and on the 7th TNM system from 2011 to June 2013. The stage considered was the tumor stage at diagnose. Regarding smoking habits, patients were classified as non-smokers, active smokers and ex-smokers (≥6 months of cessation). The overall survival was calculated using the difference between the date of death and the diagnosis date.

EGFR Mutation Screening

The EGFR mutation screening was performed by IPATIMUP (Institute of Molecular Pathology and Immunology of the University of Porto). The search of the exons 18–21 of the EGFR gene was performed through direct sequencing of Polymerase Chain Reaction (PCR) products obtained from the tumoral cells.

Statistical Analysis

Descriptive statistics (frequency, median and mean) were used to calculate the demographic and clinical characteristics. Categorical comparisons were calculated by chi-square test or by Fisher's exact test. Continuous variables were compared using the t-test.

The survival related results were obtained using the log-rank Kaplan–Meier product-limit estimates. Patients with one month or less of overall survival were excluded for the survival analysis. Statistical significance was set at $P < .05$ for all analyses. All analyses were performed using the software IBM SPSS (Statistical Package for the Social Sciences) v.21.

Results

Demographic characteristics, clinical staging and histology are summarized in Table 1. Among the 285 patients included, 186 (65.3%) were male and 99 (34.7%) female. The mean age at diagnosis was 66.3 years (standard deviation 11.7 years). The majority of the patients had some degree of smoke exposure, 88 (30.9%) were smokers, 84 (29.3%) ex-smokers and 94 (33.0%) never smoked.

Table 1
Global, EGFR Mutated and EGFR Non-mutated Population Characterization.

	Global population (n = 285)	EGFR mutated patients (n = 54; 18.9%)	EGFR non-mutated patients (n = 231; 81.1%)	*P* value
Age at the diagnose – mean (years)/standard deviation	66.31 +/– 11.679	67.41 +/– 11.908	66.06 +/– 11.636	.445[a]
Gender, no. (%)				
Male	186 (65.3%)	17 (31.5%)	169 (73.2%)	
Female	99 (34.7%)	37 (68.5%)	62 (21.8%)	<.0001[b]
Smoke habits, no. (%)				
Non-smokers	94 (33.0%)	34 (69.4%)	60 (27.6%)	
Smokers	88 (30.9%)	5 (10.2%)	83 (38.2%)	<.0001[b]
Ex-smokers [1;2 years]	4 (1.4%)	1 (2.0%)	3 (1.4%)	
Ex-smokers [2;5 years]	25 (8.8%)	1 (2.0%)	24 (9.0%)	
Ex-smokers [5;10 years]	16 (5.6%)	1 (2.0%)	15 (6.9%)	
Ex-smokers [>10 years]	39 (13.7%)	7 (2.0%)	32 (14.7%)	
Missing values	19	5	14	
ECOG PS, no. (%)				
0	125 (43.9%)	21 (41.2%)	104 (49.3%)	
1	81 (28.4%)	10 (19.6%)	71 (33.6%)	<.0001[b]
2	40 (14%)	19 (37.3%)	21 (10.0%)	
3	12 (4.2%)	0 (0.0%)	12 (5.7%)	
4	4 (1.4%)	1 (2.0%)	3 (1.4%)	
Mean/standard deviation	0.81 +/– 0.959	1.02 +/– 0.990	0.76 +/– 0.947	
Missing values	23	3	20	
Lung co-morbidities, no. (%)				
Without lung co-morbidities	203 (76%)	43 (82.7%)	164 (76.3%)	
DPOC	26 (9.1%)	4 (7.7%)	23 (10.7%)	.22[b]
Tuberculosis scars	11 (3.9%)	0 (0.0%)	11 (5.1%)	
Others	22	5	17	
Missing values	18	2	16	
Tumoral staging, no. (%)				
IV	211 (74.0%)	41 (75.9%)	170 (73.6%)	
IIIB	39 (13.7%)	2 (3.7%)	37 (16.0%)	
IIIA	17 (6.0%)	5 (9.3%)	12 (5.2%)	
IIB	6 (2.1%)	0 (0.0%)	6 (2.6%)	
IIA	3 (1.1%)	1 (1.9%)	2 (0.9%)	
IB	1 (0.4%)	1 (1.9%)	0 (0.0%)	
IA	8 (2.8%)	4 (7.4%)	4 (1.7%)	
				.003[b]
Metastization (IV stage), no. (%)				
Local	122	25	97	
Pleural effusion	42 (19.9%)	6 (14.6%)	36 (21.2%)	.471[b]
Lung metastasis	62 (29.4%)	15 (36.6%)	59 (34.7%)	
Pleural effusion and lung metastasis	14 (6.6%)	4 (9.8%)	10 (5.9%)	.482[b]
Others	4	0	4	
Distant	135	26	109	
Bone metastasis	56 (26.5%)	12 (29.3%)	42 (24.7%)	
Brain metastasis	23 (10.9%)	4 (9.8%)	20 (11.8%)	
Adrenal metastasis	12 (5.7%)	0 (0.0%)	12 (7.1%)	
Hepatic metastasis	11 (5.2%)	2 (4.9%)	9 (5.3%)	
Others/multiple metastasis	33	8	26	
Histology, no. (%)				
Adenocarcinoma	222 (77.9%)	51 (94.4%)	171 (74.0%)	
Squamous-cell carcinoma	32 (11.2%)	0 (0.0%)	32 (13.9%)	.063[b]
NOS	30 (10.5%)	3 (5.6%)	27 (11.7%)	
Neuroendocrine tumor	1 (0.4%)	0 (0.0%)	1 (0.4%)	
Sarcomatoide carcinoma	1 (0.4%)	0 (0.0%)	1 (0.4%)	
EGFR status, no. (%)				
Mutated	54 (18.9%)			
Wild-type	231 (81.1%)			

[a] *t*-Test calculation.
[b] Results from Chi-Square calculation, unless the cases where less of five patients where expected for each group, when Fisher Exact test was used.

Within ex-smokers, most (n = 39; 46.4%) quit smoking over the last ten years before diagnosis.

Regarding clinical staging, most patients (n = 250; 87.7%) were diagnosed at advanced stages of the disease and 35 (12.3%) presented with non-advanced stages with progression at the time of EGFR analysis.

Among stage IV patients, 72 (34.1%) had intrapulmonary metastasis, 85 (40.3%) distant metastasis and 50 (23.7%) both intrapulmonary and distant metastasis. The most frequent organs metastasized were bone (n = 56; 26.5%), followed by brain (n = 23; 10.9%), adrenal (n = 12; 5.7%) and liver (n = 11; 5.2%). Twenty-seven patients (12.8%) had more than one organ involved.

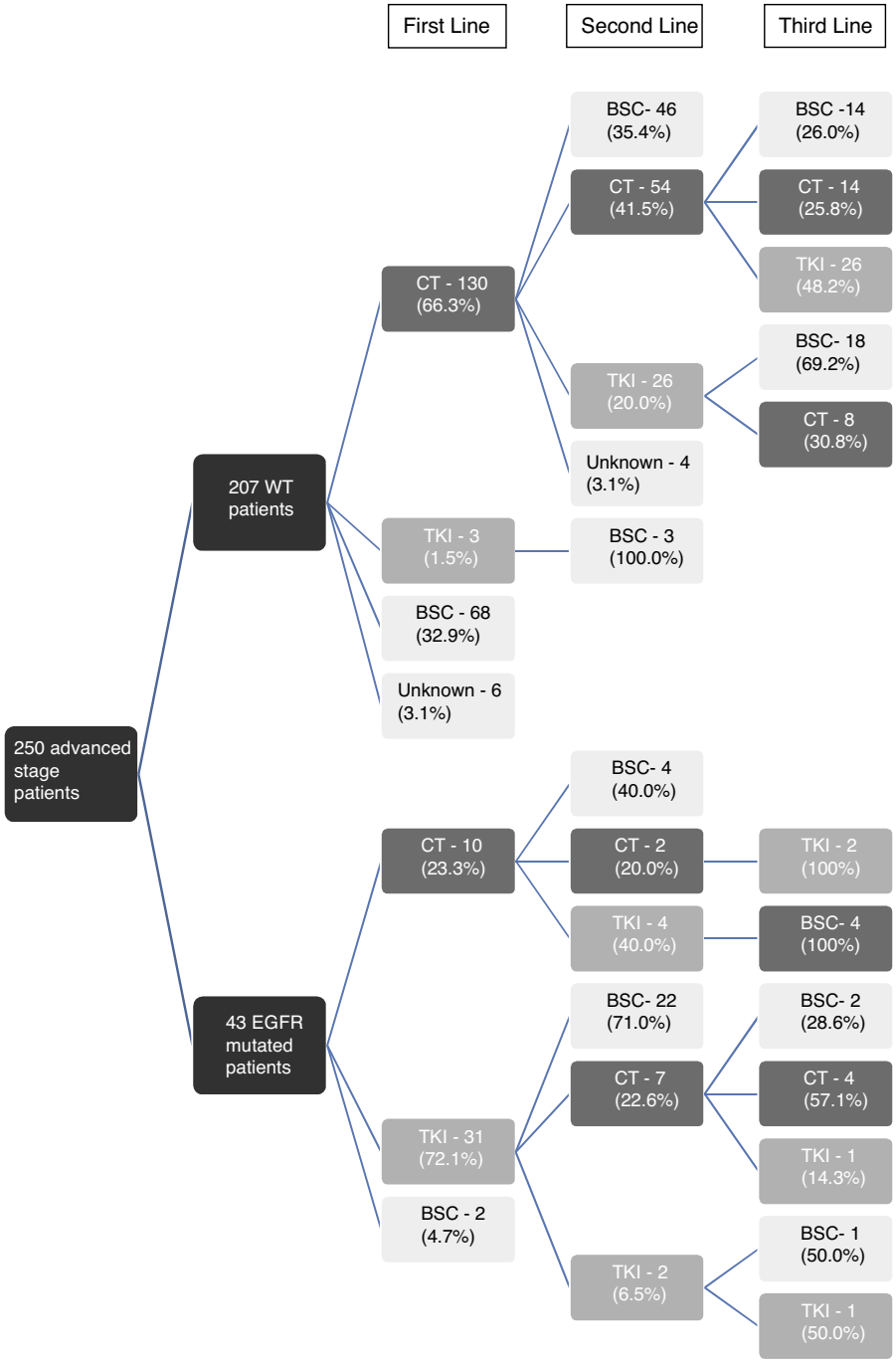

Fig. 1. Types of treatments of the 250 patients with advanced disease. EGFR – epidermal growth factor receptor; WT – wild type; CT – chemotherapy; TKI – tyrosine-kinase inhibitor; BSC – best supportive care.

From the analysis of 285 lung tissue samples, 222 (77.9%) corresponded to adenocarcinoma, 32 (11.2%) to squamous cell carcinoma and 30 (10.5%) to not otherwise specified (NOS) NSCLC. There were sporadic samples (n = 2, 0.8%) of neuroendocrine and sarcomatoid tumors.

The EGFR screening was made on samples obtained from transthoracic biopsies in 88 (30.9%) cases, bronchial biopsies in 84 (29.5%), surgical specimens in 29 (10.2%), pleural fluid in 26 (8.4%) and biopsies of metastasis in 18 (6.3%).

EGFR mutations were detected in 54 patients (18.9%), of which 25 (46.3%) were in exon 19, 20 (37.0%) in exon 21, 5 (9.3%) in exon 20, and 4 (7.4%) in exon 18 (Table 2). There were no samples with more than one mutation. The most common mutations were in-frame deletions at exon 19 (n = 25; 46.3%) ant the p.L858R missense mutation in exon 21 (n = 20; 37.1%).

In the EGFR mutated group there was a predominance of female *versus* male ($P < .001$) and of non-smokers *versus* smokers or ex-smokers ($P < .001$).

There was no statistically significant difference between groups regarding tumor progression. The most frequent *de novo* metastasis diagnosed in both groups were bone metastasis (n = 33; 37.5%) followed by brain metastasis (n = 27; 30.7%) and hepatic metastasis (n = 20; 22.7%).

Table 2
EGFR Mutations.

Exon	n	%	n (%) Total
18			
c.2156G > C (G719A)	2	50.0	4 (7.41%)
c.2117T > C	1	25.0	
c.2156G > A	1	25.0	
19			
c.2235_2249del15	11	44	25 (46.30%)
c.2236_2250del15	5	20	
c.2240_2257del18	3	12	
c.2237_2254del19insT	2	8	
c.2236_2252del17insAT	1	4	
c.2240_2248del9	1	4	
c.2240_2254del15	1	4	
2239_2248del10insC	1	4	
20			
c.2312_2313ins9	2	40	5 (9.26%)
c2307_2308ins6	1	20	
c.2308_2309ins9	1	20	
c.2308ins9	1	20	
21			
c.2573T > G (L858R)	20	100	20 (37.04%)

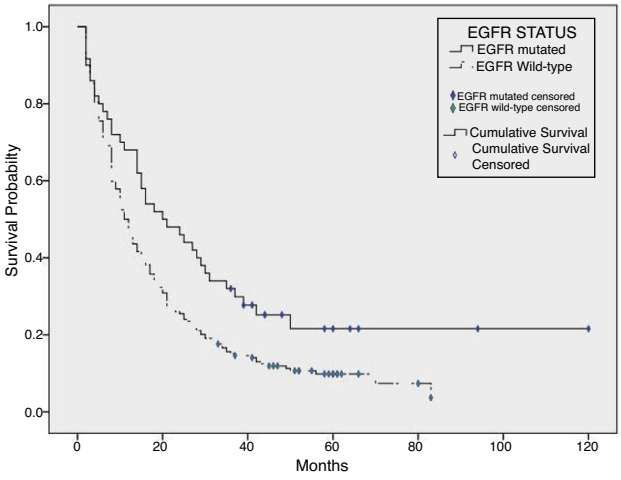

Fig. 2. Kaplan–Meier survival curve of EGFR mutated patients (continuous line) versus EGFR non-mutated patients (dotted line). *P* value = .007.

The first line treatment options, within the 43 EGFR mutated patients with advanced disease (79.6% of all EGFR mutant cases), were TKI in 31 (72.1%) patients, chemotherapy in 10 (23.3%) patients and best supportive care in 2 (4.7%) patients.

Regarding the TKI treatment, 37 (86.0%) EGFR mutant patients were exposed to TKI: 31 (83.8%) in first line of treatment, 4 (10.8%) in second line and 2 (5.4%) in third line (Fig. 1). The TKI more often used was Erlotinib in 21 (65.8%) patients and Gefitinib in 10 (27.0%). The mean duration of the TKI treatment was 8.8 ± 7.7 months (median of 8 months).

The global median overall survival (OS) was 12.0 months [95% confidence interval (CI) 9.6–14.4]. Analyzing the cohort by its EGFR mutational status, the median overall survival of the non-mutated group was 11.0 months (95% CI 9.1–12.9) and 20.0 months for the mutated group (95% CI 10.1–29.9) (*P* = .007) (Fig. 2).

In the non-mutated group, younger patients (<65 years; *P* < .001) and female sex (*P* = .03) had better overall survival (Table 3). Among the EGFR mutated group, sex (*P* = .342), age (*P* = .253), smoking habits (*P* = .666) and the EGFR exon mutated [KM comparison of 4 exons: *P* = .188, KM of TKI sensible mutations versus TKI non-sensible mutations (EGFR exon 20 insertion mutations): *P* = .362] did not have a statistical impact on OS.

Tumor staging <IIIB at diagnosis was related to better OS in both mutated (*P* = .005) and non-mutated (*P* < .001) populations.

The two-year overall survival of global population was 26.7%, corresponding to 75 patients (26.7%), 52 (20.6%) non-mutated patients and 23(42.6%) EGFR mutated. At the end of follow-up (January 2016), 32 (11.2%) patients were alive [12 (22.2%) EGFR mutated and 20 (8.7%) non-mutated patients].

Discussion

To our best knowledge, this is one of the first overall survival analysis, outside of clinical trials, comparing EGFR mutated patients to EGFR non-mutated patients with lung cancer. Our results showed a frequency of EGFR mutation of 18.9% and better overall survival in this population.

The mutation distribution was predominantly in the exons 19 and 21 [25 (46.3%) and 20 (37.0%) mutations, respectively], totalizing 45 (83.3%) of all mutations. 5 (9.3%) mutations occurred in exons 20 and 4 (7.4%) in exon 18. This distribution is similar to other studies.[17,18]

The frequency of EGFR mutations varies geographically. Higher frequencies are found in Asia, as shown in the work of Shi et al. (51.4%) that represents several regions of that continent.[19] In the Latin America intermediate frequencies are 33.2% in Argentina, Colombia, Mexico and Peru.[20] In the European region, studies revealed a frequency of 16.6%, 12.3% and 4.9% in Spain,[10] Denmark[21] and Germany.[22]

The frequency of EGFR mutations in the Portuguese population is undetermined. In the study of Mello et al., the frequency of this mutation was 16.9%[17] while in Castro et al. the global frequency was 13.1%.[23] Still, in this last work, different frequencies were determined: 16.3% and 10.4% corresponding to different inclusion criteria, from 2006 to 2009 were included patients with adenocarcinoma or without smoking habits, while in 2010 all patients with NSCLC were included.[23] Our mutation frequency of 18.9% is slightly higher to those published. Some bias related to phenotypic preselection could have occurred. The majority were stage III/IV adenocarcinoma but some patients were selected based on their characteristics such as non-smokers, female sex, younger age and progression or recurrence of tumor staged <IIIB, independently of their histology. EGFR mutations were only identified in adenocarcinoma and NOS samples, reinforcing the histologic type as criteria to the EGFR screening. EGFR mutation frequency vary along studies not only due to ethnical particularities but also to methodological discrepancies, being lower when restrictive clinical criteria were not used. To clarify this issue it is needed an epidemiologic study.

Association between EGFR mutation status and survival is difficult to estimate, particularly outside of a clinical trial setting. The obstacle to this association could be explained by the different lines of treatment and the crossover of treatments.[24,25] The median OS of the EGFR mutated group was 9 months superior to the OS of the non-mutated group (20.0 vs 11.0 months; *P* = .007). This values for OS are similar to other clinical trials,[16,24,26–28] particularly in the EURTAC trial. These OS difference could be explained by a possible influence of the mutational status in the prognosis and by the use of more efficacious drugs than the usual chemotherapy in the mutated population.

Some studies, as in the Iressa Pan-Asia Survival Study (IPASS), comparing gefitinib with paclitaxel plus carboplatin as the first-line therapy in Asian patients, have demonstrated a statistically significant higher response rate to chemotherapy in EGFR mutated patients (47.3% versus 23.5%) than patient without EGFR mutations, but the progression-free survival (PFS) and OS were not different between patients with and without EGFR mutation in the chemotherapy arm.[27] In previous studies, the survival of EGFR

Table 3
Kaplan–Meier Global Survival Analysis.

Total population (n = 254;100%)							EGFR Non-mutated population (n = 204; 80.3%)							EGFR mutated population (n = 50; 19.7)						
	n	Median[a]	CI 95%[a]		Standard deviation[a]	*P*		n	Median[a]	CI 95%[a]		Standard deviation[a]	*P*		n	Median[a]	CI 95%[a]		Standard deviation[a]	*P*
Age							Age							Age						
<65 years	113	17	13.53	– 20.47	1.77		<65 years	93	17	12.28	– 21.73	2.41		<65 years	20	16	9.43	– 22.57	3.35	
>=65 years	141	11	8.95	– 13.05	1.05		>=65 years	111	10	7.95	– 12.06	1.05		>=65 years	30	24	9.24	– 38.76	7.53	
						.031							.001							.282
Gender							Gender							Gender						
Female	91	16	10.22	– 13.78	0.91		Female	56	16	9.72	– 22.28	0.87		Female	35	16	11.38	– 20.62	8.54	
Male	163	12	12.41	– 19.59	1.83		Male	148	10	8.30	– 11.70	3.21		Male	15	35	18.26	– 51.74	2.36	
						.038							.035							.211
Smoke habits							Smoke habits							Smoke habits						
Non smoker	85	16	10.58	– 21.42	2.77		Non smoker	53	17	7.84	– 26.16	4.67		Non smoker	32	16	8.25	– 23.75	3.95	
Smoker	80	10	8.47	– 11.53	0.78		Smoker	76	9	7.49	– 10.51	0.77		Smoker	4	20	10.20	– 29.80	5.00	
Ex-smoker	76	12	10.03	– 13.97	10.03		Ex-smoker	66	12	10.56	– 13.45	0.74		Ex-smoker	10	25	0.00	– 57.54	16.60	
						.487							.771							.666
Tumoral stage							Tumoral stage							Tumoral stage						
<IIIB	32	49					<IIIB	21	49					<IIIB	11					
≥IIIB	207	10	8.06	– 11.94	0.99		≥IIIB	169	10	8.60	–11.41	0.72		≥IIIB	38	15	7.96	– 22.04	3.91	
						<.001							<.001							.008
Distant metastasis							Distant metastasis							Distant metastasis						
0	143	19	15.09	– 22.90	1.99		0	115	18	13.96	– 22.04	2.06		0	28	29	6.96	– 51.04	11.24	
1	88	8	6.38	– 9.62	0.83		1	72	8	7.18	– 8.82	0.42		1	16	20	8.24	– 3.76	6.00	
>1	19	6	4.31	– 7.69	0.86		>1	13	5	3.59	– 6.41	0.72		>1	6	7	0.00	– 20.20	6.74	
						<.001							<.001							.007
Mutational status																				
EGFR mutated	204	20	27.20	– 52.47	5.05															
EGFR wildtype	50	11	17.20	– 23.56	0.99															
						.007														
														EGFR mutated exon[b]						
														18	4	20	6.28	– 33.72	7.00	
														19	24	31	15.40	– 46.60	7.96	
														20	4	2				
														21	18	14	3.61	– 29.90	5.30	
																				.188
														18 + 19 + 21 exons	46	21	8.81	– 33.19	6.22	
														20 exon insentions	4	2				
																				.362

[a] Values in months.
[b] Efectuated in EGFR mutated patients.

mutated patients was significantly longer than those without EGFR mutations in groups of patients who received chemotherapy alone without gefitinib or erlotinib.[29,30] The study of Lin CC et al.[31] corroborated that EGFR mutations are associated with a higher tumor response rate to chemotherapy, but are not a predictive biomarker for PFS and OS. Applying the Kaplan–Meier product-limit estimates (Table 3) in the global population, the factors that were associated with a better OS were age inferior to 65 years, female sex, tumor stage <IIIB and the presence of EGFR mutation.

In the non-mutated population features like age <65 years, female sex and stage <IIIB were related to better OS. Among the EGFR mutated patients gender, age and the mutated exon did not influence the prognosis, reinforcing that the presence of EGFR mutations is a major factor associated to better OS.

Results regarding clinical factors that may influence prognosis also vary across studies. Some authors report an association between extrathoracic metastasis, in particular brain metastasis, and the presence of L858 mutation with worse survival.[32–34]

In this study the relevance of the molecular study was confirmed, permitting the identification of the EGFR mutated patients who have a distinct clinical behavior regarding overall survival and response to TKI.

The decision to screen for EGFR mutation was influenced by adenocarcinoma histology, female gender and non-smoking status. This selection bias represents a major limitation of the present study. Within the EGFR mutated group there were only a small group of patients with non-sensible TKI mutations (n = 5). For overall survival calculations, the small size of this group did not permit any conclusion in particular. Those patients were considered within the rest of the EGFR mutated patients which could represent a limitation for the study results. The study reflects the clinical results and therefore has inherent limitations such its retrospective character and a small population analyzed, but its strength is that it is one of the first real life studies aiming the OS of an EGFR mutated population.

Our data showed a better overall survival for EGFR activating mutated patients independently of the clinical characteristics, suggesting that it can be a favorable prognostic marker.

Authorship

Filipa Aguiar: study conception and design, data collection, data analysis and interpretation, statistical analysis, manuscript writing.

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Conflict of Interest

The authors declare no conflict of interest.

References

1. Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, et al. Cancer incidence and mortality worldwide: sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer. 2015;136:E359:386.
2. SEER cancer stat facts: lung and bronchus cancer. Bethesda, MD: National Cancer Institute; 2017.
3. Abernethy A, Arunachalam A, Burke T, McKay C, Cao X, Sorg R, et al. Real-world first-line treatment and overall survival in non-small cell lung cancer without known EGFR mutations or ALK rearrangements in US community oncology setting. PLoS ONE. 2017;12:e0178420.
4. Hynes NE, Lane HA. ERBB receptors and cancer: the complexity of targeted inhibitors. Nature Rev Cancer. 2005;5:341–54.
5. Mosesson Y, Yarden Y. Oncogenic growth factor receptors: implications for signal transduction therapy. Semin Cancer Biol. 2004;14:262–70.
6. Ellerbroek SM, Halbleib JM, Benavidez M, Warmka JK, Wattenberg EV, Stack MS, et al. Phosphatidylinositol 3-kinase activity in epidermal growth factor-stimulated matrix metalloproteinase-9 production and cell surface association. Cancer Res. 2001;61:1855–61.
7. Sharma SV, Bell DW, Settleman J, Haber DA. Epidermal growth factor receptor mutations in lung cancer. Nat Rev Cancer. 2007;7:169–81.
8. Pao W, Miller V, Zakowski M, Doherty J, Politi K, Sarkaria I, et al. 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 U S A. 2004;101:13306–11.
9. Thatcher N, Chang A, Parikh P, Rodrigues Pereira J, Ciuleanu T, von Pawel J, et al. Gefitinib plus best supportive care in previously treated patients with refractory advanced non-small-cell lung cancer: results from a randomised, placebo-controlled, multicentre study. Lancet. 2005;366:1527–37.
10. Rosell R, Moran T, Queralt C, Porta R, Cardenal F, Camps C, et al. Screening for epidermal growth factor receptor mutations in lung cancer. N Engl J Med. 2009;361:958–67.
11. Yu HA, Arcila ME, Hellmann MD, Kris MG, Ladanyi M, Riely GJ. Poor response to erlotinib in patients with tumors containing baseline EGFR T790M mutations found by routine clinical molecular testing. Ann Oncol. 2014;25:423–8.
12. Novello S, Barlesi F, Califano R, Cufer T, Ekman S, Giaj Levra M, et al. Metastatic non-small-cell lung cancer: ESMO clinical practice guidelines. Ann Oncol. 2016;27:1–27.
13. Ettinger DS, Wood DE, Aisner DL, Akerley W, Bauman J, Chirieac LR, et al. Non-small cell lung cancer, Version 5.2017, NCCN clinical practice guidelines in oncology. J Natl Compr Canc Netw. 2017;15:504–35.
14. PaszAres L, Soulieres D, Moecks J, Bara I, Mok T, Klughammer B. Pooled analysis of clinical outcome for EGFR TKI-treated patients with EGFR mutation-positive NSCLC. J Cell Mol Med. 2014;18:1519–39.
15. Yang JC, Hirsh V, Schuler M. Symptom control and quality of life in LUX-Lung 3: a phase III study of afatinib or cisplatin/pemetrexed in patients with advanced lung adenocarcinoma with EGFR mutations. J Clin Oncol. 2013;21:3342–50.
16. Wu YL, Zhou C, Hu C. 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. 2014;15:213–22.
17. de Mello RA, Pires FS, Marques DS, Oliveira J, Rodrigues A, Soares M, et al. EGFR exon mutation distribution and outcome in non-small-cell lung cancer: a Portuguese retrospective study. Tumour Biol. 2012;33:7.
18. Skov BG, Høgdall E, Clementsen F, Krasnik M, Larsen KR, Sørensen JB, et al. The prevalence of EGFR mutations in non-small cell lung cancer in an unselected Caucasian population. APMIS. 2015;123:108–15.
19. Shi Y, Au JS, Thongprasert S, Srinivasan S, Tsai CM, Khoa MT, et al. A prospective, molecular epidemiology study of EGFR mutations in Asian patients with advanced non-small-cell lung cancer of adenocarcinoma histology (PIONEER). J Thorac Oncol. 2014;9:154–62.
20. Arrieta O, Cardona AF, Bramuglia F, Gallo A, Campos-Parra AD, Serrano S, et al. Genotyping non-small cell lung cancer (NSCLC) in Latin America. J Thorac Oncol. 2011;6:1955–9.
21. Weber B, Hager H, Sorrensen BS, McCulloch T, Mellemgaard A, Khalil AA, et al. EGFR mutation frequency and effectiveness of erlotinib: a prospective observational study in Danish patients with non-small cell lung cancer. Lung Cancer. 2013;83:224–30.
22. Boch C, Kollmeier J, Roth A, Stephan-Falkenar S, Misch D, Gruning W, et al. The frequency of EGFR and KRAS mutations in non-small cell lung cancer (NSCLC): routine screening data for central Europe from a cohort study. BMJ Open. 2013;3:e002560.
23. Castro AS, Parente B, Gonçalves I, Antunes A, Barroso A, Conde S, et al. Estudo da mutação do recetor do fator de crescimento epidérmico, durante 5 anos, numa população de doentes com cancro do pulmão de não pequenas células. Revista Portuguesa de Pneumologia. 2013;19:7–12.
24. Mitsudomi T, Morita S, Yatabe Y. West Japan Oncology Group: gefitinib versus cisplatin plus docetaxel in patients with non-small-cell lung cancer harbouring mutations of the epidermal growth factor receptor (WJTOG3405): an open label, randomised phase 3 trial. Lancet Oncol. 2010;11:121–8.
25. Sequist LV, Yang JC, Yamamoto N. Phase III study of afatinib or cisplatin plus pemetrexed in patients with metastatic lung adenocarcinoma with EGFR mutations. J Clin Oncol. 2013;31:3327–34.
26. Rosell R, Carcereny E, Gervais R, Vergnenegre A, Massuti B, Felip E, et al. Erlotinib versus standard chemotherapy as first-line treatment for Europeanpatients with advanced EGFR mutation-positive non-small-cell lung cancer(EURTAC): a multicentre, open-label, randomised phase 3 trial. Lancet Oncol. 2012;13:239–46.
27. Mok TS, Wu YL, Thonggpraset S. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med. 2009;361:947–57.
28. Zhou C, Wu YL, Chen G. 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. 2011;12:735–42.
29. Bell DW, Lynch TJ, Haserlat SM, Harris PL, Okimoto RA, Brannigan BW, et al. Epidermal growth factor receptor mutations and gene amplification in non-small-cell lung cancer: molecular analysis of the IDEAL/INTACT gefitinib trials. J Clin Oncol. 2005;23:8081–92.

30. Eberhard DA, Johnson BE, Amler LC, Goddard AD, Heldens SL, Herbst RS, et al. 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. 2005;23:5900–9.
31. Lin CC, Hsu HH, Sun CT, Shih JY, Lin ZZ, Yu CJ, et al. Chemotherapy response in East Asian non-small cell lung cancer patients harboring wild-type or activating mutation of epidermal growth factor receptors. J Thorac Oncol. 2010;9:1424–9.
32. Park JH, Kim TM, Keam B, Jeon YK, Lee SH, Kim DW, et al. Tumor burden is predictive of survival in patients with non-small-cell lung cancer and with activating epidermal growth factor receptor mutation who receive gefitinib. Clin Lung Cancer. 2013;14:383–9.
33. Lee JY, Lim SH, Kim M, Kim S, Jung HA, Chang WJ, et al. Is there any predictor for clinical outcome in EGFR mutante NSCLC patients treated with EGFR TKIs? Cancer Chemother Pharmacol. 2014;73:1063–70.
34. Li F, Du X, Zhang H, Ju T, Chen C, Qu Q, et al. Next generation sequencing of chinese stage IV lung cancer patients reveals an association between EGFR mutation status and survival outcome. Clin Genet. 2016;91:488–93.