Int J Clin Exp Pathol 2018;11(9):4644-4649
www.ijcep.com /ISSN:1936-2625/IJCEP0080248

Original Article

Comparison of clinicopathologic characteristics between patients with EGFR exon 19 deletion and EGFR L858R mutation in lung cancer

Zhen Chen[1], Jing Zhang[1], Ke Huang[1], Qian Shen[2], Xiaodong Teng[1]

Departments of [1]Pathology, [2]Respiratory Diseases, The First Affiliated Hospital, College of Medicine, Zhejiang University, Hangzhou 310000, China

Received May 24, 2018; Accepted August 15, 2018; Epub September 1, 2018; Published September 15, 2018

Abstract: Introduction: Previous studies had demonstrated that exon 19 deletion (19Del) and L858R mutation in *EGFR* have different prognostic and predictive roles in lung cancer. We aimed to investigate whether these two mutations had different clinicopathologic characteristics in lung cancer. Methods: We enrolled 603 patients who were detected with either 19Del or L858R and collected their baseline clinical characteristics. Chi-square test was used to compare the differences. Results: There was no difference in the distribution of other clinicopathological characteristics, except age, TNM stage, and tumor location ($P < 0.05$). The age of the patients in group 19Del was significantly younger than that in group L858R (mean 59.4 y vs. 62.3 y, $P = 0.001$), and the mutation rate of 19Del decreased gradually with the increase of age. It was rare to have L858R mutation in < 40 y age subgroup. The mutation rate of 19Del was higher than L858R in left lung, and was lower than L858R in right lung ($P < 0.05$). Multivariate analysis showed age and II stage were independent predictive factors for *EGFR* mutation. Conclusion: Patients with 19Del were younger than those with L858R in lung cancer and the younger the patients are, the higher mutation rate of 19Del.

Keywords: Epidermal growth factor receptor, exon 19 deletion, L858R, lung neoplasms

Introduction

Lung cancer is one of the most common human cancers and leading cause of death due to cancer worldwide. Epidermal growth factor receptor (*EGFR*) is an important therapeutic target of lung cancer. The mutations lead to autophosphorylation of the tyrosine residues. These residues further activate downstream signaling cascades, such as the Ras-Raf-MAP-kinase, PI3K-Akt, and STAT pathways, which have strong regulatory effects on cell proliferation, differentiation, survival, and migration [1, 2]. *EGFR* mutations mainly focus on exon 18-21. The two most important mutations are L858R in exon 21 and exon 19 deletion (19Del), and they account for more than 80% of *EGFR* mutations [3], of which the presence is known to be associated with increased response to treatment with EGFR-tyrosine kinase inhibitors (EFGR-TKI) [4-6]. However, recent studies have shown that such patients with the two mutations have different survival outcomes in response to both EGFR-TKIs and chemotherapy [7-9]. Patients with 19Del mutation were more responsive to EGFR-TKI treatment and have a higher proportion of the T790M mutation compared with L858R patients [10]. However, TKI-naive patients having L858R mutation had a relatively prolonged survival compared with TKI-naive patients having 19Del mutation [11].

Although the different responses to treatment between 19Del and L858R were well-identified in previous studies, few have assessed the different clinicopathologic characteristics between 19del and L858R mutations. In the present study, our purpose was to investigate whether these two mutations had different features.

Materials and methods

Patients and samples

A total of 1375 consecutive records of patients with lung cancer who applied for *EGFR* gene

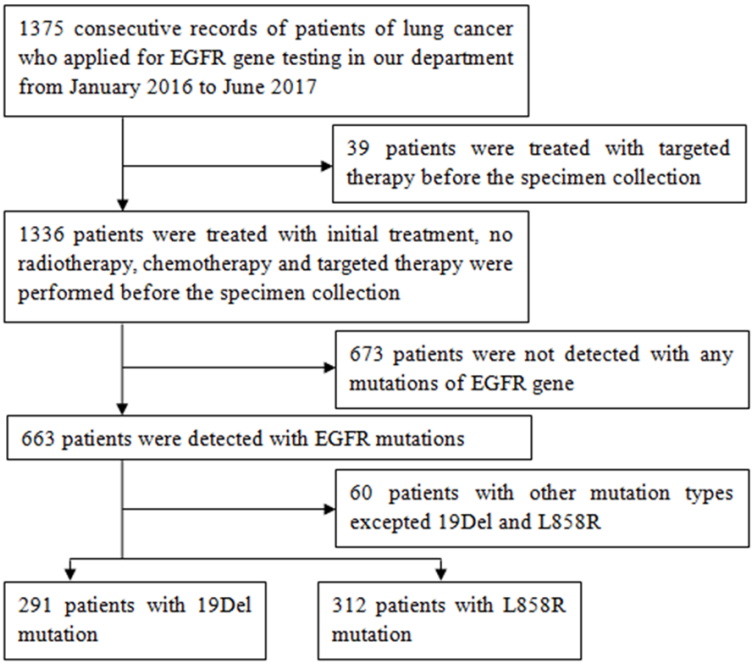

Figure 1. Flow chart of patient screening.

Table 1. Mutation types of EGFR in 1366 patients treated with initial treatment

Mutation types	No.	Mutation rate
L858R	312	23.35%
19Del	291	21.78%
20Ins	16	1.20%
L861Q	10	0.75%
G719X	9	0.67%
S768I	6	0.45%
19Del/T790M	3	0.22%
L858R/T790M	6	0.45%
S768I/L858R	5	0.37%
G719X/S768I	2	0.15%
19Del/L858R	1	0.07%
19Del/L861Q	1	0.07%
G719X/L861Q	1	0.07%

testing in the First Affiliated Hospital College of Medicine, Zhejiang University were enrolled from January 2016 to June 2017. Clinicopathologic characteristics of 603 patients with 19Del or L858R mutation who were treated with initial treatment, no radiotherapy, chemotherapy, and targeted therapy were performed before the specimen collection were collected (some data were not available), including 591 cases of adencarcinoma, 3 cases of squamous cell carcinoma, 6 cases of adenosquamous carcinoma, 1 case of carcinoma sarcomatodes, undifferentiated carcinoma and small cell carcinoma. The flow chart of patient screening was shown in **Figure 1**.

Histological evaluation

The histologic type was classified according to 2015 WHO classification of lung neoplasm by two pathologists [12], and the TNM stage of the disease was determined based on the TNM classification of the Union for International Cancer Control (UICC) [13].

EGFR mutations detection

The formalin-fixed and paraffin-embedded tissue blocks were cut in 5 μm thickness. Ten slices were collected for DNA isolation. Cytological specimens were cell blocks made from pleural fluid. All extractions were carried out using DNA extraction kit according to the manufacturers (AmoyDx, Xiamen, China). *EGFR* mutation was tested using ARMS Detection kit (AmoyDx, Xiamen, China).

Statistical analysis

Statistical analysis was performed using the SPSS statistics software (Version 17.0, Chicago). The correlation of two groups with clinicopathological characteristics was studied via Chi-square test. Multivariate analysis used logistic regression. The comparison of average age between two groups was performed by t test. P value < 0.05 was considered significant.

Results

General characteristics

In all 1336 patients who were treated with initial treatment, there were 740 males and 596 females. The age ranged from 18 to 99 years old, with an average age of 61.4 years old. There were 663 patients with *EGFR* mutation (49.6%) including 291 cases of 19Del, 312 cases of L858R and 60 cases of other mutation types (**Table 1**).

Table 2. Clinicopathological characteristics of patients with 19Del and those with L858R

	19Del		L858R		*P*
All	291		312		0.331
Gender					0.757
Male	123	(42.27%)	128	(41.03%)	
Female	168	(57.73%)	184	(58.97%)	
Age					0.002
≤60 y	151	(51.89%)	122	(39.10%)	
> 60 y	140	(48.11%)	190	(60.90%)	
Smoking history					0.832
Never-smoker	209	(71.82%)	221	(70.83%)	
Smoker	81	(27.84%)	89	(28.53%)	
Unknown	1	(0.34%)	2	(0.64%)	
Tumor location					0.016
Left lung	129	(44.33%)	109	(34.94%)	
Right lung	159	(54.64%)	201	(64.42%)	
Unknown	3	(1.03%)	2	(0.64%)	
Specimen type					0.566
Surgical specimen	126	(43.30%)	146	(46.79%)	
Biopsy specimen	137	(47.08%)	142	(45.51%)	
Cytological specimen	28	(9.62%)	24	(7.69%)	
Differentiation*					0.979
Well	39	(14.61%)	41	(14.14%)	
Moderate	103	(38.58%)	111	(38.28%)	
Poorly	125	(46.82%)	138	(47.59%)	
Lymph node metastasis					0.551
Without	94	(32.30%)	108	(34.62%)	
With	194	(66.67%)	201	(64.42%)	
Unknown	3	(1.03%)	3	(0.96%)	
TNM stage					0.034
I	72	(24.74%)	88	(28.21%)	0.350
II	25	(8.59%)	10	(3.21%)	0.005
III	36	(12.37%)	45	(14.42%)	0.472
IV	154	(52.92%)	166	(53.21%)	0.998
Unknown	4	(1.37%)	3	(0.96%)	
Histological type					0.644
Adenocarcinoma	286	(98.28%)	305	(97.76%)	
Non-adenocarcinoma	5	(1.72%)	7	(2.24%)	
Family history of cancer					0.118
Without	89	(30.6%)	66	(21.2%)	
With	55	(18.9%)	60	(19.2%)	
Unknown	147	(50.5%)	186	(59.6%)	

*Cytological specimens were not included.

Clinicopathological characteristics of the patients with 19Del and L858R

There were 603 patients with 19Del or L858R (291 and 312 cases respectively), the average age was 60.9 years old. There was no difference in the distribution of other basic characteristics (all *P* value > 0.05), except age, TNM stage and tumor location (**Table 2**). Multivariate analysis showed age and II stage were independent predictive factors for *EGFR* mutation (**Table 3**).

Comparison of age between 19Del and L858R groups

t test showed that the age of the patients in group 19Del was significantly younger than that in group L858R (mean 59.4 y vs. 62.3 y, $P = 0.001$). We also classified age group as < 40, 40-50, 50-60, 60-70, 70-80, ≥80y subgroups. Results showed that the mutation rates of 19Del were higher than L858R in < 40, 40-50 and 50-60 subgroups, lower than L858R in 60-70, 70-80, ≥80 subgroups (**Figure 2**). The mutation rate of 19Del decreased gradually with the increase of age. There was no obvious trend in L858R except the mutation rate was very low in the < 40 subgroup.

Comparison of subtypes of adencarcinoma between 19Del and L858R groups

Predominant subtypes were classified by two pathologists in 232 cases of surgically resected primary lung adenocarcinoma. Lepidic, acinar, and papillary predominant subtypes were frequently found in both of two groups, and there was no significant difference in the distribution of adenocarcinoma subtypes in two groups (**Table 4**).

Discussion

The features of *EGFR* mutation, which was frequently found in women, non-smokers, and adenocarcinoma in lung cancer, are well described [11, 14, 15]. As the most common mutation types of *EGFR*, few studies reported the difference of clinical characteristics between 19Del and L858R patients. Jackman et al [16] com-

Table 3. Multivariate logistic analysis of predictive factors for EGFR mutation

	OR	LL (95% CI)	UL (95% CI)	P
Age	1.026	10..9	1.042	0.002
Location	1.389	0.991	1.947	0.056
Stage II	0.337	0.151	0.754	0.008

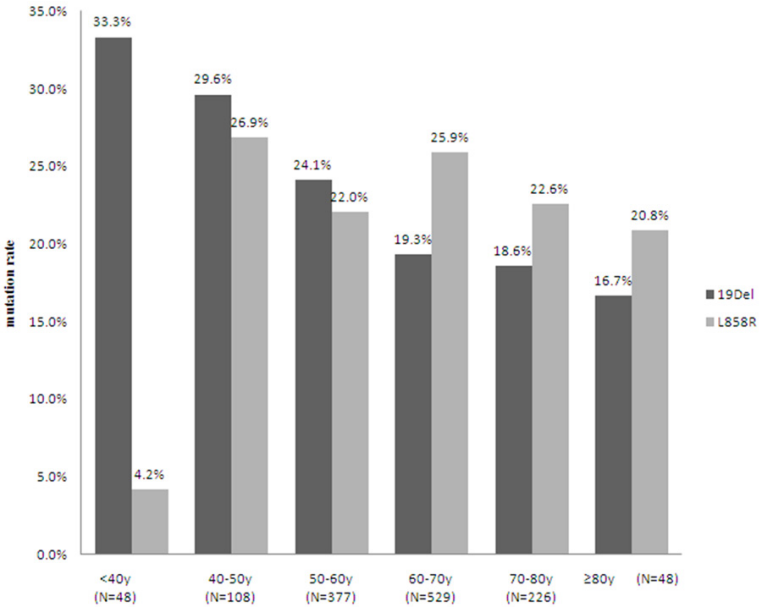

Figure 2. Mutation rate of 19Del and L858R in different age subgroups.

Table 4. Subtypes of adenocarcinoma in 19Del and L858R groups

Predominant subtypes	19Del	L858R	*P*
Total	107	125	0.176
Lepidic	26	42	0.121
Acinar	38	40	1.572
Papillary	28	30	0.704
Solid	12	9	0.288
Mucinous	1	4	0.377
Micropapillary	2	0	0.212

pared the differences between two mutations in sex, race, smoking history and histology characteristics, no significant difference was found. Similarly, Gregory [17] did not find any differences in the metastatic NSCLC, but the median age of the patients in group 19Del was lower than that in group L858R (60 y vs. 64 y). Unfortunately, the authors did not make a statistical comparison. Ke et al [10] found that the percentage of patients aged < 55 years was significantly higher in the 19Del group than the L858R group (47.5% vs. 31.8%, $P = 0.021$), confirming the results of the previous report by Zhang et al [18].

We reviewed 1336 patients with primary lung cancer and found 663 patients with *EGFR* mutations (mutation rate: 49.6%), of which 19Del and L858R occurred in 291, and 312 cases respectively, accounting for 91% of the total number of mutations. There was no significant difference in the rate of mutation between 19Del and L858R ($P > 0.05$). Like the results of previous reports [10, 18], we also found that patients with 19Del were younger than patients with L858R (mean 59.4 y vs. 62.3 y, $P = 0.001$). It is interesting that when we classified age group as six subgroups, the mutation rate of 19Del was higher than that of L858R when the age of patients was smaller than 60 y, reversed when the age of patients was more than 60 y. The mutation rate of 19Del decreased gradually with the increase of age. There was no obvious trend in L858R except the mutation rate was very low in the < 40 y subgroup, significantly lower than that of 19Del (4.2% vs. 33.3%, $P < 0.001$). It was more likely that the mutation of *EGFR* trended to be 19Del in young patients.

In addition, there were differences in the distribution of the two mutations when the location of tumor from which the specimen was collected was different. The mutation rate of 19Del was higher than L858R in left lung, and was lower than L858R in right lung ($P < 0.05$). No similar results were observed in other reports. No statistically significant differences in lymph node metastasis between the two mutations in this study which was different from Zhang' report [18]. The tumor in phase II seems to be more 19Del mutated. No similar results were observed in other reports, so this needs to be verified by large samples. Multivariate analysis showed age and II stage were independent predictive factors for *EGFR* mutation.

He et al [19] reported that higher *EGFR* mutation might be caused by family history of cancer, especially family history of lung cancer. We hypothesized that whether high 19Del mutation in young patients had relationship with a family history of cancer, but the result was negative. This mechanism needs further study.

EGFR mutation was frequently found in lepidic, acinar, and papillary subtypes of adenocarcinoma [20, 21]. 19Del and L858R also showed the same situation in this study, which lepidic, acinar and papillary predominant subtypes were frequently found. Gelatinous, fetal and intestinal adenocarcinoma were not found. There was no significant difference in the distribution of adenocarcinoma subtypes in two groups, which suggested that there is no difference in the morphology of tumors with the two mutations.

In conclusion, our investigation suggested that patients with 19Del were more likely to be younger than those with L858R, and the younger the patients are, the higher mutation rate of 19Del. The two mutations were alsolikely to have a bearing on the location of the tumor.

Acknowledgements

This work was supported by grants from the Medicine and Health Project of Zhejiang Province, China (Grant No. 2018KY061).

Disclosure of conflict of interest

None.

Address correspondence to: Xiaodong Teng, Department of Pathology, The First Affiliated Hospital of Medical College, Zhejiang University, Hangzhou 310003, China. Tel: 86-0571-87236362; E-mail: 1102069@zju.edu.cn

References

[1] Riely GJ, Politi KA, Miller VA and Pao W. Update on epidermal growth factor receptor mutations in non-small cell lung cancer. Clin Cancer Res 2006; 12: 7232-7241.

[2] Perona R. Cell signalling: growth factors and tyrosine kinase receptors. Clin Transl Oncol 2006; 8: 77-82.

[3] Zhu JQ, Zhong WZ, Zhang GC, Li R, Zhang XC, Guo AL, Zhang YF, An SJ, Mok TS and Wu YL. Better survival with EGFR exon 19 than exon 21 mutations in gefitinib-treated non-small cell lung cancer patients is due to differential inhibition of downstream signals. Cancer Lett 2008; 265: 307-317.

[4] Mitsudomi T, Morita S, Yatabe Y, Negoro S, Okamoto I, Tsurutani J, Seto T, Satouchi M, Tada H, Hirashima T, Asami K, Katakami N, Takada M, Yoshioka H, Shibata K, Kudoh S, Shimizu E, Saito H, Toyooka S, Nakagawa K, Fukuoka M; 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: 121-128.

[5] Mok TS, Wu YL, Thongprasert S, Yang CH, Chu DT, Saijo N, Sunpaweravong P, Han B, Margono B, Ichinose Y, Nishiwaki Y, Ohe Y, Yang JJ, Chewaskulyong B, Jiang H, Duffield EL, Watkins CL, Armour AA and Fukuoka M. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med 2009; 361: 947-957.

[6] Rosell R, Moran T, Queralt C, Porta R, Cardenal F, Camps C, Majem M, Lopez-Vivanco G, Isla D, Provencio M, Insa A, Massuti B, Gonzalez-Larriba JL, Paz-Ares L, Bover I, Garcia-Campelo R, Moreno MA, Catot S, Rolfo C, Reguart N, Palmero R, Sanchez JM, Bastus R, Mayo C, Bertran-Alamillo J, Molina MA, Sanchez JJ, Taron M; Spanish Lung Cancer Group. Screening for epidermal growth factor receptor mutations in lung cancer. N Engl J Med 2009; 361: 958-967.

[7] Liu Y, Ren Z, Wang J and Zhang S. Epidermal growth factor receptor-tyrosine kinase inhibitor therapy is especially beneficial to patients with exon 19 deletion compared with exon 21 L858R mutation in non-small-cell lung cancer: systematic review and meta analysis. Thorac Cancer 2016; 7: 406-414.

[8] Furuyama K, Harada T, Iwama E, Shiraishi Y, Okamura K, Ijichi K, Fujii A, Ota K, Wang S, Li H, Takayama K, Giaccone G and Nakanishi Y. Sensitivity and kinase activity of epidermal growth factor receptor (EGFR) exon 19 and others to EGFR-tyrosine kinase inhibitors. Cancer Sci 2013; 104: 584-589.

[9] Koyama N, Watanabe Y, Iwai Y, Kawamura R, Miwa C, Nagai Y, Hagiwara K and Koyama S. Distinct benefit of overall survival between patients with non-small-cell lung cancer harboring EGFR Exon 19 deletion and Exon 21 L858R substitution. Chemotherapy 2017; 62: 151-158.

[10] Ke EE, Zhou Q, Zhang QY, Su J, Chen ZH, Zhang XC, Xu CR, Yang JJ, Tu HY, Yan HH, Zhang YC, Niu FY and Wu YL. A higher proportion of the EGFR T790M mutation may contribute to the

better survival of patients with Exon 19 deletions compared with those with L858R. J Thorac Oncol 2017; 12: 1368-1375.
[11] Shigematsu H, Lin L, Takahashi T, Nomura M, Suzuki M, Wistuba II, Fong KM, Lee H, Toyooka S, Shimizu N, Fujisawa T, Feng Z, Roth JA, Herz J, Minna JD and Gazdar AF. Clinical and biological features associated with epidermal growth factor receptor gene mutations in lung cancers. J Natl Cancer Inst 2005; 97: 339-346.
[12] Cancer IAfRo. WHO classification of tumours of the lung. Pleura, Thymus and Heart 2105.
[13] Goldstraw P, Chansky K, Crowley J, Rami-Porta R, Asamura H, Eberhardt WE, Nicholson AG, Groome P, Mitchell A, Bolejack V; International Association for the Study of Lung Cancer Staging and Prognostic Factors Committee, Advisory Boards, and Participating Institutions; International Association for the Study of Lung Cancer Staging and Prognostic Factors Committee Advisory Boards and Participating Institutions. The IASLC lung cancer staging project: proposals for revision of the TNM stage groupings in the forthcoming (eighth) edition of the TNM classification for lung cancer. J Thorac Oncol 2016; 11: 39-51.
[14] Kosaka T, Yatabe Y, Endoh H, Kuwano H, Takahashi T and Mitsudomi T. Mutations of the epidermal growth factor receptor gene in lung cancer: biological and clinical implications. Cancer Res 2004; 64: 8919-8923.
[15] Paez JG, Janne PA, Lee JC, Tracy S, Greulich H, Gabriel S, Herman P, Kaye FJ, Lindeman N, Boggon TJ, Naoki K, Sasaki H, Fujii Y, Eck MJ, Sellers WR, Johnson BE and Meyerson M. EGFR mutations in lung cancer: correlation with clinical response to gefitinib therapy. Science 2004; 304: 1497-1500.
[16] Jackman DM, Yeap BY, Sequist LV, Lindeman N, Holmes AJ, Joshi VA, Bell DW, Huberman MS, Halmos B, Rabin MS, Haber DA, Lynch TJ, Meyerson M, Johnson BE and Janne PA. Exon 19 deletion mutations of epidermal growth factor receptor are associated with prolonged survival in non-small cell lung cancer patients treated with gefitinib or erlotinib. Clin Cancer Res 2006; 12: 3908-3914.
[17] Riely GJ, Pao W, Pham D, Li AR, Rizvi N, Venkatraman ES, Zakowski MF, Kris MG, Ladanyi M and Miller VA. Clinical course of patients with non-small cell lung cancer and epidermal growth factor receptor exon 19 and exon 21 mutations treated with gefitinib or erlotinib. Clin Cancer Res 2006; 12: 839-844.
[18] Zhang Y, He D, Fang W, Kang S, Chen G, Hong S, Sheng J, Zhan J, Chen N, Hu Z, Xue C, Yang Y, Ma Y, Qin T, Zhou T, Huang Y and Zhang L. The difference of clinical characteristics between patients with Exon 19 Deletion and those with L858R mutation in nonsmall cell lung cancer. Medicine (Baltimore) 2015; 94: e1949.
[19] He Y, Li S, Ren S, Cai W, Li X, Zhao C, Li J, Chen X, Gao G, Li W, Zhou F and Zhou C. Impact of family history of cancer on the incidence of mutation in epidermal growth factor receptor gene in non-small cell lung cancer patients. Lung Cancer 2013; 81: 162-166.
[20] Chen Z, Liu XY, Zhao J, Yang HJ and Teng XD. Correlation of EGFR mutation and histological subtype according to the IASLC/ATS/ERS classification of lung adenocarcinoma. Int J Clin Exp Pathol 2014; 7: 8039-8045.
[21] Song Z, Zhu H, Guo Z, Wu W, Sun W and Zhang Y. Correlation of EGFR mutation and predominant histologic subtype according to the new lung adenocarcinoma classification in Chinese patients. Med Oncol 2013; 30: 645.