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ORIGINAL ARTICLE Lung Cancer

Clinicopathological and molecular epidemiological study of lung cancer patients seen at a tertiary care hospital in Northern India

Randeep Singh, Nitesh Rohtagi[1]

Abstract

Aims: The primary objective of this study was to estimate the clinicopathological and molecular profile of lung cancer patients along with the evaluation of their clinical characteristics at a tertiary care hospital in Northern India. **Subjects and Methods:** A total of 421 patients with lung cancer histology who were treated at Max Super Speciality Hospitals were included in the study. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki and permission was obtained from the Ethics Committee before the start of the study. Clinical characteristics and molecular profiling data were collected from the patient's medical records. **Results:** There were 330 (78.4%) men and 91 (21.6%) women with a median age of 62 years (range: 30–93 years). Of the 421 patients, 388 (92.2%) patients had the nonsmall cell lung cancer (NSCLC) histology whereas 33 (7.8%) patients were of SCLC histology. Histology and gender had a significant association with NSCLC and SCLC ($P < 0.05$). Epidermal growth factor receptor (EGFR) and echinoderm microtubule-associated protein like 4-anaplastic lymphoma kinase (EML4-ALK) fusion gene testing was done in 120 and 93 patients, respectively. Of the 120 patients, 24 (20%) cases were positive for EGFR mutations whereas EML4-ALK fusion gene was present in 8 (8.6%) out of 93 patients. **Conclusions:** Our study confirms the importance of molecular testing in the NSCLC patient subgroup with an aim to identify the exact molecular targets that can benefit from the newer generation of targeted therapies.

Key words: Echinoderm microtubule-associated protein like 4-anaplastic lymphoma kinase, epidermal growth factor receptor, nonsmall cell lung cancer, small cell lung cancer

Introduction

Despite significant advancement in the overall management of lung cancer, it still remains a leading cause of cancer-related deaths worldwide. It is estimated that in the year 2012, 1.8 million new cases of lung cancer have been diagnosed globally whereas 1.6 million people succumbed to this grave disease.[1,2] Lung cancer constituted 12.9% of the total estimated cancer incidence in the year 2012 with majority of the cases (58%) occurring in less developed countries. Hungary, Serbia, and Korea are the leading countries with a lung cancer incidence of 51.6%, 45.6%, and 44.2%, respectively.[2] Globally, more men are reported to have lung cancer as compared to women with a ratio of 2.13:1. While the men-to-women ratio in the less developed WHO region is 2.38:1, the same is 1.83:1 for the more developed WHO region.[2] WHO Western Pacific Region Office accounted for the highest number of lung cancer deaths (48% in men and 45% in women), followed by WHO Europe Region (26% in men and 21% in women), WHO Americas Region Pan American Health Organization (14% in men and 23% in women), and WHO South-East Asia Region (9% each in men and women). WHO East Mediterranean Region and WHO Africa Region accounted for the lowest number of lung cancer deaths: 2% in men and 1% in women and 1% each in men and women, respectively.[1] India has a high mortality of lung cancer, and the incidence for both men and women has increased over the past three decades.[1,3,4] Adenocarcinoma is the most commonly occurring subtype of lung cancer with a very dismal prognosis.[5] Small cell lung cancers (SCLCs) are a very aggressive subtype of lung cancer that comprise approximately 15% of all lung cancer globally, whereas the other 85% are represented by non-SCLC (NSCLC).[1,6] High growth fraction, rapid doubling, and widespread metastasis are the typical hallmarks of SCLC.[7] The overall 5-year survival rate of advanced lung cancer is very poor and remains in the range of 5%–15% only.[8-12] The stage at diagnosis along with the presence of local, regional, or distant metastasis plays a key role in determining the overall 5-year survival rates.[13] Smoking is the major risk factor for lung cancer and is responsible for 80% of its incidences worldwide.[13-18] Hereditary, genetic factors, pollution, exposure to asbestos, and pesticides are some other risk factors for lung cancer.[1,14,20,21] Globally, smoking accounts for 80% and 50% of lung cancer deaths in men and women, respectively. While the prevalence of tobacco smoking has shown a downward trend in the high-income countries, the same has increased significantly in several low- and middle-income countries.[15-22]

The present study was carried out to estimate the clinicopathological and molecular profile of lung cancer patients along with the evaluation of their clinical characteristics at a tertiary care hospital in Northern India.

Subjects and Methods

Patients with lung cancer histology who were treated at Max Super Speciality Hospitals between January 2014 and June 2015 were considered for this study. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki and permission was obtained from the Ethics Committee before the start of the study. Clinical characteristics and molecular profiling data were collected from the patient's medical records. A trained pathologist performed immunohistochemistry staining using antibodies against TTF1, P53, Napsin A, and CK 5/6. Formalin-fixed, paraffin-embedded tissue blocks were used for the mutation analysis. TaqMan-based real-time polymerase chain reaction technique was used for the detection of epidermal growth factor receptor (EGFR) mutation with the help of probes that can anneal specifically to the mutant or wild-type allele. The mutations studied were in-frame deletions in exon 19,

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Department of Medical Oncology, Artemis Hospitals, [1]Department of Medical Oncology, Max Super Speciality Hospital, New Delhi, India
Correspondence to: Dr. Randeep Singh, E-mail: drrandeep@yahoo.co.in

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L858R point mutation in exon 21, and the G719C point mutation in exon 18. Anaplastic lymphoma kinase (ALK) gene rearrangement was detected by fluorescence *in situ* hybridization using the Vysis ALK Break Apart Rearrangement Probe Kit (Abbott Molecular, Abbott Park, IL, USA), which identifies all rearrangements of ALK irrespective of other fusion partners.

Descriptive statistics were used to describe demographic data. Statistical analysis was performed using Pearson's Chi-squared or Fisher's exact test, whichever was appropriate for categorical variables. Logistic regression was performed to compare the study groups. A two-sided $P < 0.05$ was considered statistically significant. Binary logistic regression with single independent variable was performed, and therefore statistical correction has not been applied to the *P* values. Statistical analysis was performed using SAS version 9.3 (SAS Institute Inc.).

Results

A total of 421 lung cancer patients were included in the study. There were 330 (78.4%) men and 91 (21.6%) women with a median age of 62 years (range: 30–93 years). Of the 421 patients, 388 (92.2%) patients had the NSCLC histology, whereas 33 (7.8%) patients were of SCLC histology. With a representation of 52.9%, adenocarcinoma was the most prominent histological subtype. Baseline patient characteristics are presented in Table 1. The site of biopsy at presentation is shown in Table 2, with lung being the primary site of biopsy both for NSCLC as well as SCLC. The association of each of the individual factor with regard to NSCLC and SCLC is shown in Table 3. Histology and gender had a significant association with NSCLC and SCLC ($P < 0.05$). The result of molecular testing is shown in Table 4. EGFR and echinoderm microtubule-associated protein like 4-ALK (EML4-ALK) fusion gene testing was done in 120 and 93 patients, respectively. Of the 120 patients, 24 (20%) cases were positive for EGFR mutations whereas EML4-ALK fusion gene was present in 8 (8.6%) out of 93 patients. The overall incidence of EGFR and EML4-ALK fusion gene in the selected NSCLC subgroup was 6.2% and 2.1%, respectively. The association of each of the individual factor with regard to EGFR and EML4-ALK fusion gene mutation is shown in Tables 5 and 6. While demography, site of disease, and gender were significantly associated with EGFR mutation ($P < 0.05$), the EML4-ALK fusion gene mutation did not show significant association with any of these factors.

Discussion

Being the leading cause of deaths worldwide, lung cancer is being seen as a very dreadful disease with a poor response to chemotherapy regimens.[1] It is estimated that by the year 2035, lung cancer deaths will double both in men and women to reach 3 million as compared to 1.6 million in the year 2012.[2] The higher men-to-women ratio of 3.62:1 and median age of

Table 1: Summary of patient demographic and tumor characteristics (*n*=421)

Characteristics	*n* (%)
Age (years)	
<40	13 (3.1)
40-60	166 (39.4)
>60	242 (57.5)
Median age, years (range)	62 (30-93)
Gender	
Male	330 (78.4)
Female	91 (21.6)
Demography	
Urban	338 (80.3)
Rural	83 (19.7)
Histology	
Adenocarcinoma	223 (52.9)
Squamous cell carcinoma	98 (23.3)
Poorly differentiated	47 (11.2)
Adenosquamous carcinoma	9 (2.2)
Small cell carcinoma	32 (7.6)
Large cell neuroendocrine carcinoma	12 (2.8)

Table 2: Site of biopsy (*n*=421)

Site	*n* (%)
Lung	284 (67.5)
Bronchial mass	57 (13.5)
Pleural fluid	24 (5.7)
Cervical lymph node	21 (4.9)
Liver	7 (1.7)
Bone	6 (1.4)
Brain	6 (1.4)
Mediastinal mass	4 (1.0)
Chest wall	2 (0.5)
Others	10 (2.4)

Table 3: Association of each of the individual factor vis-à-vis nonsmall cell lung cancer and small cell lung cancer (*n*=421)

Variable	Category	NSCLC (*n*=388), *n* (%)	SCLC (*n*=33), *n* (%)	χ^2	*P*
Age (years)	<40	13 (3.1)	0	2.0720	0.354
	40-60	150 (35.6)	16 (3.8)		
	>60	225 (53.4)	17 (4.0)		
Gender	Male	298 (70.8)	32 (7.6)	7.2995	0.006*
	Female	90 (21.4)	1 (0.2)		
Demography	Urban	311 (73.9)	27 (6.4)	0.0531	0.817
	Rural	77 (18.3)	6 (1.4)		
Histology	Adenocarcinoma	222 (52.7)	1 (0.2)	407.2194	0.000*
	Squamous cell carcinoma	98 (23.3)	0		
	Poorly differentiated	47 (11.2)	0		
	Adenosquamous carcinoma	9 (2.1)	0		
	Small cell carcinoma	0	32 (7.6)		
	Large cell neuroendocrine carcinoma	12 (2.9)	0		

*Significant value. NSCLC=Nonsmall cell lung cancer, SCLC=Small cell lung cancer

62 years in our study confirm the previously reported male dominance and the older age group of the disease in the less developed WHO region.[2] The predominance of adenocarcinoma histology (52.9%) among NSCLC patient in our study is also in line with the previously published reports.

A meta-analysis by a collaborative group has established first-line platinum-based chemotherapy regimen as a standard of care for NSCLC.[23] Another meta-analysis reported a superiority of irinotecan and carboplatin combination chemotherapy in extensive disease SCLC as compared to etoposide and cisplatin.[24] However, treatment outcomes with various agents remain poor both in SCLC as well as NSCLC.[23-25] The survival of patients has improved with the use of targeted therapies in the treatment of NSCLC. It is estimated that approximately 50%–60% of NSCLC patients harbor activated pathway with mutations in either KRAS gene, EGFR gene, or the EML4-ALK fusion protein.[26-30] However, various genetic, epigenetic, and phenotypic changes may affect the clinical behaviors of the same stage tumors with varying outcomes among patients.[31,32]

Due to the genetic heterogeneity of lung cancer among various ethnic populations, the importance of molecular characterizations has gained momentum over the past decade. The genetic heterogeneity has been reported not only across different countries but also among different ethnic regions within a country.[20,33-36] The incidence of EGFR mutation in NSCLC has been reported higher (47%) for Asian population as compared to their Caucasian (13%) counterparts.[37] A further higher incidence of EGFR mutation (23.2%–51.8%) in Indian NSCLC patients has been reported by various studies from India.[33-35] Various studies have confirmed that these patients benefit from therapy with EGFR-tyrosine kinase inhibitor (EGFR-TKI). A cumulative meta-analysis of EGFR-TKI s as first-line therapy in metastatic NSCLC has shown a significantly longer progression-free survival (PFS) in patients receiving EGFR-TKI as compared to platinum-based chemotherapy.[38] Similarly, the incidence of ALK gene rearrangement in NSCLC has been reported in the range of 3% to 13% for Caucasian, East Asian, and Indian population.[36,39,40] These patients are likely to benefit from crizotinib therapy.

Table 4: Results of molecular testing

Characteristics	*n* (%)
EGFR mutations (*n*=120)	
Positive	24 (20)
Wild type	96 (80)
EML4 ALK fusion gene (*n*=93)	
Positive	8 (8.6)
Wild type	85 (91.4)

EGFR=Epidermal growth factor receptor, EML4 ALK=Echinoderm microtubule-associated protein like 4-anaplastic lymphoma kinase

Table 5: Association of each of the individual factor vis-à-vis epidermal growth factor receptor mutations (*n*=120)

Variable	Category	EGFR		χ^2	*P*
		Mutated, *n* (%)	Wild type, *n* (%)		
Age (years)	<40	2 (1.6)	3 (2.5)	1.4433	0.485
	40-60	9 (7.5)	34 (28.3)		
	>60	13 (10.8)	59 (49.1)		
Gender	Male	6 (5.0)	72 (60.0)	21.0989	0.0000*
	Female	18 (15.0)	24 (20.0)		
Demography	Urban	24 (20.0)	79 (65.8)	4.9514	0.026*
	Rural	0	17 (14.2)		
Histology	Adenocarcinoma	21 (17.5)	71 (59.2)	8.3967	0.210
	Squamous cell carcinoma	0	8 (6.7)		
	Poorly differentiated	2 (1.6)	9 (7.5)		
	Adenosquamous carcinoma	0	3 (2.5)		
	Small cell carcinoma	0	2 (1.7)		
	Large cell neuroendocrine carcinoma	1 (0.8)	3 (2.5)		

*Significant value. EGFR=Epidermal growth factor receptor

Table 6: Association of each of the individual factor vis-à-vis echinoderm microtubule-associated protein like 4-anaplastic lymphoma kinase fusion gene mutations (*n*=93)

Variable	Category	EML4-ALK fusion gene mutations		χ^2	*P*
		Mutated, *n* (%)	Wild type, *n* (%)		
Age (years)	<40	0	5 (5.4)	2.7192	0.256
	40-60	5 (5.4)	29 (31.2)		
	>60	3 (3.2)	51 (54.8)		
Gender	Male	6 (6.5)	53 (57.0)	0.5042	0.477
	Female	2 (2.2)	32 (34.4)		
Demography	Urban	6 (6.5)	76 (81.7)	1.4561	0.227
	Rural	2 (2.2)	9 (9.7)		
Histology	Adenocarcinoma	7 (7.5)	66 (71.0)	1.9044	0.928
	Squamous cell carcinoma	0	6 (6.5)		
	Poorly differentiated	1 (1.1)	6 (6.5)		
	Adenosquamous carcinoma	0	2 (2.2)		
	Small cell carcinoma	0	1 (1.1)		
	Large cell neuroendocrine carcinoma	0	4 (4.3)		

*Significant value. EML4 ALK= Echinoderm microtubule-associated protein like 4-anaplastic lymphoma kinase

A meta-analysis on the efficacy and safety of crizotinib has shown an extended survival (1-year overall survival of 66.8% and PFS of 8.6 months) and improved response rates (61.2%) in the treatment of ALK-positive NSCLC patients.[41]

A 20% incidence of EGFR mutations in the selected population along with an overall incidence of 6.2% in our study is lower than the previously reported data on Indian and Asian population. However, our study reports a significant association between EGFR mutations vis-à-vis urban demography, female gender dominance, and lung as the primary site of disease. This observation is consistent with the previously published reports of female gender dominance in EGFR mutations.[34,35,36] The 8.6% incidence of EML4-ALK fusion gene mutations in the selected population along with an overall incidence of 2.1% in our study is well within the range of previously published reports across different ethnicities.[36,39-41] Contrary to the previous reports, our study did not show any significant association between the EML4-ALK fusion gene mutations and the female or the younger age group.[36]

Conclusions

Our study reports the clinicopathological and molecular profile of lung cancer patients at a tertiary care hospital in Northern India. Being a retrospective study, it lacked the data on smoking history, treatment outcome as well as overall survival. Since majority of the lung cancers are caused by tobacco smoking, cessation of smoking should be adopted as a strategy for reducing the global prevalence of the disease. Our study confirms the importance of molecular testing in the NSCLC patient subgroup with an aim to identify the exact molecular targets that can benefit from the newer generation of targeted therapies.

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Conflicts of interest

There are no conflicts 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:E359-86.
2. Redirect. Available from: http://www.globocan.iarc.fr. [Last cited on 2016 Aug 11].
3. Stewart BW, Kleihues P, editors. World Cancer Report. Lyon: IARC Press; 2003.
4. Behera D, Balamugesh T. Lung cancer in India. Indian J Chest Dis Allied Sci 2004;46:269-81.
5. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2013. CA Cancer J Clin 2013;63:11-30.
6. Früh M, De Ruysscher D, Popat S, Crinò L, Peters S, Felip E; ESMO Guidelines Working Group. Small-cell lung cancer (SCLC): ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol 2013;24 Suppl 6:vi99-105.
7. Kalemkerian GP, Akerley W, Bogner P, Borghaei H, Chow LQ, Downey RJ, *et al.* Small cell lung cancer. J Natl Compr Canc Netw 2013;11:78-98.
8. Murray N, Coy P, Pater JL, Hodson I, Arnold A, Zee BC, *et al.* Importance of timing for thoracic irradiation in the combined modality treatment of limited-stage small-cell lung cancer. The National Cancer Institute of Canada Clinical Trials Group. J Clin Oncol 1993;11:336-44.
9. Johnson BE, Grayson J, Makuch RW, Linnoila RI, Anderson MJ, Cohen MH, *et al.* Ten-year survival of patients with small-cell lung cancer treated with combination chemotherapy with or without irradiation. J Clin Oncol 1990;8:396-401.
10. Fry WA, Menck HR, Winchester DP. The national cancer data base report on lung cancer. Cancer 1996;77:1947-55.
11. Lassen U, Osterlind K, Hansen M, Dombernowsky P, Bergman B, Hansen HH. Long-term survival in small-cell lung cancer: Posttreatment characteristics in patients surviving 5 to 18+ years – An analysis of 1,714 consecutive patients. J Clin Oncol 1995;13:1215-20.
12. Jemal A, Siegel R, Ward E, Hao Y, Xu J, Murray T, *et al.* Cancer statistics, 2008. CA Cancer J Clin 2008;58:71-96.
13. Ries LA, Eisner MP, Kosary CL, Hankey BF, Miller BA, Clegg L, *et al.*, editors. SEER Cancer Statistics Review, 1975-2002. Bethesda, MD: National Cancer Institute; 2004. Based on November 2004 SEER Data Submission, Posted to the SEER Web Site; 2005. Available from: http://www.seer.cancer.gov/csr/1975_2002/. [Last accessed on 2016 Sep 17]
14. Thankappan KR, Thresia CU. Tobacco use and social status in Kerala. Indian J Med Res 2007;126:300-8.
15. Ezzati M, Lopez AD. Estimates of global mortality attributable to smoking in 2000. Lancet 2003;362:847-52.
16. Osler M. Tobacco control in developing countries. BMJ 2001;322:869.
17. Pesch B, Kendzia B, Gustavsson P, Jöckel KH, Johnen G, Pohlabeln H, *et al.* Cigarette smoking and lung cancer – Relative risk estimates for the major histological types from a pooled analysis of case-control studies. Int J Cancer 2012;131:1210-9.
18. Engeland A, Haldorsen T, Andersen A, Tretli S. The impact of smoking habits on lung cancer risk: 28 years' observation of 26,000 Norwegian men and women. Cancer Causes Control 1996;7:366-76.
19. Moore MA, Ariyaratne Y, Badar F, Bhurgri Y, Datta K, Mathew A, *et al.* Cancer epidemiology in South Asia – Past, present and future. Asian Pac J Cancer Prev 2010;11 Suppl 2:49-66.
20. Kirmani N, Jamil K, Naidu MU. Occupational and environmental carcinogens in epidemiology of lung cancer in South Indian population. Biol Med 2010;2:1-11.
21. Freedman ND, Leitzmann MF, Hollenbeck AR, Schatzkin A, Abnet CC. Cigarette smoking and subsequent risk of lung cancer in men and women: Analysis of a prospective cohort study. Lancet Oncol 2008;9:649-56.
22. Bilano V, Gilmour S, Moffiet T, d'Espaignet ET, Stevens GA, Commar A, *et al.* Global trends and projections for tobacco use, 1990-2025: An analysis of smoking indicators from the WHO comprehensive information systems for tobacco control. Lancet 2015;385:966-76.
23. Chemotherapy in non-small cell lung cancer: A meta-analysis using updated data on individual patients from 52 randomised clinical trials. Non-small Cell Lung Cancer Collaborative Group. BMJ 1995;311:899-909.
24. Chen Y, Chen L, Zhong D, Wang J, Peng L, Feng X. First-line chemotherapy for extensive-disease small cell lung cancer: A network meta-analysis. Zhongguo Fei Ai Za Zhi 2016;19:184-91.
25. Reck M, Heigener DF, Mok T, Soria JC, Rabe KF. Management of non-small-cell lung cancer: Recent developments. Lancet 2013;382:709-19.
26. Okamoto I, Mitsudomi T, Nakagawa K, Fukuoka M. The emerging role of epidermal growth factor receptor (EGFR) inhibitors in first-line treatment for patients with advanced non-small cell lung cancer positive for EGFR mutations. Ther Adv Med Oncol 2010;2:301-7.
27. Tsao AS, Papadimitrakopoulou VA. The future of NSCLC: Molecular profiles guiding treatment decisions. Oncology (Williston Park) 2011;25:607, 614.
28. Zhang X, Zhang S, Yang X, Yang J, Zhou Q, Yin L, *et al.* Fusion of EML4 and ALK is associated with development of lung adenocarcinomas lacking EGFR and KRAS mutations and is correlated with ALK expression. Mol Cancer 2010;9:188.
29. Sequist LV, Heist RS, Shaw AT, Fidias P, Rosovsky R, Temel JS, *et al.* Implementing multiplexed genotyping of non-small-cell lung cancers into routine clinical practice. Ann Oncol 2011;22:2616-24.
30. Kris MG, Johnson BE, Kwiatkowski DJ. Identification of driver mutations in tumor specimens from 1,000 patients with lung adenocarcinoma: the NCI's Lung Cancer Mutation Consortium (LCMC). J Clin Oncol. 2011; 29 (Suppl):Abstr CRA7506.
31. Mok TS, Wu YL, Thongprasert S, Yang CH, Chu DT, Saijo N, *et al.* Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med 2009;361:947-57.
32. Rosell R, Carcereny E, Gervais R, Vergnenegre A, Massuti B, Felip E, *et al.* Erlotinib versus standard chemotherapy as first-line treatment for European patients 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.
33. Sahoo R, Harini VV, Babu VC, Patil Okaly GV, Rao S, Nargund A, *et al.* Screening for EGFR mutations in lung cancer, a report from India. Lung Cancer 2011;73:316-9.
34. Chougule A, Prabhash K, Noronha V, Joshi A, Thavamani A, Chandrani P, *et al.* Frequency of EGFR mutations in 907 lung adenocarcioma patients of Indian ethnicity. PLoS One 2013;8:e76164.
35. Doval DC, Azam S, Batra U, Choudhury KD, Talwar V, Gupta SK, *et al.* Epidermal growth factor receptor mutation in lung adenocarcinoma in

India: A single center study. J Carcinog 2013;12:12.
36. Doval D, Prabhash K, Patil S, Chaturvedi H, Goswami C, Vaid A, *et al.* Clinical and epidemiological study of EGFR mutations and EML4-ALK fusion genes among Indian patients with adenocarcinoma of the lung. Onco Targets Ther 2015;8:117-23.
37. Sekine I, Yamamoto N, Nishio K, Saijo N. Emerging ethnic differences in lung cancer therapy. Br J Cancer 2008;99:1757-62.
38. Normando SR, Cruz FM, Del Giglio A. Cumulative meta-analysis of epidermal growth factor receptor-tyrosine kinase inhibitors as first-line therapy in metastatic non-small-cell lung cancer. Anticancer Drugs 2015;26:995-1003.
39. Soda M, Choi YL, Enomoto M, Takada S, Yamashita Y, Ishikawa S, *et al.* Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer. Nature 2007;448:561-6.
40. Wong DW, Leung EL, So KK, Tam IY, Sihoe AD, Cheng LC, *et al.* The EML4-ALK fusion gene is involved in various histologic types of lung cancers from nonsmokers with wild-type EGFR and KRAS. Cancer 2009;115:1723-33.
41. Qian H, Gao F, Wang H, Ma F. The efficacy and safety of crizotinib in the treatment of anaplastic lymphoma kinase-positive non-small cell lung cancer: A meta-analysis of clinical trials. BMC Cancer 2014;14:683.