Lung Cancer 120 (2018) 82–87

Contents lists available at ScienceDirect

Lung Cancer

journal homepage: www.elsevier.com/locate/lungcan

EGFR-TKIs plus chemotherapy demonstrated superior efficacy than EGFR-TKIs alone as first-line setting in advanced NSCLC patients with EGFR mutation and BIM deletion polymorphism

Sangtian Liu[a,1], Yayi He[a,1], Tao Jiang[a], Shengxiang Ren[a], Fei Zhou[a], Chao Zhao[b], Xuefei Li[b], Jie Zhang[a], Chunxia Su[a], Xiaoxia Chen[a], Weijing Cai[a], Guanghui Gao[a], Wei Li[a], Fengying Wu[a], Jiayu Li[a], Jing Zhao[a], Qiong Hu[a], Mingchuan Zhao[a], Caicun Zhou[a,*], Fred R. Hirsch[c]

[a] Department of Medical Oncology, Shanghai Pulmonary Hospital, Tongji University Medical School Cancer Institute, Tongji University School of Medicine, No. 507 Zhengmin Road, Shanghai, People's Republic of China
[b] Department of Lung Cancer and Immunology, Shanghai Pulmonary Hospital, Tongji University Medical School Cancer Institute, Tongji University School of Medicine, No. 507 Zhengmin Road, Shanghai, People's Republic of China
[c] Division of Medical Oncology, Department of Medicine, University of Colorado Anschutz Medical Campus, Aurora, CO, USA

ARTICLE INFO

Keywords:
BIM deletion polymorphism
Non-small-cell lung cancer
Epidermal growth factor receptor mutation
EGFR tyrosine kinase inhibitor
Chemotherapy

ABSTRACT

Background: Non–small-cell lung cancer (NSCLC) patients with both epidermal growth factor receptor (*EGFR*) positive mutation and B-cell chronic lymphocytic leukemia/lymphoma-like 11 (BIM) deletion polymorphism had a poor clinical response to *EGFR*-tyrosine kinase inhibitors (TKIs). The current study aimed to investigate the clinical efficacy and tolerability of *EGFR*-TKIs plus chemotherapy versus *EGFR*-TKIs alone as first-line treatment in advanced NSCLC patients with *EGFR* mutations and BIM deletion polymorphism.

Methods: A retrospective, non-randomized analysis was conducted. BIM deletion polymorphism was detected using polymerase chain reaction (PCR) analysis and direct sequencing of DNA from peripheral blood cells. Clinical characteristics, overall survival (OS), progress-free-survival (PFS), objective response rate (ORR) and treatment-related adverse events were compared between *EGFR*-TKIs alone versus *EGFR*-TKIs plus chemotherapy group.

Results: 65 patients were enrolled. 36 of them received *EGFR*-TKIs and 29 received *EGFR*-TKIs plus chemotherapy. *EGFR*-TKIs plus chemotherapy had significantly higher ORR than TKIs alone (65.5% vs. 38.9%, $P = 0.046$). Median PFS was significantly longer in *EGFR*-TKIs plus chemotherapy group than in TKIs group (7.2 vs 4.7 m; $P = 0.008$). Median OS was numerically longer in *EGFR*-TKIs plus chemotherapy group than in TKIs alone (18.5 vs 14.2 m; $P = 0.107$). *EGFR*-TKIs plus chemotherapy was associated with more grade 3 or 4 hematological toxic effects than *EGFR*-TKIs alone.

Conclusion: *EGFR*-TKIs plus chemotherapy conferred a significantly higher ORR, prolonged PFS and numerically longer OS in advanced NSCLC patients with *EGFR* mutation and BIM deletion polymorphism. Further prospective studies are needed to validate these findings.

1. Introduction

The epidermal growth factor receptor (*EGFR)*-tyrosine kinase inhibitors (*EGFR*-TKIs), such as gefitinib, erlotinib and osimertinib, are recommended as the first-line therapy in advanced *EGFR* mutation-positive NSCLC patients [1–12]. However, approximately 20%–30% of NSCLC patients with activated *EGFR* mutation show primary resistance to *EGFR*-TKIs [13]. One intrinsic resistance mechanism was B-cell chronic lymphocytic leukemia/lymphoma-like 11 (BIM) deletion polymorphism [14].

BIM, also called BCL2L11, is a proapoptotic protein and a member of the B-cell chronic lymphocytic leukemia/lymphoma 2 (Bcl-2) families of proteins, binding to the pro-survival Bcl-2 family members with high affinity and directly activating proapoptotic BAX and BAK to promote cell death [15–17]. Several previous studies have demonstrated that BIM was a key modulator of apoptosis triggered by *EGFR*-

* Corresponding author at: Department of Medical Oncology, Shanghai Pulmonary Hospital, Thoracic Cancer Institute, Tongji University School of Medicine, No. 507, Zheng Min Road, Shanghai, 200433, People's Republic of China.
E-mail addresses: caicunzhou_dr@163.com, tonyjiangdr@163.com (C. Zhou).
[1] Sangtian Liu and Yayi He contributed equally to the article.

https://doi.org/10.1016/j.lungcan.2018.04.004
Received 10 January 2018; Received in revised form 24 February 2018; Accepted 3 April 2018

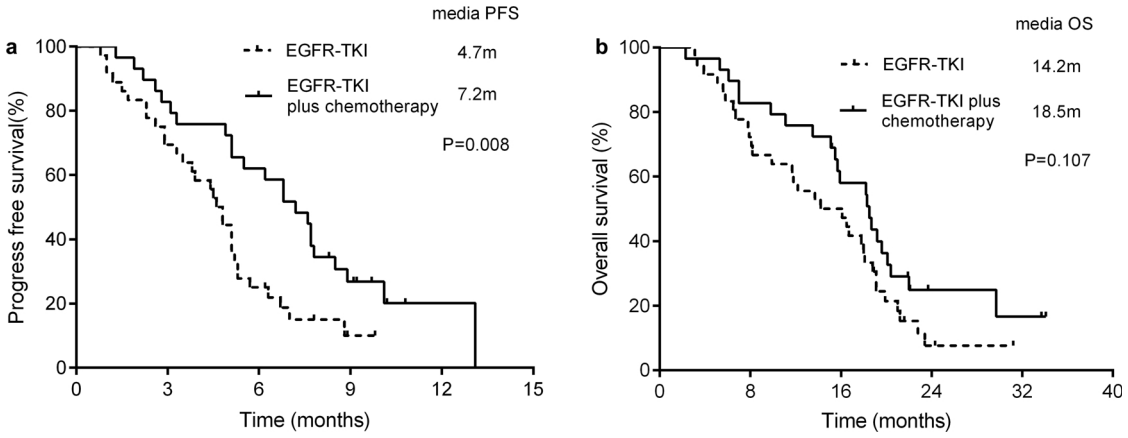

Fig 1. Survival curves for patients with different treatments. (a) progress-free survival outcomes; (b) overall survival outcomes; *EGFR*-TKI, epidermal growth factor receptor tyrosine kinase inhibitor; PFS, progress-free survival; OS, overall survival.

TKIs [18,19]. BIM deletion polymorphism, a germline alteration, was reported about 12.8% in NSCLC and was thought to be associated with intrinsic resistance to *EGFR*-TKIs [20–22]. In a study that screened 2597 healthy individuals, BIM deletion polymorphism was present in 12.3% of East Asians but absent in Africans and Europeans [14]. *EGFR*-mutant NSCLC cells with the BIM deletion polymorphism showed impaired generation of BIM with the proapoptotic BH3 domain, as well as resistance to *EGFR*-TKIs induced apoptosis [14,15,23]. JY Ma et al. reported that BIM deletion polymorphism was significantly associated with a poor response to *EGFR*-TKIs therapy in *EGFR* mutation positive NSCLC patients [24]. Another study also demonstrated that decreased BIM expression level inhibited the apoptotic response and tumor shrinkage induced by *EGFR*-TKIs therapy [22].

Our previous study found the correlation between BIM deletion polymorphism and poor response to *EGFR*-TKIs in patients with activating *EGFR* mutations [20]. Therefore, to find out a better clinical treatment strategy, we retrospectively compared *EGFR*-TKIs alone versus *EGFR*-TKIs plus chemotherapy as the first-line treatment for NSCLC patients harboring both activating *EGFR* mutations and BIM deletion polymorphism.

2. Patients and methods

2.1. Patients

This retrospective, non-randomized analysis had screened all NSCLC patients treated in Shanghai Pulmonary Hospital from June 2014 to September 2016, and only those harboring both mutated *EGFR* and BIM deletion polymorphism and their therapies were either oral *EGFR*-TKIs alone or oral *EGFR*-TKIs plus intravenous chemotherapy with cisplatin or carboplatin plus pemetrexed, docetaxel, paclitaxel or gemcitabine were enrolled. All involved patients met the following inclusion criteria: they were aged ≥18 years; they had histologically or pathologically confirmed stage IIIB or IV NSCLC and they harbored *EGFR* positive-mutation (exon 19 deletion or 21 L858R mutation) and BIM deletion polymorphism. Before starting any treatment, a complete medical history interview, physical examination, laboratory tests, and radiology examinations were carried out for each patient. Patients received *EGFR*-TKIs (gefitinib/erlotinib) or *EGFR*-TKIs together with up to 4–6 cycles of platinum-based chemotherapy until disease progression or unacceptable toxic effects. The primary endpoint was median progress-free-survival (PFS); the second endpoint included overall survival (OS), objective response rate (ORR) and treatment-emergent adverse events (TEAEs). The study was approved by the Ethics Committees of Shanghai Pulmonary Hospital Affiliated with Tongji University and was carried out in accordance with the World Medical Association's Declaration of Helsinki.

2.2. EGFR mutation detection and genotyping of the BIM deletion polymorphism

Methods to detect *EGFR* mutation and genotyping of the BIM deletion polymorphism are same as described in our previous studies [20,25]. *EGFR* detection was performed in tumor samples and BIM status was detected by peripheral blood cells. Briefly, DNA from tumor tissue was extracted using the DNeasy Blood and Tissue Kit or the QIAamp DNA FFPE Tissue Kit (both from Qiagen, Hilden, Germany) according to the manufacturer's protocol. The amplification refractory mutation system (ARMS) was used to detect *EGFR* mutations using the Human *EGFR* Gene Mutations Fluorescence Polymerase Chain Reaction Diagnostic Kit (AmoyDx, Xiamen, China). All blood samples were amplified by polymerase chain reaction (PCR) to detect BIM deletion polymorphisms using the following primer sequences: wild-type (WT) BIM forward primer, 5 0-ACTGTAAAACGACGGCCAGTCCTCATGATG AAGGCTAACTCAA-3 0; and reverse primer, 5 0-ACCAGGAAACAGCT ATGACCAACCTCTGACAAGTGACCACCA-3 0. For the BIM deletion polymorphism, the forward primer sequence was the same as that used for WT BIM, and the reverse sequence was 5 0 – ACCAGGAAACAGCT ATGACCGGCACAGCCTCTATGGAGAACA-3 0 (for details, see Fig. 1). The reaction condition was 95 °C for 10 min followed by 40 cycles at 94 °C for 30 s, 60 °C for 30 s, and 72 °C for 30 s; and a final extension at 72 °C for 10 min using the ExTaq Polymerase premix PCR Kit (TaKaRaBio Inc., Shiga, Japan). PCR products (173 base pairs for the BIM deletion polymorphism and 216 bp for WT BIM) were then separated on a 3% agarose gel with nucleic acid dye by electrophoresis and were purified before direct sequencing. Details can be provided in our previous relevant papers [26,27].

2.3. Data collection and the evaluation of clinical efficacy

We retrospectively analyzed the clinical characteristics, OS, PFS, ORR, TEAEs and multivariate cox regression analysis of PFS and OS in *EGFR*-TKIs with and without chemotherapy. TEAEs were reported according to the medical dictionary for regulatory activities (version 18.0) and were assessed according to National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.0. The PFS, OS and ORR were assessed according to the Response Evaluation Criteria in Solid Tumors (version 1.1).

2.4. Statistical analysis

SPSS statistical software (SPSS 22.0) was used to analyze the data. Characteristics were compared by the Chi-square test or Fisher exact test. PFS and OS were calculated by the Kaplan-Meier method. The survival figures were draw by Graphpad prism 7.03. Independent