ALK-rearranged lung adenocarcinoma patient with development of severe sinus bradycardia after treatment with crizotinib

A case report

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**Abstract**

**Rationale:** The anaplastic lymphoma kinase (ALK) rearrangements represent a subtype of non-small-cell lung cancer (NSCLC), and targeting ALK has radically changed the treatment of NSCLC. Crizotinib, as an ALK inhibitor, has been used in the treatment of ALK-rearranged NSCLC for several years and some adverse effects should be given attention.

**Patient concerns:** A 64-year-old woman with a no-smoking history visited hospital in November 2016 because of a persistent cough, expectoration, and bone metastases. After receiving chemotherapy for nearly 1 year, she showed progressive disease. DNA-sequencing identified an intergenic ALK rearrangement. Surprisingly, RNA-sequencing revealed the EML4-ALK fusion transcript. Subsequently, this patient switched to crizotinib therapy.

**Outcomes:** The patient achieved partial response after 1-month treatment. However, this patient suffered a severe sinus bradycardia after 4 months of treatment. When reducing the dose of crizotinib, the side effect was alleviated and this patient showed stable disease until now.

**Lessons:** Given that the severe sinus bradycardia was an unusual adverse effect, physicians should be aware of these side effects when using crizotinib. Moreover, it should be noted that this patient harbored an intergenic ALK rearrangement identified by DNA-sequencing, but EML4-ALK fusion transcript verified by RNA-sequencing. However, the mechanism remains unknown and requires further research.

**Abbreviations:** ALK = anaplastic lymphoma kinase, CT = computed tomography, ECT = emission computed tomography, HR = heart rate, LADC = lung adenocarcinoma, NSCLC = non-small-cell lung cancer, PD = progressive disease, PR = partial response, SD = stable disease, TK = tyrosine kinase.

**Keywords:** crizotinib, EML4-ALK fusion, intergenic ALK rearrangement, lung adenocarcinoma, sinus bradycardia

1. Introduction

Lung cancer is the leading cause of tumor-related deaths in the world. Non-small-cell lung cancer (NSCLC) accounts for approximately 85% of lung cancer, and adenocarcinoma is the most common histological subtype, which accounts for nearly 40% of all lung cancer cases. For certain NSCLC patients, targeted therapy has transformed treatment and improved outcomes. More importantly, the identification of genetic driver alterations, including gene mutation, rearrangement, or amplification, has developed novel potential targets for targeted therapy.

As a transmembrane receptor tyrosine kinase, anaplastic lymphoma kinase (ALK) belongs to the insulin receptor superfamily and ALK rearrangement has been identified in 5% to 6% NSCLC patients. Although increasing evidence demonstrated association of activated ALK with tumorigenesis in these rare tumors, it can be said that the current enthusiasm for ALK as a target for cancer therapy is largely due to the recent recurrence of ALK gene translocations in a significant subset of NSCLC patients. The most common ALK rearrangement in NSCLC is EML4-ALK which can be targeted by the tyrosine kinase inhibitor crizotinib.

As the first ALK tyrosine kinase inhibitor, crizotinib was approved by FDA in 2011 for ALK-rearranged NSCLCs and had achieved remarkable response in a series of clinical trials. The PROFLE 1007 trial was the first phase III trial comparing crizotinib to standard second-line chemotherapy in patients with ALK-positive lung cancer, and showed a higher objective response rate (ORR) (65% vs 20%).
study reported higher response rate (74% vs 45%) to crizotinib than standard first-line chemotherapy in previously untreated advanced ALK-positive NSCLC.\cite{6}

However, even though crizotinib has been applied to treat ALK-positive NSCLC patients for several years, there are still some adverse effects that should be paid attention. Decreases in heart rate (HR) and development of sinus bradycardia have been observed with crizotinib.\cite{7,8} Here we report a case of ALK rearrangement lung adenocarcinoma achieving partial response to crizotinib treatment but with the development of sinus bradycardia. It should be noted that DNA-sequencing identified an intergenic ALK rearrangement, whereas RNA-sequencing revealed EML4-ALK fusion transcript in this patient.

2. Case report

A 64-year-old woman with a no-smoking history visited another hospital in November 2016 because of a persistent cough.

Figure 1. Computed tomography (CT) scans. (A) Baseline of chest CT scans revealed a mass in the inferior lobe of left lung before crizotinib therapy; (B) chest CT scans showed partial response after treatment with crizotinib therapy for one month; (C) chest CT scans showed stable disease after treatment with decreased dose of crizotinib.
expectoration, and progressive dysphagia for 2 months. Clinical cytologic diagnosis of pleural effusions and basal segment mucosal biopsy of the left lower lobe revealed a primary lung adenocarcinoma (LADC). Abnormal bone metabolism in the lower scapula showed by skeletal emission computed tomography (ECT) scan and C6 vertebral abnormal signal showed by cervical vertebra MRI were suggested pleural and bone metastases in this patient. Immunohistochemical stainings were positive for TTF-1, CK7, and Ki67, and negative for P40. This patient was initially received 4 cycles chemotherapy (pemetrexed (J) 500mg/m^2, d1+carboplatin AUC=5, d1, q21d) from November 2016 to January 2017, and achieved stable disease (SD) after chemotherapy. Subsequently, she adopted 11 cycles of pemetrexed (500mg/m^2, q21d) single-agent maintenance chemotherapy from February 2017 to September 2017, and showed progressive disease (PD) in October 2017.

For further treatment, she was presented to our hospital in November 2017. Electrocardiogram (ECG) showed a sinus arrhythmia with heart rate (HR) of 85 bpm. Computed tomography (CT) scan revealed a mass about 5.8 cm × 7.2 cm × 6.1 cm in the left lung lobe oppressing inferior lobar bronchus (Fig. 1A), accompanied by mediastinal and hilar multiple lymph node metastasis, pleural metastasis with pleural effusion, and multiple liver metastases. Skeletal ECT scan showed active bone metabolism, suggesting possibility of a bone metastasis. LADC was confirmed by transthoracic needle biopsy (Fig. 2A). Immunohistochemical stainings were positive for NapsinA, Ki67, and Ventana ALK (D5F3) (Fig. 2B–D), and negative for CK5/6 and P40. The patient’s clinical stage was finally determined as T4N3M1 (stage IVb).

Next-generation DNA sequencing identified an intergenic ALK rearrangement, generated by rearrangement of intergenic region of chromosome 2p22.2 to the exons 20 to 29 of ALK (Fig. 3A), and the ALK breakpoint located in intron 19. To our surprise, RNA-sequencing revealed that the RNA transcript was EML4-ALK fusion (E6; A20) (Fig. 3B), containing a 35-bp intron which was retained from exon6 of EML4. In December 2017, the patient received crizotinib therapy (250mg, bid). After 1 month, ECG revealed a sinus arrhythmia with heart rate (HR) of 59 bpm (Fig. 4), and CT scan showed a shrunken tumor in inferior lobe of left lung and alleviative compression in inferior lobar bronchus (Fig. 1B), suggesting a partial response (PR). The patient then continued oral crizotinib (250mg, bid), and the efficacy was evaluated as SD in April 2018. ECG showed a sinus bradycardia (34 bpm) (Fig. 4), transient sinus arrest, junctional escape beat, and atrial premature beats, which may be related to side effects of crizotinib. She withdrew crizotinib therapy for 11 days and these side effects were disappeared. Considering the high costing, the patient refused to cardiac pacemaker implantation therapy. From May 2018, crizotinib was adjusted to 250mg qd for safety.
consideration. Thereafter, the patient’s ECG remained sinus arrhythmia and atrial premature beats, and the efficacy was evaluated as SD in June 2018 and August 2018 (Fig. 1C). The patient had no symptoms such as dizziness, chest distress, and shortness of breath. This study was approved by the ethics committee of Affiliated Tumor Hospital of Guangxi Medical University. Patient had provided informed consent for publication of the case.

3. Discussion

ALK is a transmembrane tyrosine kinase (TK) receptor that belongs to insulin receptor superfamily. ALK rearrangements generate an oncogenic ALK tyrosine kinase that leads to the activation of many downstream signaling pathways involving in cell proliferation and survival. More than 19 different ALK fusion partners, including EML4, KIF5B, KLC1, and TPR, have been identified in NSCLC. The EML4-ALK fusion was first discovered from a 62-year-old man with lung adenocarcinoma. The EML4-ALK fusion is the result of a chromosome rearrangement between the N-terminal portion of the EML4 gene and TK domain of the ALK gene, both of which are located in the chromosome 2p and have opposite orientations. EML4-ALK fusion can result in constitutively active ALK kinase domain and involve in oncogenic pathway in lung cancer.

In this report, we experienced a rare case of the patient who had an intergenic ALK rearrangement identified by DNA-sequencing. This ALK rearrangement was generated by the rearrangement of intergenic region of chromosome 2p22.2 to the exons 20 to 29 of ALK, retaining the ALK tyrosine kinase domain. The breakpoint located in intron 19. However, RNA-sequencing showed the EML4-ALK fusion transcript, containing a 35-bp intron which was retained from exon6 of EML4. The breakpoint also located in intron 19. To our knowledge, this is the first case with different ALK rearrangement types identified by DNA- and RNA-sequencing. However, regardless of the rearrangement types, all chimeras retain the ALK gene kinase domain responsible for the constitutive activation of ALK signaling pathways.
ALK rearrangements are sensitive to crizotinib treatment in patients with NSCLC.\textsuperscript{[12]} Crizotinib is a potent, selective, ATP-competitive ALK inhibitor. As the first-line therapy, it was demonstrated a high objective response rate of crizotinib in patients with ALK-positive NSCLC.\textsuperscript{[6]} In this case, the patient achieved a partial response to crizotinib therapy. However, crizotinib is involved in prolongation of the QT interval and sinus bradycardia. It was reported that HR decreasing is a pharmacodynamic effect of crizotinib, with an average reduction of 2.5 bpm per increasing 100 ng/mL crizotinib.\textsuperscript{[13]} In this report, the HR decreased significantly after crizotinib treatment for 1 month, and the ECG monitoring revealed the HR was 59 bpm. After 4 months of treatment, this case developed side effect with sinus bradycardia (34 bpm), transient sinus arrest, junctional escape beat, and atrial premature beats. While withdrawing crizotinib therapy for 11 days, these side effects were disappeared. Subsequently, the patient remained sinus arrhythmia and atrial premature beats when reducing the dose of crizotinib, and the efficacy was evaluated as SD. However, the mechanism that allows crizotinib to slow down HR remains unknown.

We report a rare case of adenocarcinoma harboring an intergenic ALK rearrangement identified by DNA-sequencing, but EML4-ALK fusion transcript verified by RNA-sequencing. This patient showed a partial response to crizotinib, but side effects with sinus bradycardia emerged. Therefore, when treating ALK-positive NSCLC patients with crizotinib, physicians should be aware of these severe adverse effects.

**Author contributions**

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