Author’s response to reviews

Title: A case of synchronous bilateral lung cancers: EML4-ALK positive adenocarcinoma in the right lung and adenocarcinoma in situ (the former bronchioloalveolar carcinoma) in the left lung

Authors:

Ikuo Matsuda (matsudai@hyo-med.ac.jp)
Kengo Takeuchi (kentakeuchi0416@gmail.com)
Shinjiro Mizuguchi (shinjiro123@gmail.com)
Masahide Kaji (a195108@ych.or.jp)
Kayo Ueda (a211144@ych.or.jp)
Kazuhiro Teramura (a103123@ych.or.jp)
Seiichi Hirota (hiros@hyo-med.ac.jp)

Version: 3 Date: 12 March 2013

Author’s response to reviews:

Seiichi Hirota, M.D.
Department of Surgical Pathology
Hyogo College of Medicine
1-1 Mukogawa-cho, Nishinomiya, Hyogo 663-8501
Japan
March 12, 2013
Dear Editor,

We have revised our manuscript (MS: 1935382037859010) according to the Reviewer’s suggestions. Point-by-point responses to the reviewer’s comments are provided below. Changed portions are highlighted in red in our revised manuscript. I hope that our revised manuscript is suitable for publication in the BMC pulmonary medicine with your editorial helps.

All correspondence is directed to:
Seiichi Hirota, M.D.
Department of Surgical Pathology
Hyogo College of Medicine
1-1 Mukogawa-cho, Nishinomiya, Hyogo 663-8501
Japan
FAX: +81-798-45-6671, TEL: +81-798-45-6667
E-mail: hiros@hyo-med.ac.jp
Sincerely yours,
Seiichi Hirota, M.D.

Response to reviewers’ comments

# Responses to Dr. Milica Kontic:

1) Regarding to the follow-up of the patient after the surgery, we added following description as the last paragraph in Case presentation section, which is highlighted in red in the revised manuscript: “As an adjuvant therapy, the patient has taken orally tegafur-uracil 300 mg/day since 3 months after the resection operation. Until now, she has not shown any signs of relapse or adverse effects. Eleven months after the operation, neither chest CT, nor bone scintigraphy, nor brain magnetic resonance imaging did show any signs of relapse or metastasis.”

2) Regarding to the gene expression profiling of the patient’s tumor, we added following sentences into the end of the third paragraph in Discussion and conclusion section, which is highlighted in red in the revised manuscript: “The right tumor and the left one had lepidic growth pattern in common, although the former was EML4-ALK positive, while the latter negative. The relationship between genetic alterations and histology is intriguing and it will be interesting to compare gene expression profiling of both tumors.”

# Responses to Dr. Aurelie Cazes:

1) Regarding to how the patient will be treated if any metastasis occurs, we replaced the fourth paragraph in Discussion and conclusion section with following sentences, which is highlighted in red in the revised manuscript: “The fact that the tumor of the right lung and that of the left lung harbor different genetic alterations will be useful for the follow-up of this patient. The adenocarcinoma in situ of the left lung was 15x10mm of size and pathologically at stage 0 (TisN0M0). Therefore, the possibility of presenting metastasis in the following five years is as low as nearly 0%. On the other hand, the EML4-ALK-positive tumor of the right lung was an invasive cancer. Thus, if any metastasis or relapse occurs in the future in this patient, it is more likely that the relapse derives from EML4-ALK-positive cancer of the right lung. After the confirmation that the metastasis or relapse harbor EML4-ALK translocation, ALK inhibitor such as crizotinib will be the first choice.”

2) Regarding to the comment that all the ALK-rearranged adenocarcinomas are not of the mucinous cribriform histology, and that screening for ALK fusion should not be restricted to such histology, to avoid misunderstanding as you suggested, we revised the latter half of the fifth paragraph in Discussion and conclusion section as follows, which is highlighted in red in the revised manuscript: “It was reported that these kinase gene fusion-positive lung adenocarcinomas have some histological correlates or surrogates including mucinous cribriform pattern [7]. However, as our case report illustrated, kinase gene fusion-positive lung adenocarcinomas may show different histology other than mucinous cribriform
pattern. Not only tumor histology, but also patients' sex, age, smoking habit, and so on, should be considered to suspect the involvement of kinase gene fusion in lung cancers. Although rarer than lung cancer with EGFR mutation, the identification of kinase gene fusions, including EML4-ALK, in lung cancer leads to molecularly-targeted therapy with kinase inhibitors. The identification has important implication for tractable therapy and predictable prognosis."

3) Regarding to the comments on tumor heterogeneity in lung cancers both in primary and in primary versus metastasis, and the heterogeneity in ALK staining, we revised the third paragraph in Case presentation section as follows, the revision of which is highlighted red in the revised manuscript: “Microscopic examination of the resected right lower lobe revealed a heterogenous adenocarcinoma composed of a mucinous cribriform tumor (Figure 2a) and lepidic growth (adenocarcinoma in situ) pattern (Figure 2c). At low-power microscopy, these two components were seen adjacent to each other (data not shown). As described above, the mucinous cribriform histology of the HE stained specimen made us suspicious of EML4-ALK positive lung adenocarcinoma [5-7]. To examine this possibility, immunohistochemical analysis was performed. At low-power microscopy, the tumor was homogeneously stained positive for ALK (data not shown). At high-power microscopy, the tumor cells were positively stained for ALK (Figures 2b, 2d). Fusion and split FISH analyses using probes for EML4 and ALK genes confirmed that the adenocarcinoma of the right lung was indeed EML4-ALK positive (Figures 2g-2j).”

4) Regarding to the comment on use of the term 'bronchioloalveolar carcinoma', we replaced all of the term 'bronchioloalveolar carcinoma' with 'adenocarcinoma in situ', which is highlighted in red in the revised manuscript.

5) Regarding to the comment on the sex ratio in EML4-ALK positive lung adenocarcinoma, we removed the word ‘female’ from the second paragraph in Background section, which is highlighted in red in the revised manuscript.

6) Regarding to the possibility of EGFR mutations in the EML4-ALK positive adenocarcinoma of the right lung, we confirmed by genomic sequence analysis that the EML4-ALK positive adenocarcinoma has no mutations in exons 18, 19, 20, 21 of EGFR. According to your suggestions, we added following sentences into the end of the third paragraph in Case presentation section , which is highlighted in red in the revised manuscript: “We confirmed that the EML4-ALK positive adenocarcinoma has no mutations in exons 18, 19, 20, 21 of EGFR gene (data not shown).”