

## Peer Review File

Article information: <https://dx.doi.org/10.21037/jtd-22-1783>

### Reviewer A

This is a very interesting study and it is important to better understand the efficacy of different ALK TKI combinations. Given the retrospective nature of the study, the evaluation of efficacy is challenging and often prone to bias. This should be considered in more depth to avoid quick and possibly wrong conclusions.

#### Overall comments:

- Usually, patients that start on 2nd generation ALK are still treated with another 2nd gen ALK TKI or 3rd gen ALK gen afterwards. Therefore, the comparison of PFS between the groups likely introduces bias. PFS for the sequential group is the sum of PFS under two treatments, whereas PFS for the direct 2nd generation group is the PFS from only this treatment if I understand the analysis correctly.
- Moreover, immortal time bias may be relevant to consider here when retrospectively identified groups with two treatments are compared with groups with only one treatment (since all patients in the group with two treatments must have lived to receive two treatments).
- It would be good to understand if there has been a trend over time towards direct 2nd gen ALK TKI treatment. If that is the case patients in the sequential group are generally from earlier years in the study period and patients in the direct 2nd gen group are from later years. This would then affect follow-up and could bias the results.
- The conclusions should be phrased more carefully given the possibility of bias due to the study design.

**Reply 1:** Thanks for your helpful suggestion. Your understanding is correct. In the second-generation drug treatment group, we analyzed the PFS of patients who received second-generation drugs for the first time and did not analyze their subsequent treatment. All patients in the group with two treatments must have lived to receive two treatments. The article does have the bias you mentioned, which I have added to the limitation section of the discussion.

#### Comment 2: Abstract

- the difference in patient groups should be described in the methods section

**Reply 2:** Thank you for your comment. We describe the differences between the two groups in the methods section, as detailed in the page 2, line 31-32.

**Changes in the text:** Page 2, line 31-32.

#### Comment 3:

- Are PFS, OS measures medians? Please specify.

**Reply 3:** Thank you for your comment. The PFS, OS, and CNS TTP measures medians.

**Changes in the text:** Page 2, line 33.

**Comment 4:**

- To state that PFS and OS are similar is not correct, the difference between point estimates is substantial although not statistically significant. This could, however, be due to sample size. A high p-value means you cannot reject the hypothesis that the groups are equal, it does not mean they are actually similar.

- Thus, the conclusion seems not correct, there are substantial differences in the point estimates, however not statistically significant

**Reply 4:** Thank you for your comment. We have revised the conclusion.

**Changes in the text:** Page 2, line 34-37.

**Comment 5:** Statistical analysis and follow-up:

- It would be good to include information on the median follow-up period per patient group, since differences in follow-up could affect PFS and OS results

**Reply 5:** Thank you for your comment. We increased the median follow-up period per patient group at page 6, line 130-131.

**Changes in the text:** Page 6, line 130-131

**Comment 6:**

Discussion:

- The limitation section should be developed further (see overall comments above)

**Reply 6:** Thanks for your helpful suggestion. We have supplemented the limitations of the article.

**Changes in the text:** Page 14, line 299

**Reviewer B**

The authors reported that there were no significant differences in terms of PFS and OS between the effect crizotinib followed by second generation ALK TKI and direct therapy of second generation ALK-TKI for the patients with lung cancer harboring ALK rearrangement. The authors also showed that brain metastasis and PS were independent prognostic factors of PFS and OS. Although the manuscript included the retrospective data of relatively large number of patients in a single institution, some vague points and several concerns in the manuscript.

Major concerns.

**Comment 1:** In lines 84-90 and 134-149, only 115 patients are enrolled in this study although total 556 lung cancer patients harboring ALK gene rearrangements received targeted therapy. The study flow diagram will clarify this difference, e.g., the number of patients who received only crizotinib or third generation ALK-TKIs.

**Reply 1:** Thank you for your comment. We added the flow diagram in Fig. 1.

Changes in the text: Fig. 1 (in Page 4 line 86).

**Comment 2:** In lines 154-155 and 168-170, the authors need to clarify which the drug responses were shown in sequential group. Crizotinib or the second generation TKI?

**Reply 2:** Thank you for your comment. In lines 154-155 and 168-170, the drug responses of crizotinib were shown in sequential group.

**Changes in the text:** Page 7 line 153, page 8 line 168.

**Comment 3:** Fig 3 may make readers confused, the reviewer recommends to show the results of the same analysis in both sequential group and direct second line group respectively, as well as that of the total population.

**Reply 3:** Thank you for your comment. We have supplemented the subgroup analysis by adding a new Fig. 4.

**Changes in the text:** Page 10 line 210, Fig.4.

**Comment 4:** The authors should mention what they want to tell with these data. Do authors recommend to use second generation ALK-TKIs or crizotinib in first line treatment? Please clarify.

**Reply 4:** Thanks for your helpful suggestion. Combined with previous studies, second-generation ALK TKIs have lower toxicity and higher intracranial efficacy than crizotinib, so second-generation ALK TKIs may be a better choice than first-generation ALK TKIs.

**Changes in the text:** Page 14 line 294.

Minor concerns.

**Comment 5:** Gene names should be written in italic.

**Reply 5:** Thanks for your careful review. We have modified our text as advised

**Comment 6:** Please unify the writing style of 'ALK inhibitor' and 'ALK-TKI'

**Reply 6:** Thanks for your helpful suggestion. We have a unified style of writing ALK TKI.

**Changes in the text:** the writing style of ALK TKI.