Peer Review File

Article information: https://dx.doi.org/10.21037/tlcr-23-691

Reviewer A

In this real-world retrospective analysis, the authors showed that low-dose afatinib may provide a survival benefit over 40 mg initiation in EGFR-positive NSCLC. As the authors argue, afatinib at 40 mg/day is more toxic in Asian patients, and dose-reduction strategies are commonly employed in practice. Although EGFR-TKI options vary slightly by country and region, and regional health economic circumstances must be taken into account, This interesting study could be of benefit to all thoracic oncologists prescribing afatinib. However, some points should be taken care of before publication.

General response: We would like to thank Reviewer A for his time and valuable comments and have attempted to address all the comments made.

Comment 1: The reason afatinib was started at a reduced dose is clearly stated in Table 2. If data are available, please clarify the differences in patient characteristics between the afatinib 40 mg start group and the less than 40 mg group. There may be residual bias in starting dose selection other than body size.
Reply 1: We have added the comparison between the starting dose of 40 mg and less than 40 mg dose in new Table 2. In the results section, we have added text to describe this difference (see Page 10, line 168-169).
Changes in the text: The afatinib 40 mg OD starting dose group was significantly younger and had more patients with symptomatic brain metastases than the lower dose group (Table 2).

Comment 2: In Table 4 and 5, the explanatory variables entered into the multivariate models need to be clearly stated.
Reply 2: We have identified the covariates entered into the multivariate analysis in the section on Statistical Analysis (see Page 10, line 155).
Changes in the text: Multivariate analysis was performed using binary logistic regression that included covariates that had been shown to significantly affect treatment outcome in previous studies, such as EGFR subtype, symptomatic brain metastases and ECOG performance status (15, 17,19,20).

Comment 3: Low-dose afatinib was superior in ORR, but no statistically significant difference in TTF was detected. This point needs to be specifically discussed. Is it really appropriate to strongly assert the efficacy of low-dose afatinib with this result?
Reply 3: We have rephrased the sentence on TTF results in the abstract (see Page 4, line 70-71) and results section (see Page 13, line 224-227). We have also added text in the limitation section (see Page 17, line 314-316).
Changes in the text:

- These patients had a numerically but not statistically longer median time to treatment failure (TTF).
- Patients on afatinib dose lower than 40 mg OD had a numerically but not statistically longer median TTF (12.50 months [95% CI 10.44–14.56] months) than patients on 40 mg OD (9.80 [95% CI 5.72–13.88] months) (HR: 0.76, (95% CI 0.49 – 1.20), p=0.238) (Figure 3).
- Inadequate sample size of patients on afatinib 40 mg OD could have led to the lack of statistically significant difference for TTF.

Reviewer B

The authors have demonstrated that the use of a lower dose of afatinib has better response rates when compared to the standard 40 mg dose in patients with in real-world clinical practice. Consequently, they recommend a reduced afatinib starting dose for Asian patients. While this finding appears to be relatively clear, there are a few assertions that raise questions. The current manuscript, in its present state, does not meet the standards warranting publication. Therefore, I do not recommend the publication of their manuscript.

General response: We would like to thank Reviewer B for his comments and with the corrections made, we hope that it now meets the standard for publication.

Major comments

Comment 1: The authors recommend starting treatment with a low dose of afatinib for Asian patients. However, there is uncertainty regarding whether it is appropriate to commence treatment at less than 40 mg for all Asian patients, including those with a body mass index (BMI) of 18.5 kg/m² or higher. This doubt arises because the efficacy of starting at less than 40 mg has not been investigated specifically for patients with a BMI of 18.5 kg/m² or higher. Therefore, to draw conclusive findings from this study, it is imperative to verify the efficacy of low-dose initiation specifically in patients with a BMI of 18.5 kg/m² or higher. If BMI of 18.5 kg/m² is used as a cutoff for initiating dosage, this should be explicitly added to the conclusions. Recommending a low dose for all Asian patients may lead to misconceptions and requires a modification of the title.

Reply 1: We have rewritten the article to reflect that low dose afatinib could be equally effective as standard dose (see Page 4, line 73; Page 6, line 79; Page 14, line 249-250; Page 17, line 327). We have highlighted in the limitations section that the findings should be interpreted with caution (see Page 17, line 316). In the conclusions, we recommend future studies in a more controlled and objective setting to check the validity of our current hypothesis (see Page 18, line 332-333).

Changes in the text:

- Lower afatinib doses (<40 mg OD) could be equally effective as standard dose in patients with *EGFR*-mutant advanced NSCLC and may be more suited to Asian patients, minimizing side effects that may occur at higher dosages of afatinib leading to dose interruptions and affecting treatment outcomes.
- Low dose afatinib could be equally effective as standard dose in these patients.
- Our real world study showed that lower doses of afatinib could be equally effective as standard dose in *EGFR*-mutant advanced NSCLC patients with good response and survival outcomes.
- Overall, lower doses of afatinib were equally effective as standard dose in producing good response and prolonging TTF in patients with EGFR-mutant advanced NSCLC.
- Therefore, the findings of this study should be interpreted with caution.
  We recommend future studies in a more controlled and objective setting to check the validity of our current hypothesis.

Comment 2:

The conclusion drawn in the manuscript suggests that low-dose afatinib is more suitable for Asians, leading to a longer time to treatment failure (TTF). However, there is no significant difference in TTF between low and standard doses. Moreover, no significant difference in TTF is observed between groups with dose adjustments and those without. Therefore, the conclusion that dose differences impact the treatment duration is premature due to insufficient data.

Reply 2: We agree that the recommendation is premature and has to be proven with future studies. We have added this text to our conclusions (see Page 18, line 332-33).

Changes in the text: We recommend future studies in a more controlled and objective setting to check the validity of our current hypothesis.

Minor Comments.

Comment 1: In Table 1, a large number of patients with ECOG performance status of 4 (20%) were included in the analysis. Is it common in clinical practice to administer afatinib to patients with PS 4?

Reply 1: We have added text in the discussion to explain this (see Page 16, line 302-304).

Changes in the text: One-fifth of patients had an ECOG performance status of 4. They were given afatinib in addition to standard palliative care because they were keen on active treatment after discussing with their treating clinician.

Comment 2: Detailed data on the BMI of patients who initiated treatment with a reduced dose is necessary.

Please include BMI data for each initiation dose in Table 2.
Reply 2: We have removed < 18.6 from the text (see Page 10, line 173-174). In the limitation section, we have stated that the exact BMI before starting TKI was not captured (see Page 17, line 311-313).
Changes in the text:

- The main reason for starting on lower than the recommended 40 mg dose was due to low body mass index (BMI; 85.5%).
- One major limitation is patients defined as having a low BMI in the study were based on the treating clinician's visual assessment; therefore the exact BMI was not captured in the database.

Comment 3: In Table 2, it is indicated that 31 patients (23.3%) had their afatinib dose reduced. Please describe the percentage of patients who initially started on 40mg among this group.
Reply 3: We have added text in the results section (see Page 11, line 180-181).
Changes in the text: Of 31 patients with afatinib dose reductions, 17 started on 40 mg OD (55%) while 14 started on dose <40 mg OD (45%).

Comment 4: Of the patients who did not reduce the dose of afatinib after starting treatment in Table 2, how many increased the dose to ensure efficacy?
Reply 4: We have added text in the results section (see Page 11, line 181).
Changes in the text: None of the patients had their afatinib dose increased.

Comment 5: It is described in Table 2 that five patients reduced their dosage due to financial reasons. The reviewer thinks the authors need to exclude this data from the analysis because it is not relevant to the conclusions of the current study.
Reply 5: Removing these patients may affect the overall results. Our conclusions were not focused on outcomes based on the reasons for lower doses, although we did state that lower dose may be suitable for patients who experience side effects with normal dose (see Page 17, line 330-332).
Changes in the text: -

Comment 6: 92 patients were included in the analysis of TTF in Table 6, but the authors need to explain the reasons for the exclusion of other patients.
Reply 6: We have added text in the result section to explain that these patients were excluded as they were still on afatinib (see Page 12-13, line 219-220).
Changes in the text: Only patients who discontinued afatinib (n=92) were included in the TTF analysis (Figures 1 and 2 and Table 7). Patients who were still on afatinib were excluded from this analysis.

Comment 7: Page 6, Line230-: The authors said, "Our study showed that afatinib with a dosage of less than 40 mg OD is equally effective in patients with brain metastases, as there was no significant difference in ORR between patients with baseline symptomatic brain metastases and those without." To draw this conclusion, the authors need to compare efficacy with and without brain metastases in patients group less than 40 mg of afatinib.
Reply 7: We have added data on ORR and DCR for these patients in the results section (see Page 12, line 199-202).
Changes in the text: Focusing solely on the lower dose group, the ORR was 69.6% for patients with brain metastases versus 73.3% for patients without brain metastases (p= 0.731). The DCR in this group was 78.3% for patients with brain metastases versus 86.7% for patients without brain metastases (p=0.346).

Comment 8: Page 6, Line256-: The authors said, "Secondly, drug metabolism has been demonstrated to be different amongst Asian patients as many studies have demonstrated that the metabolism of drugs in Asian patients are reduced compared to Caucasians (33-35)."

It has been reported the pharmacokinetic profile of afatinib did not exhibit statistically significant differences between Asian (including the tested subpopulations, i.e., Chinese, Japanese, Korean, Southeast Asian, Taiwanese and other Asian) and Caucasian patients.

Cancer Chemother Pharmacol. 2014 Apr;73(4):759-770.

I think the references are inappropriate.
Reply 8: We have amended the paragraph and added this reference (see Page 16, line 293-298).
Changes in the text: Asian patients may only need lower doses of afatinib for the drug to be effective, while minimizing its side effects. Many studies have demonstrated that drug metabolism in Asian patients are reduced compared to Caucasians (33-35). However, a study showed that the pharmacokinetic profile of afatinib did not exhibit statistically significant differences between Asian (including the tested subpopulations, i.e., Chinese, Japanese, Korean, Southeast Asian, Taiwanese and other Asian) and Caucasian patients (36).

Comment 9: Page 6, Line264-: The authors said, "Therefore, individualized titration of dosage of afatinib is recommended to optimally balance the risk and benefit of treatment." I think additional research is needed on how exactly to set the dosage.
Reply 9: We have added text on future studies in the Conclusions (see Page 17, line 322-324).
Changes in the text: In addition, studies should look into the efficacy of different doses according to BMI scores with objective assessments (such as PFS) in a prospective study with adequate sample size for both comparative arms.

Comment 10: There is no indication in the Results and Discussion with DCR. The authors should comment on this finding.
Reply 10: We added DCR in the results (see Page 11-12, line 197-199; Page 14, line 244-246; Page 13, line 235-237) and discussion (see Page 14, line 252) sections.
Changes in the text:

- The DCR was higher (84.3%) with afatinib starting doses of less than 40 mg dose OD compared to 40 mg OD (72.0%) but the difference was not statistically significant (p=0.149) (Table 5). There were no significant differences in DCR for mutation subtypes, ECOG performance status, tumor stage, comorbidities, afatinib starting dose, or afatinib dose adjustments.
- Patients with baseline symptomatic brain metastases on first-line afatinib were significantly less likely to achieve disease control compared to those without baseline brain metastases (71.7% versus 83.9%; OR [95% CI] = 0.37 [0.14–0.96]; p=0.041) (Table 5).
- Both groups had similar DCR.

Comment 11: Page 3, Line 120-: The authors said, "Patients with incomplete staging or treatment information were excluded from the analysis." The authors need to describe the exclusion criteria in detail.
Reply 11: We have included a flowchart of patients who met the inclusion and exclusion criteria for the study population (Figure 1) and included text in the methods section (see Page 8, line 117-118).
Changes in the text: Figure 1 shows the flowchart of patients included in the study.

Reviewer C

The authors have described that the efficacy of lower dose of afatinib in the non-small cell lung cancer patients with EGFR mutations in a real-world clinical practice, retrospectively. They concluded that lower dose of afatinib, which is less than 40mg OD, significantly resulted in higher response rate than that of 40 mg OD. The median TTF (time to treatment failure) tend to longer in lower dose of afatinib than that of 40mg OD but not significant statistically. Furthermore, they evaluated the efficacy of osimertinib as second-line treatment, exhibiting longer TTF than chemotherapy as second line treatment, significantly.
There are wonderful findings to warrant the publication of this manuscript.

General response: We would like to thank Reviewer C for his valuable comments and his recommendation for the publication of this manuscript.

Comments:

Comment 1: In FLAURA study, overall survival (OS) of osimertinib was 38.6 month (NEJM 2020, 382;41-50). Therefore, it would be very meaningful findings that OS of sequential treatment of afatinib to osimertinib in this study.

Reply 1: We have added data on cumulative OS for patients on sequential afatinib and osimertinib in the results section (see Page 13, line 236-237) and added text to describe this in the discussion (see Page 15, line 277-279).

Changes in the text:

- The cumulative OS for patients on sequential afatinib and osimertinib treatments was 25.6 (±12.3) months.
- The shorter OS with sequential treatment in our real-world study versus the FLAURA study (30) using osimertinib in the first-line setting could be due to the characteristics of our patients (poorer ECOG status, symptomatic brain metastases).

Comment 2: I agree the authors indicate that afatinib dosage of less than 40mg OD should be suited for Asian patients. Regarding Body mass index (BMI) and the afatinib efficacy, there is already reported in Annals of Oncology (2016; vol 27, 2103-2110). It would be better to include this publication in this discussion section.

Reply 2: We have included this reference in the discussion (see Page 15-16, line 284-287).

Changes in the text: In the post hoc analyses of LUX-Lung 6 trial (Asian patients), the efficacy of lower doses of afatinib was demonstrated, and there were more patients with lower BMI on a final dose of 30 mg OD compared to patients with higher BMI (32).

Reviewer D

The present study, ‘’Does dose reduction of afatinib affect treatment outcomes of patients with EGFR-mutant metastatic non-small-cell lung cancer in real-world clinical practice?“ is an interesting, but I have some important questions and confirmations.

General response: We would like to thank Reviewer D for his time and valuable comments and have attempted to address his questions.

Comment 1: Were there any differences in patient background between 40mg once daily afatinib group and lower afatinib doses group? Please consider presenting this in a table.

Reply 1: We have added the comparison between patients started on 40 mg OD versus those started on less than 40 mg OD in Table 2 and as text in the results (see Page 10, line 168-169).

Changes in the text: The afatinib 40 mg OD starting dose group was significantly younger and had more patients with symptomatic brain metastases than the lower dose group (Table 2).

Comment 2: What was the afatinib dose reduction rate in 40mg once daily afatinib group and lower afatinib doses group? Was the discontinuation rate due to adverse events high in 40mg once daily afatinib group?

Reply 2: We have added this information in the results (see Page 11, line 181-184) and discussion (see Page 14, line 256-257).

Changes in the text:

- The discontinuation rates were lower in the <40 mg OD group (16.9%) than in the 40 mg OD group (34.0%). A closer look at the reasons behind discontinuation of afatinib reveal AEs to be the major reason, but this was lower in the <40 mg OD group (57.1%) than in the 40 mg OD group (94.1%).
- In our study, patients on lower doses of afatinib had lower discontinuation rate due to AEs than those on standard dose.

Comment 3: It has been stated that there was a significant difference in response rate between 40mg once daily afatinib group and lower afatinib doses group, but was there a difference in the duration of response? Can you provide the duration of response for each group?

Reply 3: We did not include PFS as an outcome and we have included this as a limitation (see Page 17, line

315-316). We have instead used TTF as an indirect measure of the duration of response in each group.
Changes in the text: In addition, the duration of response and PFS were not captured in our database.

Comment 4: This study included a large number of cases with poor PS, and is it possible that the patient background was different from previous reports, which led to the results this time?
Reply 4: We have added text in the discussion to explain this (see Page 15, line 277-281).
Changes in the text: The shorter OS with sequential treatment in our real-world study versus the FLAURA study (30) using osimertinib in the first-line setting could be due to the characteristics of our patients (poorer ECOG status, symptomatic brain metastases). However, there was no significant difference between patients starting afatinib on 40 mg and <40 mg in terms of their ECOG performance status.

Comment 5: A detailed discussion should be added in the discussion part regarding the fact that although there was a significant difference in ORR, there was no significant difference in TTF.
Reply 5: We have added inadequate sample size as a limitation (see Page 17, line 314-315).
Changes in the text: Inadequate sample size of patients on afatinib 40 mg OD could have led to the lack of statistically significant difference for TTF.

Comment 6: Please consider adding Number at risk to the Kaplan-Meier curve.
Reply 6: We have added the number at risk for all the KM curves (Figures 2–5)
Changes in the text: -