Peer Review File

Article information: https://dx.doi.org/10.21037/tlcr-23-542

Reviewer A

Comment 1： The manuscript presents 2 cases of EGFR ex20ins mutation who gain the durable effect of combination therapy of SBRT, immunotherapy and anti-angiotherapy. Previous studies imply the poor effect of immunotherapy for those patients. Therefore, this manuscript shows that SBRT and angiotherapy could enhance the effect of immunotherapy. However, there is a lack of important information about the correlation of radiotherapy or anti-angiogenesis therapy to the effect of immunotherapy. Without the evidence based on basic science and supporting this additional effect, the value of this case report is limited.

Reply 1： Thank you very much for the comment and valuable suggestion. Although we have briefly discussed the relationship between radiotherapy and immunotherapy in the DISCUSSION section, it is clear that this is not sufficient. We have therefore added a discussion of the relationship between radiotherapy and immunotherapy.

In recent years, it has been widely recognized that radiotherapy can be used not only as a local treatment, but also to stimulate a systemic immune response because of its "distant effect", which provides a strong rationale for the combination of radiotherapy and immunotherapy(46). Radiotherapy promotes immunogenic effects and has the potential to convert irradiated tumors into in situ vaccines, thereby triggering innate and adaptive immune responses locally and systemically, and it also remodels the tumor microenvironment and exerts good immunomodulatory functions, thereby creating a therapeutically appropriate immune microenvironment that sensitizes chemotherapy and immunotherapy(47).

References:

46. Zhang Z, Liu X, Chen D, et al. Radiotherapy combined with immunotherapy: the dawn of cancer treatment. Signal Transduct Target Ther 2022;7:258.

47. Marciscano AE, Haimovitz-Friedman A, Lee P, et al. Immunomodulatory Effects of Stereotactic Body Radiation Therapy: Preclinical Insights and Clinical Opportunities. Int J Radiat Oncol Biol Phys 2021;110:35-52.

Changes in the text： see Page 8, Line15.

As for case 1

Comment 2： Please indicate the reason for discontinuation of camrelizumab and apatinib treatment.

Reply 2： Thank you for your question, we returned a phone call to the patient. She is still being treated locally with camrelizumab and apatinib, and her disease is still stable now.

Changes in the text： see Page 5, Line7.

As for case 2

Comment 3: p5 line 17: Please note crealy that the author did not conduct immunohistochemical analysis of PD-L1 or verify the negative PD-L1 study.

-Please mention the value of TMB.

-Please indicate the site of metastasis.

Reply 3: Thank you for your valuable comments. The patient in case1 was not tested for PD-L1 expression. At that time, the test for PD-L1 was not as developed as it is today, and tissue samples were limited, so we prioritized NGS testing.

-TMB is considered to be a biomarker for treatment with immune checkpoint inhibitors (ICIs). Clinical studies have indicated that high TMB is associated with a survival benefit after treatment with ICIs in single cancer types or pan-cancer types(54,55). (see Page 10, Line19)

References:

54. Lee KW, Van Cutsem E, Bang YJ, et al. Association of Tumor Mutational Burden with Efficacy of Pembrolizumab±Chemotherapy as First-Line Therapy for Gastric Cancer in the Phase III KEYNOTE-062 Study. Clin Cancer Res 2022;28:3489-98.

55. Cristescu R, Aurora-Garg D, Albright A, et al. Tumor mutational burden predicts the efficacy of pembrolizumab monotherapy: a pan-tumor retrospective analysis of participants with advanced solid tumors. J Immunother Cancer 2022;10.

The site of the patient's tumor metastasis was bilateral intrapulmonary and pleural. We have written in the text (see Page 5, Line19). Please kindly check!

Comment 4: p5 line 21: Please provide detailed information about the side effects.

Reply 4: The patient developed atrial fibrillation and second degree gastrointestinal reaction after treatment. Patients and their families requested a change in treatment.

Changes in the text：see Page 5, Line28.

Comment 5: -The author states that they demonstrate SBRT for the left lung lesion, however, figure3 shows the plan of SBRT for the right lung lesion. Which is correct?

Reply 5: Thanks for pointing out. After carefully rechecking the history data, we clarified that the patient was treated with SBRT in the right lung. We have revised it in the revised manuscript. We apologize for this mistake and ask for your forgiveness.

Changes in the text：see Page 6, Line11.

Comment 6: -Please indicate the reason for discontinuation of camrelizumab and apatinib treatment.

Reply 6: We thank the reviewer for pointing this out. The reason for discontinuation of camrelizumab and apatinib treatment was tumor progression.

On June 5, 2022, this patient was reexamined for tumor progression.

Changes in the text：Page 6, Line15.

As for discussion

Comment 7: -p8 line 21: The author mentions that the therapy of SBRT, immunotherapy and anti-angiotherapy does not depend on PD-L1, TMB, mutation type of EGFR ex20.

However, this is an overstatement because they show just 2 cases.

Reply 7: Thank you very much indeed for pointing out our incorrect wording. We apologize for causing your misunderstanding, this is not a very strict statement. But amazingly in these two patients there really was no dependence on the expression of PD-L1, TMB and the type of

EGFR ex20ins. Therefore, we think this needs to be confirmed by more prospective studies. To be clearer and more accurate, we have revised this with another expression.

The PD-L1 expression status, TMB, and EGFR ex20ins type were not significantly similar in these 2 cases. We are not sure if it means that the treatment model of SBRT combined with immunotherapy and anti-angiogenic therapy does not depend on patients' PD-L1 expression status, TMB, or EGFR ex20ins type and may benefit a wide range of patients. Of course, prospective clinical studies are needed to clarify this.

Changes in the text： see Page 9, Line16.

As for abstract

Comment 8: -p1 line 33: Please correct "grow" to "growth".

Reply 8: We thank the reviewer for pointing this out. We have changed "grow" to "growth."

Changes in the text： Page 1, Line 33.

As for Introduction

Comment 9: -p3 line 7: Please delete the comma (,) if the author intends to convey that NSCLC accounts for 80-85% of all lung cancer.

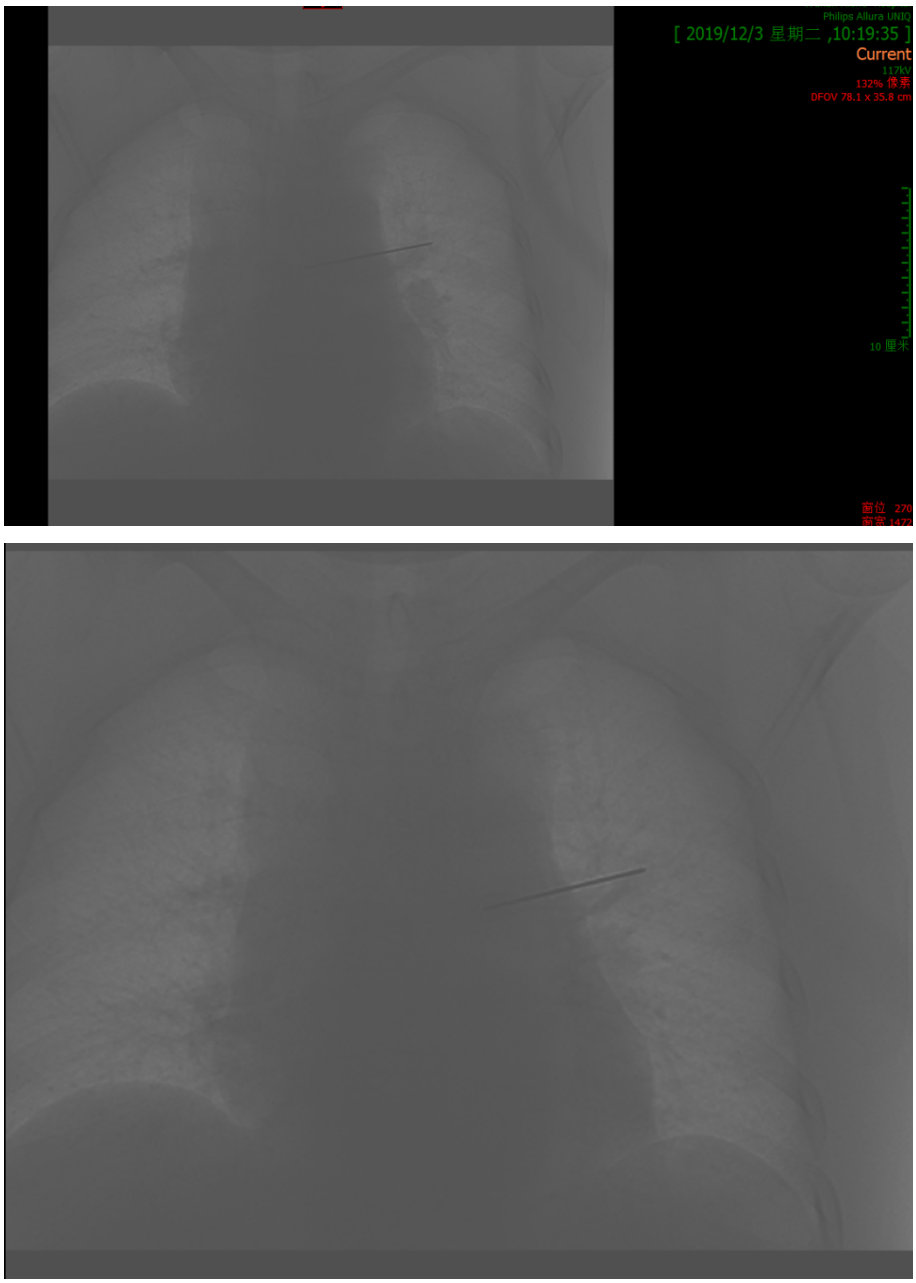
Reply 9: We thank the reviewer for pointing this out. We have deleted the comma (,)

Changes in the text： Page 3, Line 9.

As for case 2

Comment 10: -p5 line 14: Please indicate the biopsy site.

Reply 10: We thank the reviewer for pointing this out. We reviewed the records of the biopsy at that time and found the images of the biopsy, which are presented below. We hope this will answer your questions.



As for discussion

Comment 11: -p10 line 20: The data from the PEMBRO-RT trial has already been referenced in p8 line 1.

Reply 11: We thank the reviewer for pointing this out. But the content of p10 line 20 is a discussion of international experts, it is necessary for the iMDT case reporting model. Therefore, we have not modified it and would appreciate your understanding.

Reviewer B

These are 2 interesting cases about the combination of SBRT and PD-L1 combined with VEGF2-inhibition in patients with EGFR exon20 insertions.

Reply: Thank you very much for the positive comments on our work and all suggestions for its improvement.

I have minor concerns:

Comment 1: The follow-up ends in May 2022 (#1) and June 2022 (#2) with stable disease. This is >1 year ago. Please give an update of the follow-up.

Reply 1: Thank you for your valuable suggestion. The patient in case1 is still being treated locally with camrelizumab and apatinib, and her disease is still stable now. However, because this patient has been reviewed locally since May 2022, our hospital does not have her current electronic images. After our follow-up phone call she indicated that she would be returning to our hospital for a review in the near future, and after obtaining the most recent images we were happy to present them. For the patient in case 2, we regret that she was not been able to control tumor progression with subsequent therapies since disease progression in June 2022 and has reached OS in June 2023. We have added these to the revised manuscript.

Changes in the text: Thereafter, the patient continued to receive maintenance therapy with camrelizumab and apatinib locally. (see Page 5, Line 7)

But we regret that the patient has not been able to control tumor progression with subsequent therapies and has reached OS in June 2023. (see Page 6, Line18)

Comment 2: There is a mistake in the discussion / page 7: According to the results of Zhou et al. study, ... the median PFS was 8.3 (1.9–8.3) months, suggesting that camrelizumab in combination with apatinib in EGFR ex20ins NSCLC can achieve good results".

Actually, the median PFS was 2.8 months (!) and there was only a moderate benefit of this combination. In this context, it is interesting that only one of 3 patients with an EGFR ex20ins mutation responded to this therapy. PLEASE DISCUSS theses results in the context of SBRT for your patients. I find it very unlikely that SBRT completely changes the outcome for the treatment with camrelizumab and apatinib.

Reply 2: Thanks for pointing it out. We reviewed the full text of this study and we apologize that we didn't express ours idea clearly. The median PFS in this study was indeed 2.8 months, but that was for all patients. For patients with EGFR ex20ins mutation, the median PFS was 8.3 months, and it was because we noted that this treatment combination was more efficacious for patients with EGFR ex20ins mutation than all patients with regular EGFR/ALK mutation that we explored SBRT in combination with this treatment modality. In order to make it clear, we have made changes in the revised manuscript.

According to the results of Gao et al.'s study, camrelizumab in combination with apatinib in NSCLC patients with EGFR ex20ins mutation had an ORR of 33.3% (0.8–90.6%), and the median PFS was 8.3 (1.9–8.3) months, suggesting that camrelizumab in combination with apatinib in EGFR ex20ins NSCLC may achieve good results (38).

References:

38.Gao G, Ni J, Wang Y, et al. Efficacy and safety of camrelizumab plus apatinib in previously treated patients with advanced non-small cell lung cancer harboring EGFR or ALK genetic aberration. Transl Lung Cancer Res 2022;11:964-74.

Changes in the text: see Page 8, Line 5.

Comment 3: I cannot see any advantages of this kind of discussion from different perspectives and find it somewhat confusing. I strongly recommend to summarize and shorten the discussion to 3-4 main topics and emphasize the limitations.

Reply 3: We acknowledge your great contribution in improving the quality of our manuscript. We apologize for your confusion. The discussion part of our initial submission is a regular discussion, but the call for papers for iMDT Corner of this journal is in the form of iMDT, an online discussion among international experts, so we have made this modification, and we appreciate your understanding. If editor and reviewers need us to provide a regular version of discussion, we are happy to do so.

Reviewer C

Comment 1: Case 1 and case 2: what method was used for NGS and TMB? and for PD-L1 analysis? TMB is not mentioned in case 2 while it is mentioned in the discussion (page 8, line 20)

Reply 1: Thank you for your interest and suggestions. Genetic testing and TMB testing for both cases were performed using NGS panel, and immunohistochemical testing was used for PD-L1 expression. The TMB in case2 was 1.0 mutation/Mb. We have added the TMB results of case2 to the revised manuscript.

Changes in the text： see Page 5, Line 24.

Comment 2: Case 1: pt was rebiopsied: was NGS repeated on rebiopsy?

Reply 2: Thank you for your question, the patient in case 1 did not repeat NGS because her condition has been very stable.

Comment 3: page 5 line 29: CT-guided lung puncture with gold-standard implantation means what?

Reply 3: Thank you for your question. Golden marker implantation is a tool of localization for SBRT that follows tumor motion, which allows precise guidance of the radiofrequency knife beam to focus on the tumor tissue, thereby minimizing damage to surrounding normal tissue. We have changed "gold-standard implantation" to "golden marker implantation" in the revised manuscript.

Changes in the text： see Page 6, Line 4.

Comment 4: Page 9, lines 1-7; I am not sure that IMpower 010 is positive in the subgroup of EGFR mutation; not even effective CT-IO (Checkmate 722) please comment on this

Reply 4: Thank you for the noticing. The content of Page 9, lines 1-7 is the discussion section of the international experts. We added it to our manuscript at the request of the editorial board and thank you for your understanding.

Reviewer D

Although it is interesting to examine long-term survival after immunotherapy in patients with advanced non-small cell lung cancer and EGFR exon 20 insertion mutation, the discussion for this case report is insufficient.

I have some comments. I would like to give regarding this paper.

Comment 1: - Since no EGFR-TKI was administrated in these two cases, Is the author required to show Table 1 of the EGFR-TKI review for EGFR Exo20ins?
Reply 1: We appreciate the reviewer's careful review of our manuscript. Although our two patients were not treated with any EGFR-TKI, exploring effective EGFR-TKIs is undoubtedly necessary for patients with EGFR mutations. And it has demonstrated certain efficacy in the exploration of EGFR-TKIs for the treatment of NSCLC patients with EGFR ex20ins mutation. That's why we added Table 1 to the section of discussion, and we think it's necessary to show it, thank you for your understanding.

Comment 2: - The fact that immune checkpoint inhibitors are effective in EGFR ex20ins cases in Table 2 suggests that ICI alone may have been effective without concomitant angiogenesis inhibitors. The author is required to elaborate on the consideration that concomitant use of angiogenesis inhibitors is effective in EGFR ex20ins cases.
Reply 2: We thank the reviewer for pointing this out. In the IMpower150 study, chemotherapy plus Atelizumab plus Bevacizumab demonstrated the best efficacy in patients with EGFR lung cancer. This clearly demonstrates the therapeutic potential of anti-angiogenic therapy combined with immunotherapy in patients with EGFR mutations. We also found a clinical study by Prof. Zhou exploring the efficacy of camrelizumab in combination with apatinib in NSCLC patients with EGFR ex20ins mutation. These demonstrate that anti-angiogenic therapy in combination with immunotherapy is effective for patients with EGFR ex20ins lung cancer. We have added this to the revised manuscript.

According to the results of Gao et al.'s study, camrelizumab in combination with apatinib in NSCLC patients with EGFR ex20ins mutation had an ORR of 33.3% (0.8–90.6%), and the median PFS was 8.3 (1.9–8.3) months, suggesting that camrelizumab in combination with apatinib in EGFR ex20ins NSCLC may achieve good results (38). (see Page 8, Line 5)
References:
38.Gao G, Ni J, Wang Y, et al. Efficacy and safety of camrelizumab plus apatinib in previously treated patients with advanced non-small cell lung cancer harboring EGFR or ALK genetic aberration. Transl Lung Cancer Res 2022;11:964-74.

In the IMpower150 study, chemotherapy plus Atelizumab plus Bevacizumab demonstrated the best efficacy in patients with EGFR lung cancer. This clearly demonstrates the therapeutic potential of anti-angiogenic therapy combined with immunotherapy in patients with EGFR mutations(49). (see Page 8, Line 29)
References:
49. Reck M, Mok TSK, Nishio M, et al. Atezolizumab plus bevacizumab and chemotherapy in non-small-cell lung cancer (IMpower150): key subgroup analyses of patients with EGFR

mutations or baseline liver metastases in a randomised, open-label phase 3 trial. Lancet Respir Med 2019;7:387-401.

Comment 3: - It is difficult to interpret because the second half of the discussion is in an unfamiliar format.
This paper has insufficient discussions.
Reply 3: We acknowledge your great contribution in improving the quality of our manuscript. We apologize for your confusion. The discussion part of our initial submission is a regular discussion, but the call for papers for iMDT Corner of this journal is in the form of iMDT, an online discussion among international experts, so we have made this modification, and we appreciate your understanding. If editor and reviewers need us to provide a regular version of discussion, we are happy to do so.