

## Responses to Reviewers

Dear Editor and Reviewers,

Thank you for your letter and for the reviewers' comments on our manuscript, "Malignant transformation of pulmonary bronchiolar adenoma into mucinous adenocarcinoma: A case report" (Manuscript no.: 81083). These comments were all valuable and very helpful to us in revising and improving our paper. We have revised our manuscript after carefully reading all of these comments. We also employed an English-language editing service, Medjaden, to help with polishing our language and attached a certification. Below, please find our point-by-point responses to the comments. The "track changes" feature of MS Word was used show language polishing.

### Responses to the reviewers' comments:

#### Reviewer #1:

1. **Comment:** From the pathological images provided, this tumor looks like a pure invasive mucinous adenocarcinoma. The bronchiolar adenoma component cannot be clearly seen in the figures. The authors need to provide more convincing pathological images and evidence to demonstrate the bronchiolar adenoma part of this tumor. High power view of pathological images are especially necessary to show the bilayered structure of bronchiolar adenoma.

**Response:** Considering the reviewer's suggestion, we added two new high-power images (Figure 2C-D,  $\times 400$ ), and used different symbols to clearly indicate the basal cells, ciliated cells, and mucinous cells (Figure 2D). In addition, to more clearly show the absence of basal cells in certain glandular areas, we added two other high-power images (Figure 3C and 3D,  $\times 400$ ). We also replaced most of the immunohistochemical images with high-power field views (Figure 4A-F and 5C-H,  $\times 400$ ).

2. **Comment:** The ciliated cells in the bronchiolar adenoma part of this tumor cannot be well appreciated in the figures. Please provide additional images to

demonstrate the presence of ciliated cells in this tumor.

**Response:** Following your suggestion, we provided Figure 2D (×400) and highlighted the ciliated cells with orange arrows.

3. **Comment:** p40 is a more specific marker than p63, as p63 can stain some adenocarcinoma cells. It is recommended to use p40 immunostain instead of p63 to demonstrate the basal cells in this tumor.

**Response:** Thank you for the important comment. Accordingly, we presented the results of immunohistochemical staining for p40 in Figure 4B and 5B to more specifically identify basal cells.

4. **Comment:** In Section "OUTCOME AND FOLLOW-UP" The sentence "In the center of the lesion, the presence of a bilayered structure could not be ruled out" is better written as "In the center of the lesion, the presence of a bilayered structure could be observed." The sentence "The tripartite cellular components were normal in histology, without significant atypia, mitosis, or necrosis" is better written as "The tripartite cellular components were devoid of significant atypia, mitosis, or necrosis".

**Response:** Thank you for your important suggestion. We revised these sentences as suggested.

5. **Comment:** In DISCUSSION section: The authors wrote "BA/CMPT is no longer considered a benign lesion but rather a low-grade inert malignant tumor" --> This is incorrect. In current (2021) WHO classification, BA/CMPT is considered as a benign neoplasm.

**Response:** Thank you for your insightful comments. Malignant transformation of tumor cells is a complex process involving the gradual accumulation of mutations in multiple genes, finally manifesting as changes in biological behavior. There is no evidence of recurrence or metastasis in all observed cases of BA/CMPT, leading to the conclusion that these lesions are benign. However, there are a few cases of

BA/CMPT with evidence of malignant transformation. Thus, we revised this text as follows: “Although the 2021 WHO classification considers BA as a benign tumor, other evidence suggests it has malignant potential<sup>[1, 7-13]</sup>.”.

6. **Comment:** In DISCUSSION section: The authors wrote "The high prevalence of mutations in driver genes (EGFR, BRAF, ALK, and KRAS) supports the notion that these lesions are neoplastic rather than reactive, cytoplasmic, or metaplastic." -> What does "cytoplasmic" mean in this sentence?

**Response:** Thank you for the pertinent query. This was a spelling error. The word "cytoplasmic" is redundant in this sentence, and we deleted it. We are very sorry for our negligence.

7. **Comment:** In DISCUSSION section: The authors wrote "In the present case, flat, papillary, and glandular structures in the epithelial neoplasm were observed in frozen sections, making it difficult to distinguish between benign and malignant lesions." ---> Did the present case send for intraoperative frozen sections? If so, what was the diagnosis in frozen sections? This should be described in the CASE SUMMARY section.

**Response:** Yes, the intraoperative frozen sections were diagnosed as a kind of pulmonary epithelial tumor, but it was difficult to differentiate BA from invasive adenocarcinoma, indicating the need for immunohistochemical analysis of paraffin sections. We added the results of intraoperative frozen section diagnosis in the CASE SUMMARY section. Thank you for this suggestion.

8. **Comment:** Figure 2: High power view is required to better demonstrate the bilayered structure and cellular composition of the tumor.

**Response:** To more clearly show the bilayer structure of the bronchiole adenoma, we added two new figures, Figure 2C and D (×400).

9. **Comment:** Figure 2 legend: The sentence "At high power, ciliated cells or

cubic/low columnar cells and locally abundant mucinous cells are observed in the luminal epithelium, which are normal in morphology, without significant atypia and pathological mitosis." is better written as "At high power, ciliated cells or cubic/low columnar cells and locally abundant mucinous cells without significant atypia and pathological mitosis are observed in the luminal epithelium."

**Response:** Thank you very much. We revised this sentence accordingly.

**Reviewer #2:**

1. **Comment:** The study addresses very interesting idea. The manuscript has no novelty since others report the similar topic of the progress of BA to malignant transformation. The case summary reveals no abnormal detected on physical examination although the lung nodule as high as 1.7 cm. The case summary should be more informative. Please clarify!

**Response:** Considering the reviewer's suggestion, we included additional details regarding all previous chest CT examinations (from Nov 3, 2019 to Dec 22, 2021) in this section (CASE SUMMARY).

We tried our best to improve the manuscript by making revisions according to all the suggestions from the reviewers. We greatly appreciate the editors and reviewers for their earnest efforts, and hope that our revised manuscript is approved for publication.

Once again, thank you very much for these comments and suggestions.

Best regards,

Peng Li, Department of Pathology, the First Affiliated Hospital of Wenzhou Medical University, Xuefu Road, Ou Hai District, Wenzhou 325000, Zhejiang Province, P.R. China

Email: lipenglimo@163.com

Tel: +0086-13857715312