

## Peer Review File

Article information: <http://dx.doi.org/10.21037/tcr-22-1171>

### Reviewer A

In this case report by Wang et al., they described a combined small cell carcinoma with enteric adenocarcinoma component. I have the following comments:

1. Enteric adenocarcinoma typically expresses other immunohistochemical markers such as CK20, CDX2, and/or SATB2. Were those performed in this case? Otherwise, it remains unclear whether the adenocarcinoma component was truly enteric, versus other kinds of adenocarcinoma.
2. Several aspects in Figure 1 remain unclear. First, the magnifications were listed as "20x" and "40x", but it was unclear whether the authors have taken into the account the eyepiece/camera magnification, and that this may be "200x" or "400x". Please clarify. Also, based on figure 1C alone, the small cell carcinoma histology was not obvious in the image provided.
3. The authors noted that next-generation sequencing was performed on the punctured tissue. It was unclear where the site of the punctured tissue was from, and whether the NGS-sequenced tissue contains only one component or both small cell carcinoma and adenocarcinoma component. Also, given that small cell carcinoma typically harbours mutations such as TP53 and RB1, were those detected in the NGS panel? These points should be clarified.

Reply:

1. The case was considered to be a left hilar tumor with mediastinal lymph node and right lung metastasis by PET-CT examination after discovery. The clinical stage was stage IV, and there was no chance of surgical treatment, so large postoperative pathological specimens could not be obtained. Biopsy of the left lung confirmed that there were small cell carcinoma components (about 70%) and adenocarcinoma components (about 30%), and adenocarcinoma components showed pulmonary enteric adenocarcinoma differentiation. The histological features and immunophenotypes of pulmonary enteric adenocarcinoma are almost identical to those of colorectal cancer, and the diagnosis of intestinal adenocarcinoma requires the exclusion of colorectal cancer metastasis. The adenocarcinoma region of lung biopsy tissue in this patient is morphologically similar to intestinal adenocarcinoma. Immunohistochemical expression of intestinal markers (CK20, CDX2 and SATB2 were positive); In this case, no intestinal tumor was found on PET-CT examination, so the adenocarcinoma in the lung tissue of this patient supports pulmonary enteric adenocarcinoma.

- Figure 1. Magnification of pathological images were 200x and 400x.
- NGS sequencing was performed in the soft tissue lung tissue of the basal segment after puncture of the lower lobe of the left lung, and the detected tissues were reexamined to contain two kinds of small cells and adenocarcinoma components. Results: EGFR P.1861Q (EX21), BRCA2 deletion, TMB-L, MSS, PD-L1 (TPS<1%, CPS<1). In addition, the detection report showed that TP53 mutation and RB1 deletion.

### Reviewer B

- The authors have discussed about cytokeratin and synaptophysin protein expression by IHC in each tumor components. The authors need to add the IHC data of CK20, CDX2 and TTF-1.
- The authors should provide the serum data of tumor marker, such as CEA, CA19-9, and ProGRP.
- The authors should mention that they had completely excluded the possibility of lung metastasis of primary colorectal tumor by their examination.
- It could be preferable to discuss more about the mechanism of carcinogenesis in this rare disease if at all possible. The author's speculation will be also acceptable.

### Reply:

- Immunohistochemistry of this patient showed: small cell carcinoma: CKpan para-nuclear site (+), TTF-1 (+), SATB2 lesion (+), Syn (+), CgA (+), CD56 (+), Ki-67 (+) about 95%. Adenocarcinoma: CKpan (+), CK7 (+), CK20 (+), CDX-2 (+), TTF-1 (+), NapsinA (+), Ki-67 (+) about 80%. Note: Adenocarcinoma components are enteric adenocarcinoma components.
- Baseline detection of serum markers at the onset of the disease (2020-12-29) showed that CEA was 8.74ng/ml, slightly elevated; Ca19-9:27.5u /mL and ProGRP:38.01pg/ mL were both within the normal range. The specific values of the post-dynamic detection are listed as follows:

TIME	CEA (ng/ml)	CA19-9(U/mL)	ProGRP(pg/ml)	NSE (ng/ml)
2020-12-29	8.74	27.5	38.01	14.7
2021-01-25	5.17	13.3	30.43	13.4
2021-02-26	3.72	9.05	20.43	9.52

2021-03-29	3.92	9.21	37.46	11.8
2021-04-26	3.47	8.33	23.31	9.08
2021-05-26	4.44	8.38	37.09	9.32

3. In this case, the patient has blood in the stool, changes in defecation and other symptoms; Systemic PET-CT examination showed no intestinal tumor. Biopsy of the left lung confirmed the mixed type of small-cell carcinoma and adenocarcinoma, and the adenocarcinoma component showed lung-intestinal adenocarcinoma differentiation. Therefore, lung metastasis of primary colorectal tumor was ruled out.
4. At present, the pathogenesis of this rare disease is uncertain and needs further investigation.