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PD-L1 pitfalls: Emphasizing the importance of membranous localization and correlation with tumor cell and macrophage distributions

Increased utilization of PD-L1 in both clinical and research settings has led to an appropriate increase in concerns about the interpretation of this immunomarker. McCoy et al. raise an important point regarding the assessment of PD-L1 expression in tumor-associated macrophages (TAM). Their general caution regarding the technical vagaries of PD-L1 immunostaining across antibodies and platforms is critical, and should be heeded by anyone embarking on research involving this immunomarker. These concerns have been nicely summarized in their letter and thoroughly outlined elsewhere, particularly with regard to the PD-L1 immunostaining in lung carcinoma (Hirsch et al., 2017; McLaughlin et al., 2016; Scheel et al., 2016; Sholl et al., 2016). With specific regard to our manuscript, McCoy et al. note two primary concerns: first, that the membranous staining pattern observed in TAMs in our series of high grade serous ovarian carcinomas was not clearly appreciable in the provided low power images and second, that the identity of macrophages cannot be certain because PD-L1 and CD68 were performed on different sections.

We hope that we can alleviate these concerns with the attached high power figure and following clarifications. As stated in our methods, we only classified clear circumferential membranous staining as positive in both tumor cells and macrophages. We did not encounter the non-specific, cytoplasmic-only staining described by McCoy et al. using our PD-L1 detection method although we can certainly appreciate that this might be an issue using other antibodies, platforms, dilutions, etc. Rather, the TAM staining observed in our study was as illustrated in these higher power figures: membranous in localization, with minimal cytoplasmic reactivity which was, when present, significantly less prominent than the membranous component. This pattern is shown in two representative cases in this figure: in the top row, we have a case with strong membranous PD-L1 staining outlining macrophages with very minimal granular cytoplasmic positivity affecting only some cells. In the bottom row, we have fainter (but still decidedly membranous) macrophage staining without any cytoplasmic artifact (Fig. 1). In both cases, the high grade serous carcinoma adjacent to the macrophage infiltrate is PD-L1 negative. Accompanying each of these cases is the corresponding

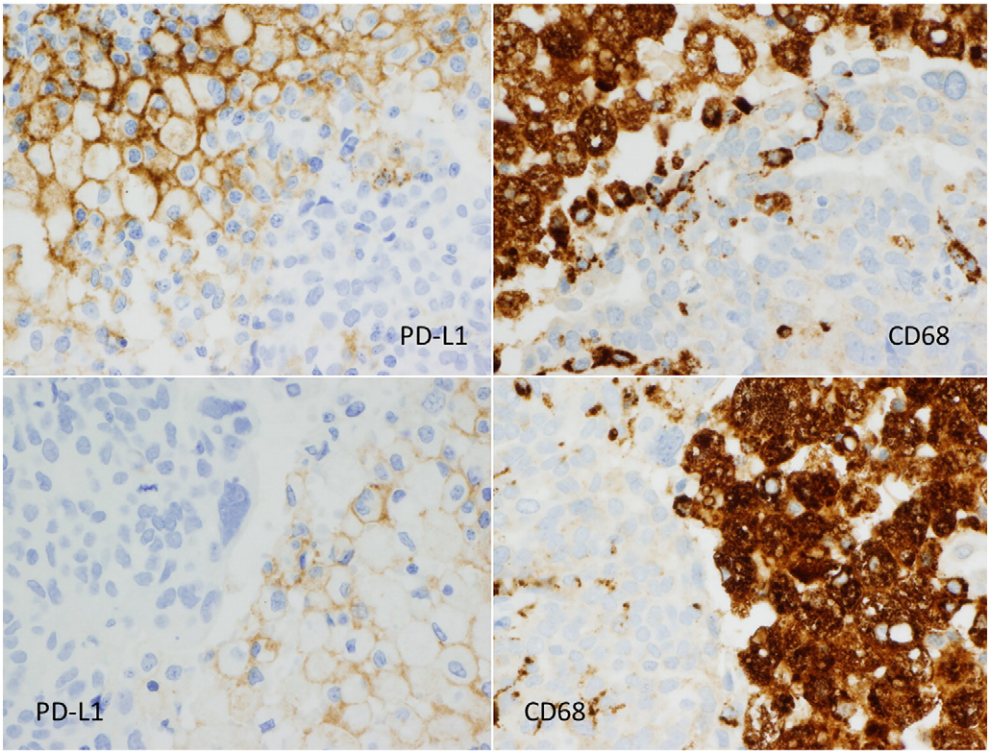

Fig. 1. Representative high power views of membranous PD-L1 expression on tumor-associated macrophages.

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CD68 stain. The TAM population is well-demarcated by this stain and corresponds with the distribution of PD-L1 positivity: in the case in the top row, both PD-L1 and CD68 are localized in the upper left portion of the image, while in the bottom row both stains demarcate the right side of the image. While we appreciate the theoretical concern that cut-through artifacts might have interfered with our interpretation, given that these represent the exact same area of tissue on consecutive 3 μm serial sections we consider that possibility extremely unlikely, particularly given the marked density of the macrophage aggregates.

A final point of emphasis regarding this topic is that one could easily imagine how TAMs might be misinterpreted as tumor cells when PD-L1 immunostains are viewed in isolation. In our experience assessing PD-L1 in a variety of tumor types this pitfall was particularly prominent in high grade serous ovarian carcinomas, which are often associated with a robust macrophage infiltrate. We therefore recommend that PD-L1 immunostains performed on ovarian serous carcinomas are interpreted in conjunction with the corresponding H&E by readers with experience in PD-L1 interpretation and with cautious attention to macrophage distributions. Furthermore, correlation with macrophage marker immunostaining is further recommended in cases for which there is morphologic uncertainty regarding cell identity. Further studies detailing true membranous PD-L1 immunostaining TAMs will be of interest, as the clinical significance of this finding with regard to prognosis and immunotherapeutic response remains largely unknown.

References

Hirsch, F.R., McElhinny, A., Stanforth, D., et al., 2017 Feb. PD-L1 immunohistochemistry assays for lung cancer: results from phase 1 of the blueprint PD-L1 IHC assay comparison project. J. Thorac. Oncol. 12 (2):208–222. http://dx.doi.org/10.1016/j.jtho.2016.11.2228.

McLaughlin, J., Han, G., et al., 2016. Quantitative assessment of the heterogeneity of PD-L1 expression in non-small cell lung cancer. JAMA Oncol. 2 (1):46–54. http://dx.doi.org/10.1001/jamaoncol.2015.3638.

Scheel, A.H., Eietel, M., Heukamp, L.C., et al., 2016. Harmonized PD-L1 immunohistochemistry for pulmonary squamous cell and adenocarcinoma. Mod. Pathol. 29 (10): 1165–1172. http://dx.doi.org/10.1038/modpathol.2016.117.

Sholl, L.M., Aisner, D.L., Allen, T.C., et al., 2016. Programmed death logand-1 immunohistochemistry—a new challenge for pathologists: a perspective from members of the pulmonary pathology society. Arch. Pathol. Lab. Med. 140 (4):341–344. http://dx.doi.org/10.5858/arpa.2015-0506-SA.

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