Background PD-(L)1 blockade has changed the landscape for advanced Merkel cell carcinoma (MCC) as over 50% of patients initially respond to therapy. There are no clinically useful approaches to assess a patient’s likelihood of response to PD-1 blockade. Understanding the underlying biology of anti-PD-(L)1 responses and identifying predictive biomarkers can inform future combination therapies for patients that do not benefit from treatment. MCC is caused by the Merkel cell polyomavirus in ~80% of cases with the remaining cases being due to extensive UV exposure. Both of these processes are highly immunogenic due to viral oncoprotein expression or UV-neoantigens, respectively. Historically, studies have focused on adaptive immunity, and little is known about innate immunity in MCC which may play an important role in immune evasion. Myeloid cells are heterogenous and have been shown to play a suppressive role in many cancer types. We sought to investigate the link between tumor-infiltrating myeloid cells and outcomes to PD-(L)1 blockade.

Methods Identification of myeloid subtypes in MCC tumors was determined by single-cell RNA sequencing (scRNAseq) of dissociated tumor samples obtained from 9 patients. Following unsupervised clustering based on differential gene signatures, we selected major markers expressed in the myeloid subtypes in MCC (CD14, CD163, S100A8) and a cytotoxic T cell marker (CD8) for multiplex immunohistochemistry (m-IHC) analysis of 51 pre-treatment patients’ tumors.

Results scRNAseq analysis discovered tumor associated macrophages in MCC tumors that express hallmarks of monocytic myeloid derived suppressor cells (M-MDSCs): the surface molecules CD14+CD11b+HLA-DRlo/- and upregulated S100A8 and S100A9 genes. Initial m-IHC analysis comparing pre-treatment tumors of 32 responders and 18 non-responders did not show differences between the response groups. However, after classifying tumors by CD8 infiltration status (intratumoral CD8+ cells/1mm – Hot score: CD8 > 60, Cold score: CD8 < 60), we observed higher ratios of S100A8-to-CD8 in patients with hot tumors that did not respond to PD-(L)1 blockade treatment (p=0.046).

Conclusions S100A8 is associated with a tumor promoting effect and is a potential prognostic biomarker for advanced melanoma.2 Our data suggests a correlation of S100A8-expressing cells in MCC tumors with response to treatment, as the ratio of S100A8-to-CD8 is higher in inflamed tumors that did not respond to immunotherapy. Future approaches to target M-MDSCs or S100A8 directly might modify the ratio of S100A8-to-CD8 and potentially improve patient responses to PD-(L)1 blockade treatment.

REFERENCES

Ethics Approval The study was approved by Fred Hutchinson Cancer Center Ethics Board, approval number 6585.