Tuberculosis following PD-1 blockade in a patient with Merkel cell carcinoma: Coincidence or Causality?

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Background

CD4 T cells are critical for containment of *Mycobacterium tuberculosis* (Mtb) infection. IFN-γ-producing Th1 cells play a major role in protection against Mtb, but other cell types, including Th17 and CD8 T cells may also contribute to control of Mtb infection. Preclinical data suggest that PD-1 blockade has a therapeutic potential to enhance T cell responses and control of a variety of chronic infections. However, PD-1 deficient mice are highly susceptible to Mtb infection and develop necrotic pulmonary lesions. This increased susceptibility to Mtb infection of PD-1 deficient mice is counter-intuitively due to increased IFN-γ-production by CD4 T cells. Therefore, preclinical animal model data raise the possibility that enhanced Th1 responses following PD-1 blockade may have unexpected deleterious consequences during Mtb infection. Here we report a case of a patient with Merkel cell carcinoma (MCC) that developed tuberculosis after initiating PD-1 blockade with Pembrolizumab.

Methods

An 83-year old man began a clinical trial of Pembrolizumab in June 2015 for advanced MCC. The patient had no risk factors and no testing for latent TB was performed. CT scan after 11 cycles revealed known sites of MCC decreasing in size or remaining stable, and a new right lower lobe pulmonary nodule was noted (1.1 x 1.6 cm). The patient underwent excision of the nodule in January 2016. Pathology revealed necrotizing granuloma and acid-fast staining of tissue sections was performed. Cryopreserved peripheral blood monocytes (PBMCs) obtained immediately prior to Pembrolizumab and at cycles 5, 8, 11, and 14 were analyzed for antigen-specific CD4 and CD8 T cell responses by intracellular cytokine staining after stimulation with purified protein derivatives (PPD). Serum samples were also analyzed for IgG responses to a panel of different Mtb antigens.

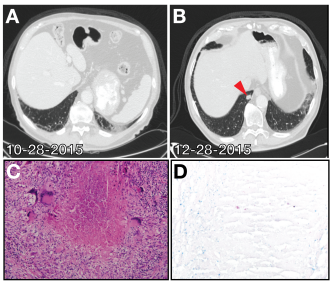

Figure 1. Clinical characteristics of a patient with Merkel cell carcinoma who developed tuberculosis after Pembrolizumab treatment. (**A**) Chest CT image of a Merkel cell carcinoma patient before onset of tuberculosis. (**B**) CT image after 11 cycles of infusion. Arrow head indicates necrotizing nodule in the right lower lobe. (**C**) H&E staining of tissue section from the excited lung nodule. (**D**) Acid-fast staining of tissue section from the nodule.

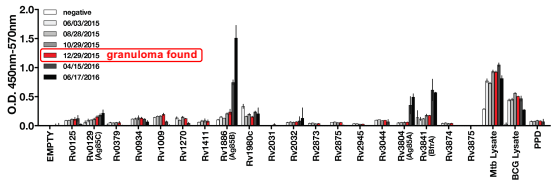

Figure 3. *M. tuberculosis*-specific antibody responses in a patient with Merkel cell carcinoma after Pembrolizumab treatment. IgG response against mycobacterial antigens was measured in sera from a patient at different points after PD-1 blockade.

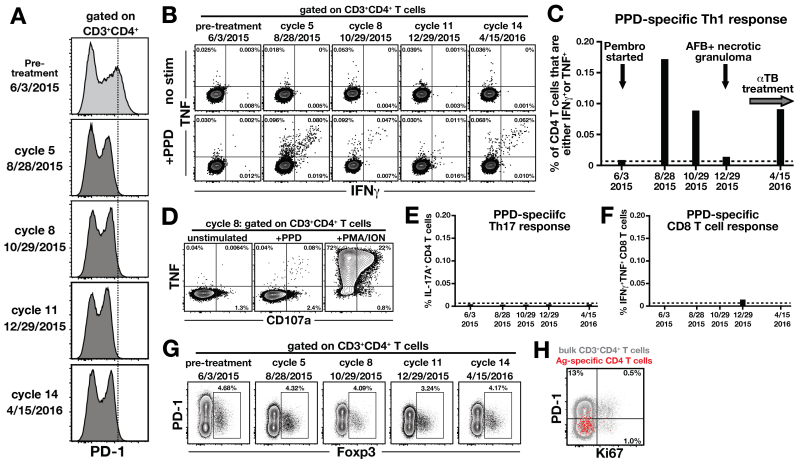

Figure 2. *M. tuberculosis*-specific T cell responses in a patient with Merkel cell carcinoma after Pembrolizumab treatment. (**A**) Flow cytometric analysis of PD-1 expression on CD4 T cells in peripheral-blood monocytes (PBMCs) from a patient at different points after Pembrolizumab treatment. Dot line denotes the baseline level of PD-1 expression on CD3+CD4+ cells before treatment. (**B**) Fluorescence-activated cell sorting (FACS) plots of CD4 T cells showing intracellular cytokine staining for tumor necrosis factor (TNF) and interferon (IFN)-γ after 6 hour stimulation with purified protein derivatives (PPD). (**C**) Summary graph of PPD-specific Th1 response in PBMCs from a patient throughout disease course. (**D**) FACS plots of CD4 T cells showing TNF and CD107a expression after 6 hour stimulation with PPD or phorbol myristate acetate and ionomycin (PMA/ION). Summary graphs of PPD-specific Th17 (**E**) and CD8 T cell (**F**) responses in PBMCs from a patient after PD-1 blockade. (**G**) Frequency of Foxp3+ regulatory CD4 T cells in PBMCs from a patient throughout disease course. (**H**) An example FACS plot showing PD-1 and Ki67 expression on bulk (gray) or PPD-specific (red) CD4 T cells in PBMCs from a patient. Overlaid PPD-specific CD4 T cells are gated on TNF+IFN-γ+ cells following stimulation.

Results

PD-1 blockade in this individual was associated with significantly increased circulating Mtb-specific Th1 responses prior to development of the necrotic pulmonary tuberculoma. However, neither Th17 cells nor CD8 T cells specific to Mtb were detectable in PBMCs at any time. Circulating Foxp3+ regulatory T cells did not change in number during Pembrolizumab treatment in this individual. Mtb-specific IgG levels were increased only after the development of the necrotic granuloma. Mtb genotyping also did not correlate with any known new clusters of TB in North America in the recent past (data not shown). Collectively, these data show that the development of TB following PD-1 blockade in this individual was selectively associated with increases in Mtb-specific Th1 responses.

Conclusion

Three previous cases of TB have been reported following PD-1 blockade. In this new case, Mtb-specific Th1 responses increased after PD-1 blockade was initiated, and clinical TB arose subsequently. Importantly, this nodule was assumed to be an MCC metastasis, and would not have been recognized as due to Mtb if an excisional biopsy had not been performed. In conjunction with animal model data suggesting a plausible mechanism, and the prior reported cases, these findings suggest that Mtb is possibly a concern following PD-1 blockade.