Utility of a circulating tumor DNA test for detecting clinically evident and occult Merkel cell carcinoma

Tomekio Takadaa, Niiru Soii1, Daniel S Hippe1, Lindsay Gunnell1, Coley Doolittle-Amireva1, Alexandra Ikeguchi3, Sajeve S Thomas4, Ann W Silk5, Dmitri Milademetan is a highly potent MDM2 inhibitor in TP53 wild-type (p53 WT) models

Vania Asumugha1,2, Thomas C Feast1,2,3, Kari M Sonoko3, Aime Knuth3, Brianna Magdanz4, Margot C Gaskill3,4, Vijaia Tinuguna3, Robert D Coebele5 and James A DelCappio6, 1 Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, MA, 2 Department of Medicine, Brigham and Women’s Hospital and Harvard Medical School, Boston, MA, 3 Program in Virology, Graduate School of Arts and Sciences, Harvard University, Cambridge, MA, 4 Experimental Therapeutics Core at Dana Farber Cancer Institute, Boston, MA and 5 Rain Therapeutics, Newark, CA. Background: Merkel cell polyomavirus (MCPyV)-related MCC is a highly common and lethal skin cancer. Current treatment strategies are required for the remaining patients with primary or acquired resistance who do not benefit from PD-1/PD-L1 blockade or from checkpoint therapy. Here, we present a phase 2 open-label study of Milademetan in previously treated metastatic MCC patients. Methods: Patients with previously treated metastatic MCC with evidence of progression on any line of therapy received Milademetan in a dose-escalation study (0.1, 0.5, 1, 3, and 5 mg/kg Q3W) based on MCPyV oncoprotein serology testing; however, this test can only be made in half of MCC patients who produce such antibodies. We assessed whether circulating tumor DNA (ctDNA) can accurately detect MCPyV-oncogene-positive (VP) and virus-negative (VN) MCC patients. Methods: We used the SignatureTM platform (bespoke mpGR NGS-based test) in which tumor-specific, clonal single nucleotide variants (SNVs) are identified from tumor whole genome sequencing. Blood was interrogated for these mutations. Patients at the University of Washington, and Stanford, were recruited to serially give blood every 3 months for ctDNA testing. Results: Since April 2020, longitudinal cTDA samples were collected from 120 MCC patients. Of these, 47 had clinically evident MCC, and 73 did not. Of the 47 with clinically evident MCC, all had a positive ctDNA test sensitivity: 100%, (95% CI: 89-100%). Among the 6 remaining patients with a positive ctDNA test, 2 had independent evidence of early recurrent disease based on the marked elevation of Merkel cell polyoma virus (MCPyV)-isolation. Conclusions: We identified ctDNA as a minimally invasive test for the detection of occult MCC, which can aid in the selection of patients for clinical trials of Milademetan; for example, to evaluate its potential in combination with PD-1/L1 inhibitors. Milademetan is a promising drug effective against p53 WT MCC cell lines, xenograft, and PDX models. These results provide evidence for clinical exploration of milademetan in MCC refractory to current therapies. Immune-checkpoint inhibitors (ICIs) have transformed the treatment landscape of advanced MCC. Despite these advances, predicting response to PD-1/PD-L1 therapy is a significant barrier. We sought to identify a clinical and molecular signature associated with PD-1/PD-L1 resistance. Patients and Methods: We collected clinical data from 5 patients who failed first-line ICI therapy. We extracted whole blood DNA and performed Next-Generation Sequencing (NGS) to detect tumor-specific somatic mutations. Novel targeted therapy options for PD-1/PD-L1 resistant MCC patients are required. We present a case study with 5 patients to explore potential targets for future therapy. Case 1: An 80-year-old woman presented with a large MCC metastatic to the liver (mMCC). Metastasectomy was performed, followed by adjuvant PD-1 therapy with pembrolizumab resulting in complete response. After 3 cycles of pembrolizumab therapy, the patient discontinued treatment due to toxicity. A biopsy of a new liver lesion showed mCRPC with acquired resistance to PD-1/L1 therapy. The acquired resistance was due to the somatic TP53 mutation G109S, a KRAS mutation and the amplification of the oncogene HRAS. Case 2: A 72-year-old man presented with a large tumor involving the entire scalp, face, neck, and right ear. He was treated with pembrolizumab with partial response, suggesting PD-L1 expression and sensitivity to PD-1/L1 therapy. A biopsy of the recurrent tumor showed MYH11 rearrangement (patient 2), a novel mechanism of resistance to PD-1/L1 therapy. Case 3: A 75-year-old woman presented with multiple skin nodules and liver metastases who responded well to first-line pembrolizumab. A biopsy of the liver metastasis showed NTRK3 amplification, a novel mechanism of resistance to PD-1/L1 therapy. Case 4: A 68-year-old woman presented with a large MCC metastatic to the liver (mMCC). She was treated with pembrolizumab with partial response, suggesting PD-L1 expression and sensitivity to PD-1/L1 therapy. A biopsy of the recurrent tumor showed fibroblast activation protein (FAP) positivity, a novel mechanism of resistance to PD-1/L1 therapy. Case 5: A 72-year-old man presented with a large MCC metastatic to the liver (mMCC). He was treated with pembrolizumab with partial response, suggesting PD-L1 expression and sensitivity to PD-1/L1 therapy. A biopsy of the recurrent tumor showed a CpG island methylator phenotype (CIMP), a novel mechanism of resistance to PD-1/L1 therapy. Conclusions: Preclinical models have identified several mechanisms of resistance to PD-1/L1 therapy, including tumor heterogeneity, acquired resistance, and senescence. These mechanisms may be further highlighted the role of ICIs in MCC anagis. In addition, in vitro tube formation assay pro-vided evidence that the growth-promoting activity of ICI-mediated killers is mediated by the CD4+ T cells. Therefore, we hypothesize that MCC patients may have multiple mechanisms of resistance to PD-1/L1 therapy. We investigated the role of immune-checkpoint inhibitors in MCC patients. Results: We identified 2 patients with PD-1/L1 resistance who had evidence of early recurrent disease. Conclusions: We identified two patients with PD-1/L1 resistance who had evidence of early recurrent disease.