After 48 hrs, supernatants (SUPs) were harvested and cytokine content quantified by ELISA. Pretreated with a CD126-IL-6 chimeric molecule and washed x 4 prior to setting up cultures. Was set up identically except that LCs were pretreated with medium alone. (3) Cultures were responsible for these observations, cultures were set up as follows: (1) BALB/c LCs were treated for 3 hrs with IL-6, then washed x 4 and co-cultured with D101 T 10 cells (which spontaneously respond to CD2-CD40L). BALB/c LCs and D101 T 10 cells (2) This group was set up identically except that LCs were pretreated with medium alone. (3) Cultures were pretreated with a CD126-IL-6 chimeric molecule and washed x 4 prior to setting up cultures. In this regard, pre-exposure of LCs to IL-6, but not pre-exposure of T cells, biased the outcome of Ag presentation in this manner. Exposure of LCs to anti-CD126 antibodies before and during IL-6 treatment inhibited much of this effect, indicating that the IL-6 receptor a-chain on LCs is involved in this process. Also, exposure of LCs to IL-6 upregulated LC IL-6 production. To examine if IL-6 trans-presentation by CD126 on LCs to T cells is responsible for these observations, cultures were set up as follows: (1) BALB/c LCs were treated for 3 hrs with IL-6, then washed x 4 and co-cultured with D101 T 10 cells (which spontaneously respond to CD2-CD40L). BALB/c LCs and D101 T 10 cells (2) This group was set up identically except that LCs were pretreated with medium alone. (3) Cultures were pretreated with a CD126-IL-6 chimeric molecule and washed x 4 prior to setting up cultures. After 48 hrs, supernatants (SUPs) were harvested and cytokine content quantified by ELISA.

In vitro genetic reprogramming increases MHC-I expression and ameliorates resistance to an antitumor immune response in Merkel cell carcinoma

M Kudy, J Green, Y Tzeng, and JC Sunshine* Biomedical Engineering, Johns Hopkins University, Baltimore, Maryland, United States and 2 Dermatology, Johns Hopkins University, Baltimore, Maryland, United States

Merkel cell carcinoma (MCC) is a rare but aggressive skin cancer with half of patients unresponsive to immune checkpoint inhibitors (ICIs). A primary resistance mechanism driving IC1 resistance in MCC in particular and cancer immune evasion in general is downregulation of tumor MHC class I (MHC-I) expression, thereby limiting cytotoxic cellular immune responses. Assessment of three patient-derived MCC cell lines, MCC13, MCC26, and UISO, demonstrated low baseline MHC-I expression in 2 of the 3 cell lines (MCC13 and UISO), with significantly increased IL-17A content compared to control cultures. Cultures in which T cells were pretreated with the chimeric molecule showed similar changes in IFNY and IL-17A production. These results strongly indicate that the effect of IL-6 treatment of LCs on biasing the outcome of Ag presentation results from trans-presentation of IL-6 by LCs to responding T cells.

Preliminary results show that MCC cell lines, with high expression of MHC-I, can be treated with IL-6 and then activate T cells through the MHC-I/TCR interaction. These results suggest that IL-6 can be used as a therapeutic agent to enhance the efficacy of MCC immunotherapy.

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