Background: Immunotherapy for the treatment of various cancers has substantially improved the clinical outcome for many patients. Yet, some cancers classified as immunologically “cold”, including high-risk neuroblastoma (HR-NBL) and melanoma (MEL), still have a poor response to immunotherapeutic intervention. These tumors are characterized by low tumor mutation burden, poor immune infiltrate, and/or low MHC-I expression. As a potential route of immune escape, these cold NBL/MEL tumors may have induced epigenetic modifications to regulate expression of MHC-I/II. Here, we investigated the ability of epigenetic modifiers (EMs), including inhibitors of DNA methyltransferases (DNMTis), histone deacetylases (HDACis), and histone methyltransferases (HMTis), to restore MHC-I/II expression in murine and human NBL/MEL models.

Methods: To determine the doses of various EM inhibitors (EMis) (n=8), human and murine NBL/MEL cell lines (n=14) were treated with various concentrations of EMis and monitored for proliferation and apoptosis via an Incucyte. Optimal doses were then used to treat NBL/MEL cells with single-agent EMis, or combinations, +/- IFNγ. Following treatment, cells were assessed by qPCR and flow cytometry for changes in MHC-I/II, PD-L1, and other genes [e.g., antigen presenting machinery (APM)].

Results: With increased doses of EMis, we observed reduced proliferation and increased apoptosis across the NBL/MEL cell lines. Using doses of EMis that we found did not alter proliferation or apoptosis (in an effort to focus on the potential immune modulation), we found that certain combinations of EMis with IFNγ restored MHC-I/II surface expression. Accordingly, we observed increased transcription of genes involved in the APM and chemokines known to influence immune cell infiltration, following guadecitabine (DNMTi) and entinostat (HDACi) treatment. Moreover, the combination of guadecitabine and entinostat resulted in co-expression of MHC-I and MHC-II on several of the cell lines tested.

Conclusions: These findings suggest that certain combinations of EMis may allow us to turn these cold NBL/MEL tumors “hot” by reversing the loss of MHC-I/II. By combining guadecitabine and entinostat with current immunotherapeutic regimens, there is potential to reinvigorate the activity of T cells in the anti-tumor response through T cell engagement with MHC-II. Future studies are aimed to investigate the in vivo ability of guadecitabine and entinostat to restore MHC-I/II expression.