TRIPLE THERAPY (BINIMETINIB, BEVACIZUMAB, PEMBROLIZUMAB) MODULATES THE TUMOR IMMUNE MICROENVIRONMENT IN MICROSATELLITE STABLE COLORECTAL CANCER PDXS IN A HUMANIZED MICE MODEL

Julie Lang*, Adrian Dominquez, Matthew Lewis, Sarah Hartmann, Jordi Lanis, Hannah Strasburger, Wells Messersmith, S. Lindsey Davis, Alexis Leal, Sunnie Kim, Tyler Friedrich Kimberly, Christopher Liu, Todd Pitts. University of Colorado Denver AMC, Aurora, CO, United States

Background Single-agent checkpoint blockade inhibitors have shown great promise in recent years but has unfortunately been limited to a subset of tumor types. The tumor mutation burden as well as the tumor microenvironment are two key important features that distinguish immunogenic, from non-immunogenic tumors. There is currently a massive effort, including a wide range of strategies, turning tumors from cold to hot including combining with small molecule inhibitors.

For colorectal (CRC) cancers, anti-PD1 therapies have proven to be successful in the clinic for microsatellite instable (MSI-high) tumors but have no efficacy in the majority of patients with CRC that have microsatellite stable (MSS) cancers. Our primary goal is to determine if the addition of binimetinib (small molecule MEK inhibitor) and bevacizumab (anti-VEGF therapeutic antibody) increases the response rate of metastatic microsatellite stable colorectal cancer to pembrolizumab in humanized patient-derived xenografts (hPDXs) of CRC. We will also identify immune and pathway modulation in MSS CRC hPDXs treated with the combination of MEK, VEGF, and immune checkpoint inhibitors.

Methods In six independent experiments, we implanted distinct MSS CRC PDXs, that were recently isolated from patients on a matching clinical trial, into the flanks of humanized BRGS (BALB/c, Rag2-/-, IL2RgC-/-, NODSIRPa) mice that had been engrafted with human hematopoietic stem cells at birth. For each PDX we generated humanized mice cohorts treated with vehicle, binimetinib, binimetinib/pembrolizumab combination, or binimetinib/pembrolizumab/bevacizumab/DC101. The human immune system in the immune organs and tumors were interrogated by flow cytometry to assess changes in the cellular composition and the activation state of the immune system as a result of treatments, and the expression of immune-related molecules were assessed on the tumor cells.

Results There were no significant differences in primary tumor growth in all treated models. However, immune modulation was observed in TILs in which we measured increased activated T cells (DR+, effector memory, TIM-3+), GrB+CD8+ and IFNg+CD8+ T cells. We also observed increased TNFa and IFNg and decreased T regulatory (FoxP3+CD25+) CD4+ T cells in the pembrolizumab/binimetinib and triple combination groups. Immune infiltrates were unique for the various PDXs.

Conclusions In this preclinical examination of combination MEK, VEGF, and PD-1 inhibition in CRC hPDXs no significant differences in tumor growth were noted, but immune modulation in TILs and tumor were observed suggesting potential immune modulation of the tumor microenvironment that may lead to greater susceptibility to immune checkpoint inhibition in patients with MSS mCRC.

Ethics Approval The human cord blood samples were generously provided as de-identified donors from Clinimmune Cord Blood Bank (Aurora, CO). All procedures and mouse husbandry were performed in accordance with IACUC protocols approved by the University of Colorado Denver Institutional Animal Care and Use Committee in the Office of Laboratory of Animal Resources (OLAR), a facility approved by the American Association for Laboratory Animal Care.