immune dysfunction, and provide a target for cancer therapeutics. Collagens are a primary component of the extracellular matrix. Abnormal levels of collagen and of the collagen-domain containing complement component 1q (C1q) in tumor microenvironments has been proposed to disrupt anti-tumor immunity. LAIR-1 is an adhesion molecule and inhibitory receptor expressed on the cell surface of several immune cell subsets. LAIR-1 binding to collagen-like domains present in collagens and C1q inhibit immune cell function. LAIR-2 is a soluble homolog of LAIR-1 that binds to and outcompetes LAIR-1 binding to collagens and C1q and serves as a natural decoy to promote immune function.

Methods Taking advantage of a natural decoy system, we designed a protein biologic, NC410, composed of LAIR-2 fused with a functional IgG1 Fc domain to target collagen-rich tumors and promote immune activation, infiltration and effector function.

Results NC410 has increased avidity due to Fc mediated dimerization, and blocks LAIR-1 interactions with ligands, and LAIR-1 signaling. In vivo administration of NC410 in humanized tumor models reduced tumor growth in a dose dependent fashion. NC410 increased the numbers of infiltrating human CD8+ and CD4+ T cells in the tumor, which is associated with increased levels of chemokines in the local tumor environment. Effector function was also enhanced, as denoted by increased levels of IFN-gamma and Granzyme B in the local tumor environment. In addition, NC410 increased specific collagen degradative products in the serum of humanized tumor-bearing mice, suggesting NC410 may promote tumor microenvironment remodeling and immune accessibility to further promote anti-tumor immunity.

Conclusions These data support NC410 as a novel therapeutic for targeting collagen-rich tumors and enabling normalization of the tumor-immune microenvironment. FIH studies have recently been initiated with NC410.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0694

695 ORAL DELIVERY OF A MICROBIAL EXTRACELLULAR VESICLE INDUCES POTENT ANTI-TUMOR IMMUNITY IN MICE

Loise Francisco-Anderson, Loise Francisco-Anderson, Mary Abdou, Michael Goldberg, Erin Troy, Alicia Ballok, Fabian Romano Chernac, Maria Sizova, Krutika Invally, Shubhra Kashyap, Audrey McBride, Jessica Tsang, Shannon Argueta, Kristie Barth, Valeria Kravitz, Holly Ponichtera, Tyler Rommel, Kevin Huynh, Tanmoy Ganguly, Mark Bodmer*, Mark Bodmer. *Evelo Biosciences, Cambridge, MA, USA*

Background The small intestinal axis (SINTAX) is a network of anatomic and functional connections between the small intestine and the rest of the body. It acts as an immunosurveillance system, integrating signals from the environment that affect physiological processes throughout the body. The impact of events in the gut in the control of tumor immunity is beginning to be appreciated. We have previously shown that an orally delivered single strain of commensal bacteria induces anti-tumor immunity preclinically via pattern recognition receptor-mediated activation of innate and adaptive immunity. Some bacteria produce extracellular vesicles (EVs) that share molecular content with the parent bacterium in a particle that is roughly 1/1000th the volume in a non-replicating form. We report here an orally-delivered and gut-restricted bacterial EV which potently attenuates tumor growth to a greater extent than whole bacteria or checkpoint inhibition.

Methods EDP1908 is a preparation of extracellular vesicles produced by a gram-stain negative strain of bacterium of the Oscillospiraceae family isolated from a human donor. EDP1908 was selected for its immunostimulatory profile in a screen of EVs from a range of distinct microbial strains. Its mechanism of action was determined by ex vivo analysis of the tumor microenvironment (TME) and by in vitro functional studies with murine and human cells.

Results Oral treatment of tumor-bearing mice with EDP1908 shows superior control of tumor growth compared to checkpoint inhibition (anti-PD-1) or an intact microbe. EDP1908 significantly increased the percentage of IFNγ and TNF producing CD8+ CTLs, NK cells, NKT cells and CD4+ cells in the tumor microenvironment (TME). EDP1908 also increased tumor-infiltrating dendritic cells (DC1 and DC2). Analysis of cytokines in the TME showed significant increases in IP-10 and IFNg production in mice treated with EDP1908, creating an environment conducive to the recruitment and activation of anti-tumor lymphocytes.

Conclusions This is the first report of striking anti-tumor effects of an orally delivered microbial extracellular vesicle. These data point to oral EVs as a new class of immunotherapeutic drugs. They are particularly effective at harnessing the biology of the small intestinal axis, acting locally on host cells in the gut to control distal immune responses within the TME. EDP1908 is in preclinical development for the treatment of cancer.

Ethics Approval Preclinical murine studies were conducted under the approval of the Avastus Preclinical Services' Ethics Board. Human in vitro samples were attained by approval of the IntegReview Ethics Board; informed consent was obtained from all subjects.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0695

696 BRENTUXIMAB VEDOTIN, A CD30-DIRECTED ANTIBODY-DRUG CONJUGATE, SELECTIVELY DEPLETES ACTIVATED TREGS IN VITRO AND IN VIVO

Bryan Grogan*, Reice James, Michelle Ulrich, Shyra Gardai, Ryan Heiser, Reice James. *Seattle Genetics, Inc., Bothell, WA, USA*

Background Regulatory T cells (Tregs) play an important role in maintaining immune homeostasis, preventing excessive inflammation in normal tissues. In cancer, Tregs hamper anti-tumor immunosurveillance and facilitate immune evasion. Selective targeting of intratumoral Tregs is a potentially promising treatment approach. Orthogonal evaluation of tumor-infiltrating lymphocytes (TILs) in solid tumors in mice and humans have identified CCR8, and several tumor necrosis family receptors (TNFRs), including TNFSFR8 (CD30), as receptors differentially upregulated on intratumoral Tregs compared to normal tissue Tregs and other intratumoral T cells, making these intriguing therapeutic targets.Brentuximab vedotin (BV) is approved for classical Hodgkin lymphoma (cHL) across multiple lines of therapy including frontline use in stage III/IV cHL in combination with doxorubicin, vinblastine, and dacarbazine. BV is also approved for certain CD30-expressing T-cell lymphomas. BV is comprised of a CD30-directed monoclonal antibody conjugated to the highly potent microtubule-disrupting agent monomethyl auristatin E (MMAE).The activity of BV in lymphomas is thought to primarily result from tumor directed intracellular MMAE release, leading to mitotic arrest and apoptotic cell death.The role CD30 plays in normal

immune function is unclear, with both costimulatory and proapoptotic roles described. CD30 is transiently upregulated following activation of memory T cells and expression has been linked to highly activated/suppressive IRF4+ effector Tregs.
Methods Here we evaluated the activity of BV on CD30-expressing T cell subsets in vitro and in vivo.
Results Treatment of enriched T cell subsets with clinically relevant concentrations of BV drove selective depletion of CD30-expressing Tregs > CD30-expressingCD4+ T memory cells, with minimal effects on CD30-expressing CD8+ T memory cells. In a humanized xeno-GVHD model, treatment with BV selectively depleted Tregs resulting in accelerated wasting and robust T cell expansion. The observed differential activity on Tregs is likely attributable to significant increases in CD30 expression and reduced efflux pump activity relative to other T cell subsets. Interestingly, blockade of CD25 signaling prevents CD30 expression on T cell subsets without impacting proliferation, suggesting a link between CD25, the high affinity IL-2 receptor, and CD30 expression.
Conclusions Together, these data suggest that BV may have an immunomodulatory effect through selective depletion of highly suppressive CD30-expressing Tregs.
Acknowledgements The authors would like to thank Michael Harrison, PharmD for their assistance in abstract preparation.
Ethics Approval Animals studies were approved by and conducted in accordance with Seattle Genetics Institutional Care and Use Committee protocol #SGE-024.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0696

697 TUMOR-TARGETED CD28 COSTIMULATORY BISPECIFIC ANTIBODIES ENHANCE T CELL ACTIVATION IN SOLID TUMORS

Michael Hedvat*, Veronica Zeng, Juan Diaz, Christine Bonzon, Kendra Avery, Rumana Rashid, Irene Leung, Norm Bartlow, Charles Bakhit, Matthew Dragovich, Liz Bogart, Umesh Muchhal, John Desjarlais, Gregory Moore. *Xencor, Inc., Monrovia, CA, USA*

Background T cells in the tumor micro-environment require TCR/MHC engagement and co-stimulatory receptor engagement to achieve complete activation. Solid tumors often lack expression of CD28 ligands, so we hypothesized that activation of CD28 signaling could be beneficial in solid tumors. We designed tumor-associated-antigen (TAA) x CD28 bispecific antibodies that conditionally costimulate CD28 only in the presence of TAA and TCR engagement. Clinical application of this class of antibodies has potential to enhance activity of either anti-PD(L)1 antibodies or TAA x CD3 T cell engagers.
Methods We designed a stability and affinity optimized anti-CD28 antibody that can be paired with TAA of choice to engage CD28 monovalently using Xencor's XmAb 2+1 and 1+1 platforms. In vitro T cell activation with these bispecifics was measured by T cell proliferation, cytokine production, and cytotoxicity, in co-cultures of human cancer cell lines mixed with primary human CD3-stimulated T cells. In vitro activity was validated in a CMV recall assay measuring CMV+ T cell proliferation of CMV+ PBMC co-cultured with cancer cell lines ectopically treated with pp65-derived NLV-peptide. In vivo anti-tumor and T cell proliferative activity of B7H3 x CD28 bispecific antibodies were determined in tumor-bearing huPBMC-NSG mice treated simultaneously with TAA x CD3 bispecific antibody. In vivo activity of PDL1 x CD28 antibodies was determined with hCD28 KI mice inoculated with MC38 tumors expressing hPDL1-antigen. Finally, safety and tolerability of B7H3 x CD28 and PDL1 x CD28 was determined in cynomolgus monkeys.
Results B7H3 x CD28 and PDL1 x CD28 antibodies enhanced T cell degranulation, cytokine secretion, and cancer cell cytotoxicity in concert with CD3 stimulation only in the presence of target antigen. B7H3 x CD28, alone or in combination with anti-PD1 antibody, enhanced proliferation of CMV+ T cells recognizing cancer cells loaded with pp65-derived NLV peptide. PDL1 x CD28 also enhanced CMV+ cell expansion but did not synergize with anti-PD1 antibody treatment. B7H3 x CD28 significantly enhanced in vivo anti-tumor activity of TAA x CD3 antibodies while also promoting greater T cell expansion. In hCD28 mice inoculated with MC38 tumors expressing hPDL1, PDL1 x CD28 antibody inhibited tumor growth greater than an anti-PDL1 antibody alone. B7H3 x CD28 and PDL1 x CD28 were well tolerated in cynomolgus monkeys.
Conclusions B7H3 x CD28 and PDL1 x CD28 bispecific antibodies show promising anti-tumor activity and warrant further development.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0697

698 TARGETING HLA-G-MEDIATED IMMUNOSUPPRESSION WITH A FIRST-IN-CLASS ANTAGONIST ANTIBODY

Douglas Hodges*, Christina Kochel, Michael Totagrande, Jeffrey Jones, Megan Welch, Bradley Spatola, Shelby Joe, John Corbin, Vanessa Soros, Courtney Beers, Achim Moesta. *Tizona Therapeutics, South San Francisco, CA, USA*

Background Human leukocyte antigen-G (HLA-G) is an immune checkpoint molecule that belongs to the non-classical HLA-class I family of receptors. HLA-G restrains immune cell activation and effector function by engaging with inhibitory receptors ILT2 and ILT4. While expression of HLA-G is highly restricted under normal healthy conditions, we have demonstrated that its expression in cancer is aberrantly upregulated and broadly detected across a variety of tumor types. Tizona Therapeutics has generated a novel, fully human antibody that specifically targets HLA-G and reverses HLA-G-mediated immunosuppression. Here we present in vitro and in vivo data demonstrating the functional impact of HLA-G blockade on immune cells and evidence to support the use of TTX-080 in the clinic to treat patients with advanced solid tumors.
Methods Evaluation of HLA-G expression in cancer was performed using immunohistochemistry, flow cytometry, and gene profiling. Expression of ILT2 and ILT4 was assessed on tumor infiltrating leukocytes by flow cytometry. To demonstrate the suppressive function of HLA-G, primary human NK cells, T cells, and monocyte-derived macrophages were cultured with target cells expressing HLA-G. TTX-080 was then evaluated for its ability to reverse this suppression. In addition, TTX-080 was investigated in vivo using a disseminated xenograft tumor model.
Results Expression of HLA-G was detected on tumor cells and tumor infiltrating leukocytes across a variety of solid tumor types. TTX-080 blocked interaction of HLA-G with both ILT2 and ILT4 and restored cytotoxicity in multiple assays using either primary NK cells or NKL cell lines. Monocyte-derived macrophages expressing ILT2 and ILT4 exhibited decreased phagocytosis of HLA-G^{+} target cells; this inhibition was reversed with an antigen-binding fragment of TTX-080. TTX-