blockade. Soluble LAG-3 may be an important biomarker for monitoring the pharmacodynamic activity of FS118 in patients.

Ethics Approval All animal experiments were conducted under a UK Home Office Project Licence and approved by an Animal Welfare and Ethical Review Board (AWERB) in accordance with the UK Animal (Scientific Procedures) Act 1986 and with EU Directive EU 86/609

REFERENCE

1. Kraman M, Faroudi M, Allen N, Kmiecik K, Gliddon D, Seal C, Koers A, Wydro M, Winnewisser J, Young L, Tuna M, Doody J, Morrow M, Brewis N. FS118, a Bispecific Antibody Targeting LAG-3 and PD-L1, Enhances T-Cell Activation Resulting in Potent Antitumor Activity. *Clin Cancer Res* 2020;**26**:3333–3344

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0715

716 CELL-BASED VIROTHERAPY FOR TARGETING CANCERS

[1]Duong Nguyen, [1]Alberto Gomez, [1]Forrest Neuharth, [1]Ashley Alamillo, [2]Thomas Herrmann, [2]Barbara Härtl, [2]Laura Schneider, [1]Ivelina Minev, [1]Boris Minev, [1]Dobrin Draganov*, [1]Antonio Santidrian. *[1]Calidi Biotherapeutics, San Diego, CA, USA; [2]StemVac GmbH, Bernried, Germany*

Background Oncolytic virotherapy has been recognized as a promising new therapy for cancer for decades but only few viruses have been approved worldwide. The therapeutic potential of oncolytic viruses can be severely restricted by innate and adaptive immune barriers making oncolytic virus clinically inefficient. To overcome this obstacle, we utilized adipose-derived stem cells (AD-MSC) loaded with tumor selective CAL1 oncolytic vaccinia virus to generate a new therapeutic agent called SNV1 (SuperNova-1).

Methods CAL1 vaccinia virus was tested for its ability to replicate and selectively kill various human cancer cell lines in vitro and in vivo. Additionally, CAL1 was loaded into adipose-derived mesenchymal stem cells to generate SuperNova1 (SNV1). Both CAL1 and SNV1 were tested for their ability to kill cancer cells in the presence of active complement and neutralizing antibodies in cell culture as well as in mice. Immune cell infiltration of the treated and untreated tumors was analyzed by flow cytometry.

Results CAL1 showed preferential amplification and killed various tested human (PC3, FaDu, MDA-MB-231, RPMI) and mouse cancer cells (CT26, EMT6, TRAMP-C2, RM1). In animals, CAL1 caused tumor regression in PC3 and CT26 mouse models without signs of toxicity. SNV1 significantly enhanced protection of CAL1 virus from clearance by the immune system as compared to naked CAL1 virus, leading to higher therapeutic efficacy in animals. Five days after SNV1 administration, tumor infiltrating lymphocytes (TILs) from both treated and untreated tumors showed increased CD4 and CD8 T-cell infiltrations. Importantly, we documented a decreased frequency of Tregs, and improved effector to Treg ratios, which was associated with inhibition of tumor growth at the treated tumor site and also at distant untreated sites.

Conclusions CAL1 is potentially used as an oncolytic agent. In addition, SNV1 cell-based platform protects and potentiates oncolytic vaccinia virus by circumventing humoral innate and adaptive immune barriers, resulting in enhanced oncolytic virotherapy. Particularly, SNV1 provided instantly active viral particles for immediate infection and simultaneous release of therapeutic proteins in the injected tumors.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0716

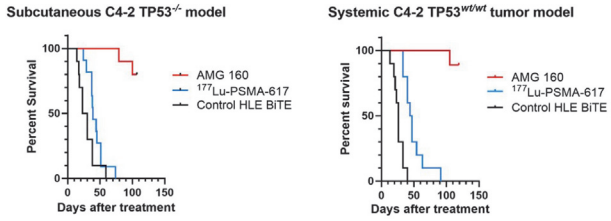
717 AMG 160, A PROSTATE-SPECIFIC MEMBRANE ANTIGEN (PSMA)-TARGETED BITE® IMMUNO-ONCOLOGY THERAPY, IS ACTIVE IN MODELS OF ADVANCED PROSTATE CANCER THAT ARE RESISTANT TO RADIOLIGAND THERAPY

[1]Katharina Lueckerath*, [2]Julie Bailis, [1]Kyle Current, [2]Mark Salvati, [1]Caius Radu, [1]Johannes Czernin. *[1]David Geffen School of Medicine at UCLA, Los Angeles, CA, USA; [2]Amgen Inc., San Francisco, CA, USA*

Background AMG 160 is an HLE BiTE® (half-life extended bispecific T-cell engager) that binds PSMA on prostate cancer cells and CD3 on T-cells and induces redirected T-cell lysis of PSMA-expressing cells. This mechanism may allow the BiTE molecule to be active in settings where other targeted or immune therapies have failed. Here, we evaluated the activity of AMG 160 in mouse models of advanced prostate cancer that are resistant to ^{177}Lu-PSMA-617, a PSMA-targeted radioligand therapy that has emerged as a promising treatment modality for metastatic castration-resistant prostate cancer (mCRPC).

Methods Two prostate cancer models were tested in 6–8-week-old male NCG mice: one cohort had established subcutaneous C4-2 TP53$^{-/-}$ tumors (C4-2 cells with TP53 knockout), and the other cohort had established systemic C4-2 TP53$^{wt/wt}$tumors that mimic metastatic lesions (intracardiac injection). PSMA levels in both models (~255,000 PSMA/cell) are sufficient for tumor growth inhibition with ^{177}Lu-PSMA-617. Mice were administered a single intravenous (IV) infusion of human T-cells. Three days later, mice were treated with 1 cycle of ^{177}Lu-PSMA-617 (30 MBq, IV), or 3 weekly doses of AMG 160 (1 mg/kg, IV) or of a control HLE BiTE molecule (1 mg/kg, IV; target not expressed on C4-2 cells). Therapeutic efficacy was assessed by tumor burden measurements, time to progression (TTP), and survival.

Results In both prostate cancer models, AMG 160 treatment significantly improved disease control (figure 1). Median TTP was not reached in the AMG 160 group ($p<0.0001$), whereas it was 31d (^{177}Lu-PSMA-617) and 23.5d (control) in the subcutaneous model, and 68d (^{177}Lu-PSMA-617) and 50.5d (control) in the systemic model. Median survival was not reached in the AMG 160 group ($p<0.0001$); it was 39d (^{177}Lu-PSMA-617) and 26.5d (control) in the subcutaneous model, and 77d (^{177}Lu-PSMA-617) and 61d (control) in the systemic model. Following treatment with AMG 160, 2/10 mice with subcutaneous and 7/9 mice with systemic tumors had not progressed at the end of the observation period (>100 days). In contrast, all mice in the ^{177}Lu-PSMA-617 and control groups succumbed to progressive disease.



Abstract 717 Figure 1 AMG 160 treatment extended survival in mouse models of advanced prostate cancer

Conclusions Our study demonstrates potent antitumor activity of AMG 160 monotherapy in models of metastatic CRPC that

are resistant to PSMA-targeted radioligand therapy with ^{177}Lu-PSMA-617. These data provide a rationale for evaluating AMG 160 in patients with mCRPC who have progressed on ^{177}Lu-PSMA-617. AMG 160 is currently being evaluated in a phase 1 study in patients with mCRPC (NCT03792841).

Acknowledgements The authors would like to acknowledge Micah Robinson, PhD of Amgen Inc. for medical writing support and Hosein Kouros-Mehr, MD, PhD of Amgen Inc. for facilitating the collaboration and helpful discussions.

Trial Registration ClinicalTrials. gov Identifier: NCT03792841

Ethics Approval All animal experimental protocols were approved by the UCLA Animal Research Committee (# 2005-090).

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0717

718 AMG 509, A STEAP1 X CD3 BISPECIFIC XMAB® 2+1 IMMUNE THERAPY, EXHIBITS AVIDITY-DRIVEN BINDING AND PREFERENTIAL KILLING OF HIGH STEAP1-EXPRESSING PROSTATE AND EWING SARCOMA CANCER CELLS

[1]Cong Li, [1]Madeline Fort, [1]Lingming Liang, [2]Gregory Moore, [2]Matthew Bernett, [2]Umesh Muchhal, [1]Tao Osgood, [1]Rodolfo Yabut, [1]Sarav Kaliyaperumal, [1]John Harrold, [1]Jude Canon, [3]Anna Rogojina, [3]Raushan Kurmasheva, [2]John Desjarlais, [3]Peter Houghton, Olivier Nolan-Stevaux*. *[1]Amgen Inc., South San Francisco, CA, USA; [2]Xencor Inc, Monrovia, CA, USA; [3]University of Texas Health San Antonio, San Antonio, TX, USA*

Background Metastatic castration-resistant prostate cancer (mCRPC) and Ewing sarcoma (EWS) are diseases for which immune therapies could potentially provide benefit. STEAP1 (Six Transmembrane Epithelial Antigen of the Prostate 1) is a cell surface protein with elevated expression in mCRPC [1] and EWS.[2]

Methods We designed AMG 509, a novel, half-life extended, STEAP1 x CD3 XmAb® 2+1 bispecific antibody to induce T cell-mediated cytotoxicity against STEAP1-expressing cancer cells. AMG 509 contains two identical anti-STEAP1 Fab domains, an anti-CD3 scFv domain, and an effectorless Fc domain that extends serum half-life. We characterized STEAP1 expression in normal and tumor tissues by immunohistochemistry, and we assessed the pharmacological properties of AMG 509 including binding, T cell-mediated redirected lysis, and in vivo antitumor activity.

Results We detected high STEAP1 surface expression on 80% of primary prostate tumors (n=88), 89% of mCRPC lesions (n=114), including 84% of mCRPC bone metastases (n=31), and 63% of EWS samples (n=35). In contrast, in normal tissues (n=72), low STEAP1 expression was detected in only six other tissues, including the normal prostate. AMG 509 bound to recombinant human CD3ϵ with a K_D of 27.6 nM, and it bound specifically to 293T cells transfected with human STEAP1 with an EC_{50} of 3.8 nM. AMG 509 triggered potent T cell-redirected lysis of STEAP1-positive cancer cells, with a median EC_{50} of 37 pM across 19 cancer cell lines that endogenously express various levels of STEAP1. AMG 509-mediated cytotoxicity was specific, as it showed no activity against prostate cancer cells in which STEAP1 was knocked out. AMG 509 was 65-fold more potent in inducing the redirected lysis of prostate cancer cells in vitro than an XmAb® molecule with a single anti-STEAP1 Fab domain. AMG 509 had greater cytotoxic activity against high STEAP1-expressing cancer cells than against low STEAP1-expressing cancer cells, and it had minimal activity against normal cells. This preferential killing of high STEAP1-expressing cells is likely driven by the avidity conferred by the dual STEAP1-binding domains, a feature that may help reduce off-target effects in the clinic. In vivo, AMG 509 induced robust anti-tumor activity in prostate cancer and EWS mouse xenograft models, with concomitant CD8+ T-cell activation and expansion in tumors.

Conclusions AMG 509 is a specific, first-in-class T cell-recruiting antibody with avidity-driven activity against STEAP1-positive malignancies. AMG 509 is currently being evaluated for safety, pharmacokinetics, and efficacy in a phase 1, first-in-human study in patients with mCRPC (NCT04221542).

Acknowledgements The authors acknowledge Micah Robinson, PhD of Amgen Inc. for medical writing support.

Trial Registration ClinicalTrials. gov Identifier: NCT04221542

Ethics Approval All animal experimental protocols were approved by an Institutional Animal Care and Use Committee (IACUC protocol number 2015-01243) and were conducted in accordance with the guidelines of the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) (Amgen) or the standards of the Public Health Service (PHS) Policy on Humane Care and Use of Laboratory Animals (IACUC protocol number 15015x) in a facility certified with an Office of Laboratory Animal Welfare (OLAW) (UTHSA).

REFERENCES

1. Grunewald TGP, Ranft A, Esposito I, Silva-Buttkus P da, Aichler M, Baumhoer D, Schaefer KL, Ottaviano L, Poremba C, Jundt G, Jurgens H, Dirksen U, Richter GHS, Burdach S. High STEAP1 expression is associated with improved outcome of Ewing's sarcoma patients. *Ann Oncol* 2012; **23**:2185–2190.
2. Hubert RS, Vivanco I, Chen E, Rastegar S, Leong K, Mitchell SC, Madraswala R, Zhou Y, Kuo J, Raitano AB, Jakobvits A, Saffran SC, Afar DE. STEAP: a prostate-specific cell-surface antigen highly expressed in human prostate tumors. *Proc Natl Acad Sci USA* 1999;**96**:14523–14528.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0718

719 CD122-DIRECTED INTERLEUKIN-2 COMPLEXES AND αPD-L1 DIFFERENTIALLY REQUIRE INNATE AND ADAPTIVE IMMUNITY TO TREAT LOCAL AND METASTATIC BLADDER CANCER

Ryan Reyes*, Yilun Deng, Deyi Zhang, Niannian Ji, Neelam Mukherjee, Karen Wheeler, Harshita Gupta, Aravind Kancharla, Myrna Garcia, Anand Kornepati, Robert Svatek, Tyler Curiel. *UT Health San Antonio, San Antonio, TX, USA*

Background αPD-L1 bladder cancer (BC) immunotherapy is effective in <30% of cases.[1] To address the large αPD-L1-unresponsive subset of patients, we tested αIL-2/IL-2 complexes (IL-2c) that block IL-2 from binding high-affinity IL-2Rα (CD25) for preferential IL-2Rβ (CD122) binding.[2] Immunosuppressive regulatory T cells capture IL-2 by CD25 whereas antitumor CD8+ T, γδ T, and NK cells use CD122. We hypothesized that the tumor microenvironment, including local immune cells in primary versus metastatic BC, differentially affects immunotherapy responses and that IL-2c effects could differ from, and thus complement αPD-L1.

Methods We used PD-L1+ mouse BC cell lines MB49 and MBT-2, for orthotopic, intravesical (i.e., in bladder) and intravenous challenge studies of local versus lung metastatic BC.

Results αPD-L1 or IL-2c alone reduced tumor burden and extended survival in local MB49 and MBT-2. Using in vivo cell depletions, we found that γδ T cells and NK cells, but strikingly not CD8+ T cells, were necessary for IL-2c efficacy in bladder. We confirmed γδ T cell requirements for IL-2c, but not αPD-L1 efficacy in γδ T cell-null TCRδKO mice. TCRβKO conventional T cell-null mice exhibited IL-2c, but