Preclinical Activity of BDC-4182, a Claudin 18.2-Targeting ISAC With Enhanced Potency and an Encouraging Safety Profile

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ABSTRACT # 1052

INTRODUCTION

Claudin (CLDN) 18.2 is a transmembrane tight junction protein with expression restricted to the gastric mucosal epithelia where CLDN18.2 protects against paracellular acid leakage and associated gastritis.[1] CLDN18.2 overexpression has been observed in several tumor types, including gastric, esophageal, and pancreatic cancer.[2,3] Loss of cell polarity in these tumors results in CLDN18.2 localization to surfaces that are more readily accessible to biologics and effector cells.[4] This expression pattern makes CLDN18.2 a compelling target for immune-stimulating antibody conjugates (ISACs) that combine the specificity of tumor-targeting antibodies with the strength of the immune system.[4,5]

BDC-4182 is an ISAC consisting of a CLDN18.2-targeting antibody covalently attached to a toll-like receptor (TLR)7/8 agonist via an intervening non-cleavable linker.[6] In preclinical models, systemic delivery of ISACs have been shown to broadly activate the innate and adaptive immune system, leading to complete tumor regression, immunologic memory, and epitope spreading. The proposed mechanism of action of BDC-4182 is depicted in **Figure 1**. Briefly, BDC-4182 binds to CLDN18.2-expressing tumor cells and triggers antibody-dependent cellular phagocytosis (ADCP) by dendritic cells (DCs) and other antigen presenting cells (APCs). Following phagocytosis, BDC-4182 localizes to the endosomal TLRs, whereby agonism of TLR7/8 drives APC activation, upregulation of antigen presenting machinery, and secretion of proinflammatory cytokines. These cytokines help activate and recruit immune effector cells to the tumor.[7-9] Activated APCs also process tumor-associated antigens and tumor neoantigens for cross-presentation to tumor-reactive T cells, which can result in a widespread and durable adaptive anti-tumor immune response.[5,6,10,11] This mechanism differs from cytotoxic antibody-drug conjugates that target individual tumor cells for destruction following internalization of the cytotoxic agent in CLDN18.2-expressing tumor cells.[12-14]

Figure 1. Boltbody™ ISAC Mechanism of Action

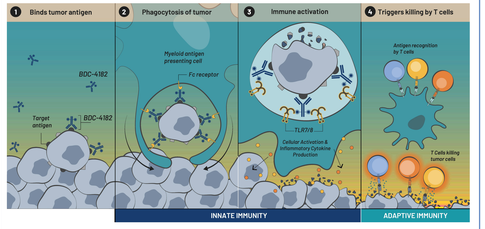
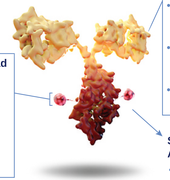


Figure 2. Key Features of BDC-4182



Immune-stimulating Payload
- Dramatically enhanced potency
- Tailored TLR specificity for key biology
- Optimized conjugation chemistry with non-cleavable linker

CLDN18.2-targeting Antibody
- Selected from a panel based on favorable in vitro and in vivo characteristics
- Subnanomolar affinity locates ISAC to antigen on surface of a tumor cell
- Active Fc region to trigger phagocytosis

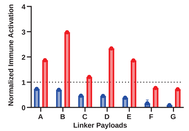
Significant Biologic Advantages
- Enhanced immune system activation
- Superior anti-tumor efficacy
- Maintains compelling safety profile

For illustrative purposes only.

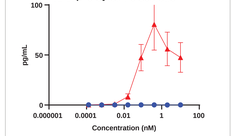
RESULTS

Figure 3. BDC-4182 Delivers Enhanced Immune Activation

P424 (CLDN18.2) ISACs Are More Potent Immune Activators Than Trastuzumab (HER2) ISACs



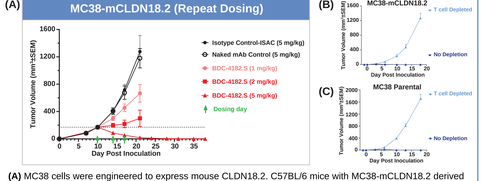
BDC-4182 Payload Has Superior Immune Activation Compared to BDC-1001 Payload



Trastuzumab (HER2) ISACs · P424 (CLDN18.2) ISACs · BDC-4182 · BDC-4182.S wi BDC-1001 Payload

(Left) Human cDCs from n=9 donors were co-cultured with Claudin 18.2-expressing PA-TU-8988S tumor cells (IHC 1+) or HER2 expressing cell line (HCC1954) and the indicated test articles for 18 hr. Cytokine secretion was measured by MSD and normalized to a control ISAC. **(Right)** Human cDC co-culture assays were run with CLDN18.2 expressing PA-TU-8988S tumor cells (IHC 1+). Cytokine secretion was measured 18 hr post incubation.

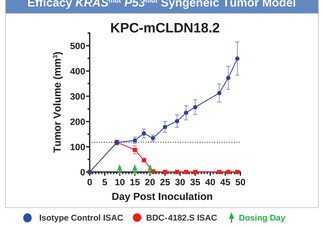
Figure 4. BDC-4182.S Inhibits Tumor Growth in Syngeneic Model

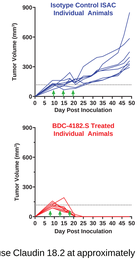


(A) MC38 cells were engineered to express mouse CLDN18.2. C57BL/6 mice with MC38-mCLDN18.2 derived tumors (N=8) were treated as indicated. In the 5 mg/kg group, all eight mice treated had no detectable tumors for > 4 weeks. Tumor free mice (28 days from final treatment with BDC-4182.S) were rechallenged with **(B)** MC38-mCLDN18.2 cells and **(C)** parental MC38 cells. Each mouse was subcutaneously inoculated with 1 million MC38-mCLDN18.2 cells on right flank and 1 million MC38 parental cells on left flank (N=8). To investigate the role of T cells, half of the mice were administered with depleting anti-CD4 and anti-CD8 antibodies (days -1, 1, 3, 7 and 10 post tumor inoculation). The depleting antibodies were given one day prior to inoculation of tumor cells. The rechallenged mice did not have detectable tumor growth for either cell lines, suggestive of epitope spreading. Depletion of T cells led to tumor growth, demonstrating a critical role of T cells in rejection and involvement of an adaptive immune response.

Figure 5. BDC-4182.S Elicits Complete Regression in Immunologically Cold KPC Model

Efficacy *KRAS*^{mut} *P53*^{mut} Syngeneic Tumor Model



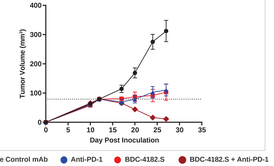


Isotype Control ISAC · BDC-4182.S ISAC · Dosing Day

KPC tumor model (*P53* mutant, *KRAS* mutant) was engineered to express mouse Claudin 18.2 at approximately 200K copies. Animal received three doses at 5 mg/kg. N=8 mice per group.

Figure 6. Combination of BDC-4182.S With Anti-PD1 Enhances *in vivo* Activity

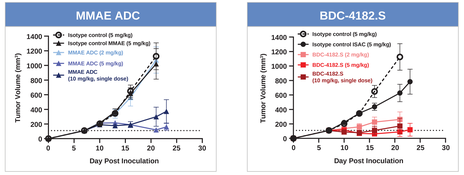
MC38 Claudin 18.2 Medium Syngeneic Model



Isotype Control mAb · Anti-PD-1 · BDC-4182.S · BDC-4182.S + Anti-PD-1

MC38 cells were engineered to express mouse CLDN18.2 (80k molecules/cell). Mice were treated as indicated when tumors reached an average of 100 mm³ (N=8). Mice were dosed with 2 mg/kg of BDC-4182.S and/or anti-PD-1 BIW for two weeks starting on day 12.

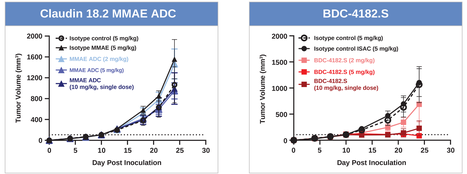
Figure 7. Activity Comparable to MMAE ADC in High CLDN18.2 Expressing Syngeneic Model



	10 mg/kg (1X)	5 mg/kg (4X)	2 mg/kg (4X)
MMAE ADC	71% TGI	88% TGI	-5% TGI
BDC-4182.S	73% TGI	86% TGI	58% TGI

The CLDN18.2 targeting antibody used on BDC-4182.S was conjugated to commercially available MMAE to a DAR4 (MMAE ADC) and tested in vivo. MC38 cells were engineered to express high level of mouse CLDN18.2 (200K molecules/cell). Treatment was initiated with 2 or 5 mg/kg BIW×4 or a single dose at 10 mg/kg when tumors reached ~100 mm³ (n=8 mice per group). TGI (%) was calculated by [1 – (mean volume of treated tumors)/(mean volume of isotype control tumors)] × 100.

Figure 8. Activity Superior to MMAE ADC in Medium CLDN18.2 Expressing Syngeneic Model

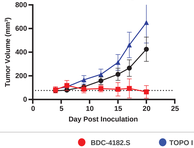


	10 mg/kg (1X)	5 mg/kg (4X)	2 mg/kg (4X)
MMAE ADC	36% TGI	39% TGI	6% TGI
BDC-4182.S	79% TGI	93% TGI	33% TGI

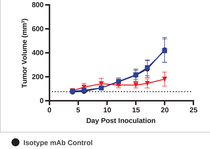
The CLDN18.2 targeting antibody used on BDC-4182.S was conjugated to MMAE to a DAR4 (MMAE ADC) and tested in vivo. MC38 cells were engineered to express medium level of mouse CLDN18.2 (80K molecules/cell). Treatment was initiated with 2 or 5 mg/kg BIW×4 or a single dose at 10 mg/kg when tumors reached ~100 mm³ (n=8 mice per group). TGI (%) was calculated by [1 – (mean volume of treated tumors)/(mean volume of isotype control tumors)] × 100.

Figure 9. BDC-4182.S Activity Superior to TOPO I ADC in Medium CLDN18.2 Expressing Syngeneic Model

5 mg/kg Dose Level



2 mg/kg Dose Level

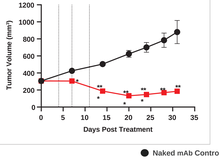


BDC-4182.S · TOPO I ADC · Isotype mAb Control

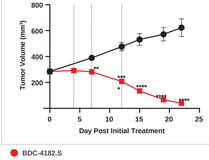
The CLDN18.2 targeting antibody used on BDC-4182.S was conjugated to deruxtecan to a DAR4 (TOPO I ADC) and tested in vivo. MC38 cells were engineered to express medium level of mouse CLDN18.2 (80K molecules/cell). Treatment was initiated with 2 or 5 mg/kg BIW×4 or a single dose at 10 mg/kg when tumors reached ~100 mm³ (n=8 mice per group). TGI (%) was calculated by [1 – (mean volume of treated tumors)/(mean volume of isotype control tumors)] × 100.

Figure 10. BDC-4182.S Inhibits Growth of Large Tumors

PA-TU-8988S (IHC1+)



NCI-N87-hCLDN18.2 (IHC1+)

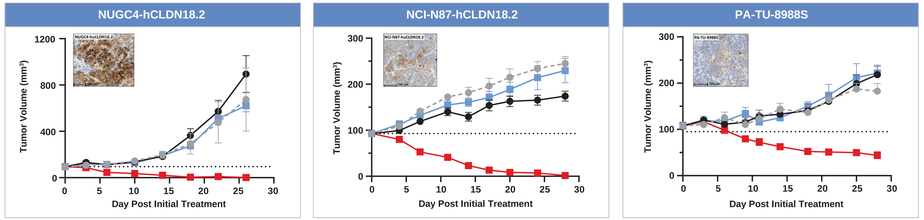


Naked mAb Control · BDC-4182.S

*P ≤0.05 **P ≤0.01 ***P ≤0.001 ****P ≤0.0001

SCID/beige mice bearing CLDN18.2 IHC1+ (A) PA-TU-8988S or (B) NCI-N87-hCLDN18.2 tumors were treated via IP with the indicated test articles at 5 mg/kg when tumors reached ~300 mm³. Animals received four doses in total (dashed lines). Data are shown as mean with SEM from n=4 mice per group and were analyzed by multiple unpaired t tests of the BDC-4182.S group compared to no treatment group. SCID/beige mice lack functional T, B and NK cells, underreporting the full activity of BDC-4182.S.

Figure 11. BDC-4182.S Inhibits Tumor Growth Across a Range of CLDN18.2 Expression



Isotype mAb Control · Isotype Control ISAC · Naked mAb Control · BDC-4182.S

	IHC Score	BDC-4182.S (TGI)	Tumor-Free Mice
NUGC4-hCLDN18.2	2+	100%	8 out of 8
NCI-N87-hCLDN18.2	1+	99%	6 out of 8
PA-TU-8988S	1+	76%	None

SCID/beige mice with NUGC4-hCLDN18.2, NCI-N87-hCLDN18.2, or PA-TU-8988S tumors (n=8 per group) were treated with BDC-4182.S at 5 mg/kg BIW (4 total doses). Tumor growth inhibition (TGI) was measured on the day 26 post initial treatment for NUGC4-hCLDN18.2 tumors and on day 28 for NCI-N87-hCLDN18.2 and PA-TU-8988S tumor models according to the following equation: %TGI = [1 – (mean volume of BDC-4182.S treated tumors) / (mean volume of isotype mAb treated tumors)] × 100. The number of tumor-free animals was recorded on the final day of the study.

BDC-4182 – Compelling Clinical Candidate Selected for Advancement

- **Robust activity in preclinical models**
  - Demonstrates superior efficacy compared to ADCs (MMAE and TOPO I)
  - Induces tumor regression in low Claudin 18.2 expressing tumors
  - Elicits epitope spreading and CD8-dependent immunologic memory
- **Tolerated in non-human primates at the highest dose tested (12 mg/kg)**
  - Findings are minor and generally transient and reversible.
  - No histological findings in the stomach.
  - Evidence of TLR7/8 activation (ie, CRP) and CLDN18.2 targeting
  - Favorable toxicology profile enables combination (eg, with chemotherapy and/or checkpoint regimens) used in first-line and second-line treatments
- **Clinical Trial Initiation in 2025**

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