Combining CD27 costimulation and PD-1 blockade into a bispecific antibody improves T cell activation and anti-tumor activity over combination of individual antibodies

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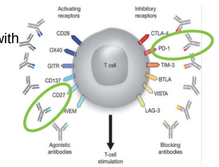
BACKGROUND

- This program builds from our experience with varlilumab (CD27 agonist mAb) in combination with nivolumab that demonstrated:
  - No additive toxicity concerns
  - Enhancement of tumor PD-L1 expression and CD8 T cells
  - Durable responses in patient populations unlikely to respond to PD-1 monotherapy
  - Best clinical activity observed with regimen that used similar doses (3 mg/kg) of each antibody administered on the same schedule



From Chen and Mellman, Immunity 2013

- CDX-527 is a bispecific antibody (BsAb) that combines blocking the PD-1 checkpoint pathway with CD27 costimulation of T cells
  - Designed from novel PD-L1 and CD27 antibodies
- Advantages of the BsAb include:
  - Cost and development advantages relative to 2 mAbs
  - Better CD27 agonist activity via PD-L1 cross-linking especially in tumor micorenvironment

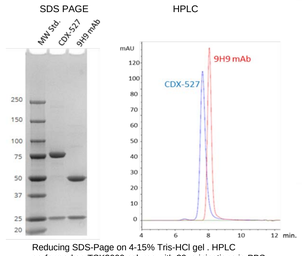
CDX-527 αPD-L1xαCD27 BsAb

Full length αPD-L1 mAb 9H9 (human IgG1κ) genetically linked to single chain variable domains of αCD27 mAb 2B3

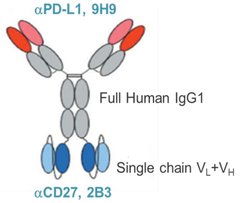
- Includes human Fc region as part of the BsAb construct
  - Retaining Fc receptor cross-linking for CD27 agonist activity
  - Retaining FcRn binding activity for antibody-like half-life (PK)
  - Enabling Protein A purification
- Tetravalent antigen binding
  - Bivalent for CD27 and PD-L1

CDX-527

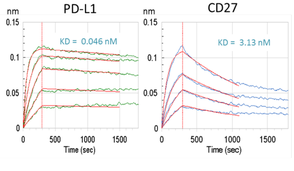
Antibody-like characteristics



Reducing SDS-Page on 4-15% Tris-HCl gel . HPLC performed on TSK3000 column with 20ug injections in PBS

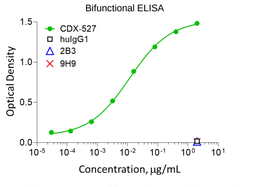


High Affinity to PD-L1 and CD27



Sensorgrams of bio-layer interferometry analysis using anti-human IgG-Fc sensors to capture CDX-527 followed by antigen

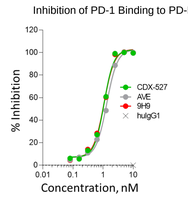
Bifunctional Binding to PD-L1 and CD27



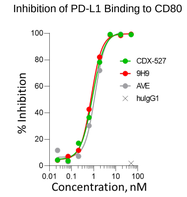
Wells coated with huCD27 and blocked, followed by Abs and then soluble msPD-L1-msFc. Detection by goat anti-ms HRP

Inhibition of PD-1 and CD80

CDX-527 Potently Inhibits PD-L1 binding to PD-1 or CD80

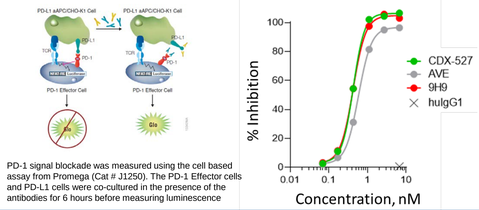


293-PD-L1 cells were incubated with samples and PD1-biotin. Binding detected with SA-PE. AVE = α-PD-L1 mAb Avelumab



Sample and PDL1-biotin incubated on CD80 coated plate. Binding detected with SA-HRP

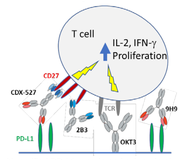
CDX-527 Efficiently Blocks PD-1 Signaling



PD-1 signal blockade was measured using the cell based assay from Promega (Cat # J1250). The PD-1 Effector cells and PD-L1 cells were co-cultured in the presence of the antibodies for 6 hours before measuring luminescence

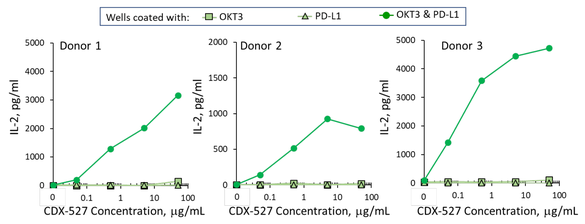
T Cell Co-Stimulation

Efficient T cell co-stimulation by CDX-527 through CD27 and TCR Signaling

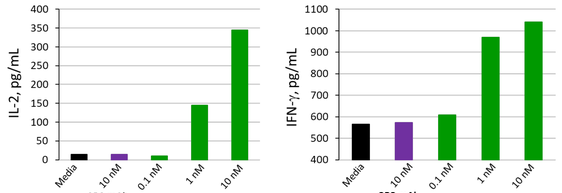


Wells are coated with purified PD-L1 protein and sub-optimal amount of anti-CD3 mAb (OKT3)
Antibodies are added, then purified T cells. IL-2/IFN-γ levels in supernatant are measured at 72 hrs

T Cell Activation With CDX-527 Requires Both PD-L1 and OKT3

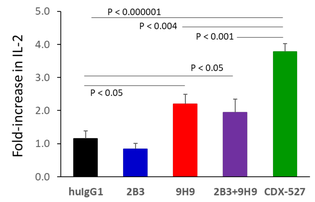


Combination of Parental Antibodies Does Not Provide Efficient T Cell Activation



Mixed Lymphocyte Reaction

CDX-527 is More Effective Than Parental Antibodies in MLR Activity



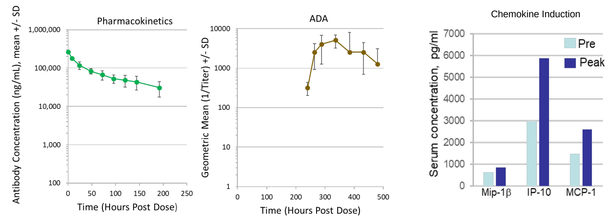
CD4 T cells and dendritic cells were prepared from independent PBMCs (n = 6). CD4 cells were incubated in the presence of allogeneic dendritic cells and mAbs or CDX-527 for 3 days. Supernatants were harvested and IL-2 levels were assessed by ELISA (R&D Systems).

Pilot Non-Human Primate Study

Mab-like pharmacokinetics and no toxicity signals in primate study

21 day study to assess pharmacokinetics and collect preliminary safety data

- Cynomolgus macaques (3) were administered a bolus injection of 7 mg/kg CDX-527
- No significant changes were observed in any clinical parameters:
  Clinical Observations, Body Weight, Body Temperature, Hematology, Coagulation, Clinical Chemistry, Urinalysis
- PK analysis suggest T1/2 ~ 110 hrs. Development of potent ADA obstructed PK assessment beyond 200 hrs
- Upregulation of chemokine levels associated with CD27 activation

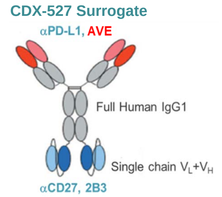


Serum levels of CDX-527 and ADA were determined by ELISA. Cytokines were measured by Luminex®. Baseline and highest on study level (peak) are shown

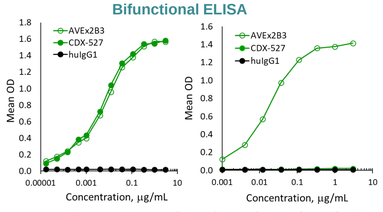
CDX-527 Surrogate BsAb

The CDX-527 mouse surrogate construct replaces the 9H9 PD-L1 CDRs with sequences from the PD-L1 mAb, avelumab (AVE). AVE binds to both human and mouse PD-L1. 2B3 does not cross-react with rodent CD27 but can be used in human CD27 transgenic mice.

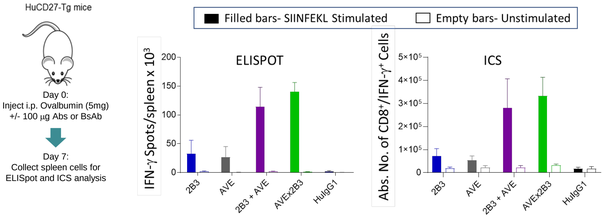
CDX-527 Surrogate



Bifunctional ELISA

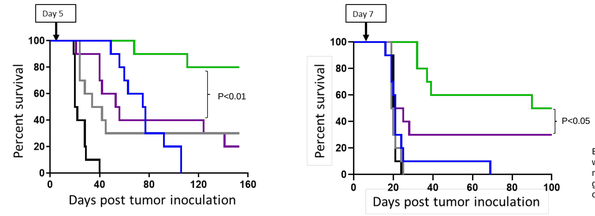


Potent Expansion of Vaccine Induced CD8 T Cell Response



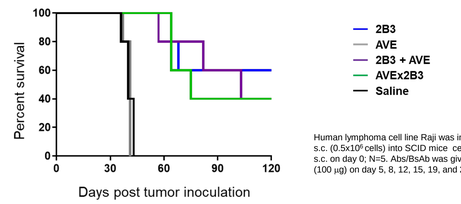
Anti-Tumor Activity

CDX-527 Surrogate is More Effective Than Parental Antibodies in BCL-1 Lymphoma Model



BCL-1 lymphoma cells (5 x 10^6) were injected i.v. into huCD27 Tg mice on day 0; Abs/BsAb were given as a single dose i.p. (200 µg) on day 5 or day 7

CDX-527 Surrogate has Direct Anti-Lymphoma Activity in Xenograft Model



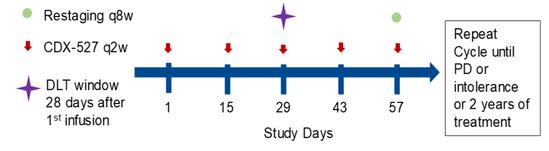
Human lymphoma cell line Raji was injected s.c. (0.5x10^6 cells) into SCID mice cell 0.5x10^6 s.c. on day 0; N=5, Abs/BsAb was given i.p. (100 µg) on day 5, 8, 12, 15, 19, and 22

Clinical Study Plans

Planned Dosing Cohorts

Study Portion	Cohort	CDX-527 Dose Level (mg/kg q2w)	Patients (n)
Dose-Escalation	1	0.3	3-6
Multiple tumor types	2	1.0	3-6
	3	3.0	3-6
	4	6.0	3-6
	5	10.0	3-6
Tumor-Specific Expansion Cohorts	6-9	Dose(s) chosen during escalation	Up to 15 per cohort

Study Schema



Summary and Next Steps

Bispecific antibodies (BsAbs) that engage two independent pathways involved in controlling immune responses to tumors are a rapidly growing area for the development of next generation PD-1 inhibitors

Our prior clinical experience with combining CD27 activation and PD-1 blockade provide the rationale for linking these two pathways into one molecule

The preclinical studies demonstrate that CDX-527 is more potent at T cell activation and anti-tumor immunity than the combination of parental monoclonal antibodies

Next steps for CDX-527 include:

- Completion of CDX-527 GMP manufacturing activities
- Completion of IND-enabling studies
- IND planned for H1 2020
- Phase 1 dose escalation trial

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