**CD40 Introduction**

CD40 is a cell surface glycoprotein in the TNF receptor (TNFR) family. CD40 is expressed constitutively and upregulated upon activation on antigen-presenting cells (APCs), including B cells, dendritic cells (DCs) and macrophages.

CD40 interacts with its ligand CD40L (aka CD154), which is rapidly induced on T cells following TCR activation, plays essential roles in the modulation of adaptive immunity. **

- Human responses:
  - B cell proliferation,
  - Immunoglobulin (Ig) production
  - Isotype switching
  - Memory B cell generation

- T-cell mediated licensing of APCs for antigen-presenting function:
  - Increased surface expression of MHC molecules
  - Upregulated costimulatory molecules to provide "second signals"
  - Release pro-inflammatory cytokines and chemokines to facilitate the polarization of Th1-type immune responses and cytotoxic T lymphocyte priming

- Enhancement of macrophage effector functions, such as phagocytosis

CD40 is expressed in a variety of malignancies, especially B cell lymphoma, being a target for antibody therapy.

**Role of CD40/CD40L in T Cell Activation**

- **CD40 Antibody for Lymphoma Therapy**
  - Stimulating CD40 signaling on B lymphoma can inhibit proliferation and promote apoptosis.
  - Engagement of CD40 on APCs can substitute for stimulation normally provided by helper T cells via CD40L, to activate and promote antitumor T-cell responses.
  - Triggering CD40 on myeloid cells, especially macrophages, can activate them with the potential to control malignancies in a T-cell-independent manner.
  - Targeting CD40 on B cells in a lymphoma with a depleting mAb can induce killing by ADCC, and ADCP.

- **CD40 mAb in combination** can sensitize other cancer-acting agents, including vaccine, cytokine drugs, etc.

- Dues to the broad expression profile of CD40 and the potency of this signaling pathway, toxicity is a critical challenging in exploiting CD40 target as antitumor therapeutics.

We set out to develop a panel of human CD40 mAbs with different levels of agonistic activity. A lead candidate, named CDX-1140, for systemic use is identified based on unique properties in agonist activity, anti-lymphoma activity in xenograft models and safety profile in non-human primates. Here we focus on the anti-lymphoma efficacy both in single agent properties in agonist activity, anti-lymphoma activity in xenograft models and safety profile.

**Potential Mechanisms of Action of Agonistic CD40 mAb on Various Immune Effectors**

- **CD40 Antigen Activity Is Fe-independent**
  - CDX-1140 binds to CD40 on human PBMCs and activates NFKB and B cell proliferation in a Fe-independent manner.

- **CDX-1140 Induces Cytokines Production**
  - The supernatant was analyzed for IL12p40 by ELISA.

- **CDX-1140 Synergizes with sCD40L**
  - CDX-1140 + Varil (Varil 5 mg/kg i.p. on day 1, 8 and 15)

- **CDX-1140 Synergizes with sCD40L**
  - CDX-1140 by itself or in combination with Varil can enhance anti-human IgG2 (specific to PE antibody).

**CD40 Introduction**

- A panel of anti-CD40 monoclonal antibodies (mAbs) were generated by immunization of non-human primate. Here we focus on the anti-lymphoma efficacy both in single agent properties in agonist activity, anti-lymphoma activity in xenograft models and safety profile.

- Targeting CD40 on B cell lymphoma with a depleting mAb can induce killing by ADCC, and ADCP.

- Triggering CD40 on myeloid cells, especially macrophages, can activate them with the potential to control malignancies in a T-cell-independent manner.

- Ligation of CD40 on APCs can substitute for stimulation normally provided by helper T cells via CD20 mAb.

- Elevation of macrophage effector functions, such as phagocytosis following TCR activation, plays essential roles in the modulation of adaptive immunity.

- **Potential Mechanisms of Action of Agonistic Immunotherapy**
  - Memory B-cell generation
  - Isotype switching

**Development and Characterization of CDX-1140**

- **CDX-1140 Anti-lymphoma Activity in Xenograph Models**
  - CDX-1140 was added and the binding to each antigen was detected

- **CDX-1140 Induces Cytokines Production**
  - Ramos cells 0.5x10^6 were s.c. inoculated on day 0; N: 5. CDX-1140 or hIgG2 0.3 mg was injected i.p. on day 1, 6, 15.

- **CDX-1140 Direct Anti-lymphoma Activity**
  - Ramos cells 0.5x10^6 were s.c. inoculated on day 0; N: 5. CDX-1140 or hIgG2 0.3 mg was injected i.p. on day 1, 6, 15.

- **CDX-1140 Synergizes with sCD40L**
  - CDX-1140 + Varil (Varil 5 mg/kg i.p. on day 1, 8 and 15)

**Summary and Next Steps**

- CD40 is a promising and powerful target for immunotherapy, but requires an appropriate balance between anti-tumor immune activation and harmful side effects of immune stimulation.

- CDX-1140 represents a novel CD40 agonist antibody with unique profile:
  - Potent agonist that functions in the format of human IgG2 isotype and independent of Fe receptor activation
  - Strong synergy with CD40L for enhanced activity

- Activates human B cells, DCs and macrocytes in vitro

- CDX-1140 has potent anti-lymphoma efficacy in xenograft models

- Direct anti-tumor activity against CD40+ lymphoma

- Enhanced anti-tumor activity when combined with human immune cells

- Activation of T cells, B cells, myeloid cells in co-cultures of PBMC with lymphoma cells

- Synergy with anti-CD20 mAb varilunab

Based on these data and a pilot study with non-human primates that confirmed a good safety profile, CDX-1140 is progressing towards clinical trials.

- A Phase 1 study with CDX-1140 in advanced cancer patients, including lymphoma patients, is planned to initiate in 2017.

- Following dose escalation of CDX-1140, combinations will be explored with immunotherapy and conventional chemotherapies.