CD40 on antigen presenting cells plays a crucial role in the induction of effective immune responses. In contrast, CD40 signaling on certain malignant cells, particularly B cell lymphomas, inhibits proliferation or triggers apoptotic cell death. Thus, two independent mechanisms provide opportunities for the use of agonist anti-CD40 monoclonal antibodies in cancer therapy: enhancement of anti-tumor immunity, and direct inhibition of tumor growth.

CDX-1140 is a human IgG2 antibody selected from a panel of fully human mAbs specific for CD40 generated by hybridoma technology from human Ig transgenic mice. We previously demonstrated the potent immune enhancing effects of CDX-1140 using in vitro models and in non-human primates. CDX-1140 was shown to activate dendritic cells and B cells in an Fc receptor independent manner. CDX-1140 does not bind to the CD40L binding site, and synergizes with CD40L in stimulation of the CD40 receptor and subsequent functional activities.

Here we further characterized the anti-tumor activity of CDX-1140 on CD40 positive tumors using xenograft models in immunodeficient mice. Using the Ramos and Raji lymphoblastoma cell lines, CDX-1140 was shown to attenuate tumor growth and prolong survival. Addition of CDX-1140 and human PBMC was highly effective at promoting complete rejection of both Ramos and Raji tumors. Importantly, the antibody CDX-1140 bladder carcinoma cell line was shown to show potent anti-tumor effects. CDX-1140 administered subcutaneously, all animals that were treated with 300 µg of CDX-1140 on days 1, 8, and 15, showed suppression of tumor growth by day 60, in comparison to saline-treated animals which developed significant tumors in 7 of 8 animals.

These data support the potential of CDX-1140 for direct anti-tumor effects on CD40-positive tumors (including epithelial tumors) that may supplement its activity as an immune activating agent. CDX-1140 is currently in a phase 1 dose-escalation study in patients with advanced solid tumors.

**INTRODUCTION**

CDX40 represents a unique target for immunotherapy due to its powerful effect on multiple immune cell types:

- CD40 activation on dendritic cells (DCs) promotes their conversion to antigen presenting cells (APCs) that are efficient for the stimulation of T cell responses.
- CD40 activation on macrophages promotes their ability to mediate effector functions such as phagocytosis.
- CD40 activation on B cells promotes proliferation and antigen presentation.
- CD40 activation on malignant B cells leads to tumor growth inhibition and rejection in xenograft models.

Functional aspects of CD40 agonist antibodies will substantially influence its activity profile:

- Blockade of natural ligand (CD40L) interaction,
- Promotes/negates Fc receptor interaction,
- Required (or require FcR for binding antigenic activity,
- Potency of agonistic activity.

CDX-1140 represents a novel fully human CD40 agonist antibody with unique properties.

**Efficacy of CDX-1140, an agonist CD40 antibody, in preclinical tumor models**

Lawrence J. Thomas, Li-Zhen He, James Testa, Anna Wasiuk, Jeffrey Weidlick, Crystal Sisson, Laura A. Vitale, Thomas O'Neill, Eric M. Forsberg, Catherine P. Gilmore, Lauren E. Gergel, Elizabeth Q. Do, James M. Boyer, April R. Barones, Mallary Rocheleau, Michelle E. Greailah, Kathleen M. Borrelli, Henry C. Marsh, Jr. and Tibor Keler

**ABSTRACT**

**CDX-1140 has Dose-dependent and Fc-independent Agonist Activity**

**Tumour Cell Lines**

<table>
<thead>
<tr>
<th>Study Group</th>
<th>CDX-1140 (µg)</th>
<th>Ramos</th>
<th>Raji</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>83</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>300</td>
<td>0.99</td>
<td>0.97</td>
<td>0.92</td>
</tr>
<tr>
<td>1000</td>
<td>0.94</td>
<td>0.93</td>
<td>0.89</td>
</tr>
<tr>
<td>3,000</td>
<td>0.89</td>
<td>0.87</td>
<td>0.83</td>
</tr>
<tr>
<td>10,000</td>
<td>0.85</td>
<td>0.83</td>
<td>0.79</td>
</tr>
<tr>
<td>30,000</td>
<td>0.79</td>
<td>0.76</td>
<td>0.72</td>
</tr>
</tbody>
</table>

**CDX-1140 Phase 1 Clinical Trial**

A Phase 1 Study of CDX-1140, a Fully Human Agonist anti-CD40 Monoclonal Antibody, in Patients with Advanced Solid Tumors (n=105)

**INTRODUCTION**

CDX-1140 is a human IgG2 anti-CD40 antibody.

- CDX-1140 does not bind to the CD40L binding site, and synergizes with CD40L in stimulation of the CD40 receptor and subsequent functional activities.
- CDX-1140 attenuated tumor growth and prolonged survival in xenograft models of the CD40-positive Ramos and Raji lymphoblastoma cell lines and the EJ138 bladder carcinoma cell line.
- An anti-CD40 surrogate, in combination with CDX-301 (FLT3L), attenuated tumor growth and prolonged survival in a syngeneic model of the CD40-negative CT26.WT murine colon carcinoma cell line.
- Two distinct mechanisms provide separate opportunities for the use of agonist anti-CD40 monoclonal antibodies in cancer therapy: enhancement of anti-tumor immunity, and direct inhibition of tumor growth.

**CDX-1140: AGONIST ACTIVITY**

- CDX-1140 binds to the CD40 receptor and is internalized, resulting in functional activation.

**Combination of CD40 AGONIST mAb with CDX-301 (FLT3L) in a SYNGENEIC TUMOR MODEL**

- CDX-1140 and CDX-301 (FLT3L) synergized to inhibit tumor growth.
- The combination of CDX-1140 and CDX-301 (FLT3L) was more effective than either agent alone.

**SUMMARY AND FUTURE DIRECTIONS**

- These data support the current Phase 1 dose-escalation study in patients with advanced solid tumors.
- Data also support adding CDX-1140 to the Phase 1 CDX-1140 study in the near future.

**CDX-1140 ACTIVITY IN XENOGRAFT BURKITT'S LYMPHOMA MODEL**

- CDX-1140 showed significant suppression of tumor growth in Ramos and Raji cells.
- The combination of CDX-1140 and FLT3L (CDX-301) showed synergistic inhibition of tumor growth.

**COMBINATION OF CD40 AGONIST mAb WITH CDX-301 (FLT3L) IN A SYNGENEIC TUMOR MODEL**

- The combination of CDX-1140 and CDX-301 (FLT3L) was more effective than either agent alone.
- The combination of CDX-1140 and CDX-301 (FLT3L) showed synergistic inhibition of tumor growth.

**CDX-1140 ACTIVITY IN A XENOGRAFT BLADDER CARCINOMA MODEL**

- CDX-1140 showed significant suppression of tumor growth in EJ138 cells.
- The combination of CDX-1140 and CDX-301 (FLT3L) showed synergistic inhibition of tumor growth.

**BINDING PROPERTIES OF CDX-1140**

- CDX-1140 has a high affinity for human CD40.
- CDX-1140 reduces but does not block CD40 ligand (CD40L) interaction.
- CDX-1140 has dose-dependent and Fc-independent agonist activity.

**CDX-1140 ACTIVITY IN XENOGRAFT BLADDER CARCINOMA MODEL**

- CDX-1140 showed significant suppression of tumor growth in EJ138 cells.
- The combination of CDX-1140 and CDX-301 (FLT3L) showed synergistic inhibition of tumor growth.

**CDX-1140 PHASE 1 CLINICAL TRIAL**

- A Phase 1 Study of CDX-1140, a Fully Human Agonist anti-CD40 Monoclonal Antibody, in Patients with Advanced Solid Tumors (n=105)