AB928, a dual antagonist of the A$_{2a}$R and A$_{2b}$R adenosine receptors for the treatment of cancer


Arcus Biosciences, 3928 Point Eden Way, Hayward, CA 94545, USA

Introduction

In many tumors, extracellular adenosine contributes to an immunosuppressed tumor micro-environment (TME) via activation of the A$_{2a}$R, expressed on lymphocytes, and the A$_{2b}$R, expressed on myeloid cells. Relative to other tissues like the brain, adenosine concentrations in the TME are much higher. Activation of A$_{2a}$R has been shown to result in suppression of T and NK cell responses, while A$_{2b}$R activation has been linked to the generation of suppressive myeloid cells.

Methods

**Human T Cell Activation**: Adenosine, or AMP, was utilized to suppress the activation of CD4 or CD8 T cells +/- AB928. Supernatants were collected for cytokine analysis after 3 days. **Monocyte differentiation**: Monocytes were differentiated using IL-4 & GM-CSF for 6 days +/- adenosine or AB928 as indicated. The resulting moDC were activated with TNF-α or LPS and cultured with CD4 T cells for 4 days, after which time proliferation and IFN-γ were measured. **AT-3 OVA Mouse Model**: AT-3 OVA cells were inoculated on the flank of C57BL/6 mice, animals were randomized to treatment groups when tumors reached 50 mm$^3$. Oxaliplatin was dosed at 10 mg/kg IP 4 times 5 days apart, AB928 was dosed 100 mg/kg PO BID.

**AB928 Human Whole Blood Potency**

<table>
<thead>
<tr>
<th>Target</th>
<th>IC$_{50}$ (nM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CD8 pCREB (against 5 μM NECA)</td>
<td>88</td>
</tr>
</tbody>
</table>

Figure 1. Adenosine is generated, via CD73, from AMP in the tumor microenvironment and results in immune suppression via A$_{2a}$ and A$_{2b}$ receptor activation on multiple cell types.

**Results**

**Immune Cell Adenosine Receptor Expression**

Figure 2. Expression of adenosine receptors 1, 2a, 2b and 3 were analyzed by qPCR in PBMC subsets from healthy donors (left panel; A$_{2a}$R and A$_{2b}$R not detected) as well as monocyte-derived DC (moDC; right panel).

**AB928 Restores CD8 T Cell Function**

Figure 3. AB928 restores CD8 T cell IFN-γ and Granzyme B production (representative data shown n = 9 donors; adenosine 6 μM).

**AB928 Restores Mouse NK Function**

Figure 4. NECA suppresses the ability of mouse IL-2 expanded NK cells to kill LLC target tumor cells. This suppression is restored by AB928. Representative data from 3 separate experiments shown.

**AB928 Restores CD4 T Cell Function**

Figure 5. AB928 restores CD4 T cell IL-2 production, representative data from 4 donor PBMC shown. Consistent with CD73 expression on T cells driving adenosine conversion, AMP inhibits T cell activation and is reversed by AB928 (adenosine and AMP at 6 μM).

**AB928 Restores Normal Dendritic Cell Maturation**

Figure 6. Adenosine (10 μM) suppresses maturation and activation of dendritic cells resulting in suppressed T cell proliferation and cytokine release in a DC-MLR. AB928 restores normal moDC maturation resulting in significantly increased T cell activation and cytokine release. Representative data from 3 separate experiments shown (*p<0.05).

**Combining AB928 & Chemotherapy Results in Significantly Reduced Tumor Growth**

Figure 7. Established AT-3 OVA tumors dosed with AB928 in combination with oxaliplatin have a significantly reduced tumor growth relative to those dosed with oxaliplatin alone (*p<0.05 AB928 vs. vehicle; ****p<0.0001 Oxaliplatin vs. Oxaliplatin + AB928). The AB928 dose was selected based upon suppression of pCREB in peripheral CD8 cells (inset panel, see poster P006 for detailed information on the pCREB assay).

**Summary**

AB928 is a potent antagonist of A$_{2a}$R and A$_{2b}$R which effectively reverses adenosine-mediated immune suppression and retards tumor growth in vivo. AB928 will enter clinical trials in 2017.