mimic physiological TME to dissect the combinational role of Nivolumab and Blinatumomab (figure 2).

**Results**

The result suggest that combinatory therapy is highly depend on the dosage of Blinatumomab and also T cell number in the TME, which might give an instruction for ongoing clinical trial design. Finally, we have employed humanized mouse models bearing Raji or Daudi tumor cells to further validate this combination treatment in vivo. Both In-vivo and In-vitro data support that Blinatumomab is dominant in activating T cell and Nivolumab can only exhibit synergistic effect under suboptimal dosage of Blinatumomab.

**Conclusions**

Successfully establish in vitro system to evaluate CD3 BiTE.

Opdivo could further promote T cell activation under the treatment of CD3 BiTE under suboptimal dosage of Blinatumomab.

**Results**

ALDHi trigger immunogenic cell death (ICD) via necroptosis and can potentiate anti-tumor immunity.

**Methods**

We performed RNA-Seq on four human OvCa cell lines (A2780, CAOV-3, OVCAR-5, OVSAHO) treated for 8 hours with two different ALDHi. To measure the impact on T cell immunity we performed flow cytometry to measure cell proliferation assays and CD4 naïve differentiation into Th1/Th17/Treg subsets. Molecular targets in the RA pathway were confirmed by western blot.

**Results**

ALDHi triggered significant changes in (i) ER stress unfolded protein response and regulators of the ER stress response, such as ATF4 and EIF2aK3 (PERK), (ii) inflammatory pathways, (iii) cell death, survival, and (iv) gene transcription-RAR signaling. Treatment of cancer cell lines with ALDHi induced expression of Phospho-elF2a, a marker for the ICD, along with increased expression of ATF3 and ATF4, and calreticulin, suggesting cancer cells undergoing ICD.

Using polyclonal stimulation of murine splenocytes and human PBMC, we observed that ALDHi promote T cell proliferation, especially of CD8 T cells. Furthermore, exposure of naïve CD4 cells to Th1 and Treg differentiation conditions leads to increased production of INFγ and reduced number of Foxp3+ iTregs, respectively. Further, in a co-culture of iTreg and stimulated splenocyte, ALDHi treatment diminishes the iTreg's capacity to induce immune suppression. Ex vivo treatment of ovarian cancer ascites cells with various ALDHi leads to significant decrease of CD14+ cells, an effect associated with downregulation of NR4A1 (NUR77), a nuclear receptor that interacts RAR/RXR, downstream of RA signaling.

**Conclusions**

ALDHi induce immunogenic cell death in cancer cells. Immune cells respond to ALDHi in a cell specific manner. ALDHi support CD8 T cell proliferation and CD4 Th1 induction, while inhibiting iTregs. Exposure to ALDHi leads to downregulation of NR4A1 and reduction in suppressive macrophage numbers. Our results support the use of ALDHi as immune modulators in ovarian cancer and adjuvants to immunotherapy.