AVA-ADR-001 SUPPresses Tumor Growth AND INDUCes ANTI-TUMOR IMMUNITY BY SELECTively INHIBITING ADAR1 P150

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Background Adenosine deaminase, RNA specific (ADAR1), catalyzes the hydrolytic deamination of adenosine (A) to inosine (I) in double-stranded (ds) RNAs. There are 2 isoforms of ADAR1 (p110 in the nucleus; p150 in cytoplasm) and both modify self dsRNA in coding and non-coding regions. The ADAR1 p150 isoform is expressed from an interferon (IFN)-response promoter and has a Z-DNA/Z-RNA binding domain at the N-terminus. ADAR1 p150 edits 3'-untranslated region dsRNAs comprising of inverted Alu repeats and thereby suppresses MDA5-MAVS-IFN signaling. ADAR1 is commonly overexpressed in multiple myeloma, breast, lung, liver, skin and esophageal cancer where it promotes cancer progression. Inhibition of ADAR1 has promising anti-tumor efficacy as monotherapy and in combination with checkpoint inhibitors, radiotherapy and chemotherapeutic modalities. Herein, we outline the discovery of a potential first-in-class ADAR1 inhibitor for cancer immunotherapy.

Methods AVA-ADR-001 was identified through a high throughput p110 knockout cell-based assay. The ability of AVA-ADR-001 to induce interferons was confirmed in various cell lines like A549 p110 KO, HCT116 and B16F10. Finally, the anti-tumor efficacy of AVA-ADR-001 was evaluated in B16F10 syngeneic melanoma mice model as monotherapy and in combination with anti-PD1.

Results We have identified a first-in-class small molecule inhibitor of ADAR1, which shows significant IFN response in vitro in an MDA5 dependent manner. In vitro binding studies have confirmed direct binding of AVA-ADR-001 with the Z-domain of ADAR1 thus confirming its selectivity to the p150 isoform. AVA-ADR-001 demonstrates micromolar EC50 and anti-tumor efficacy against B16F10 melanoma syngeneic mouse model. 100 μg of AVA-ADR-001 treatment resulted in 45% tumor growth inhibition (TGI), 1.5x superior to Anti-PD1 treatment. Combining AVA-ADR-001 with Anti-PD1 demonstrated a synergistic effect 2x superior to Anti-PD1 alone. Additionally, several interferon stimulated genes like IFIH1, IFN-β and CXCL-10 were significantly upregulated in the tumor samples of the AVA-ADR-001 monotherapy and combination groups.

Conclusions To our knowledge no selective small molecule inhibitors of ADAR1 have been reported so far and AVA-ADR-001 is the first disclosure of such an inhibitor. AVA-ADR-001 is a potent and selective first-in-class ADAR1 inhibitor which has shown significant IFN induction in various cancer cell lines and in vivo in the tumor microenvironment resulting in substantial tumor growth inhibition as monotherapy and synergistically in combination with Anti-PD1. Considering the immune-suppressive and pro-metastatic role of ADAR1, AVA-ADR-001 serves as a promising starting point for novel ADAR1 inhibitors as therapeutic modalities in cancer immunotherapy.