In vivo characterization of PD-1-001 and CD73/PD-1-001. The binding affinity of PD-1-001 to PD-L1 was determined using a kinetic binding assay. PD-L1-001 and control constructs were incubated with biotinylated human PD-L1 and the signal was measured by surface plasmon resonance (SPR). The binding assay was conducted by testing PD-1-001 and CD73/PD-1-001 to PD-L1 with MC-38 (hPD-L1) tumors (genOway, France) (n=10). MC-38 tumors were dosed IP twice weekly for 3 weeks. As a comparator, a pembrolizumab biosimilar and CD73-DFC generated modest TGIs of 14.2 and 24.8% respectively, which were not significant. However, PD-1-001 and CD73/PD-1-001 generated nearly identical tumor reductions of 60.1 and 61.5% respectively (p<0.0068). The virtually identical TGI's for CD73/PD-1-001 further suggest the maximum achievable signal in this model has been reached by PD-1-001 and CD73/PD-1-001.

Figure 4A. Efficacy of test articles in a transgenic mouse model of colorectal cancer.

Figure 5A/B. Tumor Volume (MC38, hPD-L1) in vivo characterization of PD-1-001 and CD73/PD-1-001 in a humanized mouse model. In transgenic mice expressing hPD-L1, a pembrolizumab biosimilar at 10 mg/kg had minimal effects on tumor growth (Figures 4A/B). Similarly, a CD73 only targeting DFC (CD73-DFC) did not significantly reduce tumor growth. In contrast, our proprietary anti-PD-1 inhibitor, PD-1-001, dosed at 10 mg/kg significantly reduced tumor growth (p<0.01). Robust tumor volume reductions were also achieved with the multispecific CD73/PD-1-001 construct at 1 and 10 mg/kg. Notably, the PD-1-001 and CD73/PD-1-001 constructs reduced tumor burden to a similar extent when dosed at 10 mg/kg. The lack of a dose response with PD-1-001 and CD73/PD-1-001 suggests the maximum achievable signal in this model has been reached by PD-1-001 and CD73/PD-1-001.

Figure 5A/B. Tumor Volume (hPD-1/hPD-L1 C57BL/6) in vivo characterization of PD-1-001 and CD73/PD-1-001 in a transgenic mouse tumor model improved efficacy. In a follow-up study both DFCs were effective at 1, 3, and 10 mg/kg. At the lowest test concentration (1 mg/kg) the DFCs began to differentiate, with CD73/PD-1-001 achieving a statistically significant reduction in tumor volume (p<0.05) while PD-1-001 did not (Figures 5A/B). This result highlights the exceptional potency of CD73/PD-1-001 and further demonstrates the therapeutic benefits of combining two important checkpoint targeting inhibitors on a single multispecific DFC.

Figure 5A/B. Dose range of test articles in a humanized mouse model.

Figure 6. Improved activity of CD73/PD-1-001 compared to PD-1 monotherapy.

RESULTS

PD-(L)1 checkpoint blockade has revolutionized cancer therapy. However, additional tumor immune escape mechanisms, including the adenosine pathway, play potentially important roles in generating resistance to PD-(L)1 therapy. Here we describe CD73/PD-1-001, a dual targeting DFC with potent activity against both validated immune checkpoint pathways.

CD73/PD-1-001 is a potent inhibitor of hCD73 and hPD-1 in vitro. This potent activity translated into efficacy in an in vivo model of colorectal cancer using transgenic mice. CD73/PD-1-001 reduced tumor growth by 50% with a low dose of 1 mg/kg (twice/week, x3 weeks). At 3 mg/kg TGI increases to 71.4% (p<0.01). Additionally, we demonstrated that both checkpoint pathways inhibited by CD73/PD-1-001 contributed to efficacy. Highlighting the potential clinical benefits of this first in class multispecific inhibitor.

REFERENCES

All authors are shareholders & employees of Cidara Therapeutics.

DISCLOSURE & REFERENCES

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