Human peripheral blood mononuclear cells (PBMCs) were isolated from buffy coat samples by density centrifugation with Histopaque 1077 (400 g, 30 min). Cells were washed and resuspended at a density of 2 x 10^6 cells/ml in RPMI + 10% human serum. PBMCs were stimulated with α-CD3/CD28. 1 μg/mL), CPX-016 (competitor anti-CD73 mAb, 10 μg/mL), and M). PBMCs were then stimulated after 1 hour with anti-CD3 (clone HIT3a, 1 μg/mL), anti-CD28.2, 1 μg/mL). Cells were cultured for 17.5 h and then treated with final concentrations of drugs as indicated in the figure. Cells were harvested and analyzed for gene expression by NanoString PanCancer Immune Panel. Representative data from one of >3 patients is shown above.

Inflammation

NanoString PanCancer Immune Panel. Representative data from one of >3 patients is shown above.

CD73 antagonists amplify AMP signature

CD73 antagonists inhibit adenosine formation but consequently preserve AMP. AMP amplifies adenosine-like gene signature suggesting AMP signals through adenosine.

Conclusions

- Adenosine antagonists induce a specific gene signature in human immune cells. The "Adenosine Signature" is dominated by genes involved in cell survival and proliferation.
- Updated clinical data confirms original reports that expression of the Adenosine Signature correlates with tumor regression in an ongoing Ph1/2 trial in patients with advanced melanoma.
- High Adenosine Signature expression is a statistically significant correlation with tumor response.
- Ciforadenant blocks AMP and AMPaS.
- Ciforadenant neutralizes AMP & AMPaS.
- Adenosine antagonists induce a specific gene signature in human immune cells. The "Adenosine Signature" is dominated by genes involved in cell survival and proliferation.