

Title: High-throughput flow cytometry to investigate T-cell activation and killing

Authors: David Baker¹, Anne Goeppert¹, James Robinson¹. ¹ Discovery Biology, Discovery Sciences, IMED Biotech Unit, AstraZeneca, Cambridge, United Kingdom

Abstract:

We have developed high-throughput flow cytometry assays on the Intellicyt iQue Screener Plus to interrogate primary T-cell proliferation and cytokine release. A primary human CD8⁺ T-cell proliferation assay was successfully miniaturised from a 96 well assay format on the Miltenyi MACS Quant to a 384 well assay format on the iQue, giving a 4-fold increase in throughput per amount of cells. This assay has been used to profile compounds that could be used to augment T-cell activation. Intellicyt Qbeads have been used to profile cytokine release in this assay and in a separate PBMC cell assay, and a bespoke Qbead assay has been developed in order to quantify secreted biomarker levels in cell culture supernatant . In addition to T-cell proliferation and activation markers, the iQue Screener has been used to measure tumour cell viability following co-culture with CD8⁺ T-cells in a 96 well plate format, providing vital data for selection of a T-cell donor to be used in a pooled CRISPR screen and for optimisation of final assay conditions.