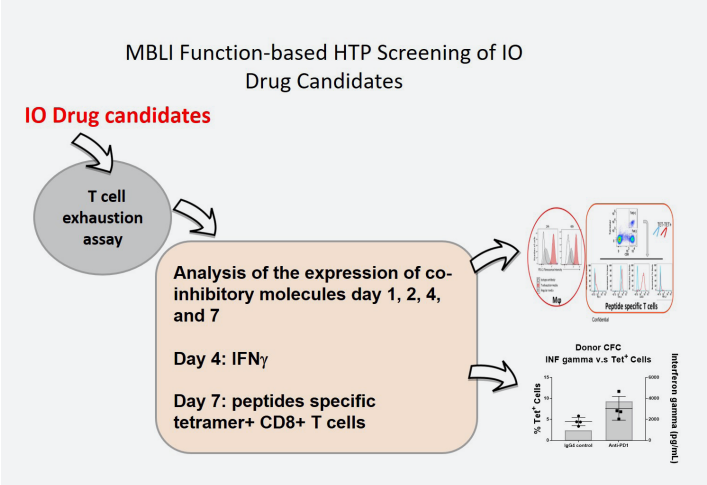
A reversible T cell exhaustion model

for functional screening of immune checkpoint drug candidates

Background: We have devised an *in vitro* model that recapitulates the main components of tumor deposits responsible for inducing T cell exhaustion. The assay consists of healthy PBMCs, cultured is a specific medium with elements responsible for cancer induced T cell exhaustion (proprietary) and stimulated with (prior-defined) specific recall antigen peptides. The T cell exhaustion is validated by the upregulation of corresponding checkpoint molecules and by reduction of antigen specific T cells is reversed in some healthy donors ("responders") upon treatment with PD-1 blockade (e.g. pembrolizumab), which serves as a positive control, and not by isotype controls. The read-out is then expression of checkpoint receptors, IFNɣ and % tetramer+ CD8 T cells. As a result, the assay identifies active immune checkpoint (or other immune-potentiating) drug candidates, in a PBMC-based setting with minimal manipulation.

T cell Exhaustion Model Assay Features:

- **Physiologic relevance:** Validated antigen-based and T cell exhaustion-like PBMC setting
- **Meaningful readouts:** drug induced further expansion of antigen specific T cells and of functional cytokine expression
- **Semi High Throughput** (96-well plate)



The Function-based Reversible T cell Exhaustion Model can potentially identify active drug candidates