

Quantitative Cell-Based Reporter Gene Bioassays to Advance Individual or Combination Cancer Immunotherapy

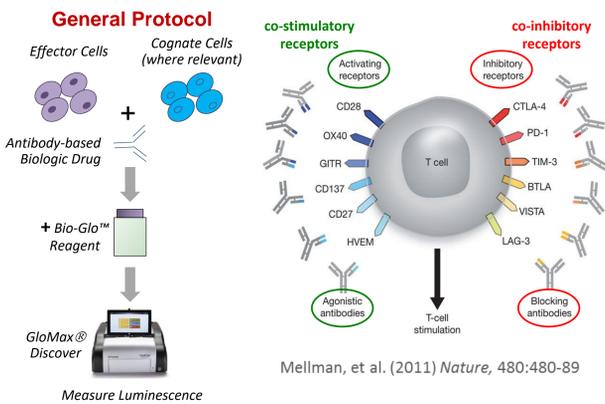
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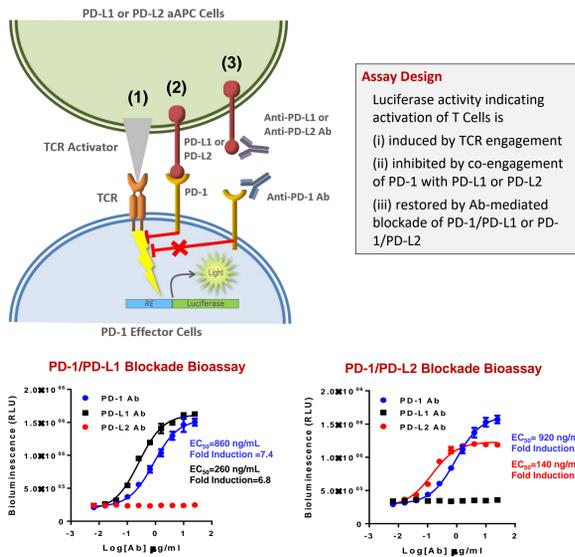


1. Introduction

A major challenge in the development of antibody-based biologics drugs is access to quantitative and reproducible functional bioassays. In contrast to the cumbersome, variable methods currently used that rely on primary cells, we have developed a portfolio of functional cell-based reporter bioassays to easily measure the activity of biologics drugs designed to target immune checkpoint receptors including co-inhibitory (e.g. PD-1, CTLA-4, LAG-3) and co-stimulatory (e.g. 4-1BB, GITR, OX40) receptors. These bioassays consist of stable cell lines that express luciferase under the precise control of receptor-mediated intracellular signals. Here we describe the application of these MOA-based bioassays for biologics drug discovery, development, potency and stability studies.

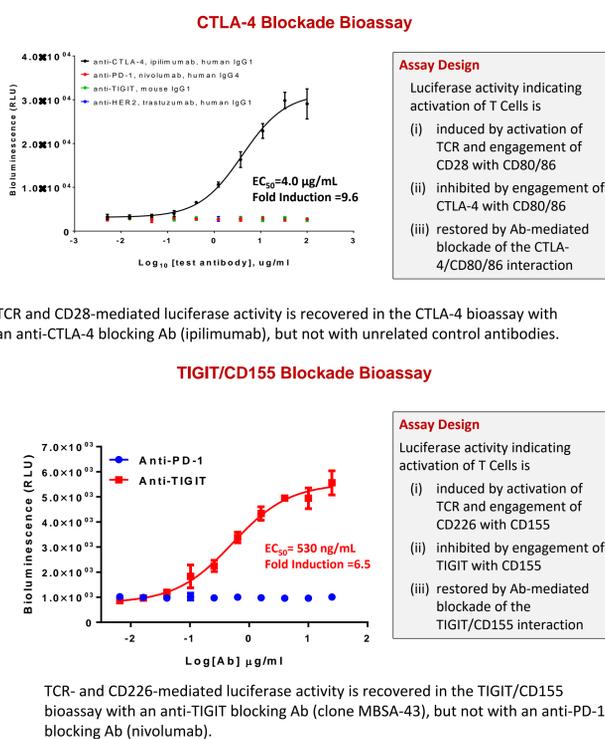


2. PD-1 Blockade Bioassays

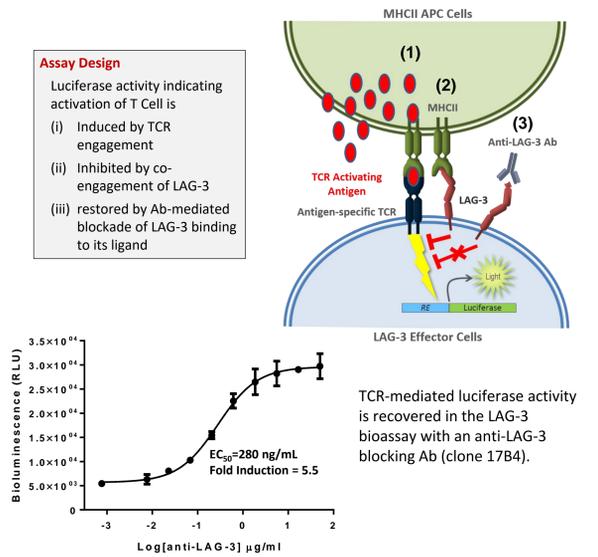


TCR mediated luciferase activity is recovered in the PD-1 Blockade Bioassay with (LEFT) anti-PD-1 and PD-L1 blocking Abs and anti-PD-1 and PD-L2 blocking Abs (RIGHT), but not with unrelated control antibodies.

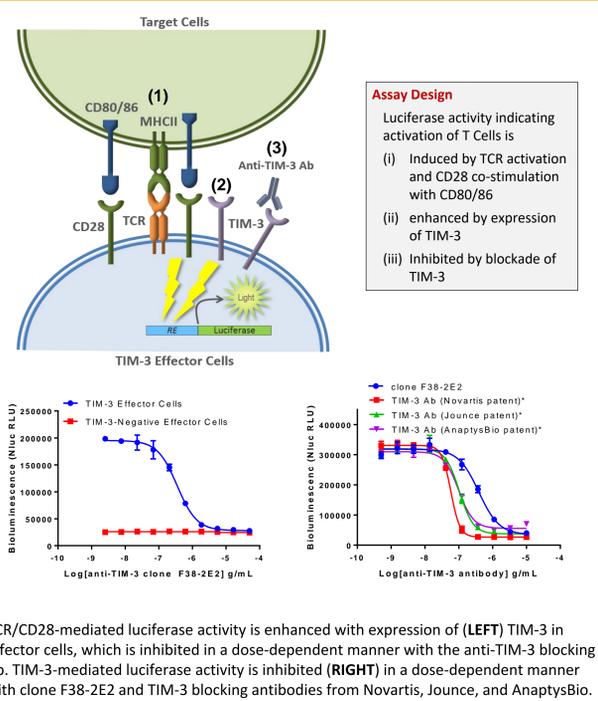
3. CTLA-4 and TIGIT Blockade Bioassays



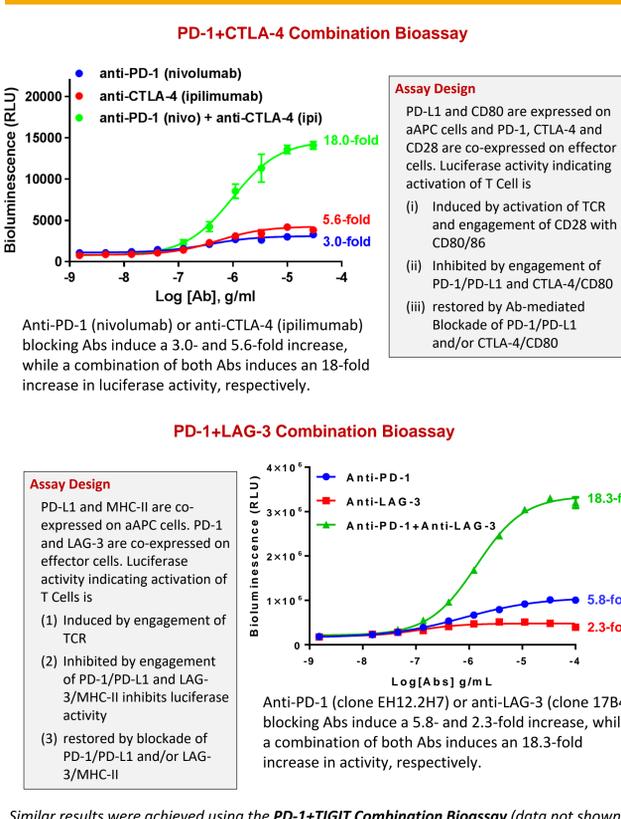
4. LAG-3/MHCII Blockade Bioassay



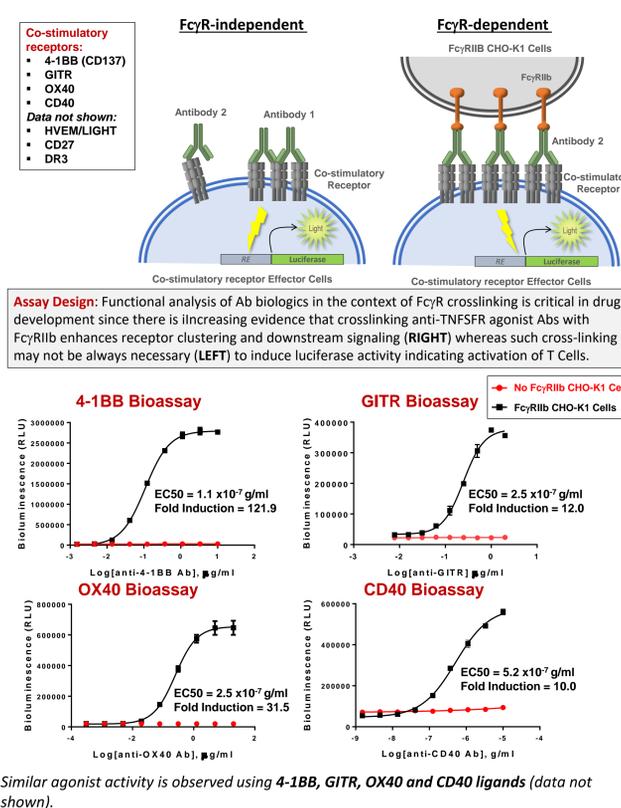
5. TIM-3 Bioassay



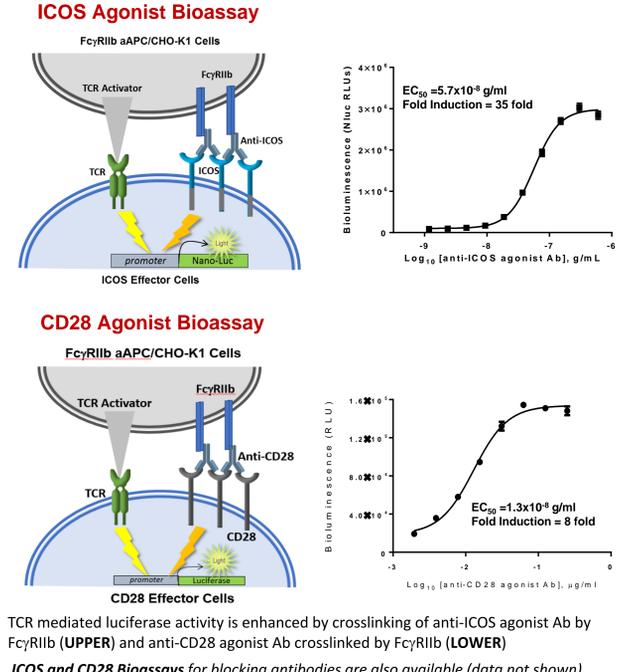
6. Combination Bioassays



7. FcγR-independent and -dependent Co-stimulatory Bioassays



8. ICOS and CD28 Bioassays



9. Conclusions

Cell-based reporter bioassays overcome the limitations of primary cell-based assays for functional characterization of antibody and other biologics drugs targeting individual or combination immune checkpoint receptors. Here we show a portfolio of MOA-based bioassays for co-inhibitory and co-stimulatory immune checkpoint receptors that can be used for antibody screening, characterization, potency and stability studies. These bioassays provide the following:

- Biologically relevant measurement of antibody MOA
- Specific immune checkpoint regulated expression of luciferase that reflects the native biology of T cell activation.
- Demonstrated ability to measure the potencies of immune checkpoint-targeted antibodies
- Consistent and reliable measure of antibody activity
- Demonstrated precision, accuracy, reproducibility, robustness
- All assays can be used as "Thaw-and-use" cell format, no cell culture required
- Functional performance suitable for development into potency, stability, and NAB assays
- Easy-to-implement
- Rapid and convenient workflow
- Amenable to standard 96-well and 384-well plate formats