Quantitative Cell-Based Bioassays for Individual and Combination Immune Checkpoint Immunotherapy Targets

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1. Introduction

The human immune system is comprised of a complex network of immune checkpoint receptors that are promising new immunotherapy targets for the treatment of a variety of diseases including cancer and autoimmune-mediated disorders.

Current methods used to measure the activity of antibody and other biologics drugs designed to target immune checkpoint receptors rely on primary human cells and measurement of functional endpoints such as cell proliferation, cell surface marker expression, and cytokine production. These assays are laborious and highly variable due to their reliance on donor primary cells, complex assay protocols, and unqualified assay reagents. As a result, these assays are difficult to establish in quality-controlled drug development settings.

To overcome these challenges, we developed a suite of cell-based biluminometric reporter bioassays for individual and combination immune checkpoint immunotherapy targets including:

- PD-1 (PD-L1 or PD-L2), CTLA-4, LAG-3, TIGIT, PD-1+TIGIT
- GITR, 4-1BB, CD40, OX40

These mechanism of action (MOA)-based bioassays are available in "thaw-and-use" format and demonstrate high specificity, sensitivity, and reproducibility. The bioassays are qualified according to ICH guidelines and demonstrate the performance required for use in antibody screening, potency testing, and stability studies.


3. PD-1/PD-L1 Blockade Bioassay: Antibody Potency and Stability Studies

4. TIGIT/CD155 Blockade Bioassay: Principle and Specificity

5. PD-1/TIGIT Combination Bioassay

6. CTLA-4 Blockade Bioassay: Specificity and Antibody Potency Study

7. LAG-3 Blockade Bioassay: Principle and Potency Study

8. GITR, 4-1BB, OX40, CD40 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 immune checkpoint bioassays 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 native biology
- Demonstrated ability to measure the activity of immune checkpoint-targeted antibodies
- Consistent and reliable measure of antibody activity
- Demonstrated precision, accuracy, reproducibility, robustness
- "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

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