

Product Sheet

Tango-H_CB1-CHO-K1 Cell Line

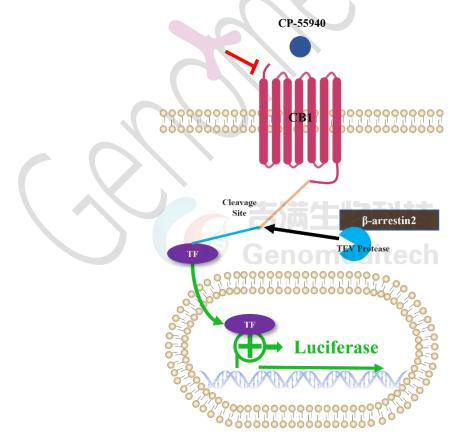
Catalog number: GM-C37720

Version 3.3.1.250328

The TANGO assay detects GPCR activation by analyzing ligand-induced effects (e.g., by small-molecule drugs or antibodies) through fluorescent signals. It offers high signal specificity and directly reflects intracellular signal transduction, and is widely used in drug screening and GPCR research

A plasmid with GPCR, a V2 tail, a TEV protease site, and the transcription factor(TF) is constructed. Upon ligand activation, GPCR recruits arrestin and TEV protease, which cleaves the TEV site, releasing TF. This factor enters the nucleus, activating a reporter gene (e.g., luciferase). Fluorescent signal intensity corresponds to GPCR activation, allowing quantitative analysis.

Tango-H_CB1-CHO-K1 Cell Line is a clonal stable CHO-K1 cell line constructed using lentiviral technology, constitutive expression of the CB1 gene constructed through Tango technology, along with signal-dependent expression of a luciferase reporter gene. When igand binds to CB1, it activates downstream signaling pathways, leading to the expression of luciferase. The luciferase activity measurement indicates the activation level of the signaling pathway and can thus be used to evaluate the in vitro effects of drugs related to CB1.





Specifications

Materials			
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.		
Safety considerations	Biosafety Level 2		
Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.		
Growth Conditions	37°C, 5% CO ₂		
Growth properties	90% FBS+10% DMSO Adherent		
Freezing Medium			
Note	None		
Growth medium	Puromycin		
	F12K+10% FBS+1% P.S+4 µg/mL Blasticidin+100 µg/mL Hygromycin+4 µg/mL		
Recovery Medium	F12K+10% FBS+1% P.S		
Storage Conditions	Liquid nitrogen immediately upon receipt		
Shipping	Shipped on dry ice		
Product Format	1 vial of frozen cells		
Quantity	5E6 Cells per vial,1 mL		

Materials

Reagent	Manufacturer/Catalogue No.
F12K	BOSTER/PYG0036
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Hygromycin	Genomeditech/GM-040403
Puromycin	Genomeditech/GM-040401
(-)-CP 55,940	GLPBIO/GC18343
Anti-H_CB1 hIgG4 Antibody(Nimacimab)	Genomeditech/GM-47472AB
GMOne-Step Luciferase Reporter Gene Ass	say Kit Genomeditech/GM-040503

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Figures

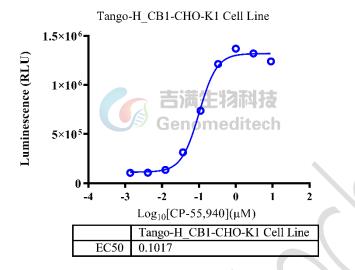


Figure 1 | Response to (-)-CP 55,940. The Tango-H_CB1-CHO-K1 Cell Line (Cat. GM-C37720) at a concentration of 1E4 cells/well (96-well format) was stimulated with serial dilutions of (-)-CP 55,940 (GLPBIO/GC18343) in assay buffer (F12K+1% FBS+1% P.S) for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [13.7]. Data are shown by drug molar concentration.

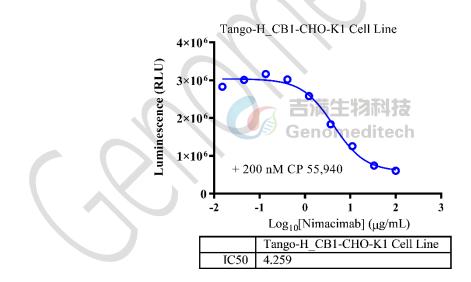


Figure 2 | Response to Anti-H_CB1 hIgG4 Antibody(Nimacimab). Serial dilutions of the Anti-H_CB1 hIgG4 Antibody(Nimacimab) (Cat. GM-47472AB) was incubated with 1E4 cells/well of the Tango-H_CB1-CHO-K1 Cell Line (Cat. GM-C37720) in a 96-well plate for 1 hour in assay buffer (F12K+1% FBS+1% P.S). Subsequently, the CP 55,940 (GLPBIO/GC18343) was added, and the coculture proceeded for an additional 16 hours. Firefly luciferase activity was then measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The results indicated maximum blocking folds of approximately [4.5]. Data are shown by drug mass concentration.

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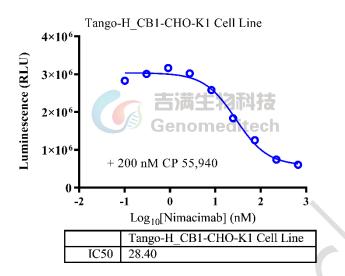


Figure 3 | Response to Anti-H_CB1 hIgG4 Antibody(Nimacimab). Serial dilutions of the Anti-H_CB1 hIgG4 Antibody(Nimacimab) (Cat. GM-47472AB) was incubated with 1E4 cells/well of the Tango-H_CB1-CHO-K1 Cell Line (Cat. GM-C37720) in a 96-well plate for 1 hour in assay buffer (F12K+1% FBS+1% P.S). Subsequently, the CP 55,940 (GLPBIO/GC18343) was added, and the coculture proceeded for an additional 16 hours. Firefly luciferase activity was then measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The results indicated maximum blocking folds of approximately [4.5]. Data are shown by drug molar concentration.

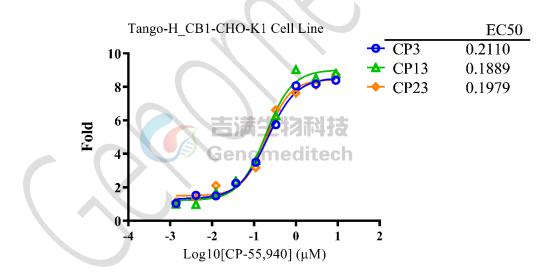
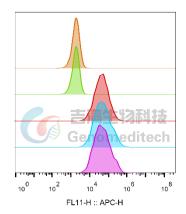


Figure 4 | The passage stability of response to (-)-CP 55,940. The passage 3, 13, and 23 of Tango-H_CB1-CHO-K1 Cell Line (Cat. GM-C37720) at a concentration of 1E4 cells/well (96-well format) were stimulated with serial dilutions of CP 55,940 (GLPBIO/GC18343) in assay buffer (F12K+1% FBS+1% P.S) for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). Data are shown by drug mass concentration.

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SampleID	Geometric Mean : FL11-H
CHO-K1 anti-CB1+APC-2nd Ab	1811
Tango-H_CB1 CHO-K1 H_IgG+APC-2nd Ab	1742
Tango-H_CB1 CHO-K1 anti-CB1+APC-2nd Ab CP6	42571
Tango-H_CB1 CHO-K1 anti-CB1+APC-2nd Ab CP16	48720
Tango-H_CB1 CHO-K1 anti-CB1+APC-2nd Ab CP26	46580

Figure 5 | Tango-H_CB1-CHO-K1 Cell Line (Cat. GM-C37720) was determined by flow cytometry using Anti-H_CB1 hIgG4 Antibody(Nimacimab) (Cat. GM-47472AB).

Cell Recovery

Recovery Medium: F12K+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70° C. Storage at -70° C will result in loss of viability.

- a) Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 3 minutes).
- b) Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- c) Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately 176 x g for 5 minutes. Discard supernatant.
- d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- e) Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- a) Centrifuge at 176 x g for 3 minutes to collect cells.
- b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
- c) Aliquot 1 mL into each vial.
- d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

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Cell passage

Growth medium: F12K+10% FBS+1% P.S+4 µg/mL Blasticidin+100 µg/mL Hygromycin+4 µg/mL Puromycin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- a) Remove and discard culture medium.
- b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- c) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 2 to 3 minutes at 37°C).
- Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach.
 Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- g) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:4 - 1:5 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) After the stabilization of the cell condition, there will be fewer dead cells post-passage, the cell growth rate will tend to stabilize, cell morphology will become uniform, and the cells will appear robust.

Related Products

CNR1(CB1)			
H_CB1 CHO-K1 Cell Line	H_CB1 HEK-293 Cell Line		
Anti-H_CB1 hIgG4 Antibody(Nimacimab)			

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