

# Product Sheet

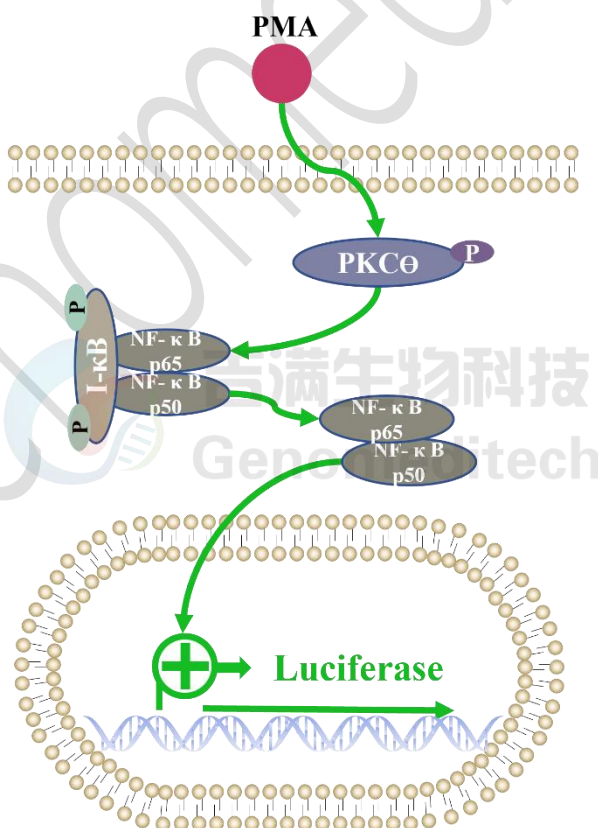
## NFKB Reporter THP1 Cell Line

Catalog number: GM-C06727

Version 3.3.1.241226

NF- $\kappa$ B is a group of transcription factors, including p65 (RelA), RelB, c-Rel, p50/p105 (NF- $\kappa$ B1), and p52/p100 (NF- $\kappa$ B2). These proteins function as dimeric transcription factors and play roles in controlling gene regulation across a wide range of biological processes, including innate and adaptive immunity, inflammation, stress responses, B cell development, and lymphoid organogenesis. Pro-inflammatory cytokines, LPS, growth factors, and antigen receptors activate the IKK complex (IKK $\beta$ , IKK $\alpha$ , and NEMO), leading to the phosphorylation of I $\kappa$ B proteins. Phosphorylation of I $\kappa$ B results in its ubiquitination and proteasomal degradation, releasing the NF- $\kappa$ B/Rel complex. The active NF- $\kappa$ B/Rel complex is further activated through phosphorylation and translocates to the nucleus to induce target gene expression.

NFKB Reporter THP1 Cell Line is a clonal stable THP1 cell line expressing a firefly luciferase under the control of the NF- $\kappa$ B response elements. PMA (phorbol 12-myristate 13-acetate) is a known activator of protein kinase C (PKC). PKC activation can lead to the activation of the NF- $\kappa$ B signaling pathway, leading to luciferase expression. Luciferase readings reflect the activation level of signaling pathways and can be used to evaluate the activation effect of NF- $\kappa$ B.



## Specifications

<b>Quantity</b>	5E6 Cells per vial, 1 mL
<b>Product Format</b>	1 vial of frozen cells
<b>Shipping</b>	Shipped on dry ice
<b>Storage Conditions</b>	Liquid nitrogen immediately upon receipt
<b>Recovery Medium</b>	RPMI 1640(ATCC)+20% FBS+1% P.S+0.05 mM $\beta$ -Me
<b>Growth medium</b>	RPMI 1640(ATCC)+10% FBS+1% P.S+0.05 mM $\beta$ -Me+0.5 $\mu$ g/mL Puromycin
<b>Note</b>	Cells should be cultured using ATCC/30-2001 RPMI 1640 medium or Growth medium from Genomeditech. The serum should be Cegrogen biotech/A0500-3010 or sourced from Gibco.
<b>Freezing Medium</b>	90% FBS+10% DMSO
<b>Growth properties</b>	Suspension
<b>Growth Conditions</b>	37°C, 5% CO <sub>2</sub>
<b>Mycoplasma Testing</b>	The cell line has been screened to confirm the absence of Mycoplasma species.
<b>Safety considerations</b>	Biosafety Level 2
<b>Note</b>	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

## Materials

<b>Reagent</b>	<b>Manufacturer/Catalogue No.</b>
RPMI 1640(ATCC)	ATCC/30-2001
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
2-Mercaptoethanol( $\beta$ -Me)	gibco/21985-023
Puromycin	Genomeditech/ <a href="#">GM-040401</a>
PMA	Sigma/880134P
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/ <a href="#">GM-040503</a>

## Figures

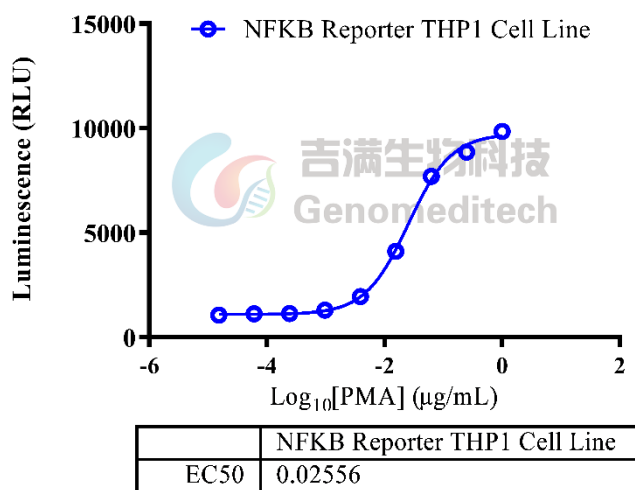


Figure 1 | Response to PMA. NFKB Reporter THP1 Cell Line (Cat. GM-C06727) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of PMA (Sigma/880134P) in assay buffer (RPMI 1640(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [9.4]. Data are shown by drug mass concentration.

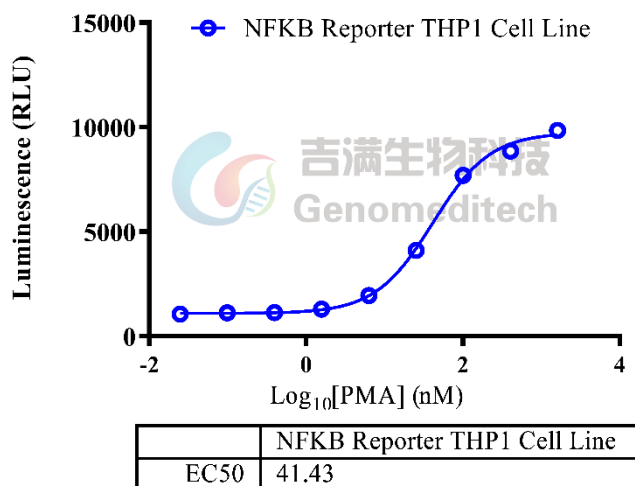


Figure 2 | Response to PMA. NFKB Reporter THP1 Cell Line (Cat. GM-C06727) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of PMA (Sigma/880134P) in assay buffer (RPMI 1640(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [9.4]. Data are shown by drug molar concentration.

## Cell Recovery

Recovery Medium: RPMI 1640(ATCC)+20% FBS+1% P.S+0.05 mM  $\beta$ -Me

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  $-70^{\circ}\text{C}$ . Storage at  $-70^{\circ}\text{C}$  will result in loss of viability.

- Thaw the vial by gentle agitation in a  $37^{\circ}\text{C}$  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).
- 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.
- Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium. And spin at approximately  $176 \times g$  for 5 minutes. Discard supernatant.
- Resuspend cell pellet with the recommended complete medium. And dispense the suspension into an appropriate culture flask and initially place the flask in an upright position after thawing.
- Incubate the culture at  $37^{\circ}\text{C}$  in a suitable incubator. A 5%  $\text{CO}_2$  in air atmosphere is recommended if using the medium described on this product sheet.

## Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- Centrifuge at  $176 \times g$  for 3 minutes to collect cells.
- Resuspend the cells in pre-cooled freezing medium and adjust the cell density to  $5 \times 10^6$  cells/mL.
- Aliquot 1 mL into each vial.
- Place the vial in a controlled-rate freezing container and store at  $-80^{\circ}\text{C}$  for at least 1 day, then transfer to liquid nitrogen as soon as possible.

## Cell passage

Growth medium: RPMI 1640(ATCC)+10% FBS+1% P.S+0.05 mM  $\beta$ -Me+0.5  $\mu\text{g}/\text{mL}$  Puromycin

During the first two passages after cell thawing, use the recovery medium. Once the cell status stabilizes, switch to growth medium containing antibiotics.

- When the cell density reaches  $8 \times 10^5$  cells/mL, subculture the cells. Do not allow the cell density to exceed  $1 \times 10^6$  cells/mL.
- It is recommended to use T-25 flasks for subculturing.
- These cells are suspension cells, and it is recommended to use the "half-medium change" method to maintain optimal cell conditions during passaging.
- During passaging, you can directly add fresh growth medium to the culture flask, gently pipette to resuspend the cells, and then transfer the cell suspension to a new T-25 flask for continued culture.

**Subcultivation Ratio: Maintain cultures at a cell concentraion between  $2.5 \times 10^5$  and  $8 \times 10^5$  viable cells/mL.**

**Medium Renewal: Every 2 to 3 days**

## Notes

- After thawing, cell growth is slow, and there will be a significant amount of cellular debris in the background. As the cells recover, the background will gradually become cleaner, with a recovery period estimated at 1 to 1.5 weeks.
- These cells are sensitive to cell density, so please ensure that cell density is maintained within an appropriate range during culture and passaging.
- The culture medium for these cells must be supplemented with  $\beta$ -mercaptoethanol. Failure to add this supplement may negatively affect cell status.
- Cells should be cultured using ATCC/30-2001 RPMI 1640 medium or complete medium purchased from Geomeditech. The serum used should be the same as specified in the manual or Gibco serum.

## Related Products

NFKB	
<a href="#">NFKB Reporter HEK-293 Cell Line</a>	<a href="#">NFKB Reporter Jurkat Cell Line</a>
<a href="#">NFKB Reporter TF-1 Cell Line</a>	

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