

# Certificate of Analysis

(Date: 5 April, 2018)



**GPC laboratory**  
Genome Program and Control

Product Name: THP-G8  
Catalog Number: GP-H01-P15  
Lot Number: 170809III31/25

TEST	RESULT
• Growth properties	Suspension
• Morphology	Lymphoblast-like
• Test for mycoplasma contamination Real-time PCR (Mycoseq Mycoplasma Detection Kit)	Negative
• Sterility test (BacT/ALERT 3D) iAST bottle (aerobic) at 32.5°C	Negative
iNST bottle (anaerobic) at 32.5°C	Negative

## Quality control testing (Refer to OECD TG442E, In vitro skin sensitisation: IL-8 Luc assay)

- For testing, THP-G8 cells are seeded at a density of  $2$  to  $5 \times 10^5$  cells/mL, and pre-cultured in culture flasks for 48 to 96 hours. On the day of the test, cells harvested from the culture flask are washed with RPMI-1640 containing 10% FBS without any antibiotics, and then, resuspended with RPMI-1640 containing 10% FBS without any antibiotics at  $1 \times 10^6$  cells/mL.
- Luminescence is measured using a 96-well microplate luminometer, Phelios (ATTO, Tokyo, Japan), equipped with optical filters.
- 100 $\mu$ L of pre-warmed Tripluc<sup>®</sup> Luciferase assay reagent is transferred to each well of the plate containing the cell suspension treated with or without chemical.
- Bioluminescence is measured for 3 sec each in the absence (F0) and presence (F1, 600nm long pass filter) of the optical filter.

## Results

Positive control: 4-NBB (4-nitrobenzyl bromide)

Negative control: LA (lactic acid)

(n=4, mean $\pm$ SD)

Chemical concentration (dilution)	0	4-NBB (20 mg/mL)			LA (20 mg/mL)		
		1/8,192	1/4,096	1/2,048	1/8,192	1/4,096	1/2,048
IL8LA	4,428 $\pm$ 303	21,425 $\pm$ 1,470	28,429 $\pm$ 1,029	287 $\pm$ 31	4,282 $\pm$ 133	3,970 $\pm$ 311	2,860 $\pm$ 219
GAPLA	6,732 $\pm$ 128	6,097 $\pm$ 342	5,710 $\pm$ 323	1,070 $\pm$ 104	6,235 $\pm$ 269	6,228 $\pm$ 509	6,004 $\pm$ 242
nIL8LA	0.658 $\pm$ 0.043	3.521 $\pm$ 0.029	4.986 $\pm$ 0.235	0.273 $\pm$ 0.057	0.687 $\pm$ 0.022	0.644 $\pm$ 0.101	0.477 $\pm$ 0.046
Inh-GAPLA	1.000 $\pm$ 0.019	0.906 $\pm$ 0.051	0.848 $\pm$ 0.048	0.159 $\pm$ 0.015	0.926 $\pm$ 0.040	0.925 $\pm$ 0.076	0.892 $\pm$ 0.036
Ind-IL8LA	1.000 $\pm$ 0.065	5.352 $\pm$ 0.440	7.579 $\pm$ 0.357	0.415 $\pm$ 0.087	1.045 $\pm$ 0.033	0.978 $\pm$ 0.153	0.725 $\pm$ 0.070

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