



# SZABO SCANDIC

Part of Europa Biosite

## Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

Weitere Information auf den folgenden Seiten!  
See the following pages for more information!



### Lieferung & Zahlungsart

siehe unsere [Liefer- und Versandbedingungen](#)

### Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



6042 Cornerstone Court West, Suite B  
San Diego, CA 92121  
**Tel:** 1.858.202.1401  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

## Data Sheet

### ***NF- $\kappa$ B Reporter (Luc) – Raw 264.7 Cell line*** **Catalog #:79978**

#### **Product Description**

The NF- $\kappa$ B reporter (Luc)-Raw 264.7 cell line is designed for monitoring nuclear factor Kappa B (NF- $\kappa$ B) signal transduction pathways. It contains a firefly luciferase gene driven by four copies of the NF- $\kappa$ B response element located upstream of the minimal TATA promoter. After activation by pro-inflammatory cytokines or stimulants of lymphokine receptors, endogenous NF- $\kappa$ B transcription factors bind to the DNA response elements, inducing transcription of the luciferase reporter gene.

#### **Applications**

- Screen for activators or inhibitors of NF- $\kappa$ B signaling pathway
- Screen for activators or inhibitors of RANKL/RANK pathway.

#### **Format**

Each vial contains 2 X 10<sup>6</sup> cells in 1 ml of 10% DMSO.

#### **Storage**

Store in liquid nitrogen immediately upon receipt.

#### **General Culture Conditions**

**Thaw Medium 11 (BPS Bioscience #79976):** DMEM medium (Hyclone #SH30024.01) supplemented with 10% heat-inactivated FBS (Gibco #26140-079), 1% GlutaMAX (Gibco #35050-061), 1% Penicillin/Streptomycin (Hyclone #SV30010.01).

**Growth Medium 11A (BPS Bioscience #79977):** Thaw Medium 11 (BPS Bioscience #79976) plus 700  $\mu$ g/ml of Genecticin (Life Technologies #11811031).

Cells should be grown at 37°C with 5% CO<sub>2</sub> using Growth Medium 11A.

NF- $\kappa$ B reporter (Luc)-Raw 264.7 cells should exhibit a typical cell division time of 24 hours.

**To thaw the cells,** it is recommended to quickly thaw the frozen cells from liquid nitrogen in a 37°C water-bath, transfer to a tube containing 10 ml of Thaw Medium 11 (**no Genecticin**), spin down cells, resuspend cells in pre-warmed Thaw Medium 11 (**no Genecticin**), transfer resuspended cells to T25 flask and culture in 37°C CO<sub>2</sub> incubator. At first passage switch to Growth Medium 11A (**contains Genecticin**). Cells should be split before they reach complete confluence.

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)



6042 Cornerstone Court West, Suite B  
San Diego, CA 92121  
**Tel:** 1.858.202.1401  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

**To passage the cells**, rinse cells with phosphate buffered saline (PBS) and enzyme-free cell dissociation buffer (ThermoFisher #13151014), detach cells from culture vessel with enzyme-free cell dissociation buffer, add Growth Medium 11A and transfer to a tube, spin down cells, re suspend cells and seed appropriate aliquots of cell suspension into new culture vessels. Subcultivation ratio: 1:10 every 3-4 days.

**To freeze down the cells**, rinse cells with phosphate buffered saline (PBS) and enzyme-free cell dissociation buffer (ThermoFisher #13151014), detach cells from culture vessel with enzyme-free cell dissociation buffer. Add Growth Medium 11A and transfer to a tube, spin down cells, and resuspend in freezing medium (10% DMSO + 90% FBS). Place at -80°C overnight and place in liquid nitrogen the next day. Alternatively, vials may be placed directly in liquid nitrogen.

### **Mycoplasma testing**

The cell line has been screened using the PCR-based VenorGeM™ Mycoplasma Detection kit (Sigma-Aldrich, #MP0025) to confirm the absence of *Mycoplasma* species.

### **Assay performance**

The following assays are designed for 96-well format. To perform the assay in different tissue culture formats, cell number and reagent volume should be scaled appropriately.

### **Materials Required but Not Supplied**

- mouse RANKL (R&D Systems #462-TEC-010)
- mouse TNF $\alpha$  (R&D Systems #390-TN-010)
- Assay Medium: Thaw Medium 11 (BPS Bioscience #79976)
- Growth Medium 11A (BPS Bioscience #79977)
- 96-well tissue culture treated white clear-bottom assay plate (Corning #3610)
- ONE-Step™ luciferase assay system (BPS Bioscience #60690) or other luciferase reagents for measuring firefly luciferase activity
- Luminometer

### **A. RANKL (or TNF $\alpha$ ) dose response**

1. Harvest NF- $\kappa$ B reporter (Luc)-Raw 264.7 cells and seed cells at a density of 30,000 cells per well into white opaque 96-well microplate in 90  $\mu$ l of assay medium. Incubate cells at 37°C with 5% CO<sub>2</sub> overnight.
2. Add threefold serial dilution of mouse RANKL (or mouse TNF $\alpha$ ) in 10  $\mu$ l of assay medium to RANKL (or TNF $\alpha$ ) -stimulated wells.
3. Add 10  $\mu$ l of assay medium to the unstimulated control wells (for measuring uninduced level of NF- $\kappa$ B reporter activity).
4. Add 100  $\mu$ l of assay medium to cell-free control wells (for determining background luminescence).

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)

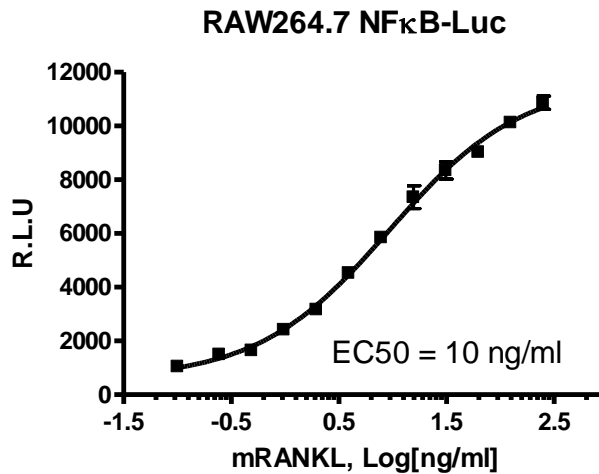


6042 Cornerstone Court West, Suite B  
San Diego, CA 92121  
Tel: 1.858.202.1401  
Fax: 1.858.481.8694  
Email: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

5. Incubate at 37°C with 5% CO<sub>2</sub> for 5-6 hours.
6. Prepare ONE-Step™ Luciferase Assay reagent as directed and add 100 µl per well. Incubate at room temperature for ~5 minutes and measure luminescence using a luminometer. Subtract background luminescence value from all measurements.

**Figure 1. Mouse RANKL dose response in NF-κB reporter (Luc)-RAW 264.7 cells.** The results are shown as relative luminescence units of luciferase reporter expression.

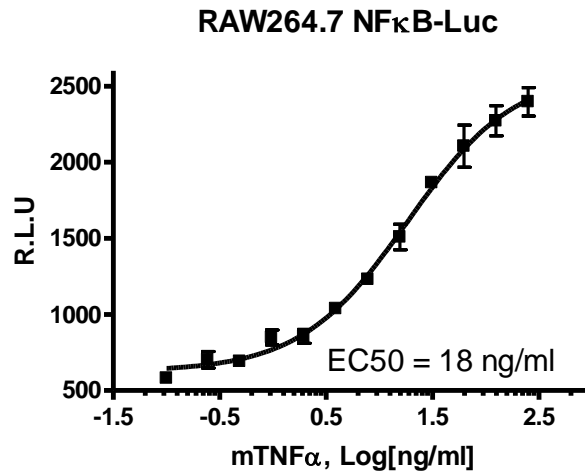
The EC<sub>50</sub> of mouse RANKL in this cell line is ~10 ng/ml.



OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.  
To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**  
Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)  
Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)

**Figure 2. Mouse TNF $\alpha$  dose response in NF- $\kappa$ B reporter (Luc)-RAW 264.7 cells.** The results are shown as relative luminescence units of luciferase reporter expression.

The EC<sub>50</sub> of mouse TNF $\alpha$  in this cell line is ~20 ng/ml.



## References

1. Penninger, J. M., *et al.* (2006) RANKL–RANK signaling in osteoclastogenesis and bone disease. *Trends Mol. Med.* **12(1)**:17-25.
2. Baeuerle, P.A. (1998) Pro-inflammatory signaling: last pieces in the NF- $\kappa$ B puzzle? *Curr Biol.* **8(1)**:R19-R22.

## License Disclosure

Purchase of this cell line grants you with a 10-year license to use this cell line in your immediate laboratory, for research use only. This license does not permit you to share, distribute, sell, sublicense, or otherwise make the cell line available for use to other laboratories, departments, research institutions, hospitals, universities, or biotech companies. The license does not permit use of this cell line in humans or for therapeutic or drug use. The license does not permit modification of the cell line in any way. Inappropriate use or distribution of this cell line will result in revocation of the license and result in an immediate cease of sales and distribution of BPS products to your laboratory. BPS does not warrant the suitability of the cell line for any particular use, and does not accept any liability in connection with the handling or use of the cell line. Modifications of this cell line, transfer to another facility, or commercial use of the cells may require a separate license and additional fees; contact [sales@bpsbioscience.com](mailto:sales@bpsbioscience.com) for details. Publications using this cell line should reference BPS Bioscience, Inc., San Diego.

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)



6042 Cornerstone Court West, Suite B  
San Diego, CA 92121  
**Tel:** 1.858.202.1401  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

### **Related Products**

<u>Product</u>	<u>Cat. #</u>	<u>Size</u>
Human Tumor Necrosis Factor-alpha	90244-A	10 µg
RANKL, His-Tag (Human)	71051	100 µg
Thaw Medium 11	79976	100 ml
Growth Medium 11A	79977	500 ml
NF- κB Reporter (Luc) – THP-1 Cell Line	79645	2 vials
NF- κB Reporter (Luc) – NIH/3T3 Cell Line	79469	2 vials
NF- κB Reporter (Luc) – Jurkat Cell Line	60651	2 vials
NF- κB Reporter (Luc) – A549 Cell Line	60625	2 vials
NF- κB Reporter (Luc) – HCT116 Cell Line	60623	2 vials
NF- B Reporter (Luc) – CHO-K1 Cell Line	60622	2 vials
NF-κB Reporter Kit	60614	500 rxns.
CD27/NF-κB Reporter-Jurkat Cell Line	79509	2 vials
TLR8/NF-κB Reporter-HEK293 Cell Line	60684	2 vials
GITR/NF-κB Reporter-Jurkat Cell Line	60546	2 vials

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.202.1401** Fax **1.858.481.8694**

Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

Please visit our website at: [www.bpsbioscience.com](http://www.bpsbioscience.com)