



# 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 W, Ste B  
San Diego, CA 92121  
**Tel:** 1.858.829.3082  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

## Data Sheet

### NF- $\kappa$ B-Luciferase Reporter HCT-116 Cell Line

### Catalog #60623

#### **Description**

NF- $\kappa$ B luciferase reporter construct is stably integrated into the genome of HCT-116 cells. The firefly luciferase gene is controlled by 4 copies of NF- $\kappa$ B response element located upstream of the TATA promoter. Following activation by stimulants, endogenous NF- $\kappa$ B transcription factors bind to the DNA response elements to induce transcription of the luciferase gene.

#### **Application**

The NF- $\kappa$ B-luciferase/HCT-116 cell line is suitable for monitoring the activity of NF- $\kappa$ B signaling in response to stimulants such as the cytokines TNF $\alpha$  and IL-1 $\beta$ , pathogen-associated molecular pattern (PAMP) (i.e. flagellin) or endogenous damage-associated molecular pattern (DAMP) molecules (i.e. NOD1 ligand) (see application references). It is also suitable for establishing cell-based screens for inhibitors that target specific NF- $\kappa$ B stimulating molecules. This cell line can be further modified to allow investigation of downstream NF- $\kappa$ B activities as a result of targeted genetic mutation(s).

#### **Host Cell**

HCT-116 Human Colorectal Carcinoma Cell line. Adherent epithelial cells.

#### **Format**

Each vial contains  $\sim 3 \times 10^6$  cells in 1 mL of 10% DMSO in FBS.

#### **Storage**

Store in liquid nitrogen immediately upon receipt.

#### **Culture Medium**

**Thaw Medium 7 (BPS #60185):** McCoy's 5A medium (Hyclone #SH30200.01) with 10% FBS (Life technologies #26140-079), 1% Penicillin/Streptomycin (Hyclone SV30010.01)

**Growth Medium 7A (BPS #79543):** Thaw Medium 7 (BPS Cat. #60185) plus 1 mg/ml Geneticin (G418) (Thermo Fisher, Cat. #11811031).

#### **Recommended Culture conditions**

*Frozen Cells:* Prepare T-75 culture flask with 20 ml of pre-warmed Thaw Medium 7 (**no G418**). Quickly thaw cells in a 37°C water bath with constant and slow agitation. After cleaning the outside of the vial with 70% ethanol, immediately transfer the entire content to Thaw Medium 7 (**no G418**). Avoid pipetting up and down, and gently rock the flask to distribute the cells. Incubate the cells in a humidified 37°C incubator with 5% CO<sub>2</sub>. 24-48 hours after incubation,

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

To place your order, please contact us by Phone **1.858.829.3082** 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 W, Ste B  
San Diego, CA 92121  
**Tel:** 1.858.829.3082  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

change to fresh Growth Medium 7A (**contains G418**), without disturbing the attached cells. Continue to change medium every 2-3 days until cells reach desired confluency. If slow cell growth occurs during resuscitation, increase FBS to 15% for the first week of culture.

*Subculture:* When cells reached 90% confluency, remove the medium and wash twice with PBS (without Magnesium or Calcium). Treat cells with 2-3 ml of 0.25% trypsin/EDTA and incubate for 2-3 minutes at 37°C. After confirming cell detachment by light microscopy, add 10 mL pre-warmed medium and gently pipette up and down to dissociate cell clumps. Transfer cells to a 15 ml conical tube and centrifuge at 200 x g for 5 minutes. Remove the medium and resuspend cells in 10 ml pre-warmed growth medium. Dispense 2 mL of the cell suspension into a new T75 flask containing pre-warmed 18 ml complete medium (a subcultivation ratio of 1:2 to 1:10 is recommended). Incubate cells in a humidified 37°C incubator with 5% CO<sub>2</sub>. To freeze cells, re-suspend cell pellet in freezing medium (10% DMSO in FBS). Cells have been demonstrated to be stable for at least 15 passages; BPS recommends preparing frozen stocks so cells are not used beyond passage 20.

### **Mycoplasma Testing**

This cell line has been screened using the MycoAlert™ Mycoplasma Detection Kit (Lonza, Cat. #LT07-118) to confirm the absence of Mycoplasma contamination. MycoAlert Assay Control Set (Lonza, Cat. #LT07-518) was used as a positive control.

### **Application References**

1. Samuel T *et.al.* (2014) Variable NF-κB pathway responses in colon cancer cells treated with chemotherapeutic drugs. *BMC Cancer* **14**: 599.
2. Arabi A *et.al.* (2012) Proteomic screen reveals Fbw7 as a modulator of the NF-κB pathway. *Nature Communication* **3**: 976.
3. Clemo NK *et.al.* (2008) BAG-1 is up-regulated in colorectal tumour progression and promotes colorectal tumour cell survival through increased NF-κB activity. *Carcinogenesis* **29**: 849.
4. Tikhvatulin AI *et.al.* (2011) An *In Vitro* and *In Vivo* Study of the ability of NOD1 Ligands to Activate the Transcriptional Factor NF-κB. *Acta Naturae* **3**: 77.

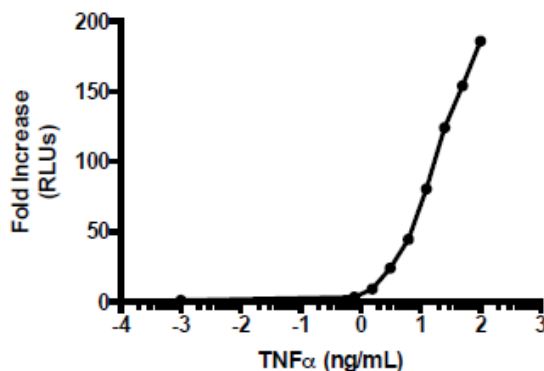
OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

To place your order, please contact us by Phone **1.858.829.3082** Fax **1.858.481.8694**

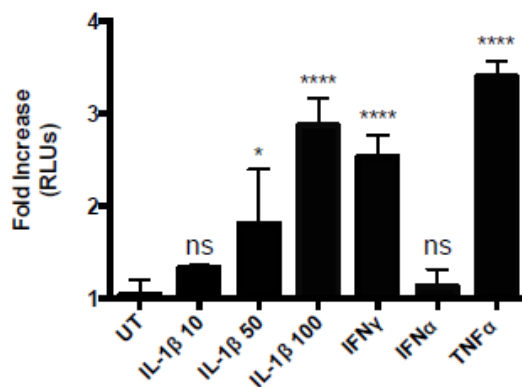
Or you can Email us at: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

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

### Quality Assurance and Functional Analysis



**Figure 1. Analysis of NF-κB / HCT-116 reporter activity in response to TNFα.** NF-κB-Luciferase HCT-116 cells were seeded on a white opaque 96-well plate overnight at 5000 cells/well in complete growth medium. Cells were treated with human TNFα in growth medium and incubated for 7 hours at 37°C before the addition of luciferin according to manufacturer’s protocol (ONE-Step™ Luciferase assay system, BPS Bioscience, Cat. #60690-2). Luminescence was read using a luminometer and readings were normalized to wells that only contain medium to obtain the Relative Luminescence Units (RLUs). Error bar = standard deviation (SD), n=3.



**Figure 2. Analysis of NF-κB / HCT-116 reporter activity in response to cytokine stimulation.** NF-κB-Luciferase HCT-116 cells were seeded on a white opaque 96-well plate overnight at 5000 cells/well in complete growth medium. Cells were treated with human cytokines in growth medium (IL-1β, 10, 50, or 100 ng/ml; IFNγ, 2 μg/ml; IFNα, 10<sup>4</sup> U/ml; TNFα, 0.8 ng/ml) and incubated for 7 hours at 37°C, followed by the addition of luciferin according to manufacturer’s protocol (ONE-Step™ Luciferase assay system, BPS Bioscience, Cat. #60690-2). Luminescence was read using a luminometer and readings were normalized to wells containing only medium to determine the Relative Luminescence Unit (RLU). Fold increase is calculated with respect to untreated control cells (UT). Error bar = standard deviation (SD), n=3. \* P < 0.05, \*\*\*\* P < 0.0001, ns = not significant. One way ANOVA.

OUR PRODUCTS ARE FOR RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.  
 To place your order, please contact us by Phone **1.858.829.3082** 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 W, Ste B  
San Diego, CA 92121  
**Tel:** 1.858.829.3082  
**Fax:** 1.858.481.8694  
**Email:** [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

### Vector

NF- $\kappa$ B-Luciferase was cloned into the MCS of pCDNA3.1™ (+) vector (Invitrogen, Cat. #V79020).

### Related Products

<u>Product</u>	<u>Cat. #</u>	<u>Size</u>
Thaw Medium 7	60185	100 ml
ONE-Step™ Luciferase Assay System	60690-1	10 ml
ONE-Step™ Luciferase Assay System	60690-2	100 ml
NF- $\kappa$ B (Luc) Reporter CHO-K1 Cell Line	60622	2 vials
NF- $\kappa$ B reporter (Luc) - HEK293 Cell line	60650	2 vials
NF- $\kappa$ B Reporter Kit (NF- $\kappa$ B Signaling Pathway)	60614	500 rxns

### 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 **1.858.829.3082** 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)