

# Product Datasheet

## Human NFkB Secreted Alkaline Phosphatase Reporter SEAP - (SEAPorter™) Stable Reporter Cell Line NBP2-26260

Unit Size: 1 Vial

Store in gas phase of liquid nitrogen.

[www.novusbio.com](http://www.novusbio.com)



[technical@novusbio.com](mailto:technical@novusbio.com)

### Publications: 7

Protocols, Publications, Related Products, Reviews, Research Tools and Images at:  
[www.novusbio.com/NBP2-26260](http://www.novusbio.com/NBP2-26260)

Updated 2/10/2021 v.20.1

Earn rewards for product  
reviews and publications.

Submit a publication at [www.novusbio.com/publications](http://www.novusbio.com/publications)

Submit a review at [www.novusbio.com/reviews/destination/NBP2-26260](http://www.novusbio.com/reviews/destination/NBP2-26260)



**NBP2-26260****Human NFkB Secreted Alkaline Phosphatase Reporter SEAP - (SEAPorter™) Stable Reporter Cell Line**

<b>Product Information</b>	
<b>Unit Size</b>	1 Vial
<b>Concentration</b>	Concentration is not relevant for this product. Please see the protocols for proper use of this product.
<b>Storage</b>	Store in gas phase of liquid nitrogen.
<b>Reconstitution Instructions</b>	Complete Growth Medium: DMEM with 4.5 g/L glucose + 10% FBS + 4 mM L-glutamine + 1 mM sodium pyruvate + 100 units/ml penicillin + 0.1 mg/ml streptomycin + 0.5 mg/ml G418 (Geneticin).
<b>Product Description</b>	
<b>Description</b>	<p>Placental alkaline phosphatase is one of the most stable isoenzyme, only existing in the placenta of higher primates. These characteristics make placental alkaline phosphatase suitable to use as a reporter gene for the analysis of promoter activity and gene expression in cell culture and animal serum. The natural form of placental alkaline phosphatase (PLAP) is membrane anchored. The recombinant form of placental alkaline phosphatase (secreted alkaline phosphatase, SEAP) is used for reporter gene function. SEAP is created by inserting a translational terminator after amino acid 489 (Berger, et al., Gene 66 (1): 10 (1988). This mutation converts the membrane-bound PLAP protein into the secreted protein.</p> <p>As a major transcription factor, NF-kB plays a key role in regulating genes responsible for the innate and adaptive immune responses. In unstimulated cells, the NF-kB dimers are held in the cytoplasm by IκBs that masks the nuclear localization signals of NF-kB. Upon cell stimulation, which leads to IκB degradation, NF-kB quickly translocates to the nucleus and activates various genes that have DNA-binding sites for NF-kB.</p> <p>The NF-kB/SEAP stable HEK 293 cell line is designed to measure NF-kB activation using SEAP protein secreted to the culture media as a read-out with our SEAPorter™ Assay kit (NBP2-25285). The NF-kB/SEAP stable cells are not only useful in helping with the identification of pro or anti-inflammatory substances, but also can help to assay for proteasome activity since the activation of NF-kB results in the degradation of IκB through the proteasome-dependant pathway.</p> <p>Contents: 3~4 x 10<sup>6</sup> cells Biosafety Level: 2</p>
<b>Host</b>	HEK293
<b>Target Species</b>	Human
<b>Reporter Gene</b>	Secreted alkaline phosphatase (SEAP)
<b>Species</b>	Human
<b>Reactivity Notes</b>	Human reactivity reported in scientific literature (PMID:33201697)
<b>Selection Agent</b>	G418
<b>Specificity/Sensitivity</b>	NF-kB/SEAP
<b>Immunogen</b>	The NF-kB/SEAP is a reporter gene assay for the detection of NF-kB activation and contains a cell line modified for this purpose. This cell line is derived from HEK 293 (Human Embryonal Kidney) cells. It is stably transfected with the SEAP (secreted alkaline phosphatase) reporter gene under the transcriptional control of an NF-kB response element. The recombinant cell line is provided frozen.

<b>Notes</b>	<p>Assume all cultures are hazardous since they may harbor latent viruses or other organisms that are uncharacterized. The following safety precautions should be observed.</p> <ul style="list-style-type: none"> <li>- Use pipette aids to prevent ingestion and keep aerosols down to a minimum.</li> <li>- No eating, drinking or smoking while handling cells.</li> <li>- Wash hands after handling cultures and before leaving the lab.</li> <li>- Decontaminate work surface with disinfectant or 70% ethanol before and after working with cells.</li> <li>- All waste should be considered hazardous.</li> <li>- Dispose of all liquid waste after each experiment and treat with bleach.</li> </ul>
<b>Lysate Type</b>	Cell
<b>Product Application Details</b>	
<b>Applications</b>	Immunocytochemistry/ Immunofluorescence, Ligand Activation
<b>Recommended Dilutions</b>	Immunocytochemistry/ Immunofluorescence, Ligand Activation
<b>Application Notes</b>	Use in ICC/IF reported in scientific literature (PMID:33201697)



## Publications

Jackson PA, Schares HAM, Jones KFM et al. Synthesis of Guaianolide Analogues with a Tunable alpha-Methylene-gamma-lactam Electrophile and Correlating Bioactivity with Thiol Reactivity Nat Commun 2020-11-13 [PMID: 33201697] (ICC/IF, Human)

MohanKumar, K;Namachivayam, K;Song, T;Jake Cha, B;Slate, A;Hendrickson, JE;Pan, H;Wickline, SA;Oh, JY;Patel, RP;He, L;Torres, BA;Maheshwari, A; A murine neonatal model of necrotizing enterocolitis caused by anemia and red blood cell transfusions Nat Commun 2019-08-02 [PMID: 31375667]

Zurcher C, Sauter KS, Schweizer M. Pestiviral Erns blocks TLR-3-dependent IFN synthesis by LL37 complexed RNA. Veterinary Microbiology. 2014-10-13 [PMID: 25457366]

Signorino G, Mohammadi N, Patane F et al. Role of TLR13 in innate immune recognition of group B streptococci. Infect. Immun. 2014-09-15 [PMID: 25225249] (In vitro, Human)

### Details:

Human NFkB Secreted Alkaline Phosphatase SEAP - (SEAPorter™) Stable Reporter Cell Line was transiently transfected with full-length mouse TLR13 and stimulated with heat-killed bacteria and bacterial RNA or with phorbol myristate acetate /PMA followed by quantification of NFkB Secreted Alkaline Phosphatase using ELISA kit for the latter.

Rockman S, Dyson A, Koernig S et al. Evaluation of the bioactivity of influenza vaccine strains in vitro suggests that the introduction of new strains in the 2010 Southern Hemisphere Trivalent Influenza Vaccine is associated with adverse events. Vaccine. 2014-06-24 [PMID: 24928062] (ELISA, Human)

### Details:

Figs 5,6: The HEK293 NF-kB/SEAP reporter cells were stimulated with various influenza strains from the 2005-2012 flu season. Note: Recombinant TNF-alpha, endotoxin, and TLR ligands poly I:C and R848 were used as positive stimulation controls for activating the reporter cell line. The authors also did some additional characterization of the cell line, and the cell line tested positive for the endogenous presence of TLR3, TLR8, MDA5, and RIG-I but not TLR7 by RT-PCR.

Laguet N, Bregnard C, Hue P et al. Premature activation of the SLX4 complex by Vpr promotes G2/M arrest and escape from innate immune sensing. Cell. 2014-01-16 [PMID: 24412650] (Human)

### Details:

AP-1-Luciferase activity, Figs 6D, F, G

Andre CM, Greenwood JM, Walker EG et al. Anti-inflammatory procyanidins and triterpenes in 109 apple varieties. J Agric Food Chem. 2012-10-24 [PMID: 23013475]

### Details:

Product cited, NF-kB/SEAPorter HEK293 Cell Line (IML-101): Fig 3, Table 1, S2. Readout assay: The study evaluated the potential of apple varieties to reduce inflammation. Phenolic and triterpenes from apple flesh were analyzed for their ability to inhibit

## Procedures

### Product Handling Protocol (NBP2-26260)

To ensure the highest cell viability, it is strongly recommended that one should thaw the vial and initiate the cell culture as soon as possible upon receipt. If continued storage of the frozen vial upon receipt is necessary, it should be immediately stored in liquid nitrogen but not at -80C. Storage at -80C will lead to significant loss of cell viability. Please read the entire data sheet before thawing the cells. It is recommended that users follow good cell culture practice when using the cells. The cells are sterile and all work should be performed under sterile conditions.

1. Prepare a sterile 15-ml tube with 9 ml fresh medium without selection agents pre-warmed at 37C.
2. Thaw the frozen cell vial quickly in a 37C water bath, keeping the cap portion out of the water to avoid any possible contamination.
3. Upon thawing, take the vial out of the water and clean it with 70% ethanol to decontaminate.
4. Transfer contents to the 15-ml tube (Step 1) and mix with medium by gentle inversion of tube.
5. Centrifuge at 1,000 RPM for 5 minutes.
6. Remove supernatant and resuspend cells in 10 ml of fresh medium without selection agents. It is important to grow the cells at this stage without any selection agents.
7. Transfer cells into a 25-cm<sup>2</sup> tissue culture flask and incubate at 37C in a 95% air-5% CO<sub>2</sub> mixture.
8. After cells settle down (in 1-3 days), remove the medium containing minor floating cells and replace with fresh complete growth medium containing selection agents.
9. Whenever the cells are 70-80% confluent, detach the cells by trypsinization and split into new flasks with fresh complete growth medium.
10. Freeze IML cell lines at 3-4x10<sup>6</sup> cells/ml per cryogenic vial. For optimal cell viability after freezing, freeze cells when they have reached log phase growth (95-98% confluency). Detach cells by trypsinization at 37C for 5 min, and harvest cells by mixing with 3 volumes of fresh medium followed by centrifugation (Step 5). Re-suspend the cell pellet in freeze media (FBS with 10% DMSO). Add cell suspension to cryogenic vials in 1 ml aliquots. Place cryogenic vials, in a tissue culture approved cryogenic vial container, in -80C freezer for 24-48 hours. After 24-48 hours, move the vials into liquid nitrogen storage.

### MSDS (NBP2-26260)

#### IDENTIFICATION

Product Name HEK 293 cells (human embryonic kidney), HeLa cells (human epithelial carcinoma) or RAW cells (mouse

macrophage) stably transfected; Dimethyl sulfoxide

Synonyms Methyl sulfoxide; DMSO; Sulfinylbis (methane)

#### COMPOSITION, INFORMATION ON INGREDIENTS

CAS# none

Name Cells, human origin

CAS# 67-68-5

Chemical Name Dimethyl Sulfoxide

Percent 10

#### HAZARDS IDENTIFICATION

#### EMERGENCY OVERVIEW

Appearance: clear liquid. May be absorbed through intact skin. Hygroscopic (absorbs moisture from the air). May cause liver and kidney damage. CAUTION! Causes eye and skin irritation. Causes respiratory tract irritation.

Target Organs: Kidneys, liver, eyes, skin, mucous membranes.

Potential Health Effects



**Eye:** Produces irritation, characterized by a burning sensation, redness, tearing, inflammation, and possible corneal injury. May cause chemical conjunctivitis.

**Skin:** May cause irritation with burning pain, itching and redness. Substance is rapidly absorbed through the skin.

**Ingestion:** May cause gastrointestinal irritation with nausea, vomiting and diarrhea. May cause liver and kidney damage. May cause garlic smell on the breath and body.

**Inhalation:** May cause respiratory tract irritation. Can produce delayed pulmonary edema.

**Chronic:** Prolonged or repeated skin contact may cause dermatitis. May cause liver and kidney damage. Effects may be delayed.

## FIRST AID MEASURES

**Eyes:** Immediately flush eyes with plenty of water for at least 15 minutes, occasionally lifting the upper and lower eyelids. Get medical aid.

**Skin:** Get medical aid. Flush skin with plenty of water for at least 15 minutes while removing contaminated clothing and shoes. Wash clothing before reuse.

**Ingestion:** Never give anything by mouth to an unconscious person. Get medical aid. Do NOT induce vomiting. If conscious and alert, rinse mouth and drink 2-4 cupfuls of milk or water.

Product Name HEK 293 cells (human embryonic kidney), HeLa cells (human epithelial carcinoma) or RAW cells (mouse

macrophage) stably transfected; Dimethyl sulfoxide

Synonyms Methyl sulfoxide; DMSO; Sulfinylbis (methane)

**Inhalation:** Remove from exposure and move to fresh air immediately. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical aid. Do NOT use mouth-to-mouth resuscitation.

**Notes to Physician:** Treat symptomatically and supportively.

## FIRE FIGHTING MEASURES

**General Information:** As in any fire, wear a self-contained breathing apparatus in pressure-demand, MSHA/NIOSH (approved or equivalent), and full protective gear. Vapors may form an explosive mixture with air. During a fire, irritating and highly toxic gases may be generated by thermal decomposition or combustion. Use water spray to keep fire-exposed containers cool. Vapors may be heavier than air. They can spread along the ground and collect in low or confined areas. Containers may explode when heated.

**Extinguishing Media:** Cool containers with flooding quantities of water until well after fire is out. Use water spray, dry chemical, carbon dioxide, or appropriate foam.

**Flash Point:** 95 deg C ( 203.00 deg F)

**Autoignition Temperature:** 215 deg C ( 419.00 deg F)

**Explosion Limits, Lower:** 2.6 vol %

**Upper:** 42 vol %

**NFPA Rating:** (estimated) Health: 1; Flammability: 1; Instability: 0

## ACCIDENTAL RELEASE MEASURES

**General Information:** Use proper personal protective equipment as indicated in Section 8.

**Spills/Leaks:** Absorb spill with inert material (e.g. vermiculite, sand or earth), then place in suitable container. Do not flush into a sewer. Clean up spills immediately, observing precautions in the Protective Equipment section. Provide ventilation.

## HANDLING AND STORAGE

**Handling:** Avoid contact with eyes, skin, and clothing. Keep container tightly closed. Avoid ingestion and inhalation. Use with adequate ventilation. Wash clothing before reuse.

**Storage:** Keep away from heat, sparks, and flame. Store in a tightly closed container. Keep from contact with oxidizing materials. Store in a cool, dry, well-ventilated area away from incompatible substances.

## EXPOSURE CONTROLS, PERSONAL PROTECTION

**Engineering Controls:** Facilities storing or utilizing this material should be equipped with an eyewash facility and a safety shower. Use adequate ventilation to keep airborne concentrations low.

**Exposure Limits**

Chemical Name Dimethyl Sulfoxide

ACGIH None listed

NIOSH None listed

OSHA - Final PELs None listed

OSHA Vacated PELs: Dimethyl sulfoxide: No OSHA Vacated PELs are listed for this chemical.

Personal Protective Equipment Eyes: Wear chemical goggles.

Skin: Wear appropriate protective gloves to prevent skin exposure.

Clothing: Wear appropriate protective clothing to prevent skin exposure.

Respirators: A respiratory protection program that meets OSHA's 29 CFR 1910.134 and ANSI Z88.2 requirements or European Standard EN 149 must be followed whenever workplace conditions warrant a respirator's use.

## PHYSICAL AND CHEMICAL PROPERTIES

Physical State: Liquid

Appearance: clear

Odor: slight odor - sulfurous odor - garlic-like odor

pH: Not available.

Vapor Pressure: 0.4 mm Hg at 20 Vapor Density: 2.7 (air=1) Evaporation Rate: Not available. Viscosity: 1.1cp @ 27

deg C Boiling Point: 189 deg C Freezing/Melting Point: 18.4 deg C

Decomposition Temperature: > 200 deg C

Solubility: Soluble.

Specific Gravity/Density: 1.1010g/cm<sup>3</sup> Molecular Formula: C<sub>2</sub>H<sub>6</sub>OS Molecular Weight: 78.13

## STABILITY AND REACTIVITY

Chemical Stability: Stable at room temperature in closed containers under normal storage and handling conditions.

Conditions to Avoid: Excess heat.

Incompatibilities with Other Materials: Strong oxidizing agents, strong acids, strong bases. Hazardous Decomposition

Products: Carbon monoxide, oxides of sulfur, carbon dioxide. Hazardous Polymerization: Has not been reported.

## TOXICOLOGICAL INFORMATION RTECS#

CAS# 67-68-5: PV6210000

### LD50/LC50

CAS# 67-68-5:

Draize test, rabbit, eye: 100 mg;

Draize test, rabbit, eye: 500 mg/24H Mild; Draize test, rabbit, skin: 500 mg/24H Mild; Oral, mouse: LD50 = 7920 mg/kg;

Oral, rat: LD50 = 14500 mg/kg; Skin, rat: LD50 = 40 gm/kg;<BR.

### Carcinogenicity

CAS# 67-68-5: Not listed by ACGIH, IARC, NIOSH, NTP, or OSHA.

Epidemiology: No information available. Teratogenicity: No information available. Reproductive Effects: No information available. Neurotoxicity: No information available.

Mutagenicity: No information available.

Other Studies: See actual entry in RTECS for complete information.

## ECOLOGICAL INFORMATION

Ecotoxicity: No data available. No information available.

Environmental: Terrestrial: Expected to be mobile in soil, due to its high water solubility. Some volatilization from dry soil and surfaces may be expected. Aquatic: Dimethyl sulfoxide disproportionates in water to dimethyl sulfide and dimethyl sulfone, a reaction catalyzed by light. Atmospheric: Exists primarily in the vapor phase and be removed by both wet and dry deposition. It will react with photochemically-produced hydroxyl radicals with a half-life of about 7 hr. DMSO is very difficult to biodegrade. Physical: No information available.

Other: For more information, see "HANDBOOK OF ENVIRONMENTAL FATE AND EXPOSURE DATA."

## DISPOSAL CONSIDERATIONS

Chemical waste generators must determine whether a discarded chemical is classified as a hazardous waste. US EPA guidelines for the classification determination are listed in 40 CFR Parts 261.3. Additionally, waste generators must consult state and local hazardous waste regulations to ensure complete and accurate classification.

RCRA P-Series: None listed





RCRA U-Series: None listed

#### SPECIAL PRECAUTIONS

Store at 2-8 degrees C in well-sealed container. Store away from strong oxidizing agents. This product is intended for research use only.

#### DISCLAIMER

For R&D use only. Not for drug, household or other uses.

#### WARRANTY

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. The information in this document is based on the present state of our knowledge and is applicable to the product with regard to appropriate safety precautions. It does not represent any guarantee of the properties of the product. NOVUS, shall not be held liable for any damage resulting from handling or from contact with the above product. See reverse side of invoice or packing slip for additional terms and conditions of sale. Copyright 2008 NOVUS License granted to make unlimited paper copies for internal use only.







### **Novus Biologicals USA**

10730 E. Briarwood Avenue  
Centennial, CO 80112  
USA  
Phone: 303.730.1950  
Toll Free: 1.888.506.6887  
Fax: 303.730.1966  
nb-customerservice@bio-techne.com

### **Bio-Techne Canada**

21 Canmotor Ave  
Toronto, ON M8Z 4E6  
Canada  
Phone: 905.827.6400  
Toll Free: 855.668.8722  
Fax: 905.827.6402  
canada.inquires@bio-techne.com

### **Bio-Techne Ltd**

19 Barton Lane  
Abingdon Science Park  
Abingdon, OX14 3NB, United Kingdom  
Phone: (44) (0) 1235 529449  
Free Phone: 0800 37 34 15  
Fax: (44) (0) 1235 533420  
info.EMEA@bio-techne.com

### **General Contact Information**

[www.novusbio.com](http://www.novusbio.com)  
Technical Support: nb-technical@bio-techne.com  
Orders: nb-customerservice@bio-techne.com  
General: novus@novusbio.com

### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Reporter Cell Lines are guaranteed for 1 year from date of receipt.

For more information on our 100% guarantee, please visit [www.novusbio.com/guarantee](http://www.novusbio.com/guarantee)

Earn gift cards/discounts by submitting a review: [www.novusbio.com/reviews/submit/NBP2-26260](http://www.novusbio.com/reviews/submit/NBP2-26260)

Earn gift cards/discounts by submitting a publication using this product:  
[www.novusbio.com/publications](http://www.novusbio.com/publications)

