Product Datasheet

RIG-I/NF-kB Luciferase - (LUCPorter™) Stable Reporter Cell Line

NBP2-32289

Unit Size: 1 Vial

Store in gas phase of liquid nitrogen.

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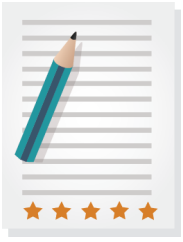

NBP2-32289

RIG-I/NF-kB Luciferase - (LUCPorter™) Stable Reporter Cell Line

Product Information	
Unit Size	1 Vial
Concentration	Concentration is not relevant for this product. Please see the protocols for proper use of this product.
Storage	Store in gas phase of liquid nitrogen.
Reconstitution Instructions	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 (Note: The selection agents for this cell line are blasticidin at 5 ug/ml and puromycin at 3 ug/ml).

Product Description	
Description	The RIG-I/NF-kB LUCPoter(TM) reporter cell line is designed to monitor the RIG-I-mediated NF-kB induction activity, and can be used for studying RIG-I signaling pathways as well as screening of agonists, antagonists or signaling inhibitors for RIG-I. Contents: 3-4 x 10^6 cells Biosafety Level: 2
Host	HEK293
Growth Properties	Adherent Morphology : Epithelial
Selection Agent	Blasticidin at 5 mg/ml and Puromycin at 3 mg/ml
Immunogen	The RIG-I/NF-kB LUCPorter(TM) reporter cell line is a stably transfected HEK 293T cell line which expresses human retinoic acid-induced protein-I (RIG-I) and an optimized Renilla luciferase reporter gene (RenSP) under the transcriptional control of an NF-kB response element. As a dsRNA helicase enzyme, RIG-I is encoded by the DDX58 gene. RIG-I is one of the RIG-I-like receptors (RLRs) that are a family of DExD/H box RNA helicases including RIG-I, MDA5 and LPG2, which play a major role in pathogen sensing of RNA virus infection to initiate and modulate antiviral immunity. RLR expression is typically maintained at low levels in resting cells but is greatly increased during inflammation, specifically with IFN exposure and after virus infection. RIG-I detects cytoplasmic dsRNA generated during viral replication unlike Toll-like receptor 3 (TLR3) which can detect phagocytosed dsRNA in endosomes. RIG-I also responds to poly(I:C), the synthetic analog of viral dsRNA. As shown in Figure 1, the RIG-I/NF-kB LUCPorter(TM) reporter cell line was specifically activated, in a dose response manner, by the poly(I:C) packed with Lipofectamine 2000, in which Lipofectamine mediated the cytoplasmic translocation of poly(I:C) (Figure 1).

Product Application Details	
Applications	Ligand Activation
Recommended Dilutions	Ligand Activation

Application Notes	Refer to the following preparation protocol for the poly(I:C) ligand packed with Lipofectamine 2000: 1. A 500 ug (50 ul, 10 mg/ml) of poly(I:C) (NBP2-25288) was preincubated in 50 ul Opti-MEM (Life Technologies) for 5 min. Similarly, a 20 ul Lipofectamine 2000 (Life Technologies) was preincubated in 80 ul Opti-MEM for 5 min. After 5 min, they were combined together as a total volume of 200 ul and further incubated for 20 min at room temperature. 2. The poly(I:C) packed with Lipofectamine 2000 (A 200 ul total at 2.5 mg/ml) was then used to stimulate the RIG-I/NF-kB LUCPorter(TM) HEK 293T cell line as described in Figure 1A.

Procedures

Product Handling Protocol (NBP2-32289)
Note: 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. It is recommended that users follow good tissue culture practice. The reporter line is 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. Note: It is important to grow the cells at this stage without selection agents.

7. Transfer cells into a 25-cm2 tissue culture flask and incubate at 37C in a 95% air-5% CO2 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 the reporter cell line at 3-4 x 10^6 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 (NBP2-32289)
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 contain- ers 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/cm3 Molecular Formula: C2H6OS 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.

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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

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