

Product Datasheet

HRE Luciferase - (LUCPorter™) Stable Reporter Cell Line NBP2-32798

Unit Size: 1 Vial

Store in gas phase of liquid nitrogen.

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NBP2-32798**HRE 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 is puromycin at 1 ug/ml).

Product Description	
Description	<p>The HRE/LUCPorter(TM) reporter cell line is designed to monitor the induction of HRE and can be used for screening of agonists, antagonists or signaling inhibitors of HRE induction (Hypoxia) as well as for studying the hypoxia-related signaling pathways.</p> <p>Contents: 3-4 x 10⁶ cells Biosafety Level: 2</p>
Host	HeLa
Growth Properties	Adherent Morphology : Epithelial
Selection Agent	Puromycin at 1 ug/ml
Immunogen	The HRE/LUCPorter(TM) reporter cell line is a stably transfected HeLa cell line which expresses an optimized Renilla luciferase reporter gene (RenSP) under the transcriptional control of the hypoxia response element (HRE). In response to hypoxia (low oxygen), HREs of target genes are recognized and regulated by the hypoxia-inducible factors (HIFs) which belong to the family of basic helix-loop-helix transcription factors and form heterodimeric complex comprising the alpha subunit (HIF-1 alpha, HIF-2 alpha and HIF-3 alpha) and beta subunit (Arnt1, Arnt2 and Arnt3), among which HIF-1 alpha and HIF-2 alpha are predominant isoforms. Activation of HIFs can also be mediated by chemical hydroxylase inhibitors as hypoxia mimetics including the iron chelator desferrioxamine and cobalt chloride. As also shown in Figure 1, the HRE activity in the HRE/LUCPorter(TM) reporter cell line was induced by cobalt chloride in a dose response manner (Figure 1).

Product Application Details	
Applications	Ligand Activation
Recommended Dilutions	Ligand Activation

Procedures

Product Handling Protocol (NBP2-32798)

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 $3-4 \times 10^6$ 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.

Product Handling Guide (NBP2-32798)

Assume all cultures are hazardous since they may harbor latent viruses or other organisms that are uncharacterized. The following safety precautions should be observed.

Use pipette aids to prevent ingestion and keep aerosols down to a minimum.

No eating, drinking or smoking while handling cells.

Wash hands after handling cultures and before leaving the lab.

Decontaminate work surface with disinfectant or 70% ethanol before and after working with cells.

All waste should be considered hazardous.

Dispose of all liquid waste after each experiment and treat with bleach.





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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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