BOSTER BIOLOGICAL TECHNOLOGY
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RIG-I/NF-kB Luciferase Reporter-HEK293T Cell Line

Catalog number: RC1020

This package insert must be read in its entirety before using this product.
For research use only. Not for use in diagnostic procedures.

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Catalog Number: RC1020, **Storage:** Immediately upon receipt, store in liquid nitrogen. (Ship on dry ice.)

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Contents: Each vial contains 2 ~ 3 x 10^6 cells in 1 ml of 90% FBS + 10% DMSO.
Description: The RIG-I Luciferase reporter cell line is a stably transfected HEK 293T cell line which expresses human retinoic acid-induced protein-I (RIG-I) and Renilla luciferase reporter gene under the transcriptional control of the 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. The RIG-I activation by poly (I:C) is shown in Figure 1.
Applications: Functional Assay
Application Notes: Functional Assay, detecting the transcriptional activity of RIG-I/NF-kB
Application Details:

Application:

Monitor the RIG-I signaling pathway activity.Screen for activators or inhibitors of the RIG-I signaling pathway.

Culture conditions:

Cells should be grown at 37°C with 5% CO2 using DMEM medium supplemented with 10% FBS and 1% Pen/Strep, plus 2 µg/ml Puromycin and 5 µg/ml blasticidin. It is recommended to quickly thaw the frozen cells upon receipt or from liquid nitrogen in a 37°C water-bath, transfer to a tube containing 10 ml of growth medium without Puromycin and Blasticidin, spin down cells, resuspend cells in pre-warmed growth medium without Puromycin and Blasticidin, transfer resuspended cells to T25 flask and culture in 37°C-CO2 incubator. Leave the T25 flask in the incubator for 2~4 days without disturbing or changing the medium until cells completely recover viability and become adherent. Once cells are over 90% adherent, remove growth medium and passage the cells through trypsinization and centrifugation. At first passage, switch to growth medium containing Puromycin and Blasticidin. Cells should be split before they reach complete confluence. To passage the cells, detach cells from culture vessel with Trypsin/EDTA, add complete growth medium and transfer to a tube, spin down cells, resuspend cells and seed appropriate aliquots of cells suspension into new culture vessels. Subcultivation ration = 1:10 to 1:20 weekly.

Functional validation:

A. Response of RIG-I HEK293T cells to Poly (I:C).1. Harvest RIG-I HEK293T cells and seed cells into a white solid-bottom 96-well microplate in 100

RIG-I/NF-kB Luciferase Reporter-HEK293T Cell Line (RC1020) Images

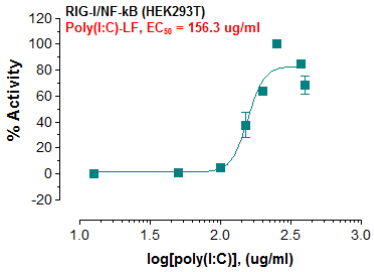

Fig-1: Induction of RIG-I activity by poly(I:C) prepacked with lipofectamine in RIG-I HEK293T cells.

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