HSV-60c/LyoVec™

Viral DNA motif complexed with LyoVec™- Negative Control

Catalog # tlrl-hsv60cc

For research use only

Version # 13D08-MM

PRODUCT INFORMATION

Content:

- 100 µg HSV-60c/LyoVec™

Note: Each vial contains 25 µg of HSV-60c complexed with 50 µg LyoVec™.

- 10 ml endotoxin-free water

Sequence:

5'-TAAGACACGATGCGATAAAATCTGTTTGTAAAATTTATTA--AGGGTACAAATTGCCCTAGC-3'

Storage:

- HSV-60c/LyoVec™ is provided lyophilized and shipped at room temperature. Store lyophilized product at -20˚C. Lyophilized product is stable for 12 months when properly stored.
- Upon resuspension, store HSV-60c/LyoVec™ at 4˚C. Resuspended product is stable 1 week when properly stored.

DESCRIPTION

Intracellular DNA from pathogens is recognized by multiple cytosolic DNA sensors (CDSs), which display contextual preferences for the recognition of DNA[1]. HSV-60 derives from the herpes simplex virus 1 genome[2]. Transfected HSV-60 was shown to potently induce interferon-beta (IFN-β) in a TLR-, DAI and RNA Pol III-independent, but STING-, TBK1- and IRF3-dependent manner. HSV-60 is recognized by the CDSs, DDX41[3] and IFI16[2].

HSV-60c is a control for HSV-60. HSV-60c is a single-stranded oligonucleotide, which unlike its double-stranded counterpart does not induce type I IFNs. HSV-60c is complexed with the cationic lipid LyoVec™ to facilitate its uptake.

CDS ligands, including transfected HSV-60, trigger type I IFN production and the induction of interferon stimulated genes (ISG) through interferon regulatory factors (IRFs). In order to facilitate their study, InvivoGen has developed stable reporter cells in two well established immune cell models, the human monocytic THP-1 cell line and the murine RAW 264.7 macrophages. These cells express a reporter gene, either SEAP or Lucia®, a secreted luciferase, under the control of an IRF-inducible promoter. For more information visit http://www.invivogen.com/cds-cell-lines

METHODS

Preparation of stock solution (50 µg/ml)

- Add 500 µl endotoxin-free water (provided) per vial of 25 µg HSV-60c/LyoVec™. Mix gently. Allow at least 15 minutes for complete solubilization.
- Store at 4°C. Do not store for more than 1 week.

Below is a protocol for determining type I IFN induction with a CDS ligand. Please note that HSV-60c is a single-stranded oligonucleotide that does not induce type I IFNs. Use HSV-60c/LyoVec™ at the same concentration as the double-stranded oligonucleotide HSV-60/LyoVec™.

Induction of type I IFNs in THP1-Lucia ISG cells

Induction of type I IFNs can be studied in a variety of cells. The human monocytic cell line THP-1 has been shown to express all the CDSs[2-4], with the exception of DAI[5]. A protocol for the induction of type I IFNs using THP1-Lucia™ ISG cells, an IRF-luciferase reporter cell line, is given below:

- Resuspend HSV-60c/LyoVec™, as described above.
- Stimulate cells with 300 ng/ml - 10 µg/ml HSV-60c/LyoVec™ for 16 - 48 hours.
- Monitor induction of type I IFNs by measuring the levels of IRF-induced Lucia® in the cell culture supernatant using QUANTI-Luc™, a Lucia® detection reagent.

1. Sharma S. & Fitzgerald KA. 2011. Innate immune sensing of DNA. PLoS Pathog. 7(4):e1001310. **2. Unterholzner L. *et al.*, 2010.** IFI16 is an innate immune sensor for intracellular DNA.Nat Immunol. 11(11):997-1004. **3. Zhang Z. *et al.*, 2011.** The helicase DDX41 senses intracellular DNA mediated by the adaptor STING in dendritic cells. Nat Immunol.12(10):959-65. **4. Arakawa R. *et al.*, 2010.** Characterization of LRRFIP1. Biochem Cell Biol. 88(6):899-906. **5. Lippmann J. *et al.*, 2010.** IFN beta responses induced by intracellular bacteria or cytosolic DNA in different human cells do not require ZBP1 (DLM-1/DAI). Cell Microbiol. 10(12):2579-88.

RELATED PRODUCTS

Product	Catalog Code
THP1-Lucia™ ISG cells	thpl-isg
Raw-Lucia™ ISG cells	rawl-isg
QUANTI-Luc™	rep-qlc1
CDS ligands	
HSV-60/LyoVec™	tlrl-hsv60c
ISD/LyoVec™	tlrl-isdc
pCpGfree-giant/LyoVec™	tlrl-cpgfc
VACV-70/LyoVec™	tlrl-vav70c

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