

3'3'-cGAMP Fluorinated

Fluorinated cyclic diadenylate monophosphate: a STING ligand

Catalog # ttrl-nacgaf

<http://www.invivogen.com/cgamp-fluorinated>

For research use only

Version # 17F01-MM

PRODUCT INFORMATION

Content:

- 100 µg of lyophilized 3'3'-cGAMP Fluorinated

Note: 3'3'-cGAMP Fluorinated is sterile filtered prior to lyophilization.

- 1.5 ml endotoxin-free water

Storage and stability:

- Product is shipped at room temperature and should be stored at -20°C. Lyophilized product is stable for 1 year when properly stored.

- Upon resuspension, prepare aliquots of 3'3'-cGAMP Fluorinated and store at -20°C. Resuspended product is stable for 6 months when properly stored. Avoid repeated freeze-thaw cycles.

Quality control:

- Purity and structure has been determined by LC/MS and NMR: ≥ 95%
- The ability of 3'3'-cGAMP Fluorinated to induce type I interferon (IFN) has been confirmed in THP1-Blue™ ISG cells.
- The absence of bacterial contamination (e.g. lipoproteins & endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

3'3'-cGAMP Fluorinated (c-[2'FdGMP]-[2'FdAMP]) is a synthetic analog of cyclic guanosine monophosphate- adenosine monophosphate (cyclic GMP-AMP, cGAMP) with a fluorine atom at 2' position of the nucleosides. 3'3'-cGAMP is a cyclic di-nucleotide produced by bacteria. It is also referred to as "canonical" cGAMP due the presence of the classical 3'-5' phosphodiester linkages between the guanosine and the adenosine. It has been reported that cGAMP binds STING (stimulator of IFN genes) and subsequently induces TBK1-IRF3-dependent production of IFN-β¹.

The incorporation of fluorine into biologically active molecules is commonly used in medicinal chemistry to improve their metabolic stability or to modulate physicochemical properties such as lipophilicity^{2, 3}. Moreover, the introduction of a fluorine atom can change the biological activities of a molecule. Interestingly, when used at low concentrations in various cellular assays, 3'3'-cGAMP Fluorinated induces higher levels of type I IFNs than does cGAMP.

STING ligands such as cGAMP induce type I IFNs and activate interferon stimulated genes (ISG) through IRFs. To facilitate their study, InvivoGen has developed stable reporter cells in two well established immune cell models: THP-1 human monocytes and RAW 264.7 murine macrophages. These cells express a reporter gene (SEAP or Lucia luciferase), under control of an IRF-inducible promoter.

1. Zhang X. et al., 2013. Cyclic GMP-AMP containing mixed phosphodiester linkages is an endogenous high-affinity ligand for STING. Mol Cell.51(2):226-35. 2. Liu P. et al., 2008. Fluorinated Nucleosides: Synthesis and biological implication. J Fluor Chem. 129(9): 743-766. 4. Böhm HJ. et al., 2004. Fluorine in medicinal chemistry. Chembiochem. 5(5):637-43.

CHEMICAL PROPERTIES

Source: Synthetic

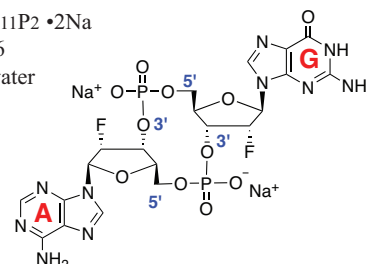
Synonyms: 2'Fluoro-cyclic GMP-AMP, c-(2'FdGMP)-(2'FdAMP), F-c-GpAp sodium salt

Formula: C₂₀H₂₀F₂N₁₀O₁₁P₂ •2Na

Molecular weight: 722.36

Solubility: 50 mg/ml in water

Structure:



METHODS

Preparation of stock solution (100 µg/ml):

Stimulation of STING can be achieved with 100 ng-10 µg/ml 3'3'-cGAMP Fluorinated.

1. Briefly centrifuge the vial before opening to dislodge any lyophilized material that may be dispersed on the wall or cap of the vial. Carefully open the vial lid to avoid any loss of product.
2. Add 1 ml of endotoxin-free water to 100 µg of 3'3'-cGAMP Fluorinated.
3. Vortex until completely dissolved.

Induction of type I IFNs in THP1-Lucia ISG cells

Induction of type I IFNs with 3'3'-cGAMP Fluorinated can be studied in a variety of cells. The human monocytic cell line THP-1 has been shown to express all the cytosolic DNA sensors, with the exception of DAI. A protocol for the induction of type I IFNs using THP1-Lucia™ ISG cells, an IRF-luciferase reporter cell line, is given below:

1. Resuspend 3'3'-cGAMP Fluorinated, as described above.
2. Stimulate cells with 0.1-100 µg/ml 3'3'-cGAMP Fluorinated for 16-48 hours.
3. Monitor induction of type I IFNs by measuring the levels of IRF-induced Lucia luciferase in the cell culture supernatant using QUANTI-Luc™, a Lucia luciferase detection reagent.

RELATED PRODUCTS

Product	Catalog Code
RAW-Lucia™ ISG cells	rawl-isg
RAW-Lucia™ ISG-KO-STING cells	rawl-kostg
THP1-Blue™ ISG cells	thp-isg
THP1-Lucia™ ISG cells	thpl-isg
Other STING ligands	
3'3'-cGAMP	ttrl-nacga
c-di-GMP	ttrl-nacd
c-di-GMP Fluorinated	ttrl-nacd

TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873

InvivoGen USA (International): +1 (858) 457-5873

InvivoGen Europe: +33 (0) 5-62-71-69-39

InvivoGen Hong Kong: +852 3-622-34-80

E-mail: info@invivogen.com