

Caution: For Laboratory Use. A research reagent for research purposes only

Human Anaphylatoxin C3a Receptor, γ -Irradiated Frozen Cells

Product No.: ES-730-CF

Lot No.: 1001

Material Provided

Cells:	1 x 1 mL frozen aliquot
Format:	~2.5 x 10 ⁶ cells / mL in Ham's F12, 10% FBS with 10 % DMSO

Product Information

Cellular Background:	CHO-K1
Frozen cells info:	Frozen recombinant, γ -irradiated CHO-K1 cells expressing the human Chemotactic Peptide C3a receptor.
DNA Sequence:	Identical to coding sequence of GenBank NM_004054.2.
Corresponding Protein Sequence:	Identical to GenBank NP_004045.1.
Storage Conditions:	Store in liquid nitrogen (vapor phase) immediately upon receipt, or maximum 15 days at -80°C. cAMPZen [®] is designed for single use only. Do not refreeze.

Quality Control

EC₅₀ for a reference agonist is determined using a LANCE[®] cAMP assay (Figure 1). Mycoplasma test is performed using MycoAlert[®] Mycoplasma detection kit. We certify that these results meet our quality release criteria.

(TRP63, TRP64)-C3A (63-77) - (EC₅₀):	0.3 nM
Mycoplasma:	This cell line tested negative for Mycoplasma.

Recommended Use and Thawing Conditions

Assay Medium: HBSS 1X (without phenol red); 5 mM HEPES; BSA-stabilizer (7.5% solution); 0.5 mM IBMX (pH 7.4).

Thawing Cells: Using appropriate personal protective equipment, place the frozen aliquot in a 37°C water bath and agitate until its content is thawed completely. Immediately remove from water bath, transfer content to 10 mL assay medium and centrifuge (150 x g, 5 min). Resuspend cell pellet in appropriate volume of assay medium for use in the selected LANCE[®] cAMP assay (see LANCE[®] cAMP Assays section).

Typical Product Data

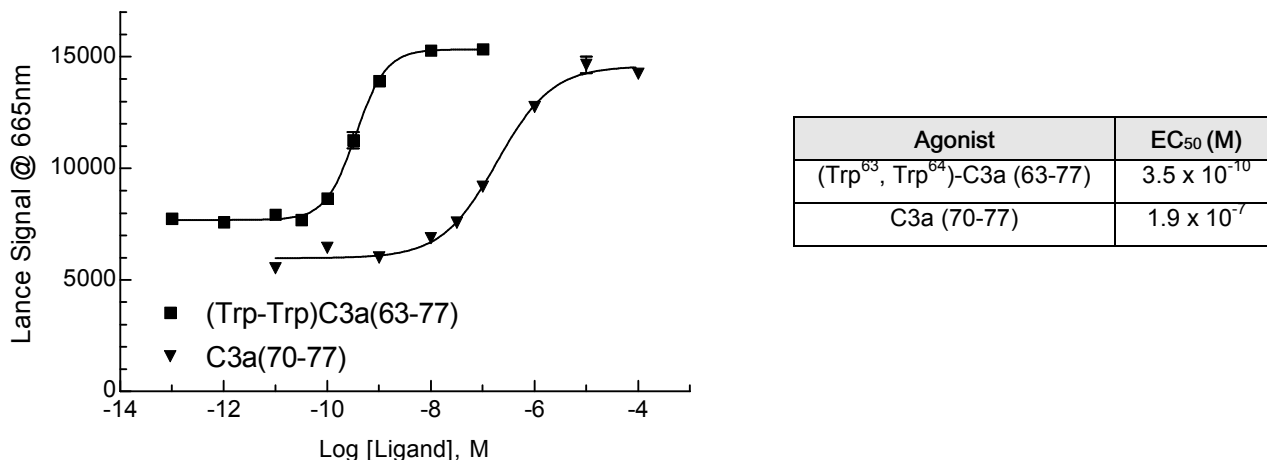


Figure 1: Agonist Response in LANCE[®] cAMP assay

An agonist dose-response experiment was performed in 384-well format using 2 500 cells/well. Cell stimulation was performed for 30 min at room temperature with 5 μM Forskolin (FK) and the indicated agonist concentrations. Reader: EnVision[®]. Data from a representative experiment are shown. The Z'-factor was calculated for (Trp⁶³, Trp⁶⁴)-C3a (63-77) with at least 16 background and 16 maximal signal points (Z' = 0.89).

Table 1. References of compounds used for functional characterization

Name	Provider	Cat n°	Working Stock Solution
(Trp ⁶³ , Trp ⁶⁴)-C3a (63-77)	Bachem	H-1264	10 mM in water
C3a (70-77)	Bachem	H-1645	10 mM in water

LANCE[®] cAMP Assays

LANCE[®] Ultra cAMP - please refer to the user manual included with the LANCE[®] Ultra cAMP kits:

TRF0262 – 1 000 Assay points

TRF0263 – 10 000 Assay points

TRF0264 – 50 000 Assay points

LANCE[®] cAMP - please refer to the user manual included with the LANCE[®] cAMP kits:

AD0262 – 500 Assay points

AD0262E – 1 000 Assay points

AD0263 – 10 000 Assay points

AD0264 – 50 000 Assay points

Suggested Materials and Instrumentation

Please visit our website

www.perkinelmer.com/CellLines

This product is not for resale or distribution except by authorized distributors.

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