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PRODUCT DATASHEET

ChemiScreen™ Membrane Preparation Recombinant Human FPRL1 Formylpeptide Receptor

CATALOG NUMBER:	HTS056M	**QUANTITY:**	200 units
LOT NUMBER:	010714	**VOLUME/CONCENTRATION:**	1 mL, 1 mg/mL

BACKGROUND: FPRL1 (formyl peptide receptor-like 1, also known as FPR2) is a GPCR that belongs to the N-formyl peptide receptor family. Initially described as a receptor for lipoxin A4, FPRL1 has been shown to bind to a synthetic peptide WKYMVm, amyloid beta peptides, and N-formylated mitochondrial peptides and mediates phagocyte chemotaxis (Fiore *et al.*, 1994; Le *et al.*, 1999; Rabiet *et al.*, 2005; Iribarren *et al.*, 2005). FPRL1 membrane preparations are crude membrane preparations made from our proprietary stable recombinant cell lines to ensure high-level of GPCR surface expression; thus, they are ideal HTS tools for screening of FPRL1 interactions with its ligands.

APPLICATIONS: GTPγS binding assay.

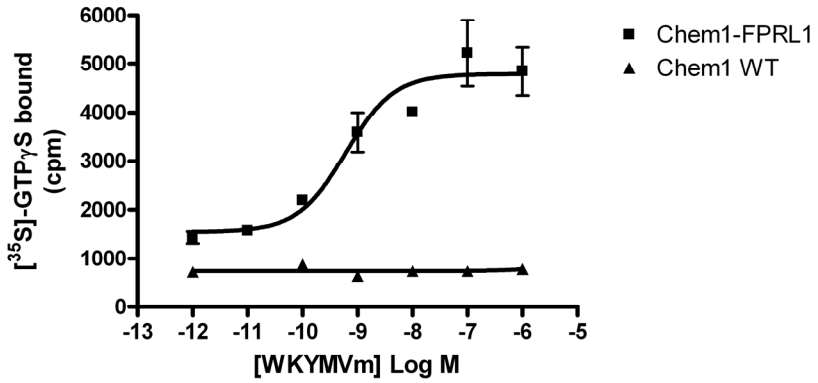

Figure 1. Binding of [^{35}S]-GTPγS to FPRL1 membrane preparation. FPRL1 Membrane Preparation (HTS056M) or wild-type Chem-1 membrane preparation (5 μg/well) was incubated with 0.3 nM [^{35}S]-GTPγS and increasing amounts of unlabeled WKYMVm. Bound radioactivity was determined by filtration and scintillation counting. Sample data from a representative lot.

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SPECIFICATIONS: 1 unit = 5 μg
EC50 for WKYMVm: 1 nM
Signal:background: ≥ 1000 cpm

Species: Human FPRL1 (Accession number NM_001462)

HOST CELLS: Chem-1, an adherent mammalian cell line without any endogenous FPRL1 expression.

RECOMMENDED ASSAY CONDITIONS: Membranes are permeabilized by addition of saponin to an equal concentration by mass, then mixed with [^{35}S]-GTPγS (final concentration of 0.3 nM) in 20 mM HEPES, pH 7.4/100 mM NaCl/10 mM $MgCl_2$/0.5 μM GDP in a nonbinding 96-well plate. Unlabeled WKYMVm is added to the final concentration indicated in Figure 1 (final volume 100 μL), and incubated for 30 min at 30°C. The binding reaction is transferred to an FB filter plate (EMD Millipore MAHF B1H) pre-wetted with water, and washed 2 times (1 mL per well per wash) with cold 10 mM sodium phosphate, pH 7.4. The plate is dried and counted.

One vial contains enough membranes for at least 200 assays (units), where one unit is the amount of membrane that will yield greater than 1000 cpm specific WKYMVm stimulated [^{35}S]-GTPγS binding when final concentration of 0.3nM [^{35}S]-GTPγS was used.

PRESENTATION:

Liquid in packaging buffer: 50 mM Tris pH 7.4, 10% glycerol and 1% BSA with no preservatives.
Packaging method: Membrane proteins were adjusted to the indicated concentration in 1 ml packaging buffer, rapidly frozen, and stored at -80°C.

STORAGE/HANDLING:

Store at –70°C. Product is stable for at least 6 months from the date of receipt when stored as directed. Do not freeze and thaw.

REFERENCES:

Fiore S *et al.* (1994) Identification of a human cDNA encoding a functional high affinity lipoxin A receptor. *J. Exp. Med.* 180: 253-260.

Iribarren P *et al.* (2005) Role of formyl peptide receptor-like 1 (FPRL1/FPR2) in mononuclear phagocyte responses in Alzheimer disease. *Immunol. Res.* 31: 165-76.

Le Y *et al.* (1999) Utilization of two seven-transmembrane, G protein-coupled receptors, formyl peptide receptor-like 1 and formyl peptide recptor, byt the synthetic hexapeptide WKYMVm for human phagocyte activation. *J. Immunol.* 163: 6777-6784.

Rabiet MJ *et al.* (2005) Human mitochondria-derived N-formylated peptides are novel agonists equally active on FPR and FPRL1, while Listeria monocytogenes-derived peptides preferentially activate FPR. *Eur. J. Immunol.* 35: 2486-95.

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