

**ChemiScreen™ CALCIUM-OPTIMIZED STABLE CELL LINE
HUMAN RECOMBINANT NTR1 NEUROPEPTIDE RECEPTOR**

CATALOG NUMBER:	HTS034C	QUANTITY:	2 vials, 1 mL each
LOT NUMBER:		CONCENTRATION:	2 x 10 ⁶ cells/mL

BACKGROUND:

Neurotensin is a 13 amino acid peptide that induces hypothermia and mediates analgesia when administered centrally, and also mediates the behavioral effects of antipsychotic drugs in rodents (Kinkead and Nemeroff, 2004). In addition, several intestinal functions, such as contractility and inflammation, are mediated by neurotensin (Castagliuolo et al., 1999). The actions of neurotensin are mediated by two GPCRs, NTR1 (or NTS1) and NTR2 (or NTS2). NTR1 knockout mice are resistant to the hypothermic, analgesic and contractile effects of neurotensin, indicating that NTR1 is the primary mediator of these effects (Pettibone et al., 2002). Small molecule antagonists, including SR48692 and SR142948A, that are partially selective for NTR1 also interfere with the biological effects of neurotensin (Labbé-Jullié et al., 1998).

Chemicon's cloned human NTR1-expressing cell line is made in the Chem-1 host, which supports high levels of recombinant NTR1 expression on the cell surface and contains high levels of the promiscuous G protein G α 15 to couple the receptor to the calcium signaling pathway. Thus, the cell line is an ideal tool for screening for antagonists of interactions between NTR1 and neurotensin.

APPLICATIONS:

Calcium flux assay, ligand binding assays

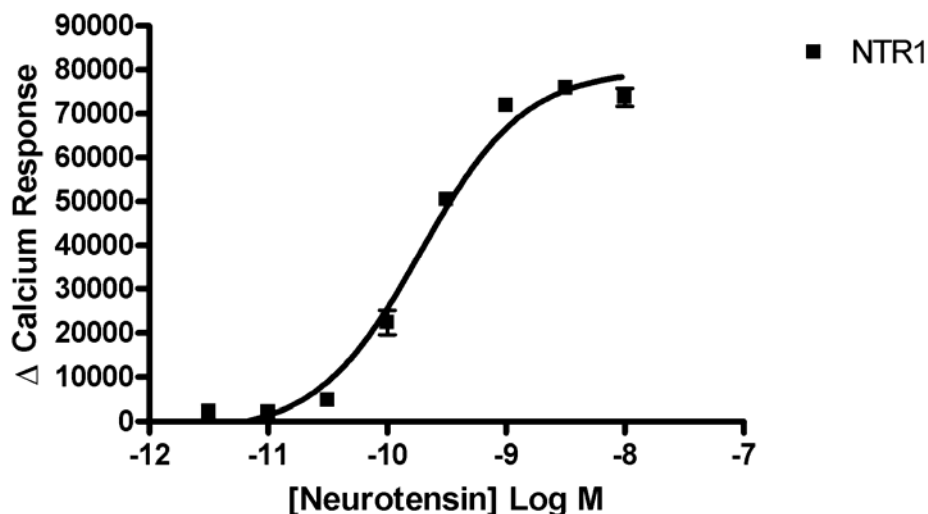


Figure 1. Calcium flux in NTR1-expressing Chem-1 cell line induced by Neurotensin. NTR1-expressing Chem-1 cells were loaded with Fluo-4 and calcium flux in response to human Neurotensin (10⁻⁸ to 10^{-11.5} M) was determined in triplicate on a Molecular Devices Flex Station. A signal:background ratio of greater than 15 was obtained.

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SPECIFICATIONS: EC50 for calcium mobilization by Neurotensin: ~ 0.2 nM

HOST CELLS: Chem-1

TRANSFECTION: Full-length human NTR1 cDNA (Accession Number: NM_002531)

GROWTH MEDIA: DMEM containing 4.5 g/L glucose/10% heat inactivated fetal bovine serum/1x nonessential amino acids/10 mM HEPES/0.25 mg/ml Geneticin (G418)/100 U/ml each penicillin and streptomycin

PRESENTATION:

Cells are frozen at 2×10^6 cells/mL in DMEM/20% fetal bovine serum/100 U/ml penicillin and streptomycin/10% DMSO. Cell line tests negative for mycoplasma.

STORAGE/HANDLING:

Place cells in liquid nitrogen immediately upon receipt. Maintain frozen in liquid nitrogen for up to 5 years. Thaw cells rapidly by removing from liquid nitrogen and immediately immersing in a 37°C water bath. Immediately after ice has thawed, sterilize the exterior of the vial with 70% ethanol. Transfer contents of the vial to a T75 flask containing growth media, and replace media after 8-24 h. Cells are typically passaged 1:10 every 3-4 days, and should be passaged at least once after thawing prior to use in calcium flux assays.

REFERENCES:

Castagliuolo I., *et al.* (1999) Neurotensin is a proinflammatory neuropeptide in colonic inflammation. *J. Clin. Invest.* 103: 843-849.

Kinkead B. and Nemeroff C. (2004) Neurotensin, schizophrenia, and antipsychotic drug action. *Int. Rev. Neurobiol.* 59: 327-349.

Labbé-Jullié C., *et al.* (1998) Mutagenesis and modeling of the neurotensin receptor NTR1: Identification of residues that are critical for binding SR48692, a nonpeptide neurotensin antagonist. *J. Biol. Chem.* 273: 16351-16357.

Pettibone D.J., *et al.* (2002) The effects of deleting the mouse neurotensin receptor NTR1 on central and peripheral responses to neurotensin. *J. Pharmacol. Exp. Ther.* 300: 305-313.

Important Note: *During shipment, small volumes of product will occasionally become entrapped in the seal of the product vial. For products with volumes of 200 μ L or less, we recommend gently tapping the vial on a hard surface or briefly centrifuging the vial in a tabletop centrifuge to dislodge any liquid in the container's cap.*

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HUMAN RECOMBINANT NTR1 NEUROPEPTIDE RECEPTOR**

Product No. HTS034C

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