Neurotensin(8-13)

Cat. No.: HY-P0251

CAS No.: 60482-95-3

Molecular Formula: $C_{38}H_{64}N_{12}O_8$

Molecular Weight: 816.99

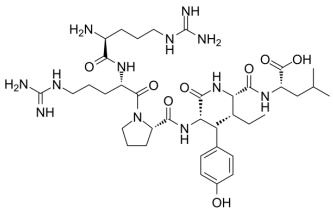
Sequence: Arg-Arg-Pro-Tyr-Ile-Leu

Sequence Shortening: RRPYIL

Target: Neurotensin Receptor

Pathway: GPCR/G Protein; Neuronal Signaling

Storage: Sealed storage, away from moisture and light
Powder -80°C 2 years
-20°C 1 year
* In solvent : -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture and light)



SOLVENT & SOLUBILITY

In Vitro

H_2O : 50 mg/mL (61.20 mM; Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	1.2240 mL	6.1200 mL	12.2401 mL
	5 mM	0.2448 mL	1.2240 mL	2.4480 mL
	10 mM	0.1224 mL	0.6120 mL	1.2240 mL

Please refer to the solubility information to select the appropriate solvent.

In Vivo

1. Add each solvent one by one: PBS
Solubility: 33.33 mg/mL (40.80 mM); Clear solution; Need ultrasonic

BIOLOGICAL ACTIVITY

Description

Neurotensin (8-13) is an active fragment of Neurotensin. Neurotensin(8-13) results in a decrease in cell-surface NT1 receptors (NTR1) density.

IC_{50} & Target

NTR1[1]

In Vitro

Receptor internalization induced by Neurotensin(8-13) results in a decrease in cell-surface NT1 receptors (NTR1) density. The receptor downregulation in response to high extracellular concentrations of the peptide has been described for Neurotensin (NT) in HT-29 cells and in rat primary cultured neurons. Reappearance of the receptors on the cell surface is also different[1].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Kinase Assay [1]

Binding assays are performed on whole HT-29 cells at confluence. A day before the assay, cells (10^6 cells/0.4 mL, equivalent to 0.3 mg protein) are placed in 48-well plates. A special binding buffer that includes protease inhibitors (50 mM HEPES, 125 mM NaCl, 7.5 mM KCl, 5.5 mM $MgCl_2$, 1 mM EGTA, 5 g/L bovine serum albumin, 2 mg/L chymostatin, 100 mg/L soybean trypsin inhibitor, 50 mg/L bacitracin, pH 7.4) is used for the experiments. In inhibition studies, cells are incubated for 1 h at 37°C in triplicate with 25,000 cpm of ^{125}I-NT and variable concentrations (0.001-3,000 nM) of unlabeled NT(8-13), unlabeled NT-VIII, or NT-VIII labeled with ^{nat}Re (final volume of 0.2 mL per well). The cells are then washed twice with cold binding buffer and afterward are solubilized with 1N NaOH at 37°C (0.4 mL per well). The activity is determined in a γ-counter. In saturation studies, cells are incubated in triplicate with increasing concentrations (0.1-10 nM) of ^{99m}Tc(CO)3NT-VIII for 1 h at 37°C (final volume, 0.2 mL per well). The concentrations of total technetium ($^{99+99m}$Tc) are equivalent to 0.2-20 MBq ^{99m}Tc activity per well. After 2 washings with the same binding buffer as before, the cells are then solubilized with 1N NaOH at 37°C (0.4 mL per well). The bound radioactivity is measured in the γ-counter. Nonspecific binding is determined with 1 μM unlabeled NT(8-13)[1].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

REFERENCES

[1]. García-Garayoa E, et al. Preclinical evaluation of a new, stabilized neurotensin(8--13) pseudopeptide radiolabeled with (99m)tc. J Nucl Med. 2002 Mar;43(3):374-83.

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