Recombinant Human NT-3 GMP

Catalog Number: 267-GMP

DESCRIPTION

Source	*Spodoptera frugiperda*, *Sf* 21 (baculovirus)-derived human NT-3 protein Tyr139-Thr257 (K196R) Accession # P20783 Manufactured and tested under cGMP guidelines.
N-terminal Sequence Analysis	Tyr-Ala-Glu-His-Lys-Ser-His-Arg-Gly-Glu
Predicted Molecular Mass	13.7 kDa (monomer)

SPECIFICATIONS

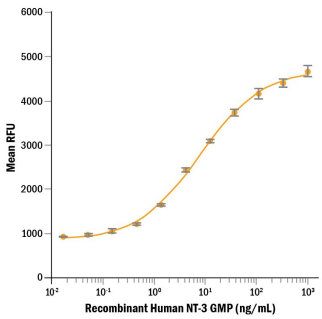
Activity	Measured in a cell proliferation assay using BaF mouse pro-B cells transfected with TrkB. The ED_{50} for this effect is 1-10 ng/mL. The specific activity of Recombinant Human NT-3 is approximately 2.1 x 10^3 units/µg, which is calibrated against recombinant human NT-3 Standard (NIBSC code: 98/718).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>97%, by SDS-PAGE with silver staining.
Mycoplasma	Negative when tested in a ribosomal RNA hybridization assay.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 100 µg/mL in PBS.
Shipping	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • A minimum of 6 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 3 months, ≤ -20 °C under sterile conditions after reconstitution.

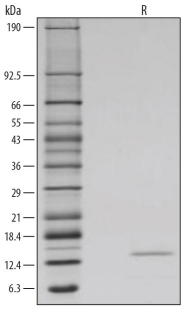
DATA

Bioactivity



GMP-grade Recombinant Human NT-3 (Catalog # 267-GMP) stimulates proliferation of the BaF mouse pro-B cell line transfected with TrkB. The ED_{50} for this effect is 1-10 ng/mL.

SDS-PAGE



1 µg/lane of GMP-grade Recombinant Human NT-3 (Catalog # 267-GMP) was resolved with SDS-PAGE under reducing (R) conditions and visualized by silver staining, showing a band at 14 kDa.

BACKGROUND

Neurotrophin-3 (NT-3) is a member of the NGF family of neurotrophic factors (also named neurotrophins) that are required for the differentiation and survival of specific neuronal subpopulations in both the central as well as the peripheral nervous systems. The neurotrophin family is comprised of at least four proteins including NGF, BDNF, NT-3, and NT-4/5. These secreted cytokines are synthesized as prepropeptides that are proteolytically processed to generate the mature proteins. All neurotrophins have six conserved cysteine residues that are involved in the formation of three disulfide bonds and all share approximately 55% sequence identity at the amino acid level. Similarly to NGF, bioactive NT-3 is predicted to be a non-covalently linked homodimer.

The NT-3 cDNA encodes a 257 amino acid residue precursor protein with a signal peptide and a proprotein that are cleaved to yield the 119 amino acid residue mature NT-3. The amino acid sequence of mature NT-3 is identical in human, mouse and rat. NT-3 transcripts have been detected in the cerebellum, hippocampus, placenta, heart, skin, and skeletal muscle. NT-3 primarily activates the TrkC tyrosine kinase receptor. In addition, NT-3 can activate Trk and TrkB kinase receptors in certain cell systems. NT-3 can also bind with low affinity to the low affinity NGF receptor.

References:

1. Eide, F.F. *et al.* (1993) Exp. Neurol. **121**:200.
2. Snider, W.D. (1994) Cell **77**:627.
3. Barbacid, M. (1994) J. Neurobiol. **25**:1386.

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MANUFACTURING SPECIFICATIONS

GMP Proteins

R&D Systems, a Bio-Techne Brand's GMP proteins are produced according to relevant sections of the following documents: WHO TRS, No. 822, 1992 Annex 1, Good Manufacturing Practices for Biological Products; USP Chapter 1043, Ancillary Materials for Cell, Gene and Tissue-Engineered Products and USP Chapter 92, Growth Factors and Cytokines Used in Cell Therapy Manufacturing.

R&D Systems' quality focus includes:

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- Personnel training programs
- Raw material testing and vendor qualification/monitoring
- Fully validated equipment, processes and test methods
- Equipment calibration schedules using a computerized calibration program
- Facility maintenance, safety programs and pest control
- Material review process for variances
- Monitoring of stability over product shelf-life

R&D Systems strives to provide our customers with the analytical characteristics of each product so that customers may determine whether our products are appropriate for their research. The Certificate of Analysis provided contains the following lot specific information:

- N-terminal amino acid analysis, SDS-PAGE analysis, and endotoxin level (as determined by LAL assay) performed on each bulk QC lot, not on individual bottlings of each QC lot
- Post-bottling lot-specific bioassay results (compliance with an established range) and results of microbial bioburden testing (using broth culture, Sabourand's dextrose and blood agar plates with results reported at 3 days and at 7 days)
- Mycoplasma testing by ribosomal RNA hybridization assay

Additional testing and documentation requested by the customer can be arranged at an additional cost. Testing may include, but is not limited to, USP <61> bioburden testing, positive identity testing, testing for adventitious agents and testing for residual host cell content.

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