

**DESCRIPTION**

<b>Source</b>	<i>Spodoptera frugiperda</i> , Sf 21 (baculovirus)-derived BDNF protein His129-Arg247 Accession # P23560 100% sequence homology with Mouse, Rat, Canine, Equine and all other mammalian proteins examined. Manufactured and tested under cGMP guidelines.
<b>N-terminal Sequence Analysis</b>	His <sub>129</sub> -Ser-Asp-Pro-Ala-Arg-Arg-Gly-Glu-Leu
<b>Predicted Molecular Mass</b>	13.5 kDa

**SPECIFICATIONS**

<b>SDS-PAGE</b>	13-14 kDa, reducing conditions
<b>Activity</b>	Measured in a cell proliferation assay using BaF mouse pro-B cells transfected with TrkB. The ED <sub>50</sub> for this effect is 0.2-2 ng/mL.  The specific activity of Recombinant Human BDNF is approximately 1.3 x 10 <sup>3</sup> units/μg, which is calibrated against recombinant human BDNF WHO Standard (NIBSC code: 96/534).  Measured by its binding ability in a functional ELISA. When Recombinant Human TrkB Fc Chimera (Catalog # 688-TK) is coated at 1 μg/mL, recombinant human BDNF binds with an apparent K <sub>d</sub> <1 nM.
<b>Endotoxin Level</b>	<0.10 EU per 1 μg of the protein by the LAL method.
<b>Purity</b>	>97%, by SDS-PAGE with silver staining, under reducing conditions.
<b>Mycoplasma</b>	Negative when tested in a ribosomal RNA hybridization assay.
<b>Formulation</b>	Lyophilized from a 0.2 μm filtered solution in Sodium Citrate and NaCl. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 100 μg/mL in PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>A minimum of 6 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date.</li> <li>1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>3 months, ≤ -20 °C under sterile conditions after reconstitution.</li> </ul>

**DATA**

<p><b>Bioactivity</b></p> <p>GMP-grade Recombinant Human BDNF (Catalog # 248-GMP) stimulates proliferation in the BaF mouse pro-B cell line transfected with TrkB. The ED<sub>50</sub> for this effect is 0.2-2 ng/mL.</p>	<p><b>SDS-PAGE</b></p> <p>1 μg/lane of GMP-grade Recombinant Human BDNF (Catalog # 248-GMP) was resolved with SDS-PAGE under reducing (R) conditions and visualized by silver staining, showing a band at 14 kDa.</p>
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## BACKGROUND

Brain-derived neurotrophic factor (BDNF) 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 system. 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 (1, 2). 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 BDNF is predicted to be a non-covalently linked homodimer.

BDNF cDNA encodes a 247 amino acid residue precursor protein with a signal peptide and a proprotein that are cleaved to yield the 119 amino acid residue mature BDNF. The amino acid sequence of mature BDNF is identical in all mammals examined. High levels of expression of BDNF have been detected in the hippocampus, cerebellum, fetal eye and placenta. In addition, low levels of BDNF expression are also found in the pituitary gland, spinal cord, heart, lung and skeletal muscle. BDNF binds with high affinity and specifically activates the TrkB tyrosine kinase receptor (3).

### 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.

## 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.

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- Equipment calibration schedules using a computerized calibration program
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- Monitoring of stability over product shelf-life

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- 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

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