

Product Information

Nerve Growth Factor-Beta (NGF- β) from Mouse Submaxillary Glands

Product No. **N 2393**

Product Description

Nerve Growth Factor (NGF) was first discovered in 1953 by Levi-Montalcini, Hamburger and Cohen¹⁻³ in two mouse sarcomas, and was described as a diffusible agent which strongly promotes fiber outgrowth of sensory neurons in chick embryos. Cohen purified NGF from snake venom⁴ and from mouse salivary glands.⁵ NGF is a neurotrophic agent thought to be provided by peripheral tissues for the guidance and sustenance of outgrowing embryonic sympathetic and sensory neurons.⁶ NGF induces the formation of neurite-like filaments from chick embryo dorsal root ganglia² and from rat PC12 pheochromocytoma cells.⁷ *In vivo* NGF may be involved in fetal development^{8,9} and nerve regeneration.¹⁰ NGF may also play a physiological role within the central nervous system.^{8,11,12} Cellular receptors for NGF have been found in a variety of cell lines¹³ and tissues, including cholinergic neurons of the brain^{14,15} and Schwann cells of damaged nerve axons.¹⁰ Two kinetic types of NGF receptors have been identified from peripheral neurons,¹⁶ neuroblastoma cells,¹⁷ and PC12 cells¹⁸ and are designated as type I (high affinity) and type II (low affinity). The signal transduction mechanism of the receptor has not been clearly identified.

Nerve Growth Factor isolated from mouse submaxillary glands under non-dissociative conditions (NGF-7S, Product No. N 0513) has a sedimentation coefficient of 7.1S.^{19,20} It is generally believed that NGF-7S is a 130 kDa protein composed of 5 non-covalently linked subunits (2α , 1β , 2γ), although there is recent evidence for a different endogenous form of high molecular weight NGF.²¹ After dissociation of purified NGF-7S by acidic or basic pH, only the β subunit of NGF (NGF- β , Product No. N 2269) has neurotrophic activity.⁶ NGF- β is a 26.5 kDa dimer of identical 118-residue chains held together tightly by noncovalent bonds. A form of NGF- β , NGF-2.5S²² (Product No. N 6009), initially isolated under dissociative conditions, is often slightly different from NGF- β due to proteolysis incurred during its purification⁶. Apparently the 7S complex protects the amino- and carboxy-terminals of NGF- β from hydrolytic enzymes present in the submaxillary gland extract²³. Both NGF-2.5S and NGF- β have comparable bioactive potencies²⁴. NGF- β is isolated by HPLC from purified NGF-7S of mouse submaxillary glands by a modification of the method of Varon, et al.²⁰

Product Information

Purity: >95% by SDS-PAGE (combined monomeric and dimeric β -NGF).

Biological Activity: Optimal at 0.3 - 3 ng/ml.

Immunological Identity: Single band at 13 kDa (monomeric β -NGF) with minor band at 26 kDa (dimeric β -NGF) by SDS-PAGE and Western blot using Anti-NGF (2.5S).

Sequence Identity: N-terminal sequence analysis β -NGF.

Mass/vial: ≥ 10 μ g by amino acid analysis.

Lyophilization buffer: 125 μ l of 0.2 μ m-filtered 20 mM sodium phosphate, pH 7.0.

Carrier Protein: None

Endotoxin: <10 endotoxin units/vial.

Reconstitution and Use

To prepare a stock solution, reconstitute the vial contents in a solution that contains 0.1-1.0% BSA or 1-10% serum in buffered saline or tissue culture media. This may be diluted to the final working concentration of NGF- β , generally 0.1 to 10 ng/ml. Additional filtration of the stock solution is not recommended and may result in product loss due to adsorption onto the filter membrane.

Storage/Stability

Prior to reconstitution store vial below 0 °C. After reconstitution, the product may be stored for two weeks at 2-8 °C or may be stored as aliquots at -20 °C. Prolonged storage of product or repeated freezing and thawing is not recommended.

Performance Characteristics

Nerve Growth Factor- β has been tested for bioactivity using 8-day chick embryo dorsal root ganglia²⁵. The product induced an optimal degree of neurite-like filament growth when ganglia were exposed to NGF- β concentrations between 0.1 and 10 ng/ml. Filament growth was not observed from ganglia without added NGF- β . The EC₅₀ is defined as the effective concentration of growth factor that elicits a 50% increase in cell growth in a cell based bioassay.

References

1. Levi-Montalcini, R., *Science*, **237**, 1154 (1987).
2. Levi-Montalcini, R., et al., *Cancer Res.*, **14**, 49 (1954).
3. Cohen, S., et al., *Proc. Natl. Acad. Sci. USA*, **40**, 1014 (1954).
4. Cohen, S., *J. Biol. Chem.*, **234**, 1129 (1959).
5. Cohen, S., *Proc. Natl. Acad. Sci. USA*, **46**, 302 (1960).
6. Server, A. and Shooter, E., *Adv. Protein Chem.*, **31**, 339 (1977).
7. Greene, R. and Tischler, A., *Proc. Natl. Acad. Sci. USA*, **73**, 2424 (1976).
8. Ayer-Lelievre, C., et al., *Med. Biol.*, **61**, 296 (1983).
9. Taniuchi, M., et al., *Proc. Natl. Acad. Sci. USA*, **83**, 4094 (1986).
10. Thornburn, G., et al., *Growth and Maturation Factors*, Vol. 3 (G. Guroff, ed.) John Wiley & Sons, NY, p.175 (1985).
11. Ebendal, T., *Prog. Growth Factor Res.*, **1**, 143 (1989).
12. Dreyfus, C., *Trends Pharmacol. Sci.*, **10**, 145 (1989).
13. Eveleth, D., *In Vitro Cell. Dev. Biol.*, **24**, 1148 (1988).
14. Hefti, F., et al., *Neurosci. Lett.*, **69**, 37 (1986).
15. Raivich, G. and Kreutzberg, G., *Neuroscience*, **20**, 23 (1987).
16. Godfrey, E. and Shooter, E., *J. Neurosci.* **6**, 2543 (1986).
17. Marchetti, D. and Perez-Polo, J., *J. Neurochem.*, **49**, 475 (1987).
18. Buxser, S., et al., *J. Biol. Chem.*, **265**, 12701 (1990).
19. Varon, S., et al., *Biochemistry*, **6**, 2202 (1967).
20. Varon, S., et al., *Biochemistry*, **7**, 1296 (1968).
21. Young, M., et al., *Biochemistry*, **27**, 6675 (1988).
22. Angeletti, R. and Bradshaw, R., *Proc. Natl. Acad. Sci. USA*, **68**, 2417 (1971).
23. Moore, J., et al., *Biochemistry*, **13**, 833 (1974).
24. Mobley, W., et al., *Biochemistry*, **15**, 5543 (1976).
25. Nomura, J., et al., *Meth. Neurochem.*, **3**, 203 (1972).

Pcs1/00

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.