

Cat. No.: 13257-019

Lot No.:

Size: 10 µg

Storage Conditions: -20°C

REFERENCES:

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STORAGE -20°C. **DO NOT freeze and thaw repeatedly.**

PRODUCT DESCRIPTION This preparation of Nerve Growth Factor (NGF 2.5S) was purified from the submaxillary glands of male mice and was filtered through a 0.22 micron filter. It is supplied at a concentration of 100 µg/ml in 10 mM sodium acetate (pH 5)

BACKGROUND NGF was the first growth factor to be identified. It is a trophic factor *in vivo* and *in vitro*, and regulates the development and maintenance of sympathetic and some sensory neurons.¹ NGF elicits a wide variety of responses from its target cells including metabolic changes in gene expression and chemotactic effects.²⁻⁵ NGF exists naturally as a complex referred to as 7S NGF which contains two molecules of the arginine esteropeptidase γ -subunit, two molecules of the acidic protein α -subunit, 1 or more zinc ions bound to the γ -subunit and the biologically active 2.5S subunit, also referred to as the β -subunit. The 2.5S subunit is an approximately 26 kDa protein with an isoelectric point of 9.3 and is composed of two identical polypeptide chains held together by noncovalent forces.^{4,5} The 2.5S subunit is entirely responsible for the biological activities of NGF,⁶ being the only part of the 7S complex that promotes nerve growth and maintenance of adrenergic neurons *in vitro*.⁷ The additional subunits that comprise 7S NGF appear to protect the 2.5S subunit from proteolytic cleavage.⁵

This product is distributed for laboratory research use only. CAUTION: Not for diagnostic use. The safety and efficacy of this product in diagnostic or other clinical uses has not been established.

The rat pheochromocytoma cell line PC12⁸ has been a primary model that has been used to study the *in vitro* actions of NGF because it responds to NGF by acquiring properties of sympathetic neurons. Over a period of several days NGF stimulates various neuron-like changes in PC12 including neurite outgrowth,⁸ increased electric excitability,⁹ changes in neurotransmitter synthesis,¹⁰ and increased levels of acetylcholine receptors.¹¹ NGF also induces rapid changes in PC12 cells such as changes in morphology of the cell surface¹¹ and in the levels of phosphorylation of several proteins,¹² increased levels of intracellular Ca²⁺ associated with expression of *c-fos* and β -actin genes,¹³⁻¹⁵ rapid redistribution of F-actin,¹⁷ cessation of cell division,⁸ increased cell-substratum and cell-cell adhesion,¹⁸ PC12 cells do not require NGF for survival,¹⁶

APPLICATIONS This preparation of NGF can be used for studies of nerve cell growth and differentiation, and for biochemical studies of the basis of the various immediate and delayed biological activities of NGF. It can also be used as a medium supplement to support the growth or survival of neuronal cells in culture. The biological effects of NGF are usually observed at 5 to 100 ng/ml (0.2 to 4 nM) Examples include:

- Induction of neurite outgrowth⁸
- Maintenance of neural cell viability in culture⁷
- Effects on morphology and differentiation^{8,11}
- Stimulation of protein tyrosine phosphorylation¹²

DIRECTIONS FOR USE Dilute immediately before use. Thaw vial, centrifuge briefly and place on ice. To avoid loss, prepare dilute solutions in appropriate buffer or medium containing at least 0.1% BSA. **DO NOT store in dilute solution.** At first thawing, aliquot into single-use portions in polypropylene vials and store at -20°C to avoid repeated freeze-thaw cycles. In applications requiring long-term use of this growth factor, it is advisable to filter-sterilize the dilute solutions through a 0.22 micron low protein-binding filter.

QUALITY CONTROL

PURITY AND IDENTITY

- SDS-PAGE: 10% polyacrylamide gel stained with Coomassie blue.
Result: >95% in band at 13 kDa

FUNCTIONAL QUALIFICATION

- Neurite outgrowth in PC12 cells.
Result: Half-maximal effect at 50 ng/ml NGF