
Product Data Sheet

Product Name: HRP
Cat. No.: GP21810
Batch No.: 1

Product Data

Purity	>98%	Source	Root extracts of horseradish.
Physical Appearance	solid	Shipping Condition	Shipped at Room temp.
Synonyms	Horseradish Peroxidase; HRP; EC 1.11.1.7.		
Solubility	It is recommended to reconstitute the lyophilized HRP in sterile 18MΩ-cm H ₂ O not less than 100 µg/ml or more than 10 mg/ml solutions.		
Formulation			

Introduction

The enzyme horseradish peroxidase, found in horseradish, is used extensively in molecular biology and in antibody amplification and detection, among other things. For example, "In recent years the technique of marking neurons with the enzyme horseradish peroxidase (HRP) has become a major tool. In its brief history, this method has probably been used by more neurobiologists than have used the Golgi stain since its discovery in 1870." Horseradish peroxidase is also highly used in techniques such as Western blotting and ELISAs. HRP is widely used as an enzymatic label in immunoassays. Usually, the enzyme is coupled to antibodies, lectins or haptens. Coupling to antibodies etc. may be performed through the carbohydrate side chains of the HRP.

Biological Activity

276 U/mg (25°C, guaiacol as the hydrogen donor, pH-7 and H₂O₂ as substrates).

Stability

Lyophilized HRP although stable at room temperature for 3 weeks, should be stored desiccated below -18°C. Upon reconstitution HRP should be stored at 4°C between 2-7 days and for future use below -18°C. Please prevent freeze-thaw cycles.

Protocol

Caution: Product has not been fully validated for medical applications. For research use only.

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Cell experiment [1]:

Cell lines	graphene oxide and RGO samples
Preparation Method	To each sample containing HRP (0.35 μ M) and etoposide (200 μ M), H ₂ O ₂ (80 μ M) was added, and either a full ESR spectra or the time course of the EPR signal was recorded.
Reaction Conditions	0.35 μ M
Applications	The assay revealed that at least a portion of HRP retained enzymatic activity in the presence of RGO; this observation was also confirmed by electron paramagnetic resonance spectroscopy.

Animal experiment [2]:

Animal models	Twenty-eight albino and two non-albino guinea pigs
Preparation Method	After right retroauricular skin incision, a small hole was made in the exposed tympanic bulla and 0.15 ml of HRP was injected into the middle ear cavity. Two animals were injected once and the survival times were 3 and 7 hours, respectively. Five animals were injected once in 24 hours. Two animals were injected twice during a period of 48 hours.
Dosage form	0.15 ml of HRP; i.v.

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Applications

The normal round window membrane resisted HRP penetration from the middle ear side, but when it became pathological after repeated applications, its permeability increased. HRP deposits were found in the cochlear and vestibular sensory cells and in the lumen of the endolymphatic sac.

References:

[1] Kotchey GP, et al. The enzymatic oxidation of graphene oxide. ACS Nano. 2011 Mar 22;5(3):2098-108.

[2] Saijo S, Kimura RS. Distribution of HRP in the inner ear after injection into the middle ear cavity. Acta Otolaryngol. 1984 May-Jun;97(5-6):593-610.

Background

HRP, as a retrograde and anterograde tracer of neuronal connections, increased the sensitivity for detecting the chromagin.^[1]

In vitro, treatment with 1 ug of complexed HRP, macrophages interiorized complexes formed in a wide range of HRP/anti-HRP ratios, while FDC's associated with complexes formed in HRP excess only. ^[2] In vitro kinetic experiment shown that the soluble HRP had more affinity toward guaiacol and H₂O₂ than immobilized HRP. ^[5]

In vivo, mice were injected 100 mg/ml HRP after 30 sec, enzyme reaction products were weakly detected in interstitium around some thick blood vessels of corticomedullary boundary areas, but within capillaries of cortical areas. After injection 30 min,

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phagocytosis of HRP by macrophages was scattered throughout the interstitium, which was accompanied by decrease of HRP reaction intensity in interstitial matrices.^[3] In vivo test it demonstrated that WGA-HRP (horseradish peroxidase conjugates of either the lectin wheat germ agglutinin) injection resulted in labeling of primary afferents mainly in the substantia gelatinosa of the trigeminal subnucleus caudalis. While B-HRP (cholera toxin) injection, it was found labeling only existed in the magnocellular zone and innermost part of the substantia gelatinosa.^[4] In vivo efficacy test it exhibited that the HRP-diaminobenzidine reaction products were heavily distributed in the OVLN and surrounding brain tissues 10 and 60 min after the injection of HRP (50 mg/kg, i.v.) in rabbits, and also were retained in the parenchymal tissues at 24 h post-injection.^[6]

References:

- [1] Wilczynski W, et al. Transcellular transfer of HRP in the amphibian visual system. *Brain Res.* 1982 May 6;239(1):29-40.
- [2] Chen LL, et al. Distribution of horseradish peroxidase (HRP)-anti-HRP immune complexes in mouse spleen with special reference to follicular dendritic cells. *J Cell Biol.* 1978 Oct;79(1):184-99.
- [3] Wu B, et al. Immuno- and Enzyme-histochemistry of HRP for Demonstration of Blood Vessel Permeability in Mouse Thymic Tissues by "In Vivo Cryotechnique". *Acta Histochem Cytochem.* 2014;47(6):273-88.
- [4] Robertson B, et al. Transganglionic transport of wheat germ agglutinin-HRP and cholera toxin-HRP in rat trigeminal primary sensory neurons. *Brain Res.* 1985 Nov 25;348(1):44-51.
- [5] Alshawafi WM, et al. Immobilization of horseradish peroxidase on PMMA nanofibers incorporated with nanodiamond. *Artif Cells Nanomed Biotechnol.* 2018;46(sup3):S973-S981.
- [6] Yamaguchi K, Sieber NC. The capillary of the organum vasculosum laminae terminalis (OVLN) in rabbits is more permeable to horseradish peroxidase (HRP) than that in rats. *J Electron Microsc (Tokyo).* 2000;49(6):783-91.

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