Hy-Proteinase K

Cat No. N9016

Size: 100mg

Contents: Hy-Proteinase K 100mg

Specific Activity: 40 U/mg protein

Form: Contains 100 mg Hy-Proteinase K, Lyophilized Powder

Description

Hy-Proteinase K is an endolytic protease that cleaves peptide bonds at the carboxylic sides of aliphatic, aromatic or hydrophobic amino acids. The Hy-Proteinase K is classified as a serine protease. The smallest peptide to be hydrolyzed by this enzyme is a tetrapeptide.

Features

Active in a wide range of reaction products

Applications

• Isolation of genomic DNA from cultured cells and tissues
• Removal of DNases and RNases when isolating DNA and RNA from tissues or cell lines
• Determination of enzyme localization
• Improving cloning efficiency of PCR products

Storage

For long time storage, store the Proteinase K powder at 4ºC and Delution Buffer at -20ºC separately. For use, dissolved solution should be stored at -20ºC.

Quality Control

DNase Activity: None detectable enzyme activity with λDNA after 6 hrs incubation at 37ºC.

RNase Activity: None detectable ribonuclease activity after 16 hrs incubation at 25ºC.

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Definition of Activity Unit

One unit of the enzyme liberates Folin-positive amino acids and peptides corresponding to 1 µmol tyrosine in 1min at 37°C, pH 7.5 using denatured hemoglobin as substrate. Enzyme activity is assayed in the following mixture: 0.08 M potassium phosphate (pH 7.5), 5 M urea, 4 mM NaCl, 3 mM $CaCl_2$ and 16.7 mg/ml hemoglobin.

Source

Pichia pastoris cells with a cloned gene encoding Tritirachium album endolytic protease (Proteinase K).

Molecular Weight

28.9 kDa monomer.

Dilution Buffer

50mM Tris-HCl (pH 7.5), containing 5mM calcium chloride and 50% (v/v) glycerol.

Inhibition and Inactivation

Inhibitors: Proteinase K is not inactivated by metal chelators, by thiol-reactive reagents or by specific trypsin and chymotrypsin inhibitors. Phenylmethylsulfonyl fluoride and diisopropyl phosphorofluoridate completely inhibit the enzyme. Inactivated by heating at 95°C for 10 minutes.

Note

- Optimum activity at 50-55°C.
- Rapid denaturation of enzyme occurs at temperatures above 65°C.
- The recommended working concentration for Hy-Proteinase K is 0.05-1 mg/ml. The activity of the enzyme is stimulated by 0.2-1% SDS or by 1-4 M urea.
- Ca2+ protects Proteinase K against autolysis, increases the thermal stability and has a regulatory function for the substrate binding site of Hy-Proteinase K.
- Stable over a wide pH range: 4.0-12.5, optimum pH 7.5-8.0.

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