

Technical Data Sheet

Proteinase K, recombinant

for molecular biology

Order number: 2114

Proteinase K (also called Protease K or Endopeptidase K) EC 3.4.21.64 is a broad-spectrum serine protease. The enzyme was discovered in 1974 in extracts of the fungus *Engyodontium album* (formerly *Tritirachium album*). Proteinase K is able to digest native keratin (hair), hence, the name "Proteinase K". The predominant site of cleavage is the peptide bond adjacent to the carboxyl group of aliphatic and aromatic amino acids with blocked alpha amino groups.

Proteinase K is intended for the digestion of proteins in biological samples. The enzyme exhibits extremely effective degradation of both native and denatured proteins. Proteinase K is widely recommended for quick deactivation of endogenous RNases and DNases within the first steps of nucleic acid isolation.

Product details

Proteinase K for molecular biology is a recombinant, DNA free, serine protease of a highest available purity grade. The enzyme (cloned from fungus *Engyodontium album*) is produced in *Pichia pastoris*. Two chromatographic steps during the production process allow for complete removal of DNA, RNA, RNases and DNases.

Specific activity¹: min. 30 U/mg

Unit definition: One unit will hydrolyze urea denatured hemoglobin to produce color (by Folin Ciocalteu reagent) equivalent to 1.0 µmol of Tyrosine per min at pH 7.5 at 37 °C.

Storage and Stability

Proteinase K is an extremely stable protein that can withstand both high temperatures and strong fluctuations in the pH range. The pH optimum is 7.5-8.0, but pH values of 4.0 to 12.5 are also tolerated without damage. Proteinase K withstands denaturing conditions (urea, SDS) and temperatures up to 65°C.

The presence of Ca²⁺ ions (1-5 mM) in storage and reaction buffers prevent autolysis.

The lyophilized Proteinase K should be stored at 2 – 8°C or -20°C. Repeated freeze thaw cycles should be avoided. In solution, Proteinase K is stable for approx. 6 - 12 months at -20°C.

¹Anson, M.L. (1938) J. Gen. Physiol. 22, 79-98



Application

Dissolve 20 mg Proteinase K lyophilisate in 1 ml of DEPC-treated water to obtain a clear and colorless Proteinase K solution. Stock solutions (10 - 20 mg/ml) may be prepared in 10 mM CaCl₂ or 50 mM Tris·HCl, pH 8.0; 1 mM CaCl₂ with or without 50 % glycerol. As a preservative, 0.02 % Sodium azide might be added.

Common usage:

- x Purification of target material from contaminating proteins
- x Removal of DNases and RNases when isolating DNA and RNA
- x Prion (TSE) diagnostics
- x Mitochondria isolation
- x Protease foot printing

Inactivation

Proteinase K can be irreversibly denatured by heat treatment. Due to its high temperature tolerance, 20 minutes at 75°C is required. Proteinase K is also inhibited by Hg²⁺ ions, phenol, Diisopropyl fluorophosphate (DFP) and Phenyl methane sulfonyl fluoride (PMSF).

EDTA, sulfhydryl reagents and trypsin or chymotrypsin inhibitors, on the other hand, show hardly any inhibitory effect.

Related products

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| 1110 | Agarose Basic for biochemistry |
| 1057 | Agarose low EEO for molecular biology |
| 1531 | DNA Marker 1 kb (lyophilized) |
| 1530 | DNA Marker 100 bp (lyophilized) |
| 1254 | Ethidium bromide - Solution 0.07 % dropping bottle for electrophoresis |
| 1280 | Glycerol anhydrous for molecular biology |
| 1121 | DNase I for molecular biology |
| 1263 | RNase A (DNase-free) for molecular biology |
| 1303 | TBE buffer (10X) for electrophoresis |
| 1134 | Ethanol absolute for molecular biology |

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