

Active Recombinant Proteinase K, Molecular Biology Grade

Cat. No. Proteinase K-25E **Lot. No.** (See product label)

SPECIFICATION

Product Overview	From yeast cells with cloned gene encoding genetically engineered Engyodontium album (<i>Tritirachium album</i>) endolytic protease
Species	Engyodontium album
Source	Yeast
Description	Proteinase K is a broad-spectrum serine protease originally isolated from fungus Engyodontium album. The protease was named "Proteinase K" for its ability to digest Keratin. Crystal and molecular structure studies suggest that the enzyme belongs to the subtilisin family characterized with a catalytic triad (Asp39-His69-Ser224) in active site. Proteinase K has no pronounced cleavage specificity and preferential cleavage site is the peptide bond adjacent hydrophobic amino acids.
Form	Lyophilized powder
Bio-activity	≥ 34 units/mg of protein
Molecular Mass	29.3 kDa
Applications	Proteinase K is widely used in molecular biological applications to remove protein contamination from preparations of highly native, undamaged nucleic acids. It rapidly and effectively inactivates nucleases that could degrade DNA or RNA, even in the presence of denaturing agents. Proteinase K remains active in 1% Triton X-100 and fully active in 0.5% (w/v) SDS, which helps denature protein substrates and increase

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digestion rates. The enzyme works optimally at concentrations of 50-200 µg/mL, within a pH range of 7.5-11.0, and at temperatures between 37-70 centigrade. It is usually denatured by subsequent phenol extractions. Incubation times can vary from 30 minutes to 18 hours. Proteinase K also exhibits autolysis, a process where the enzyme digests itself, particularly during prolonged incubation.

Storage

At -20 centigrade Recommended. The shelf life of proteinase K lyophilized powder is 36 months after delivery when stored dry below 4 centigrade. The shelf life of proteinase K solution is 12 months after delivery when stored below 4 centigrade. Long term storage at -20 centigrade and keep dry for lyophilized powder to maintain stable for at least 3 years. Enzyme solutions prepared as recommended maintain stable at room temperature for 12 months. Proteinase K lyophilized powder and solution are extremely stable. Short term shipping and storage at ambient temperatures do not cause activity loss.

CAS No.

39450-01-6

Unit Definition

One unit is defined as the enzyme activity that produces 1 µmol of tyrosine per minute from casein at 37 centigrade at pH 7.5. Refer to the Certificate of Analysis (CoA) for specific values for the present lot.

EC No.

3.4.21.64

pI

8.9

pH range

4.5-12.0, Optimum pH range 7.5-11.5

Solubility

This product is soluble in water (1 mg/mL), yielding a clear colorless solution. Enzyme solutions prepared as recommended maintain stable at room temperature for 12 months.

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Bioburden	Microorganisms count: < 5 cfu/g
Advantages	Recombinant Proteinase K is a mutant form of the native protease, offering improved specific activity, higher yield, and a broader pH and temperature range for optimal activity. The large-scale recombinant production ensures lot-to-lot consistency, superior purity, and cost-efficiency. Its DNA-free nature makes it ideal for isolating DNA and RNA templates. Recombinant Proteinase K is widely used in molecular biology, molecular diagnostics, and biopharmaceutical applications for general protein digestion and chemo-enzymatic peptide synthesis. Its broad temperature profile facilitates protein unfolding, enhancing Proteinase K's ability to break down proteins effectively.
Dilution Buffer	1mM calcium chloride, 20mM Tris-HCl, pH 7.4;
Storage Buffer	1mM calcium chloride, 20mM Tris-HCl, 50% Glycerol, pH 7.4.
Temperature profile	Maximum activity at 70 centigrade 37-70 centigrade recommended
Quality Control	DNase is not detected in quality control procedure of incubation 40 µg Proteinase K with 1 µg λ DNA at 37 centigrade for 6 hours. DNase Contamination Assay: RNase is not detected in quality control procedure of incubation 40 µg Proteinase K with 2 µg RNA at 37 centigrade for 2 hours. RNase Contamination Assay: Each lot was tested to ensure the absence of Nucleases, DNA and RNA.
Activators	1-5 mM Ca ²⁺ To stimulate proteinase K activity, 1-5 mM Ca ²⁺ can be added. Optimization using activators can increase proteinase activity significantly. Enzyme activity will be reduced by 25% when calcium is removed by addition of EDTA. Enzyme activity will be reduced by 80% if the EDTA-Ca ²⁺ complex is removed from the enzyme solution by gel filtration, while it can be partially restored by addition of excess Ca ²⁺ .

Inhibitors

DIFP or PMSF The enzyme is inactivated by DIFP or PMSF (PMSF used at final concentration 5 mM.). However, it is not inhibited by EDTA, iodoacetic acid, trypsin-specific inhibitor TLCK, chymotrypsin-specific inhibitor TPCK, and p-chloromercuribenzoate.

GENE INFORMATION**Official Symbol**

Proteinase K

Synonyms

Peptidase K; Endoproteinase K; Endopeptidase K

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