

PRODUCT INFORMATION

Proteinase K, recombinant NGS grade

Cat. no. 33757

Product Description:

General	<p>Proteinase K from <i>Parengyodontium album</i> (<i>Tritirachium album</i>) is a Subtilisin-related serine protease. It is a broad-spectrum endopeptidase with very high specific activity widely used for digestion of proteins, including DNases and RNases, during nucleic acid preparations without compromising integrity of isolated DNA or RNA.</p> <p>Proteinase K is active under wide range of reaction conditions, including elevated temperatures and presence of SDS. This recombinant enzyme is expressed in <i>Pichia pastoris</i>, and undergoes extensive purification to yield the highest quality product. An extra purification step results in significantly increased solubility (2.5 fold), increased specific activity, and decreased DNA content.</p>
Application	<ul style="list-style-type: none"> • Inactivation of RNases and DNases during isolation of DNA or RNA from tissues, cell lines or cultured cells • Removal of protein impurities • Improving cloning efficiency of PCR products • Determination of enzyme localization on membranes • Removal of nucleases for <i>in situ</i> hybridization
Features	<ul style="list-style-type: none"> • Supplied as highly purified lyophilized powder • Specific activity*: ≥ 45 U/mg protein, ≥ 35 U/mg lyophilizate • DNA content: ≤ 0.1 pg/mg by qPCR • Free of DNase and RNase activity • Exhibits broad substrate specificity • Solubility in water: ≥ 50 mg/ml
Storage	Recommended temperature for long-term storage: - 20 °C
Activation	The enzyme is stimulated by addition of denaturing agents 0.2 - 1 % (w/v) SDS or 4 M urea. It exhibits prolonged stability due to the presence of Ca^{2+} (1 - 6 mM), which protects enzyme from autolysis and increases its thermal stability.
Inhibition	Diisopropylfluorophosphate, Phenylmethysulfonyl fluoride and mercury ions. Proteinase K is unaffected by metal-chelating agents and sulfhydryl inhibitors.
Stock solution preparation	<p>Solution 20 - 50 mg/ml: Use purified water for immediate use, or 50 % (v/v) glycerol in purified water for long-term storage at -20°C.</p> <p>Solution ≥ 20 mg/ml: Use 50 mM Tris/HCl, pH 7.8, 3 mM CaCl_2 for immediate use, or 50 mM Tris/HCl, pH 7.8, 3 mM CaCl_2, 50 % (v/v) glycerol for long-term storage at -20°C.</p>
Reaction conditions	<p>Proteinase K is typically used at 50 to 200 $\mu\text{g/ml}$ in nucleic acid preparations at pH 7.5 to 8.0 and 37 °C to 55 °C. Incubation times vary from 30 minutes to 18 hours.</p> <p>Working pH range: 4.0 - 12.0 (optimum activity at pH 7.5 - 8.5) Working temperature range: 20 °C to 65 °C (optimum at 50 °C to 56 °C)</p>
Note	<i>If necessary, to help the product to be fully soluble before use, heat the solution to 55 °C temperature and maintain the it for 15 – 45 min. After that cool and store at -20 °C. The product will retain its full activity.</i>

*Unit definition: 1 U is defined as the amount of enzyme that liberates Folin-positive amino acids and peptides, corresponding to 1 μmol tyrosine under assay conditions in 1 minute using haemoglobin as substrate.

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