

**Product Insert**

**Proteinase K**

<b>Cat. No.</b>		
<b>EA61020</b>	<b>Proteinase K Powder</b>	<b>20mg</b>
<b>EA61200</b>	<b>Proteinase K Powder</b>	<b>200mg</b>
<b>EA62020</b>	<b>Proteinase K Solution</b>	<b>20mg</b>
<b>EA62200</b>	<b>Proteinase K Solution</b>	<b>200mg</b>

**Shipping: On Dry/Blue Ice**

**Store at -20°C**

**Features**

- Ready-to-use solution
- Active in a wide range of reaction products

**Description**

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

**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

**Quality Control**

The absence of endo-, exodeoxyribonucleases and ribonucleases confirmed by appropriate quality tests.

**Source**

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

**Molecular Weight**

28.9 kDa monomer (6).

**Definition of Activity Unit**

One unit of the enzyme liberates Folin-positive amino acids and peptides corresponding to 1  $\mu$ mol tyrosine in 1 min at 37°C using denatured hemoglobin as substrate. Enzyme activity is assayed in the following mixture: 0.08M potassium phosphate (pH 7.5), 5 M urea, 4 mM NaCl, 3 mM CaCl<sub>2</sub> and 16.7 mg/ml hemoglobin.

**Storage Buffer**

The enzyme is supplied in: 50 mM Tris-HCl (pH 7.5), containing 5 mM 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 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.
- Ca<sup>2+</sup> protects Proteinase K against autolysis, increases the thermal stability and has a regulatory function for the substrate binding site of Proteinase K.
- Stable over a wide pH range: 4.0 - 12.5, optimum pH 7.5 - 8.0.

**Technical Support**

If the troubleshooting guide does not solve the difficulty you are experiencing, please contact Technical Support with details of reaction setup, cycling conditions and relevant date.

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