

Quick-Start Protocol

Proteinase K – Solution

The Proteinase K (cat. nos. RP107B-1 and RP107B-5). A recombinant Proteinase K from *Parengyodontium album* (*Tritirachium album*) is a 28.9 kDa protein expressed in *Komagataella phaffii* (*Pichia pastoris*). It is a broad-spectrum endopeptidase with an extraordinary specificity allowing highly effective digestion of proteins in many laboratory applications. Upon arrival, Proteinase K should be stored at -20°C , $+4^{\circ}\text{C}$, or room temperature. Do not store above 25°C . Avoid direct UV radiation/sunlight. Proteinase K maintains activity ≥ 800 U/mL for at least 24 months when stored in its original, unopened container.

Further information

- Safety Data Sheets: www.qiagen.com/safety
- Technical assistance: support.qiagen.com

Notes before starting

- One unit of Proteinase K hydrolyzes urea-denatured hemoglobin producing color equivalent of 1 μmol tyrosine per 1 min at 37°C and pH 7.5 (Folin & Ciocalteu's method), 1 U = 1 mAnsonU.
- Storage buffer: 10 mM Tris-HCl, pH 7.5; 1 mM $(\text{CH}_3\text{COO})_2\text{Ca}$; 50% glycerol

Considerations for use

1. The enzyme is typically used at 50–200 $\mu\text{g}/\text{mL}$ nucleic acid preparations at pH 7.5–8.5 and 37 – 55°C . Incubation times vary from 30 minutes to 18 hours.

2. The Proteinase K cleaves proteins preferably behind hydrophobic amino acids. The smallest peptide to be hydrolyzed is a tetrapeptide.
3. Working pH range is 4.0–12.0 with optimum activity at pH 7.5–8.5. Full activity is maintained over several hours over a pH range of 6.5–10.0.
4. Working temperature range is 20–65°C with optimum activity at 50–56°C.
5. The enzyme is stimulated by addition of denaturing agents 0.2–1% SDS or 1–4 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.
6. The enzyme is not inactivated by chelating agents (e.g., EDTA), chaotropic salts, detergents (e.g., 1% SDS, 1–4 M urea), metal ions, thiol reagents, or trypsin-specific inhibitors.
7. Proteinase K is usually denatured by subsequent phenol extractions. It can be also inactivated by heating above 65°C or using inhibitors such as PMSF or DIFP.

Document Revision History

Date	Changes
August 2023	Initial release

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual.

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