# PRODUCT INFORMATION SHEET

Mo Bi Tec
MOLECULAR BIOLOGISCHE TECHNOLOGIE

## IMMOBILIZED PROTEINASE K G3M # P3502

Proteinase K from Tritirachium album. Chromatographically purified, free of ribo- and deoxyribonucleases.

Proteinase K is an unspecific serine protease with strong proteolytic activity on denatured (in SDS) and high molecular weight native proteins. It cleaves peptide bonds mostly after the carboxyl group of N-substituted hydrophobic, aliphatic and aromatic amino acids.

**G3m:** 25 µg proteinase K immobilized on matrix G3m per CR-column.
0.7 mAnson units immobilized per CR-column.
This CR-column cuts at least 370 µg BSA per application.

**Nr. 5 Storage buffer:** 50 mM Tris/HCl, pH 7.5
**Nr. 16 Reaction buffer:** 50 mM Tris/HCl, 5 mM NaCl, pH 8.0
**Nr. 17 Washing buffer:** 50 mM Tris/HCl, 1.0 M NaCl, pH 8.0

The columns are more active in 0.1% SDS and at 40°C. Also active in PBS buffer (20 mM Na-phosphate, 150 mM NaCl at pH 7.6).

## Protocol

For more details see MoBiTec-CRC-Handbook.

**1. Dilute delivered buffers** (at least 2 ml each) with sterile doubly distilled water.
For 1 application you need:
1 ml 10x reaction buffer and 9 ml doubly distilled water
2 ml 5x washing buffer and 8 ml doubly distilled water
1 ml 10x storage buffer and 9 ml doubly distilled water
The substrate should be in reaction buffer

**2. Equilibrate the CR-column with 10 ml reaction buffer.**
Fill 10 ml reaction buffer into a syringe, let the reaction buffer run through the column by gravity to the upper filter. In case the buffer runs very slowly, apply pressure by a syringe.

**3. Load substrate solution in reaction buffer.**
Small volumes (< 70 µl): spin the CR-column 5 seconds in a benchtop centrifuge (2000 rpm are sufficient). Let the substrate solution enter the matrix material.
Larger volumes: Let the substrate solution run through the column.
Flow-rate: up to 70 µl/minute
Keep the substrate in the column for about 1 minute at room temperature. Higher turn-over is obtained when the substrate is applied to the column again or incubated for longer times.

**4. Elute the product solution.**
Small volumes (< 70 µl): centrifuge the product out of the column.
Larger volumes: Let the substrate run through the column and spin the residual solution out of the matrix
Notice: Molecules < 700 Dalton have to be eluted with 7 ml reaction buffer.
It does not harm the columns if they run dry.

**5. Wash the column with 10 ml washing buffer.**

**6. Equilibrate the column with 10 ml storage buffer.**
Store the column at 4°C. Never freeze a CR-column!



**MoBiTec** GmbH, Lotzestraße 22a, D-37083 Göttingen, Tel.: ++49 551-70 722- 0, Fax: ++49 551-70 722-22
E-mail: info@mobitec.de or order@mobitec.de, INTERNET: http://www.mobitec.de