



# PRODUCT INFORMATION SHEET

## IMMOBILIZED TRYPSIN G3M

# P3702

Trypsin from bovine pancreas. TPCK treated (free of a-chymotrypsin activity).

Trypsin hydrolyzes proteins, peptides, amides and esters specifically at the carboxyl groups of the basic amino acids L-arginine or L-lysine.

**G3m:** 25 µg trypsin per CR-column, immobilized on dextran.

260 ST-units immobilized per CR-column.

This CR-column cuts at least 50 µg tubulin per application.

- Nr. 15 Storage buffer:** 50 mM Tris/HCl at pH 8.0  
**Nr. 67 Reaction buffer:** 50 mM phosphate at pH 8.0 (Sørensen)  
**Nr. 68 Washing buffer:** 50 mM phosphate at pH 8.0, 1 M NaCl

## Protocol

For more details see MoBiTec-CRC-Handbook

### 1. Dilute delivered buffers (at least 2 ml each) with 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.

Load in a syringe 10 ml reaction buffer, 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!