

PRODUCT INFORMATION

Collagenase NB 6 GMP Grade

Cat. No. 17458

Product Description:

General

Collagenase NB 6 GMP Grade is designed for dissociation of human tissue for isolation of different cell types which are intended for transplantation into humans.

The aseptic production process complies with the requirements of Annex 18 to the EU-Guide to Good Manufacturing Practices "GMP for active pharmaceutical ingredients". It therefore guarantees for reproducibility and traceability of batches. The documentation comprises all production, control and distribution records.

Documentation concerning collagenase (e.g. viral and TSE safety, stability data) is available for approval of the cell isolation procedure by the authorities.

The Collagenase NB 6 producing strain of *Clostridium histolyticum* has been carefully selected for producing a collagenase product that is non-toxic according to the requirements of the European Pharmacopoeial Test for Abnormal Toxicity ("General Safety Test"). A balanced ratio of collagenase and other proteases guarantees for high yields of viable cells.

Specification	Collagenase activity	≥ 0.1 U/mg (PZ acc. to Wunsch)
	Sterility (Ph. Eur.)	Must comply
	<i>Clostridia</i>	Must comply
	Abnormal Toxicity	Must comply

Application

Collagenase NB 6 GMP Grade is exclusively designed for dissociation of different human tissues to isolate cells for transplantation into humans.

The enzymes are not intended for use in humans.

Responsibility for clinical use and the methods to isolate, purify and transplant cells lies solely with the providing physician/researcher.

Storage conditions

Collagenase NB 6 GMP Grade is provided as a lyophilized powder and should be stored **in a dry state** at +2 to +8 °C (Stability data acc. to ICH guidelines for 5 years).

The dissolved enzyme (in Tris buffer, pH 7.1) is stable at below -20 °C (Stability data acc. to ICH guidelines for 1 year available).

Instructions for use:

General Collagenase NB 6 GMP Grade is designed for dissociation of different tissues to isolate various cell types, e.g. chondrocytes, muscle cells, skin fibroblasts, hepatocytes, adipocytes. For isolation of each cell type a specific isolation procedure has to be applied to receive optimal yields of viable cells.

Required enzymatic activity For establishing of the procedure the following concentrations of Collagenase NB 6 can be used as starting point:

Adipocytes (adult)	0.1 - 0.3 PZ-U/ml
Chondrocytes	0.36 PZ-U/ml
Fibroblasts (foreskin)	0.45 PZ-U/ml
Endothelial cells (vessel)	0.1 PZ-U/ml

If digestion is incomplete or if cells are damaged, collagenase concentration, time or temperature should be increased or decreased, respectively.

For establishing of the procedure Collagenase NB 4 Standard Grade can be used, which has the same enzymatic composition as Collagenase NB 6 GMP Grade.

Stock solution Collagenase NB 6 easily dissolves in a concentration of up to 150 mg/ml in all buffers, which are generally used for cell isolation. Keep the dissolved enzyme on ice. As Collagenase and some of the secondary proteases depend on calcium, 2 mM Ca²⁺ should be added, and no calcium chelating agents (e.g. EDTA) should be used. Collagenase NB 6 is pharmaceutical sterile therefore no sterile filtration is necessary if sterile buffers and reagents are used. Sterile filtration might result in loss of protein/activity.

Working solution Stock solution is diluted with dissociation buffer to achieve the required enzymatic activity (see above). Dissociation can be terminated by diluting the enzyme solution with buffer and cooling the system.
