

PRODUCT INFORMATION

Collagenase AF-1 GMP Grade

Cat. No. N0003554

General Collagenases from *Clostridium histolyticum* are proteolytic enzymes that cleave peptide bonds in the triple helical collagen molecules of human or animal tissue *in situ*.

For this reason collagenases are widely used for isolation of various cell types by tissue dissociation.

Description Collagenase AF-1 GMP Grade is manufactured according to GMP guidelines using a production process completely free of animal-based components. In this way the introduction of any potential animal-derived pathogen is excluded.

Collagenase AF-1 GMP Grade is chromatographically highly purified leading to a very high collagenolytic activity. It is largely free from additional enzymatic activities like clostripain, trypsin-like activity and neutral protease, as well as endotoxins.

The absence of any animal-derived ingredients and thorough microbial analysis provide the highest possible safety.

Specification	Collagenase activity	≥ 3.000 U/mg (PZ acc. to Wunsch) ≥ 2000 U/vial (PZ acc. to Wunsch)
	Neutral protease activity	≥ 0.050 U/mg (DMC)
	Trypsin-like activity	status
	Clostripain, native state	status
	TAMC	≤ 10/vial
	TYMC	≤ 10/vial
	Bacterial endotoxins	≤ 10.0 EU/mg

Application Collagenase AF-1 GMP Grade is suitable for cell isolation from several tissue types intended for clinical applications.

It is often used in combination with Neutral Protease AF GMP Grade (Cat. No. N0003553).

Storage conditions Collagenase AF-1 GMP Grade is available as a lyophilized powder. It should be stored at +2 to +8 °C in a dry environment.

Under these conditions the product is stable until the date stated on the certificate of analysis if repeated opening and closing of the vial is avoided.

Documents For each lot a customer-specific certificate of analysis is provided.

For additional documentation (e.g. stability data, GMP certificate, certificate of origin, etc.), please contact our product management team at collagenase@nordmark-biochemicals.com.

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Instructions for use:

General Collagenase AF-1 GMP Grade, often in combination with Neutral Protease AF GMP Grade (Cat. No. N0003553), is suitable for the dissociation of sensitive cells from several tissues, such as pancreas, liver and dental pulp. It has been shown to be highly effective for the isolation of islets of Langerhans from human pancreas intended for transplantation.

Tissue dissociation Recommended starting concentrations for isolation of islets of Langerhans from human pancreas:

Collagenase AF-1 GMP Grade:	15 – 20 PZ U/g tissue
Neutral Protease AF GMP Grade:	0.8 – 1.5 DMC U/g tissue

Collagenase AF-1 GMP Grade is provided in vials with ≥ 2000 PZ U which is usually sufficient for dissociation of one human pancreas.

The appropriate collagenase concentration depends on the tissue type and origin as well as on the applied isolation procedure.

Collagenase activity is at an optimum at 37 °C and pH 7.4.

If you have more questions concerning your application, you are welcome to contact the product management team at collagenase@nordmark-biochemicals.com.

Stock solution A stock solution of Collagenase AF-1 GMP Grade can be prepared by dissolving the enzyme in buffer or water. The enzyme solution should be constantly stored on ice.

Since collagenase depends on calcium, absolutely no calcium chelating agents (e.g. EDTA) should be present at all. If desired, a buffer with ≥ 2 mM Ca^{2+} can be used.

Reconstituted Collagenase AF-1 GMP Grade can be filtered, aliquoted and stored at -20 °C. Repeated freezing and thawing should be avoided. For 0.22 μm filtration filters with low protein binding properties (e.g. cellulose acetate, PVDF, or PES) are recommended.

Working solution To prepare a working solution, the stock solution is diluted to achieve the desired collagenase concentration.

The working solution should be constantly stored on ice until use.

If Collagenase AF-1 GMP Grade solution is mixed with neutral protease solution, the blend should be used immediately.

Inactivation and inhibitors The dissociation process can be reduced, e.g. by cooling down or diluting the enzyme solution.

Collagenase is reversibly inactivated at high pH values and irreversibly inactivated at low pH values. Inhibitors of collagenase include cysteine or chelating agents like EDTA.

Important note Collagenase AF-1 GMP Grade is not intended for direct application in humans.