

For life science research only.
Not for use in diagnostic procedures.



Chymotrypsin Sequencing Grade from bovine pancreas

 **Version: 10**

Content Version: November 2020

Lyophilized

Cat. No. 11 418 467 001 4 x 25 µg

Store the product at +2 to +8°C.

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1. General Information

1.1. Contents

Vial / Bottle	Label	Function / Description	Content
1	Chymotrypsin Sequencing Grade	<ul style="list-style-type: none"> Highly purified and specific protease. Salt-free 	4 vials, 25 µg each

1.2. Storage and Stability

Storage Conditions (Product)

When stored at +2 to +8°C, the product is stable through the expiry date printed on the label.

Vial / Bottle	Label	Storage
1	Chymotrypsin Sequencing Grade	Store at +2 to +8°C.  Store dry.

1.3. Additional Equipment and Reagent required

For reconstitution of lyophilizate

- HCl

For digestion of proteins

- Tris-HCl*
- CaCl₂

1.4. Application

Use Chymotrypsin for the hydrolysis of proteins, alone or in combination with other proteases. The enzyme is suitable for:

- Peptide mapping
- Fingerprinting
- Sequencing analysis

2. How to Use this Product

2.1. Before you Begin

General Considerations

General handling

The content of one vial may be used for several simultaneous digests.

⚠ Use a new vial when repeating a digest in order to minimize the risk of contamination or autolysis.

Activity determination

Denaturing agent	Concentration	Enzyme activity in [%]
SDS (1 hour incubation)	0.01% (w/v)	50 to 60
	0.1% (w/v)	0
	1% (w/v)	0
Urea (5 hours incubation time)	1.0 M	80
Guanidine hydrochloride (5 hours incubation time)	1.0 M	80
Acetonitrile (5 hours incubation time)	5% (v/v)	80

Safety Information

Laboratory procedures

- Handle all samples as if potentially infectious, using safe laboratory procedures. As the sensitivity and titer of potential pathogens in the sample material varies, the operator must optimize pathogen inactivation by the Lysis / Binding Buffer or take appropriate measures, according to local safety regulations.
- Do not eat, drink or smoke in the laboratory work area.
- Do not pipette by mouth.
- Wear protective disposable gloves, laboratory coats and eye protection, when handling samples and kit reagents.
- Wash hands thoroughly after handling samples and reagents.

Waste handling

- Discard unused reagents and waste in accordance with country, federal, state, and local regulations.
- Safety Data Sheets (SDS) are available online on dialog.roche.com, or upon request from the local Roche office.

Working Solution

Solution	Preparation/ Composition	Storage and Stability	For use in...
Chymotrypsin Sequencing Grade	Dissolve lyophilizate in 1 mM HCl.	Store 1 week at +2 to +8°C.	Digestion mixture of proteins.
Digestion buffer	100 mM Tris-HCl*, 10 mM CaCl ₂ , pH 7.8.	-	Dissolution of the proteins.

2.2. Protocols

Incubation

i See section, **Working Solution** for information on preparing solutions.

- 1 Dissolve the proteins to be sequenced in Digestion buffer.

- 2 Dilute protein solution with buffer, see section, **General Considerations** to determine the enzyme activity depending on the denaturing agent in the digest.
 - i* The recommended amount of enzyme is 1/100 to 1/20 of the protein by weight.

- 3 Choose an incubation time between 2 and 18 hours at +25°C, depending on the amount of enzyme.

2.3. Parameters

Contaminants

Trypsin <0.07%

EC-Number

EC 3.4.21.1

Inhibition

Aprotinin, DFP, PMSF, phenothiazine-N-carbonyl chloride, TPCK, ZPCK, α2-macroglobulin, α₁-antitrypsin, soybean trypsin inhibitor, and chymostatin.

i No inhibition by APMSF.

Molecular Weight

25 kDa

pH Optimum

pH 7.0 to 9.0

Purity

The enzyme is free of impurities which might interfere in the separation range of peptides in reversed-phase HPLC (detection at 215 nm).

Function and purity control by HPLC and SDS polyacrylamide gel electrophoresis ensures a constant quality with each lot.

Specificity

Serine endopeptidase. At pH 7.0 to 9.0, it specifically hydrolyzes peptide bonds at the C-termini of Tyr, Phe, and Trp.

- Leu, Met Ala, Asp, and Glu are cleaved at a lower rate.
- The specificity of Chymotrypsin Sequencing Grade is tested with melittin as substrate.

Working Concentration

Amount of enzyme is 1/200 to 1/20 of the quantity of protein by weight.

3. Supplementary Information

3.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols

i *Information Note: Additional information about the current topic or procedure.*

⚠ Important Note: Information critical to the success of the current procedure or use of the product.

① ② ③ etc. Stages in a process that usually occur in the order listed.

① ② ③ etc. Steps in a procedure that must be performed in the order listed.

* (Asterisk) The Asterisk denotes a product available from Roche Diagnostics.

3.2. Changes to previous version

Layout changes.

Editorial changes.

Update to include new safety Information to ensure handling according controlled conditions.

3.3. Ordering Information

Product	Pack Size	Cat. No.
Reagents, kits		
Tris hydrochloride	500 g	10 812 846 001

3.4. Trademarks

All product names and trademarks are the property of their respective owners.

3.5. License Disclaimer

For patent license limitations for individual products please refer to:

List of biochemical reagent products.

3.6. Regulatory Disclaimer

For life science research only. Not for use in diagnostic procedures.

3.7. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

3.8. Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site.**

To call, write, fax, or email us, visit **sigma-aldrich.com**, and select your home country. Country-specific contact information will be displayed.

