

## Certificate of Analysis

### Pepsin:

<b>Part No.</b>	<b>Size</b>
V195A	250mg

**Description:** Pepsin preferentially cleaves at the C-terminus of phenylalanine, leucine, tyrosine and tryptophan (1–4). This protease can be used alone or in combination with other proteases for protein analysis by mass spectrometry and other applications.

**Biological Source:** Porcine stomach.

**Molecular Weight:** 34.6kDa.

**Form:** Lyophilized.

**Storage Conditions:** See the Product Information Label for storage conditions and expiration date.

**Optimal pH:** 1.0–3.0 (4–6).

**Activators:** Hydrochloric acid (HCl), trifluoroacetic acid (TFA).

**Inactivators:** pH greater than 6.0.

#### Usage Notes:

1. Resuspend Pepsin in double-distilled water (pH 5.5 or lower) to a final concentration of 1mg/ml. Store reconstituted Pepsin at 4°C for up to 1 month.
2. Specificity for cleavage at Phe and Leu is best at pH 1.0 and decreased above pH 2.0. Pepsin irreversibly inactivates above pH 6.0.

## Quality Control Assays

This lot passes the following Quality Control specifications:

**Activity:** Digestion reactions using insulin as a substrate are performed at a protease:substrate ratio of 1:20 and analyzed by reverse-phase HPLC. Intact substrate is undetectable after incubation for 15 minutes at 37°C.

#### Usage Information on Back

Signed by:



R. Wheeler, Quality Assurance

Part# 9PIV1959

Revised 8/16



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## Promega

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## 1. In-Solution Digestion Protocol

1. Resuspend the protein in buffer at pH 7.5.
2. Transfer the protein solution to a microcentrifuge tube.
3. Add 1N HCl to the solution to a final concentration of 0.04N.
4. Resuspend Pepsin in double-distilled water.  
**Note:** Resuspending the enzyme in buffer at neutral or higher pH will destabilize or irreversibly inactivate it.
5. Add Pepsin to protein solution; mix. We recommend using enzyme:protein ratios of 1:20 to 1:100 (w:w).
6. Mix and centrifuge briefly.
7. Incubate 1–18 hours at 37°C.
8. Stop the reaction by heating at 95°C for 10 minutes.

## 2. References

1. Aiten, A. *et al.* (1989) *Protein Sequencing: A Practical Approach*, IRL Press, Oxford, UK, 43.
2. Christenses, L.K. (1955) *Arch. Biochem.* **57**, 163–73.
3. Sweeney, P.J. and Walker, J.M. (1993) In: *Enzymes of Molecular Biology*, Burrell, M.M., ed., Humana Press, Totowa, NJ, 290–1.
4. Dixon, M. *et al.* (1979) *Enzymes*, Academic Press, New York, NY, 262.
5. Schlamowitz, M. *et al.* (1959) *J. Bio. Chem.* **234**, 3137–45.
6. Cornish-Bowden, A.J. and Knowles, J.R. (1969) *BioChem. J.* **113**, 353–62.

## 3. Related Products

Product	Size	Conc.	Cat. #
Asp-N, Sequencing Grade	2µg		V1621
Arg-C, Sequencing Grade	10µg		V1881
Chymotrypsin, Sequencing Grade	25µg		V1061
	100µg (4 × 25µg)		V1062
Elastase	5mg		V1891
Endo H	10,000u	500u/µl	V4871
	50,000u	500u/µl	V4875
Endoproteinase Lys-C, Sequencing Grade	5µg		V1071
Fetuin	500µg	10mg/ml	V4961
Glu-C, Sequencing Grade	50µg (5 × 10µg)		V1651
Immobilized Trypsin	2ml		V9012
	4ml (2 × 2ml)		V9013
PNGase F	500u	10u/µl	V4831
ProteaseMAX™ Surfactant, Trypsin Enhancer	1mg		V2071
	5 × 1mg		V2072
Protein Deglycosylation Mix	20 reactions		V4931
rLys-C, Mass Spec Grade	15µg		V1671
Sequencing Grade Modified Trypsin	100µg (5 × 20µg)		V5111
Sequencing Grade Modified Trypsin, Frozen	100µg (5 × 20µg)		V5113
Thermolysin	25mg		V4001
Trypsin Gold, Mass Spectrometry Grade	100µg		V5280
Trypsin/Lys-C Mix, Mass Spec Grade	20µg		V5071
	100µg		V5072
	100µg (5 × 20µg)		V5073