# Sigma-Aldrich.

Product Information

## Pepsin from porcine gastric mucosa

Lyophilized powder

#### P7012

## **Product Description**

CAS Registry Number: 9001-75-6

Enzyme Commission (EC) Number: 3.4.23.1

Molecular Mass: 34,620 (calculated from amino acid

sequence)1

pI:2,3 2.2-3.0; 2.2, 2.8

λ<sub>max</sub>: 278 nm<sup>4</sup>

Extinction coefficient:  $E^{M} = 51,300^{4}$ 

Synonym: Pepsin A

Pepsin, unlike some other peptidases, hydrolyzes only peptide bonds, not amide or ester linkages. The cleavage specificity includes peptides with an aromatic acid on either side of the peptide bond, especially if the other residue is also aromatic or a dicarboxylic amino acid. Increased susceptibility to hydrolysis occurs if there is a sulfur-containing amino acid close to the peptide bond, which has an aromatic amino acid. Pepsin will also preferentially cleave at the carboxyl side of Phe and Leu, and to a lesser extent at the carboxyl side of Glu residues. Pepsin will not cleave at Val, Ala, or Gly linkages. Some good substrates of pepsin include:

- Z-L-tyrosyl-L-phenylalanine
- Z-L-glutamyl-L-tyrosine
- Z-L-methionyl-L-tyrosine

Amidation of the C-terminal carboxyl group prevents hydrolysis by pepsin.<sup>5,6</sup>

Pepsin is commonly used in the preparation of Fab fragments from antibodies. The optimal pH for the pepsin reaction is 1.5-2.5, which will not be detrimental to the antibody if it is not exposed for long durations to the low pH. Solutions should be adjusted to neutral pH for storage. The control of pepsin digestion of antibodies has been reported.<sup>7</sup>

For general protein digestion, suggested conditions are a 0.4% solution of pepsin in 10 mM HCl, and digestion for 30-90 minutes at 37 °C. Pepsin has an optimal activity with native proteins at pH  $\sim$ 1.0. However, with some denatured proteins, the optimal activity is in the range of pH 1.5-3.5.8,9 Pepsin is inhibited by several Phe-containing peptides.10

Several theses<sup>11-15</sup> and dissertations<sup>16-22</sup> have cited use of product P7012 in their protocols.

## Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## **Preparation Instructions**

Pepsin is soluble in deionized water at 10 mg/mL, and 4 mg/mL in cold 10 mM HCl.

## Storage/Stability

In lyophilized form as sold, this product should be stored at  $-20~^{\circ}\text{C}$ .

Solutions of pepsin at pH 4.4 are stable at -20 °C for about 2-3 months.<sup>23</sup> Pepsin is not active when not at an acidic pH and a solution is stable at pH 6-7. Bringing the pH up to 8, however, will irreversibly inactivate pepsin. Pepsin is irreversibly denatured at pH 8.5-11 at room temperature.<sup>24</sup>

#### References

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