



SG-ChymotrypsinTM

INTRODUCTION

SG-Chymotrypsin^{$^{\text{IM}}$} is a sequencing grade serine endopeptidase, which predominantly cleaves peptide bonds on the carboxy side of tyrosine, phenylalanine and tryptophan. In addition, it has a low catalytic activity against the carboxy side of leucine, methionine, alanine, aspartic and glutamic acids, although at a much lower rate. It is therefore recommended to always use the shortest digestion time possible. SG-Chymotrypsin^{$^{\text{IM}}$} is first treated with TLCK and then subjected to an extensive purification process to remove contaminating protease and chymotryptic autolysis by-products. The highly purified enzyme is then chemically modified to increase its resistance to autolysis and increase its stability. The modified enzyme retains >80% of its activity after 6 hours incubation at 30°C in reaction buffer and >70% of activity after 24 hours incubation under the same conditions. The chemically modified SG-Chymotrypsin^{$^{\text{IM}}$} is stable in denaturing agents (see Table) and therefore can be used to digest difficult to solubilize proteins.

Denaturing Agent	Concentration	% Enzyme Activity Retained
Control	-	100
Urea	0.50M	100
Urea	1.00M	100
Urea	2.00M	100
Urea	3.00M	100
Urea	4.00M	100
Guanidine-HCl	0.05M	100
Guanidine-HCl	0.10M	100
Guanidine-HCl	0.25M	100
Guanidine-HCl	0.50M	11

ITEM(S) SUPPLIED

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Description	Size		
SG-Chymotrypsin TM	2 vials, 25µg/vial		
Digestion Buffer (CHY)	2ml		

STORAGE CONDITIONS

It is shipped at ambient temperature. Upon arrival, store at 2-8°C and is stable for 1 year.

PREPARATION BEFORE USE

NOTE: SG-Chymotrypsin is supplied lyophilized, $5\mu g/vial$.

Reconstitute the enzyme with 25 μ l Digestion Buffer (CHY) to produce a concentration of 1.0 μ g/ml. Reconstituted enzyme is stable for 1 month at -70 °C, repeated freeze thawing is not recommended.

PROTOCOL

For optimal digestion make sure protein sample is either prepared or equilibrated in 50mM Tris-HCl, pH8.0, 0.1mM CaCl₂.

- 1. For protein fragmentation, SG-ChymotrypsinTM is typically added to the protein at a ratio of 1:200 to 1:50 enzyme to protein, by weight.
- 2. The incubation is allowed to proceed at 25-30°C for 1-10 hours, but can be extended to 24 hours in some applications.

<u>NOTE</u>: It is recommended to choose a ratio of enzyme to protein to allow for the shortest incubation time possible, to reduce or eliminate the catalyzed hydrolysis of peptide bonds with non-aromatic amino acid residues.

RELATED PRODUCTS

NOTE: For other related products, visit our web site at <u>www.GBiosciences.com</u> or contact us.

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