



Turbo TEV Protease

recombinant, E. coli

Cat. Nº.	Amount
□ ENZ-102S	1.000 U
□ ENZ-102M	10.000 U
□ ENZ-102L	50.000 U

Unit Definition: One unit of TEV protease cleaves >85% of 3 µg of control substrate in one hour at pH 8.0 at 30 °C.

Storage Buffer

The enzyme is supplied in: 50 mM Tris-HCl (pH 8.0), 50 mM NaCl, 1 mM EDTA, 5 mM DTT and 50% (v/v) glycerol.

Concentration:

1 mg/mL (5 U/μL)

For in vitro use only!

Shelf Life:

12 months

Shipping: Shipped on blue ice

Storage Conditions:

Store at -20 °C

Additional Storage Conditions: Avoid freeze/thaw cycles.

Origin:

Tobacco Etch Virus

Description:

Turbo TEV Protease is an enhanced form of Tobacco Etch Virus (TEV) protease that is highly site-specific, active, and more stable than native TEV protease. Turbo TEV Protease recognizes the seven-amino-acid sequence Glu-Asn-Leu-Tyr-Phe-Gln-Gly and cleaves between Gln and Gly with high specificity. The protease is used to cleave affinity tags from fusion proteins. The optimal temperature for cleavage is 30°C; however, the enzyme is active over wide ranges of temperature and pH (pH 6.0-8.5). Following digestion, Turbo TEV Protease is easily removed from the cleavage reaction by affinity chromatography using the polyhistidine tag at the N-terminus of the protease.

Removal of Turbo TEV Protease after Cleavage

The Turbo TEV protease contains a polyhistidine tag at the Nterminus. After cleavage of the fusion protein, remove Turbo TEV Protease from the cleavage reaction by affinity chromatography on a nickel chelating resin.

Reaction Conditions:

Prepare fresh dialysis buffer. Dialysis buffer should be optimized for target protein solubility and contain no protease inhibitors. The dialysis buffer should also be compatible with downstream purification processes, e.g. minimal amount of EDTA or DTT if a IMAC column will be used to remove the cleaved His-tag. Example of suitable dialysis buffer: 25 mM Tris-HCl pH 8.0, 150 -500 mM NaCl, 14 mM mercaptoethanol.

The standard reaction buffer for Turbo TEV Protease is 50 mM Tris-HCl pH 8.0, 0.5 mM EDTA, 1 mM DTT. Save a small aliquot as a control for PAGE analysis.

Protocol:

- Add Turbo TEV protease to target protein ratio of 1:100 (w/w) or 10,000 unit (1 mg) TEV protease for 100 mg of target protein. There is no need to calculate the molar ratio.
- Turbo TEV protease can be added directly to the target protein. There is no need to change buffer or dilute Turbo TEV protease. The optimal ratio should be determined empirically.
- A Protease-to-target protein ratio (w/w) of 1:50 to 1:200 should provide an affective range for most target proteins.
- Dialyze against the dialysis buffer at 4 °C ~ 16 hrs. Dialysis or desalting chromatography are intended to remove imidazole in order to remove the cleaved tag or TEV protease after cleavage.
- Typically, 1 mg of TEV protease will cleave >90% of 100mg of a control protein at 4 °C in 16 hours.