

## **TEV** protease

### Cat# BB-E0030 (0.25mg)

**Description:** This cysteine protease, which originates from the *Tobacco Etche Virus* (TEV) is highly site specific and is expressed in *E. coli*. It recognizes the sequence Glu-Asn-Leu-Tyr-Phe-Gln-(Gly/Ser) (ENLYFQ (G/S)) and cleaves between the Gln and Gly/Ser residues. Although the optimal temperature for activity is 30° C, it can work over a broad temperature range from 4° C to 30° C.

## Reagents supplied:

TEV Protease: 250μl (5U/μl)

20X TEV Reaction buffer (1 M Tris-HCl, pH 8.0, 10 mM EDTA)

**Storage Buffer:** 25 mM Tris-HCl pH 7.5, 200mM NaCl, 0.5 mM EDTA, 5mM DTT, 50% glycerol.

**Storage instructions:** -70°C for long term storage and -20°C for < 6 months. Always store TEV protease at -20°C and return to -20°C immediately after use. If precipitation occurs, spin at 4°C at full speed for 5 minutes in a bench top centrifuge to collect the precipitate and use the cleared supernatant for your experiment.

# Example of a time course experiment where the amount of TEV and temperature are held constant is presented:

Add the following components to a microcentrifuge tube:

Fusion Protein: 20 µg 20X TEV Buffer: 5 µl 0.1 M DTT: 1 µl

TEV Protease: 2 µl (10 units)

Water up to 100 µl

Incubate at 30°C. Remove 20µl aliquots at 1, 2, 4 and 6 hours. Check in SDS-PAGE.

(Recommended Time for digestion: 6hrs at 30°C or overnight at 4°C).

#### Notes:

 If the fusion protein sample contains >2 M urea, >0.5 M Guanidine hydrochloride, >50 mM imidazole, with pH values below 6 or above 9, or cysteine protease inhibitors, then it will be necessary to dialyze the fusion protein into TEV protease reaction buffer before TEV Protease cleavage.

TEV protease is inhibited by reaction buffers containing >40% Glycerol.

Expiry: After 6 months upon receiving at proper storage condition as mentioned in datasheet.