

Product Information

Order: # PR-ETA20010

HRV3C PROTEASE (TURBO3C), RECOMBINANT

Description

Human rhinovirus 3C protease (HRV3C Protease) is a cysteine protease that recognizes the cleavage site of Leu-Glu-Val-Leu-Phe-Gln-Gly-Pro, commonly referred to as the PreScission Site. It cleaves between Gln and Gly. The recombinant form of the HRV3C protease is a restriction grade protease that has robust activity at 4 °C with high specific activity and great stability. It does not require any special buffer for its activity and can be used in a buffer most suitable for the target protein. This HRV3C Protease is a 47 kDa protein with both GST and His tags so it can be easily removed by either Ni-chelating or Glutathione (GSH) resin along with the cleaved tag.

Component and Formulation

HRV3C Protease: 2 mg/ml in 25 mM Tris-HCl, pH8.0, 50 mM NaCl, 1 mM TCEP, and 50% glycerol

Storage and Stability

Store HRV3C Protease at -20 °C. HRV3C Protease is stable at room temperature for at least two week without loss of any activity. It retains full activity after incubation at 37 °C for one week.

Activity and Specificity

The activity of HRV3C Protease is tested using a control target protein. One (1) µg of HRV3C Protease has at least 1 unit activity conventionally used by other suppliers (1 unit of HRV3C protease cleaves >95% of 100 µg of control target protein at 4 °C for 16 hours). No non-specific activity has been observed under the same condition with HRV3C Protease to control target protein ratio of 1:10. Prolonged incubation (several days) under the same condition does not show any non-specific cleavage.

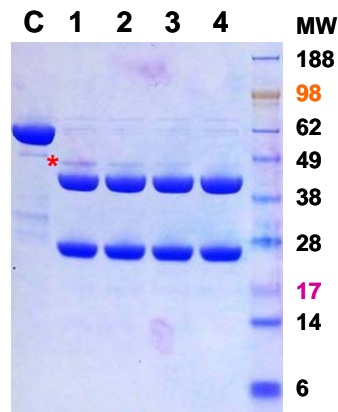


Figure 1

A 68 kDa GST-fusion protein (lane C) at 1 mg/ml is incubated with HRV3C Protease (*) at a ratio of (lane 1) 1:50, (lane 2) 1:100, (lane 3) 1:200, (lane 4) 1:400 (w/w) in a buffer of 25 mM Tris-HCl, pH8.0, 150 mM NaCl, 14 mM β-mercaptoethanol at 4 °C for 16 hours. The cleaved products are 42 kDa and 26 kDa.

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Cleavage Condition

It is recommended to use HRV3C Protease at a protease-to-target protein ratio of 1:100 (w/w) or 1 unit of HRV3C Protease to 100 µg of target protein in a buffer suitable for the target protein at 4 °C overnight, with the target protein concentration at 1-2 mg/ml. In most cases, target proteins are completely cleaved with a protease-to-target protein ratio of 1:50 to 1:400 or 1 unit HRV3C Protease to 50-400 µg of target protein (as shown in Figure 1). The efficiency of cleavage may vary due to the sequences around the cleavage site, the conformation and the solubility of the target protein. Due to its high specificity, more HRV3C Protease (at 1:10 ratio) or longer cleavage time (over a weekend) at higher temperature (37 °C) can be used to achieve high cleavage efficiency without non-specific cleavage of target proteins.

Removal of HRV3C Protease after Cleavage

The HRV3C Protease contains both GST and His tags. After cleavage of the target protein, HRV3C Protease can be easily removed along with the tags from the cleavage reaction by affinity chromatography on a Ni-chelating resin for His-tagged target protein or GSH resin for GST-tagged target protein.

Order Information & Storage

Order #	Product	Amount
PR-ETA20010-01	HRV3C Protease (Turbo3C), recombinant, GST- & His-tag	1 mg (1.000 U)
Store at -20 °C		