

Recombinant Enterokinase

Cat.NO.:SGZ 01003

PRICE:EUR64.00/100U

DESCRIPTION

Enterokinase (EK) is a specific protease that cleaves a protein after lysine residue at its recognition site: Asp-Asp-Asp-Asp-Lys. Enterokinase will not work if the recognition site is followed by proline. If a fusion tag is placed at the N-terminus with an enterokinase site, enterokinase will be able to remove the fusion tag and to generate the protein exactly as you need without adding any unwanted residues. ShineGene **Enterokinase, Light Chain, Porcine** is a highly purified recombinat porcine enterokinase (Light Chain) expressed from *P. Pastoris* GS115.

COMPONENTS

100 U Recombinant Enterokinase, 2 U/μl, 50 μl in all.
2 ml 1X EK Dilution/Storage Buffer
1 ml 10X EK cleavage buffer
10 μg Cleavage control protein, lyophilized powder on the bottom of the tube.

MOLECULAR WEIGHT

Theoretical MW: 21,880 Da; The apparent MW on SDS-PAGE: about 40,000 Da.

FORMULATION

ShineGene Enterokinase, Light Chain, Porcine has been formulated using a proprietary technology, and the enzyme can be shipped at room temperature or stand at 37°C for 7 days without losing any activity.

UNIT DEFINITION

One unit is defined as the amount of enzyme needed to cleave 50 μg of fusion protein in 16 hours to 95% completion at 22°C in a buffer containing 25 mM Tris-HCl, pH 8.0.

GENERAL PROTOCOL

Preliminary small scale digestion is recommended in order to find an optimal cleavage condition and enzyme:target protein ratio. Scale up the reaction using the optimized conditions.

1. Make a serial dilution of Enterokinase (2 U/μl) in 8 tubes so that there is 1, 0.1, 0.01, 0.001, 0.0001, 0.00001, 0.000001 and 0 U enzymes in 8 different tubes, respectively.
2. Set up the digestion reactions in the 8 labeled tubes with 50 μg of protein in a total volume of 50 μl per tube.
3. Incubate the reactions at room temperature (20°C - 22°C) for 16 hours.
4. 10 μl from each tube is loaded on a SDS-PAGE to determine the extent of cleavage. (See Fig.1.).

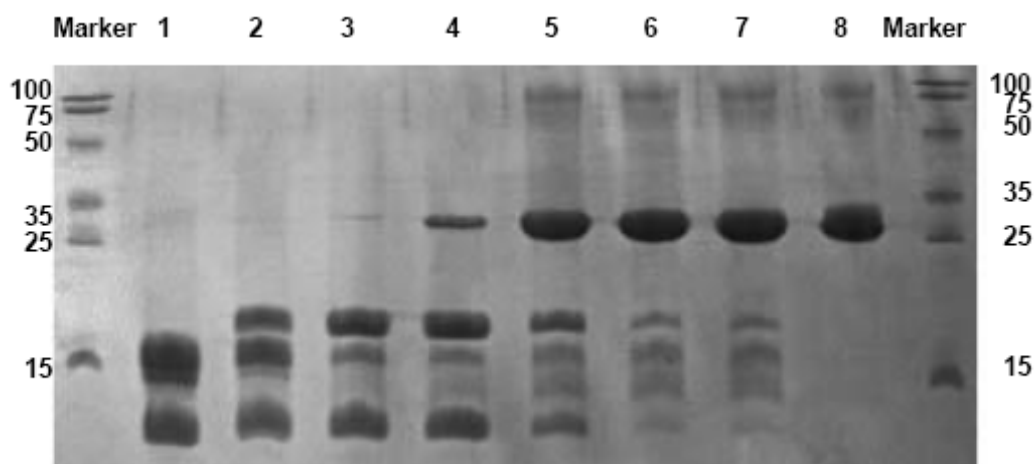


Fig.1. Each reaction contains 50 µg of purified IL-8 fusion protein and varying amount of ShineGene rPorcine Enterokinase. The reactions were incubated at 22°C for 16 hours and analysed on a coomassie-stained SDS-PAGE (15%). Units of enzyme used per reaction is listed below:

Lane 1.:1 U	Lane 2: 0.1 U	Lane 3: 0.01 U	Lane 4.:0.001 U
Lane 5: 0.0001 U	Lane 6: 0.00001 U	Lane 7: 0.000001 U	Lane 8: 0 U

STORAGE

Store at -20°C after receiving.

For Research Use Only!



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