RapidEK™ (RECOMBINANT ENTEROKINASE)

www.parasbiopharma.com

Paras Biopharmaceuticals Finland Oy

What is Protein Purification

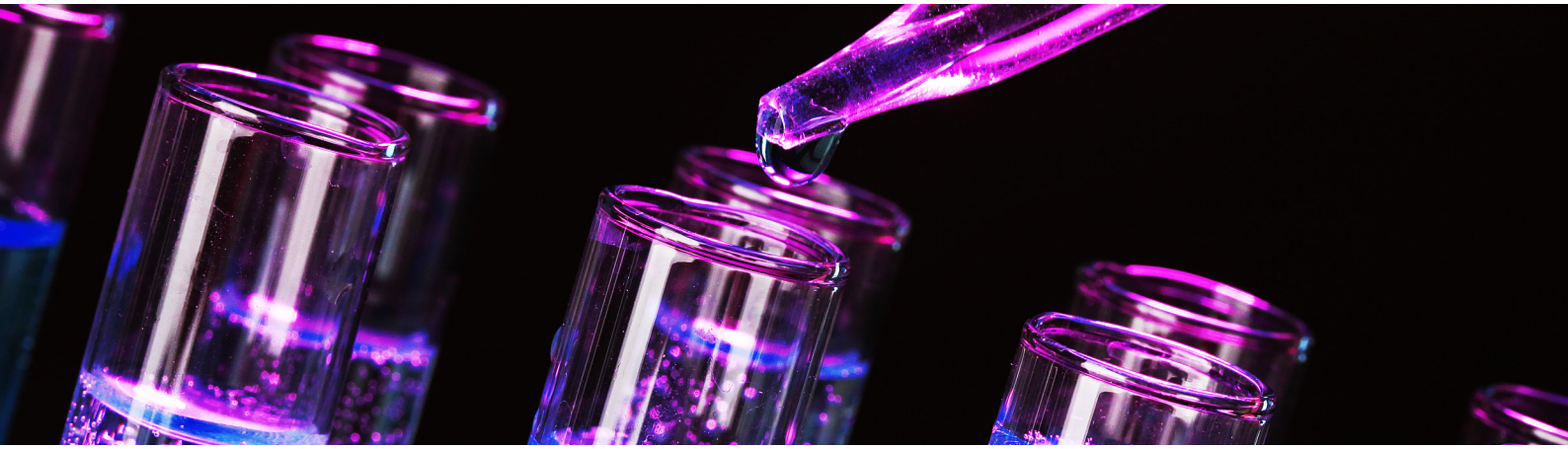
Protein purification is a series of processes intended to isolate one or more proteins from a complex structure. It is vital for the characterisation of the function, structure and interactions of the protein of interest.

One effective technique is the tagging of proteins to engineer an antigen peptide tag onto a protein, and then purify the protein through a column. Once finished, the tag can be cleaved from the protein by a protease.

What are Protein Tags?

Protein tags are peptide sequences genetically grafted onto a recombinant protein which are often removable by chemical agents or by enzymatic means. One such example are affinity tags which are appended to proteins so that they can be purified from their crude biological source using an affinity technique.

Enterokinase can be used to cleave a fusion protein containing a C-terminal affinity tag to produce a target protein following protein purification.



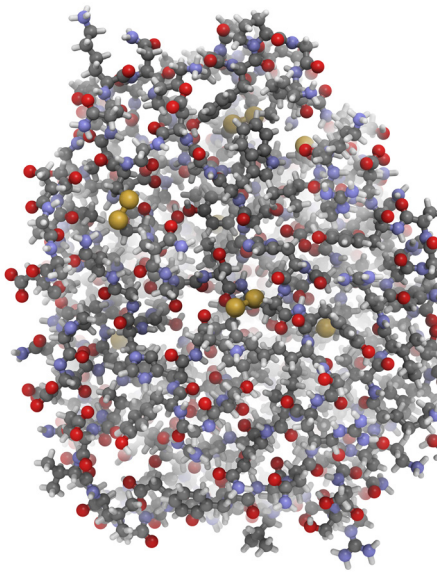
Recombinant Enterokinase

Enterokinase is a type 2 transmembrane serine protease produced by cells in the duodenum which are involved in the body's digestive system. It is responsible for the initial activation of pancreatic proteolytic proenzymes that catalyses the conversion of trypsinogen to trypsin. **Enterokinase's specificity makes it an ideal tool for biotechnological and biochemical removal of fusion proteins and tags.**

- As a human enzyme, enterokinase is 1019 amino acids in length containing 14 disulfide bonds, 18 potential N-glycosylation sites and a N-myristoyl lipdation site as the N-terminus.
- Enterokinase's catalytic light chain is 235 amino acids in length and contains 4 disulfide bonds.
- Specifically cleaves the bond between Lys23 and Ile24 in human trypsin-1 and the equivalent peptide bond in other trypsin family members.
- Enterokinase deficiency is a life threatening intestinal malabsorption disorder.

Molecular weight of enterokinase varies from 82 - 140 kD (113 kD protein core) and 35 - 62 kD for its disulfide-linked heavy-chain and light-chain, respectively, depending on the organism.

DISCOVERED BY NOBEL PRIZE WINNER IVAN PAVLOV, IT IS THE FIRST KNOWN ENZYME TO ACTIVATE OTHER ENZYMES.



Action

• RapidEK™ cuts the following sequence

Asp-Asp-Asp-Asp-Lys - X - X - X - X

• It has minimal requirements for specific amino acids in the P1' and P4' positions*

* Natural substrates have a propensity for Gly or Ser at P3' and/or P4' to ensure regular secondary structure formation does not limit accessibility to the cleavage site.

• This independence makes RapidEK™ ideal for the removal of fusion proteins and/or tags with the subsequent generation of an authentic N-terminus.

• Many companies monitor activity by following the cleavage of a protein. The fusion partners used vary widely.

• The assay conditions employed also vary widely and so, the units of activity are often defined differently.

• Paras uses one of the most credible industry standard methods for recording the activity of the enzyme.

RAPIDEK™
PRODUCT SUMMARY

As a type 2 transmembrane serine protease, enterokinase is ideally suited for the biotechnological and biochemical removal of fusion proteins and cleavage of tags after purification. Produced by cells, enterokinase is responsible for the initial activation of pancreatic proteolytic proenzymes that catalyses the conversion of trypsinogen to trypsin. This means enterokinase is a very important enzyme in the biopharma and nutrition industry.

- BRAND NAME: RapidEK™
- ACTIVE INGREDIENT: Recombinant Enterokinase (also known as Enteropeptidase)
- DESCRIPTION: Type 2 transmembrane serine protease
- TARGET GROUP: Nutrition Industry and for the removal of fusion proteins and tags
- PRODUCT PATENT EXPIRY: No patents exist

Paras Biopharmaceuticals' biologics candidates / technologies under development

- Biosimilar candidate to Forteo® (Teriparatide)
- Biosimilar candidate to Kineret® (Anakinra)
- Biosimilar candidate to Novolog® (Insulin Aspart)
- Biosimilar candidate to Nplate® (Romiplostim)
- Biosimilar candidate to Elitek® (Rasburicase)

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