

## 41. Title: Production of recombinant serratiopeptidase protein using *E. coli* as an expression system

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**Keywords:** Serratiopeptidase, Proteolytic

**Domain:** Life Sciences (Biotech)

### Summary:

Serratiopeptidase, a proteolytic enzyme, has many clinical and biotechnological applications such as active ingredient in drug combinations, treatment of several diseases, health supplement to prevent morbidity, additive in detergents, leather and paper industry, food processing, activity against biofilm formation, amyloid degradation, etc. Serratiopeptidase is produced using wild and mutant strains of enterobacterium *Serratia marcescens* ATCC 21074. This approach is source organism dependent and has narrow scope of optimization, hence limiting the yield. The organism is also an opportunistic pathogen. Recombinant expression of serratiopeptidase is a viable solution but has various difficulties in terms of poor expression levels, stability issues and inactive protein due to inclusion bodies. Also, heterologous expression of proteases causes critical cellular stress leading to toxicity and death of host cells. Purifying proteins from inclusion bodies is a tedious job and adds to the cost.

The invention relates to an optimized recombinant overexpression methodology for high-level expression of serratiopeptidase in *E. coli*. The enzyme is initially expressed in its non-toxic, inactive, zymogenic version which is segregated into the inclusion bodies resulting in proper cell growth and survival of the recombinant host cells. The protocol is optimized with respect to various physicochemical parameters such as nutrient composition, induction point, inducer concentration, temperature, post-induction duration, etc., leading to significant enhancement in yields. The inclusion bodies are solubilized and the protein is processed and purified to obtain the mature active enzyme.

### Advantages:

- » *E. coli* is the most convenient and well-studied host system; Production is simple, less time consuming and cost-effective; Simple down-stream processing
- » By using *E. coli*, health hazards associated with *S. marcescens* are overcome
- » High yield of functionally active protein
- » Product is biosimilar and compatible to the commercial / wild type serratiopeptidase

**Applications:** Pharma and drugs

**Scale of Development:** Lab-scale production with high protein yield achieved, biocompatibility studies done

**Technology Readiness Level:** 4

**IP Status:** Indian Patent Application 201911032068, Indian Patent Application 201811029173