

Purification and Partial Characterization of Human Pancreatic Elastase

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Summary:

1) An elastolytic enzyme has been isolated from human pancreatic juice. The concentration of the enzyme was about 0.6 mg/ml corresponding to about 10% of the protein concentration of the juice.

2) The purification procedure included chromatography on Sephadex G-25 followed by ion-exchange chromatography on SP-Sephadex C-50 at pH 6.0, affinity chromatography on Trasylol-Sepharose 4-B and a final ion-exchange chromatography on SP-Sephadex at pH 7.6. The yield was about 50%.

3) The elastase isolated was homogeneous in analytical disc electrophoresis and showed a single protein component with a molecular weight of 26 300 in dodecylsulfate-electrophoresis.

There are similarities in the amino acid compositions of human and porcine pancreatic elastases.

4) The human enzyme has a lower activity on elastin than porcine elastase but similar activities on casein and fibrin. The K_m value for Boc-Ala-ONp was $5.13 \times 10^{-4} M$. The elastase isolated was 98% active, as judged from active site titration.

5) The results of immunodiffusion studies of activated pancreatic juice and duodenal juice with specific elastase-directed antibodies indicate that the purified enzyme was the single elastolytic enzyme present.

6) It is a cationic protein without any carbohydrate. The activation of the pro-elastase resulted in the formation of an active enzyme with a higher isoelectric point than the zymogen.

Reinigung und partielle Charakterisierung der Elastase aus menschlichem Pankreas

Zusammenfassung:

1) Aus menschlichem Pankreassaft wurde ein elastolytisch wirksames Enzym isoliert. Die Konzentration dieses Enzyms im Pankreassaft betrug etwa 0.6 mg/ml was einem Anteil von etwa 10% am Proteingehalt des Saftes entspricht.

2) Die Isolierung des Enzyms erfolgte durch Chromatographie an Sephadex G-25, SP-Sephadex C-50 bei pH 6.0, Trasylol-Sepharose 4-B (Affinitätschromatographie) und schließlich SP-Sephadex bei pH 7.6. Die Gesamtausbeute lag bei 50%.

Enzyme:

Elastase (EC 3.4.21.11).

Abbreviations:

Ac- = acetyl-; Boc- = tert.-butyloxycarbonyl-; -NH-N(CH₃)CO-ONp = 2-methyl-2-(p-nitrophenoxycarbonyl)hydrazide; -NHnp = p-nitroanilide; Suc- = 3-carboxypropionyl-.