SUMMARY OF THE WORK

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1. An enzyme preparation, containing proteolytic activity, has been made from the bladders of Utricularia aurea Lour., a species of insectivorous plant, by homogenization with sea sand, in presence of cysteine and phosphate buffer, 0.02 M pH 7.0.

2. The impurities and cell debris have been eliminated from the homogenate by adjusting the pH at 9.0.

3. The supernatant, obtained from the pH adjusted homogenate, has been fractionated by ammonium sulphate at 40% and 40-80% saturations, and the collected precipitates have been called Fraction I and Fraction II, respectively.

4. The proteolytic activities of the crude homogenate and of the prepared fractions have been studied using human blood plasma as substrate. Fraction I has been found to be most active.

5. By varying the pH of the incubation medium it has been observed that the prepared Fraction I is active between pH ranges 3.5 to 8.0, and the same fraction is maximally active at pH 4.5.

6. By varying the pH of the incubation medium it has been observed that Fraction II is active between pH ranges 5.5 to 9.0, and has maximum activity at pH 8.0.

7. By varying the temperature of incubation, it has been observed that both the prepared Fractions I and II are maximally active at about 37 to 38^{o}.

8. While studying the effect of varying period of incubation on enzyme activity, it has been observed that Fraction I is maximally active between 60-120 minutes.

9. While studying the effect of varying period of incubation on enzyme activity it has been observed that Fraction II is maximally active between 30-60 minutes.

10. Studies on thermal sensitivity of the prepared fractions have been made, and it has been observed that pre-incubation temperature treatment of both the fractions, at 40^{o} for 5 minutes, shows maximum proteolytic activities to both the fractions.

11. It has been observed that cysteine, Mg^{2+}, Zn^{2+}, BrO_3^- and KCN (1×10^{-3} M) activate both the fractions, Moreover, Mn^{2+} activates Fraction II.

12. It has been observed that Hg^{2+}, Ca^{2+}, CrO_3^{3-}, $As_2O_5^{3-}$ and PCMB (1×10^{-3} M) inhibit proteolytic activities of both the fractions.

13. A few synthetic substrates, like glycylglycine, carbobenzoxyglutamyltyrosine and benzoylglycinamide (0.25mM) are hydrolyzed by both the fractions. Moreover, leucylglycylglycine is hydrolyzed only by Fraction II, and benzoylglycine

is hydrolyzed only by Fraction I.

14. Benzoylglycinamide has been used as co-substrate with each of carbobenzoxyglutamyltyrosine, benzoylargininamide, glycylglycine and hippuric acid; Fraction I hydrolyzes all these co-substrates. Benzoylglycinamide-leucylglycylglycine co-substrate is not hydrolyzed by Fraction I. Carbobenzoxyglutamyl-tyrosine has been used as co-substrate with each of glycylglycine and leucylglycylglycine; Fraction I hydrolyzes both these co-substrates. But, carbobenzoxyglutamyltyrosine when used as co-substrate with each of hippuric acid and benzoylargininamide, is not hydrolyzed by Fraction I. Leucylglycylglycine has been used as co-substrate with each of benzoylargininamide, glycyl-gycine and hippuric acid; Fraction I hydrolyzes all these co-substrates. But benzoylargininamide when forms co-substrate with each of carbobenzoxyglutamyltyrosine, glycylglycine and hippuric acid, is not hydrolyzed by Fraction I.

15. Carbobenzoxyglutamyltyrosine has been used as co-substrate with each of benzoylglycinamide, glycylglycine, leucylglycylglycine and hippuric acid; Fraction II hydrolyzes all these co-substrates. Benzoylargininamide has been used as co-substrate with each of benzoylglycinamide, hippuric acid and leucylglycylglycine; Fraction II hydrolyzes all these co-substrates. But benzoylargininamide when used as co-substrate with any of carbobenzoxyglutamyltyrosine and glycylglycine, is not hydrolyzed by Fraction II. Leucylglycylglycine has been used as co-substrate with each of hippuric acid, glycylglycine and benzoylargininamide; Fraction II hydrolyzes all these

co-substrates. Benzoylglycinamide when used as co-substrate with any of leucylglycylglycine, glycylglycine and hippuric acid, is not hydrolyzed by Fraction II.

16. Further purification of the Fractions I and II has been done by ECTEOLA-cellulose column chromatography. Four peaks have been obtained by fractionating each of Fractions I and II at 0.005 M, 0.1 M, 0.5 M, 1.0 M and 0.02 M, 0.05 M, 0.5 M, 1.0 M, buffers of pH 4.5 and 8.0, respectively.

17. The effluents of Fraction I at 0.005 M, and that of Fraction II at 0.5 M buffer concentrations, have shown maximum proteolytic activities towards haemoglobin as substrate.

18. Histochemical studies have been done with the bladders of Utricularia aurea, using azo-dye staining methods of Nachlas et al., and Gomori et al. The tufts of 3-5 pronged hairy glands, situated on the inner walls of bladders are stained reddish pink indicating the site of proteolytic activity.