Jirapa Hinsui 2009: Purification, Characterization and Application of Trypsin and Chymotrypsin from Viscera of Nile Tilapia (*Oreochromis niloticus* Linneaus). Doctor of Philosophy (Fishery Products), Major Field: Fishery Products, Department of Fishery Products. Thesis Advisor: Associate Professor Wanchai Worawattanamateekul, Ph.D. 146 pages.

Nile tilapia enzyme was extracted with 0.05 M Tris-HCl, contained 0.5 M NaCl and 0.02 M $CaCl_2$ in ratio 1:5 (w/v) at 4 °C. Spleen was found to be the best source for trypsin with the highest activity at 1.68 units/ ml, followed by intestine, liver, mixed viscera and stomach, respectively. The best source for chymotrypsin was intestine, giving the highest activity at 0.17 units/ ml, followed by spleen, mixed viscera, liver and stomach, respectively. Specific activity of intestine trypsin (0.17 U/ mg protein) is closer to spleen trypsin (0.19 U/ mg protein). The quantity of intestine is more than spleen, so intestine was selected source for enzyme extraction. The enzymes were purified by ammonium sulfate precipitation, followed by chromatography columns. There were 4 enzyme fractions in hydrochloric acid fraction, S711, S712, S721 and S722. The highest concentration of enzymes was S711. The molecular weight was estimated to be 26,000 Da by SDS-PAGE. Enzyme activity and stability were in pH range of 7.0 -10.0. The optimum temperature was 70 °C. It was unstable at the temperatures higher than 50 °C. The activity was inhibited by serine protease inhibitor (PMSF and aprotinin), trypsin inhibitor (SBTI and TLCK) and chymotrypsin inhibitor (TPCK). It showed that it should be trypsin-like enzyme. There were 4 enzyme fractions in Tris fraction, S511, S512, S521 and S522. The highest concentration of enzymes was S521. The molecular weight was estimated to be 51,800 Da. The pH and temperature optimum was 9.0 and 70 °C, respectively. It was stable in pH range of 7.0 -10.0, but it was unstable at the temperatures higher than 60 °C. The activity was inhibited by PMSF and aprotinin, SBTI and TPCK. It showed that it should be chymotrypsin-like enzyme. Nile tilapia trypsin was used to extract carotenoprotein from head of giant freshwater prawn at pH 8.0 and 25 °C, optimum condition for enzyme, then found that 3 hours was suitable time to extract, giving the highest yield (54.02%) , followed by bovine trypsin (48.3%) and no enzyme (45.76%), respectively.

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