PEMANFAATAN TUMBUHAN *FICUS* SEBAGAI SUMBER ENZIM FISIN DAN APLIKASINYA PADA PRODUK PANGAN

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Disertasi

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Summary

Ismed. Utilization of Ficus Plants as a Source of Ficin Enzyme and Its Application in Food Products. Supervised by Rina Yenrina, Hasbullah, Daimon Syukri, and Yusniwati.

Ficus spp. are plants in the form of trees, shrubs, or bushes commonly found in tropical and subtropical regions. In Indonesia, Ficus species are distributed across several islands including Sumatra, Kalimantan, Papua, and Sulawesi. In West Sumatra, 20 species of Ficus have been identified in conservation forests, with the Padang Aro area of South Solok also known for its local name associated with Ficus. The latex of Ficus plants contains a cysteine protease enzyme called ficin. Understanding the characteristics and purification process of ficin plays a crucial role both scientifically and practically, particularly in the development of biotechnology and its application in the food industry. This study aims to: (a) obtain information on the characteristics of crude ficin enzymes from Ficus spp.; (b) isolate ficin enzymes using the Three Phase Partitioning (TPP) method optimized through variations in salt concentration (ammonium sulfate), crude enzyme extract, organic solvent (tert-butanol), pH, and temperature; (c) determine ficin characteristics including pH, temperature, cofactors, activators, inhibitors, enzyme kinetics, enzyme and substrate concentration, molecular weight, and drying methods; (d) apply ficin in meat tenderization and milk-clotting processes. The research employed a qualitative approach with descriptive and experimental methods using a completely randomized design (CRD), response surface methodology (RSM), and a factorial design. Key findings include: (a) Crude ficin enzymes extracted from the fruit and latex of *Ficus aurata* (Miq.), *Ficus padana* Burm.f., and *Ficus racemosa* L. showed total enzyme activity between 7.35–24.01 U, total protein content ranging from 1.64–4.83 mg, and specific activity between 1.87–5.41 U/mg. Latex generally demonstrated higher catalytic activity. Partial purification using 60% ammonium sulfate and dialysis increased specific activity of ficin from *Ficus aurata* latex. Proximate analysis revealed high protein content in *Ficus aurata* fruit. (b) The Three Phase Partitioning (TPP) method optimized by RSM recovery the best conditions at 60% ammonium sulfate saturation and a tert-butanol to enzyme ratio of 1:1.25, resulting in a specific activity of 33.33 U/mg. (c) Optimal purification was achieved at 40°C and pH 6.5, with a specific activity of 42.25 U/mg. TPP effectively enhanced both purity and activity, increasing specific activity from 4.62 U/mg to 42.25 U/mg, a recovery of 227.89%, and a 9.15-fold purification. Further analysis showed that cysteine significantly increased enzyme activity, whereas $MgCl_2$ and $CaCl_2$ inhibited it at high concentrations. $MnSO_4$ enhanced activity in a concentration-dependent manner, while KCl increased total activity at low concentrations but reduced specific activity at higher concentrations. EDTA acted as an inhibitor, and $CuSO_4$, $FeSO_4$, and $BaCl_2$ increased activity at low concentrations but inhibited it at high levels, with $BaCl_2$ showing the weakest inhibitory effect. Substrate testing showed that casein significantly enhanced enzyme activity, while gelatin, ISP, and BSA produced lower activity. Ficin was more effective in hydrolyzing casein due to its

compatible amino acid composition. Enzyme kinetics indicated that total activity increased with substrate concentration, peaking at 0.3 mM, while specific activity remained constant at around 40 U/mg. Freeze drying was more effective than solar drying in preserving amino acid content and enzyme quality. SDS-PAGE analysis showed a protein band around 35 kDa, suggesting the presence of proteolytic fractions. (d) In meat tenderization, higher temperatures reduced protein content and water holding capacity (WHC), whereas lower temperatures with higher ficin concentrations improved WHC and meat tenderness. The best results were achieved at 80°C with 5% ficin, producing tender meat with optimal texture, although high temperatures reduced brightness and increased redness. In milk-clotting applications, 40°C was the optimal temperature, recovering the fastest coagulation time and highest clotting activity. This study also highlights the potential of Ficus species as conservation plants capable of preventing erosion and rehabilitating marginal lands, making them relevant for application in tropical agroforestry systems to support sustainable agriculture.