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Partial Purification of Hen Egg White Lysozyme by Ethanol Precipitation Method and Determination of the Thermal Stability of Its Lyophilized Form

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Abstract: Lysozyme was partially purified from hen egg white by precipitation of nonlysozyme protein impurities during incubation in the presence of ethanol. The thermal stability of the obtained partially purified enzyme was also characterized. The incubation of diluted egg white for 2-8 h in the presence of 20% ethanol was not very effective for the partial purification of lysozyme by precipitation of major egg white proteins; however, 4- to 6-h or 6- to 8-h incubation of diluted egg white in the presence of 30% and 40% ethanol could be employed more effectively for partial purification of lysozyme. Without applying the incubation period, the highest specific activity was obtained by the treatment of egg white with 40% ethanol. Thus, ethanol at this concentration could be used for a continuous process of partial purification. For batch lysozyme purification, on the other hand, incubation in the presence of 30% ethanol was more appropriate. The activities and protein contents of dialyzed and lyophilized enzymes obtained by 6 h-incubation in the presence of 20%, 30%, and 40% ethanol precipitations were 1878, 6669, and 6115 U/mg powder, and 0.98, 0.90, and 0.93 mg protein per mg powder, respectively. The ranges of thermal inactivation parameters, such as D (D_{80 °C} = 29.2-59 min, D_{90 °C} = 8.8-21 min) and z ($z_{80-90 °C}$ = 17.4-22.3 °C) values of the enzyme, clearly indicated the moderate and variable heat stability of lyophilized lysozymes obtained from different batches of egg white.

Key Words: Lysozyme, hen egg white, partial purification, ethanol precipitation, thermal stability

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