Purification and Partial Characterization of Ostrich Muscle Collagenases.

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Collagenases are a group of collagen degrading enzymes which belong to a family of calcium- and zinc- dependent endopeptidases called the matrix metalloproteinases (MMPs). The main function of MMPs is degradation of the extracellular matrix and connective tissue components. Collagen has been reported to contribute largely to background toughness of connective tissue (skin, tendon, blood vessels, bone, etc) in the mammalian muscle (1); hence its degradation may lead to meat tenderization. Collagenases can be obtained from both eukaryotes and prokaryotes in latent forms and they catalyze the cleavage of triple-helical portions of the classical types of collagen. These enzymes have found a widespread application in the isolation of specific cell types from attendant connective tissue. Since these enzymes are released by specific cells involved in remodeling processes as zymogens, they can be activated *in vitro* by a variety of means, proteolytically and non-proteolytically.

Since collagenases have not been studied in the ostrich, these enzymes have been isolated from ostrich muscle by extraction and subsequently purified by ammonium sulfate precipitation (25-55% saturation), followed by DE-52, hydroxyapatite, and Superdex G-200 column chromatography. Aminophenylmercuric acetate was used to activate the latent enzyme after each purification step and assayed for activities using a fluorogenic synthetic peptide substrate. The purified enzyme will be characterized by N-terminal amino acid sequencing and will be tested for pH and temperature optima. Finally, its contribution to meat tenderization will also be investigated.

[1]Sylvestre, M.N., Balcerzak, D., Feidt, C., Baracos, V.E., and J. Bellut.(2002). Elevated rate of collagen solubilization and post-mortem degradation in muscle of lambs with high growth rates: possible relationship with activity of matrix metalloproteinases.*J. Anim. Sci.* 80, 1871-1878.