Partial Purification and Enzymatic Characterization of Hyaluronidase from Crotalus durissus collilineatus Snake Venom

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Abstract: Ophidic accidents represent a serious health problem in tropical countries, where south rattlesnakes, known by Crotalus genus in Brazil, are responsible for the highest indexes of lethality. This kind of poisoning is characterized mainly by systemic effects, as acute renal and respiratory failures, neurotoxicity, cardiotoxicity, myotoxicity and hemorrhage. Hyaluronidase is a non toxic enzyme known as "spreading factor", because it degrades the extra cellular matrix and, at the same time, improve the diffusion of the other toxins during the poisoning. This work shows the partial purification of hyaluronidase from Crotalus durissus collilineatus snake venom. The hyaluronidasic activity of crude venom and fractions was assayed by turbidity and zymography. The crude venom showed high specific activity of approximately 1850 turbiditmetric reducing units (TRU/mg). Only 5 µg of crude venom was able to hydrolyse hyaluronan by zymography, presenting an apparent molecular weight of 65 kDa. Hyaluronidase enzyme was partially purified by an ion-exchange chromatography on CM-Sepharose C-25 with a yield of purification of 0.21%. It was obtained 3 proteic peaks and hyaluronidase was found in the baseline in the end of the fractioning. A next step of purification is needed to isolate the enzyme, which can enable further investigations about its function and structure.

Keywords: Crotalus durissus collilineatus, hyaluronidase, spreading factor.

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