Purification and Partial Characterization of a New Acidic Phospholipase A₂ from Bothrops leucurus Snake Venom

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Phospholipases A₂ are enzymes defined by the ability to catalyze the hydrolysis of the sn-2-acyl bond of glycerophospholipids releasing free fatty acids and lysophospholipids. In this work an acidic phospholipase A₂ was isolated from Bothrops leucurus snake venom and its enzymatic and pharmacological characteristics were determined. It was highly purified through by two chromatographic steps on CM-Sepharose and Phenyl-Sepharose CL-4B. The purity assay of the enzyme was determined by PAGE-SDS analysis and reverse phase chromatography on Schimadzu C18 column. The enzyme was designated by BI-PLA₂ and showed a single chain protein of 16,3kDa and revealed high homology with others Asp49 acidic PLA₂s from snake venoms. The phospholipase A₂ activity of BI-PLA₂ was determined upon egg yolk emulsion, which contains phosphatidylcholine as substrate and by indirect hemolysis method, using washed mice erythrocytes and hen's egg-yolk emulsion as substrate. It displayed high phospholipase A_2 activity (158.7 U/mg) when compared with crude venom (69.3 U/mg). On the other hand, indirect hemolysis was not so expressive. It was also able to induced low myotoxic and edema, corroborating with majority of the acidic PLA₂s isolated from bothropic snake venoms, which showed low toxicity. Thus, Bl-PLA₂ becomes a good target for studies on cytotoxic potential, opening prospects for its use in developing new drugs, but further studies are necessary.

Keywords: acidic phospholipases A₂; *Bothrops leucurus*; snake venom.

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