EXPRESSION OF A FUNCTIONAL HOMOLOGUE OF A PHOSPHOLIPASE A2 INHIBITOR FROM THE BLOOD PLASMA OF THE SOUTH AMERICAN RATTLESNAKE

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Inhibitors of phospholipases A₂ (PLIs) are commonly found in the blood plasma of snakes with the primary role of protection against the toxic action of their own venom. Considering that phospholipases A₂ participate in a series of physiopathological processes in humans, including envenoming by snake bites, PLIs may provide interesting models to the development of new therapeutical drugs. This potentiality, however, has been underexplored due to insufficient amounts of native material. In order to check for the viability of obtaining functional recombinant homologues of PLIs in bacteria, we started by expressing CNF, a PLI that is present in the plasma of C. d. terrificus. Native CNF is an oligomer formed by a mixture of glycosylated and non-glycosylated monomers with identical protein structure. The cDNA encoding for CNF monomer was directionally subcloned into pCP vector and protein expression was induced in transformed bacteria. Recombinant CNF (CNFr) was obtained as a fusion protein with a ZZ domain which allowed a first-step purification by affinity chromatography in IgG-sepharose. CNFr was active as a PLI inhibitor suggesting that glycosylation may be not a prerequisite for the functionality of CNF.

KEY WORDS: PLA2 inhibitor, PLI, Crotalus durissus terrificus, CNF.

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