ENZYMATIC CHARACTERIZATION OF TWO ACIDIC PHOSPHOLIPASES A_2 ISOFORMS ISOLATED FROM *Lachesis muta* SNAKE VENOM

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In previous reports, we showed that phospholipases A_2, denoted LM-PLA_2-I and LM-PLA_2-II isolated from *Lachesis muta* snake venom showed indirect hemolytic activity, inhibition on platelet aggregation, edema and myotoxicity. Phospholipase A_2 (PLA_2) enzymes (EC 3.1.1.4) are able to hydrolyze phospholipids generating fatty acids and lysophospholips, that regulate cellular physiology. Here, we investigated which phospholipid substrates (phosphatidylcholine, phosphatidylserine, phosphatidylglycerol, phosphatidic acid) is better hydrolyzed by them as well as which lysophospholipids formed are important on platelet effects. We observed that both PLA_2s did not hydrolyze phosphatidylserine or phosphatidic acid. But, for phosphatidylcholine and phosphatidylglycerol activities with different potencies and affinities were achieved, 30 % of activity upon phosphatidylglycerol compared to phosphatidylcholine hydrolysis. Both enzymes required Ca^{++} for activity and the replacement of it by other divalent cations reduced PLA_2 activity. Only when enzymes were mixed with phosphatidylcholine, they inhibited collagen-induced platelet aggregation, in spite of enzymes hydrolyzed phosphatidylglycerol. The commercial lysophosphatidylcholine inhibited platelet aggregation whereas lysophosphatidylglycerol did not. As expected, enzymes treated with p-bromophenacyl bromide, the PLA_2 activity and inhibition on platelet were lost.
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