

Supporting Information

Catalytically Active Snake Venom PLA₂ enzymes: An Overview of Its Elusive Mechanisms of Reaction

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Table S1. The sequence identity matrix of six different sPLA₂ resulted from the multiple sequence alignment of a human synovial (UniProtKB AC: P14555), bovine pancreatic (UniProtKB AC: P00593), *B. asper* (UniProtKB AC: P20474), *E. carinatus* (UniProtKB AC: Q7T3S7), *N. atra* (UniProtKB AC: P00598) and *N. sputatrix* (UniProtKB AC: Q92085). Percent sequence identity matrix was created by ClustalW. ¹ Higher percentages are represented with darker colors.

Synovial (GIIA)	Pancreatic (GIB)	<i>B. asper</i> (GIIA)	<i>E. carinatus</i> (GIIA)	<i>N. atra</i> (GIA)	<i>N. sputatrix</i> (GIA)	
100	37	45	47	35	33	Synovial
	100	36	35	54	52	Pancreatic
		100	52	33	32	<i>B. asper</i>
			100	35	33	<i>E. carinatus</i>
				100	95	<i>N. atra</i>
					100	<i>N. sputatrix</i>

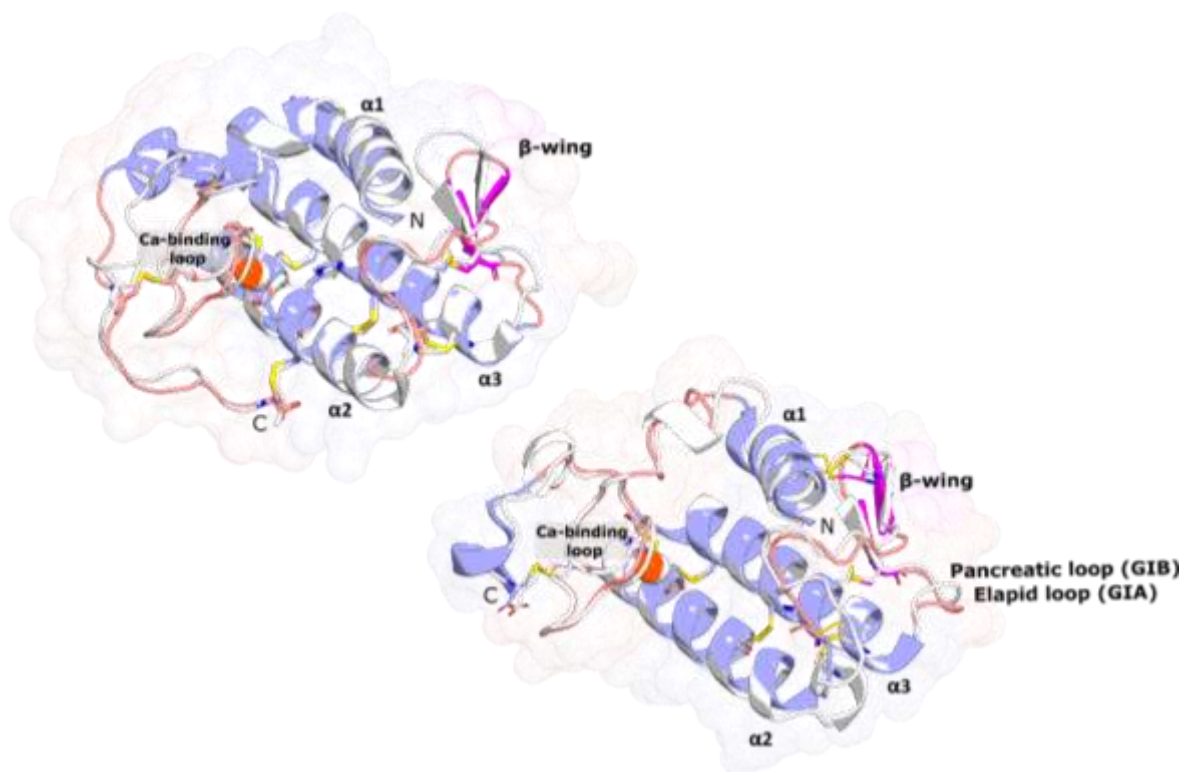


Figure S1. Cartoon representation of PLA₂ tertiary structure. (left) Superimposition of a bovine pancreatic (PDB 1MKV) (white) and snake venom GI-PLA₂ from the Chinese cobra (*Naja atra*) (PDB 1POA); (right) Superimposition of a human synovial (white) (PDB 1POE) and snake venom GII-PLA₂ from the Indian saw-scaled viper (*Echis carinatus*) (PDB 1OZ6). All PLA₂ have highly similar tertiary structures: Helices (lightblue), loops (salmon) and β -sheets (magenta) cartoon. The Ca²⁺ is show as an orange sphere and the disulfide bonds as yellow lines. The Ca²⁺-binding loop, the N- and C-terminal are also identified. The PyMOL molecular graphics software package was used to generate the representations.

Table S2. A selection of the reported X-ray crystallographic structures of snake venom, human, porcine and bovine GI/GII PLA₂.

Group	Source	Species	PDB	Ligands	Resolution	Ref.
GIA	Elapid snake venom	<i>Naja atra</i>	1POB	holo	2.00 Å	2
GIA	Elapid snake venom	<i>Naja atra</i>	1POA	apo	1.50 Å	3
GIB	Bovine pancreas	<i>Bos taurus</i>	1BP2	apo	1.70 Å	4
GIB	Bovine pancreas	<i>Bos taurus</i>	1MKV	holo	1.89 Å	5
GIB	Porcine pancreas	<i>Sus scrofa</i>	5P2P	holo	2.40 Å	6
GIIA	Human synovial fluid	<i>Homo sapiens</i>	1KVO	holo	2.00 Å	7
GIIA	Human synovial fluid	<i>Homo sapiens</i>	1POE	holo	2.10 Å	8
GIIA	Viperid snake venom	<i>Crotalus atrox</i>	1PP2	apo	2.50 Å	9
GIIA	Viperid snake venom	<i>Bothrops asper</i>	5TFV	apo	2.54 Å	10
GIIA	Viperid snake venom	<i>Gloydius halys</i>	1PSJ	apo	2.00 Å	11
GIIA	Viperid snake venom	<i>Echis carinatus</i>	1OZ6	apo	2.60 Å	12

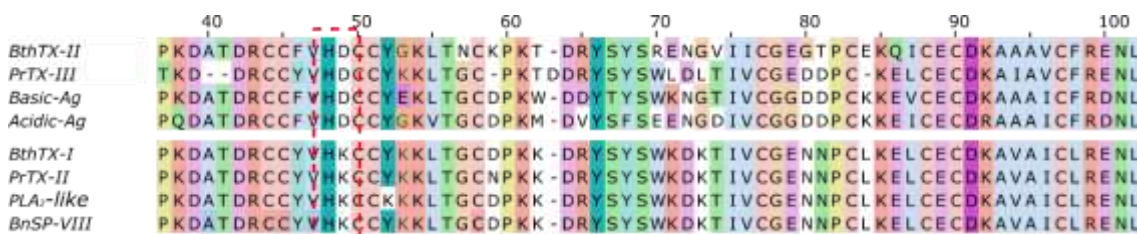


Figure S2. Sequence alignment of four classic PLA₂ (BthTX-II, PrTX-III, Basic-PLA₂ and Acidic-PLA₂ - UniProtKB ACs: P45881, P58464, O42187 and P14418, respectively) and four PLA₂-like proteins (BthTX-I, PrTX-II, PLA₂-like and BnSP-VIII - UniProtKB AC: Q90249, P82287, I6L8L6 and Q9IAT9, respectively). The replacement of the Asp residue by a Lysine at position 49 is highlighted in a red dashed box. High sequence identity is also evidenced. Amino acids are colored according to the ClustalX color scheme: hydrophobic (blue), positive charge (red), negative charge (magenta), polar (green), cysteines (pink), glycines (orange), prolines (yellow), aromatic (cyan) and unconserved (white). Created with Jalview 2.11 software.¹³

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