Evidences of venom metalloprotease in Pseudonaja textilis.

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Snake venom metalloproteases are of extreme relevance in snake envenoming, disrupting the endothelium integrity and affecting platelet aggregation and blood clotting. However, these toxins and their action are frequently neglected in accidents involving Elapidae, since the major symptoms involve mostly neurotoxicity. Preliminary data indicated the presence of a metalloprotease in the venom of Pseudonaja textilis, at the transcript level, but no further characterization has been performed to our knowledge. P. textilis metalloprotease transcripts were cloned from a venom gland cDNA library built using In-Fusion SMARTer cDNA library construction kit (Clontech). PCR reaction was performed using 20-mers primers, designed based on highly conserved, untranslated regions (UTR) from metalloprotease mRNA from Elapidae, Colubridae and Viperidae species. The presence of metalloprotease in the P. textilis venom was also checked by western-blot of crude venom and metal affinity purified toxins, using an anti-jararhagin policional antibody. Both assays were positive. The PCR enabled the detection of 2 separate bands from the P. textilis venom gland cDNA. Noteworthy is the fact that the same set of primers also enabled the amplification of transcripts from cDNA from Bothrops erythromelas, a remotely related species, suggesting that these primers might be used as universal probes for the detection of metalloproteases in snake venoms.

Word Keys: cDNA library, metalloprotease, PCR reaction, *Pseudonaja textilis*.

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