

MS/MS. In line with previous proteomic and transcriptomic analyses showing that snake venom proteins belong to only a few major protein families, the venom proteomes of *Bitis gabonica rhinoceros*, *Bitis nasicornis*, and *Bitis caudalis*, comprise, respectively, toxins from 11, 9, and 8 toxin families. Dimeric disintegrins, PLA<sub>2</sub> molecules, serine proteinases, a CRISP, C-type lectin-like proteins, L-amino acid oxidases, and snake venom metalloproteases are present in the three *Bitis* snake venoms, though they depart from each other in the composition and the relative abundance of their toxins. The venom composition appears to keep information on the evolutionary history of congeneric taxa. Protein similarity coefficients used to estimate the similarity of venom proteins of the *Bitis* taxa sampled here and in previous studies

(eg. *Bitis arietans* and *Bitis gabonica gabonica*) support the monophyly of the three West African taxa (*B.g. gabonica*, *B.g. rhinoceros*, and *B. nasicornis*) based on genetic distance reconstructions, the lack of alliances between *B. arietans* and any other *Bitis* species, and are consistent with the taxonomic association of *Bitis caudalis* within the differentiated group of small *Bitis* species. The low level of venom toxin composition similarity between the two conventionally recognized subspecies of *Bitis gabonica*, *B. g. gabonica* and *B. g. rhinoceros*, supports the consideration by some authors of *B. g. rhinoceros* as a separate species, *Bitis rhinoceros*. Moreover, our proteomic data fit better to a weighted phylogram based on overall genetic distances than to an unweighted maximum-parsimony tree.

## Snake venomics of Central American species from the *Atropoides* and *Bothriechis* genera

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Venoms produced by snakes of the family Viperidae (vipers and pitvipers) contain proteins that interfere with the coagulation cascade, the haemostatic system and tissue repair, and human envenomations are often characterized by clotting disorders, hypofibrinogenemia and local tissue necrosis. In addition to understanding how venoms evolve, characterization of the protein/peptide content of snake venoms also has a number of potential benefits for basic research, clinical diagnosis, development of new research tools and drugs of potential clinical use, and for antivenom production strategies. In addition, venom composition may retain information on its evolutionary history, and may thus have a potential taxonomical value. We report the venom composition of Central American snakes from *Atropoides nummifer*, *A. picadoi*, *Bothriechis lateralis* and *B. schlegelii* as part of a larger snake venomics project. Our results show that the 30-40 protein fractions collected from each venom correspond to molecules of molecular masses in the range of 1-110 kDa, which belong to only a few toxin families. The major toxins of *A. nummifer* be-

long to the PLA<sub>2</sub> (relative abundance, 36.5%) and the serine proteinase (22%) families, whereas the most abundant *A. picadoi* toxins are Zn<sup>2+</sup>-dependent metalloproteinases (66.4%). Their distinct venom toxin compositions provide clues for rationalizing the low hemorrhagic, coagulant, and defibrinating activities, and the high myotoxic and proteolytic effects evoked by *A. nummifer* snakebite in comparison to other crotaline snake venoms, and the high hemorrhagic activity of *A. picadoi*. On the other hand, the large degree of compositional variation between the venoms of *A. nummifer* and *A. picadoi* supports the need of reassessing the evolutionary relationships of these snakes. Whereas *Atropoides* snakes are primarily terrestrial, *Bothriechis* genus comprises arboreal species that spend their time in the thick foliage of forest trees and shrubbery. Phylogenetic analyses separate the pitviper of the New World from those of the Old World, and the arboreal Asiatic species from the terrestrial Asiatic species. Our first venomics characterization of arboreal snakes may aid in understanding their ecology and the biological action of their venom.