Exploring Bothrops snake venom variability by proteomic approaches

Solange M.T. Serrano

Laboratorio Especial de Toxinologia Aplicada, CAT/CEPID, Instituto Butantan, São Paulo.

Viperid venom proteomes are the most complex among the venomous snakes and offer an intriguing challenge in terms of understanding the proteome variability and the pathological outcomes of envenomation. In this work, the venom complexity of eight species of Bothrops was analyzed using two-dimensional electrophoresis (2-DE) and LC/MS/MS and the sub-proteomes of proteinases were analyzed by 2D-immnostaining and 2D-gelatin zymography. The venom profiles obtained demonstrated the similarity as well as the diversity among venom proteinases. We also explored the sub-proteome of heparin-binding toxins of Bothrops venoms by heparin-Sepharose chromatography. Toxins with high-affinity for heparin varied in abundance among the venoms and were identified as serine proteinases and C-type lectins. The variability of *B. jararaca* venom was also assessed concerning two stages of the snake life: newborn and adult. 2-DE revealed distinct profiles between newborn and adult venoms illustrated by a higher abundance of acidic proteins of high molecular mass in the newborn venom. The adult venom showed clear differences in spots at low molecular range, most of which were absent in the newborn venom. The analysis by isobaric tag peptide labeling (iTRAQ) revealed that metalloproteinases, serine proteinases and growth factors are among the proteins with higher expression in adult venom. We also analyzed *B. jararaca* venom protein maturation in the venom gland. The analysis of venom milked at day zero, day three and day nine by 2-DE showed clear differences indicating variability in the proteomic profile along the time. Analysis using iTRAQ identified forty one proteins, however, no significant quantitative difference was detected suggesting that protein maturation in the venom gland is related to post-translational modification and processing of precursors. These results underscore the complex protein composition and variability of snake venom analyzed by different proteomic approaches.
Support: FAPESP