Cross-Reactivity of Polyclonal Anti-Neuwiedase Antibodies against Different Bothrops Venoms

<u>Souza, D.L.N.¹</u>, Ferreira, F.B.¹, Castanheira, L.E.¹,; Mendes, M.M.¹, Nunes, D. C. O.¹, Izidoro, L.H.M.², Cardoso, R.¹, Hamaguchi, A.¹, Homsi-Brandeburgo, M. I.¹, Rodrigues, V. M.¹

¹ Instituto de Genética e Bioquímica, Universidade Federal de Uberlândia, Minas Gerais, Brasil.

² Faculdade de Ciências Integradas do Pontal, Universidade Federal de Uberlândia, Minas Gerais, Brasil.

Snake venoms are compounds that cause several effects, due to different classes of enzymes including proteases, hemorrhagic factors and phospholipases A2. Hemorrhage is an effect resulting from action of snake venom metalloproteinase (SVMPs). This work shows the cross-reactivity between polyclonal antibodies anti-Neuwiedase against different snake venoms and the hemorrhagic neutralization of Bothrops pauloensis (Bp) snake venoms. Neuwiedase was previously purified from Bothrops neuwiedi (Bn) snake venom and the immunization assays were performed by 50µg of Neuwiedase and Freud's adjuvant complete or incomplete injections in Balb-c mice at ratio 1:1 (immunogenic solution/ adjuvant, v/v). The mice were bleed after 7 days of last immunization and serum was kept at -20°C. Antibodies were obtained after protein-A Sepharose chromatography. The fractions contained immunoglobulin G (IgG) were frozen, lyophilized and stored at -20°C. The cross-reactivity of antibodies anti-Neuwiedase against venom Bp, Bothrops alternatus (Ba) and Bothrops moojeni (Bm) were determined by enzymelinked immunosorbent assay (ELISA). The anti-Neuwiedase antibodies showed cross-reactivity against Bp, Ba and Bm snake venoms at 1:1600 dilution. The anti-Neuwiedase antibodies were able to inhibit 100% the hemorrhagic activity induced by Bp snake venom after incubation for 30 min at 37°C at ratio 1:50 (venom:antibody, w/w). Our results suggest that anti-Neuwiedase antibodies can recognize similar epitopes presented in the different venoms, mainly which are present on hemorrhagic metalloproteinases. This toxin may be used as immunogen for stimulate the production of antibodies effective to assist the envenomation treatment.

Key-words: Antibody polyclonal, *Bothrops,* Neuwiedase, SVMPs.

Support: FAPEMIG, CNPq, UFU