that the iBAv is significantly more efficient in neutralizing the venom of *Bothrops jararaca* than BAv. This reverse antivenomics approach may contribute to the characterization of current antivenoms and help in the planning of a new generation of improved antivenoms.

LOXOSCELES GAUCHO VENOM GLAND PROTEOME: A NEW PERSPECTIVE ON LOXOSCELES VENOM BIOCHEMICAL COMPOSITION

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In Brazil, Loxosceles are considered to be the most important arthropods for public health, these animals can cause important clinical local or systemic manifestations in human. The cutaneous loxoscelism is characterized by severe inflammation at the site of the bite, that commonly courses with purple colored points forming a marmoreal aspect that spreads gravitationally, that after evolves with a extensive necrotic area. Systemic loxoscelism occurs mainly in children, and presents hemostasis disturbance, renal failure and intravascular hemolysis. The treatment for loxoscelism includes mainly antivenom and corticosteroids for systemic symptoms, and in nowadays tetracycline ointment is used to treat the local lesion. The antivenom efficacy depends on the identification of antigenic toxins that could lead to a neutralization of toxic activities by antiserum use. Proteomic analysis of Loxosceles venom is an very important tool, that bring new strategies to antivenom production and could clarify the differences between envenomation caused by different species of Loxosceles. For this work, the venom gland of three specimens of Loxosceles gaucho were extracted and analysed by 1D-SDS-PAGE and by shotgun proteomics (LC-MS) using three different enzymes (Trypsin, Chymotrypsin and Pepsin). Electrophoresis presented the major bands in the range of 35 kDa characteristic of dermonecrotic D Phospholipases (PDL). On the proteome were observed a wide diversity of Phospholipase D sicaritoxins, belongings to different families: alpha-2, C1, B1, I1 and Beta A1. Specific toxins from L. gaucho were identified LOXN2 and LOXN1/7 and good coverage for LgRec1 protein (69%) and LvSicTox-alpha C1 (54%) were obtained. The proteomic analysis showed 12 differents PDLs, 2 Astacin-Metalloprotease and firstly described the presence of an insecticidal toxin Sicaritoxin-Lia1 on the L.gaucho venom.

HYPANUS AMERICANUS MUCUS: A NEW POINT OF VIEW ABOUT STINGRAY IMMUNITY AND TOXINS

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Fish skin plays important biological roles, such as the control of the osmotic pressure gradient, protection against mechanical forces and microorganism infections. The mucus, on the other hand, is a rich and complex fluid, important for the fish acting as innate immunity system, swimming and nutrition. The elasmobranch epidermis is characterized mainly by mucus secretory cells, and marine stingrays have already been described to present secretory glands spread throughout the body. Little is known about the biochemical composition of the stingray mucus, but recent studies denoted the importance of mucus in the envenomation process. Stingrays venom are largely studied due the human medical importance of envenoming caused by sting puncture, that evolve with local inflammation and necrosis, and these toxic events can be correlated to the chemical composition of the sting skin, according to the literature. Aiming to analyse the mucus composition, a new non-invasive mucus collection method was developed that focused on peptides and proteins, and biological assays were performed to analyze preliminary toxic and immune activities of the *Hypanus americanus* mucus. Pathophysiological characterization showed the presence of peptidases on mucus, as well that the induction of edema and leukocyte recruitment in mice. The fractionated mucus improved phagocytosis on macrophages and showed antimicrobial activity against *T. rubrum, C. neoformans* and *C. albicans in vitro*. The proteomic analyses showed the presence of immune-related proteins like actin, histones, hemoglobin, and ribosomal proteins. This protein pattern is similar to those reported for other fish mucus and stingray venom. This is the first report depicting the *Hypanus* stingray mucus composition, highlighting its biochemical composition and importance for the stingray immune system and the possible role on the envenomation process.

SEARCH FOR PROTEASES AND PROTEASE INHIBITORS IN THE WAXY SECRETION THAT COVERS THE TICK *AMBLYOMMA SCULPTUM'S* EGGS (ACARI: IXODIDAE)

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Ticks are hematophagous mites that can feed on various animal species. The *Amblyomma sculptum* tick is widely found parasitizing animals, including humans, in Brazil, and it is recognized as a vector of important pathogens relevant to public health. For their perpetuation, females of ixodids when engorged, can lay up to 10,000 eggs. Protease inhibitors (PIs) and proteases present in organs and other secretions of ticks have been described, but there is no data in the literature on the presence of these molecules in the wax that covers *A. sculptum* eggs (EW). Therefore, this work had as objective the study and the characterization of PIs and proteases present in EW. The eggs were collected and the EW's aqueous extract was submitted to a 10 kDa molecular weight cut off membrane, and the high molecular weight pool (HMWP) and the low molecular weight pool (LMWP) were obtained. The LMWP was fractionated on a C18-RP-HPLC, and 24 peaks were manually collected and tested as elastase inhibitor, using a FRET substrate. Four peaks inhibited about 50% of the elastase catalytic activity, and their molecular contents and primary sequences are being analyzed by mass spectrometry. Analysis of HMWP by SDS-PAGE showed proteins of 100-260 kDa, in addition to two bands with about 40 kDa. In a proteolytic activity assay, using metallo and serine peptidases inhibitors and FRETs substrates, it was observed the presence of metallopeptidases in the pool. This is an important result and will be deepened, since the inhibition of metalloproteases present in eggs of other species of arthropods prevented their hatching. In addition, the search for bioactive molecules with biotechnological potential in EW's *A. sculptum* showed to be promising, according the results obtained in this work.

CROSS-SPECIES AND GEOGRAPHIC POTENTIAL OF B-CELL EPITOPE STRINGS IDENTIFIED FOR GENERATION OF AN AFRICA-SPECIFIC SNAKE VENOM-INDUCED NECROSIS THERAPEUTIC

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Every year 1.8-2.7 million people suffer snakebite envenoming, of these, an estimated 137'000 die and 400'000 are left disfigured or disabled. Fundamental to alleviating this burden is improving antivenom efficacy. Linear B-cells epitopes are short peptides displayed by antigen presenting cells to elicit an immune response. Multiple toxin epitopes can be identified and arranged as 'beads on a string', increasing the probability of a broadly-neutralising immune response. Prior studies have shown epitope-string immunogens are able to generate antibodies capable of neutralising haemotoxic venom proteins. I intend to adopt this approach to create snakebite necrosis specific serotherapies. Snakebite necrosis causes significant physical, mental, and economic distress. Whilst there is a debate