## Alternative approach to immunotherapy against scorpion envenomation using detoxified venom associated with alum adjuvant: inflammatory response assessment.

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Scorpion envenomation is a public health problem in several parts of the world. An approach to treat this accidental pathology could be the vaccination to complete the used immunotherapy. Aluminum hydroxide adjuvant is an immunologic adjuvant used in human and veterinary vaccines. In this study, we used detoxified venom with Alum adjuvant in order to develop an effective vaccine against the toxic effects induced after scorpion envenomation.

Detoxified *Androctonus australis hector* venom was associated to Alum adjuvant and used in immunization of rabbits. The animals were inoculated three times at one month interval. During the immunization protocol, blood samples were collected weekly after each injection. Cell count, serum peroxidase activities (MPO, EPO) and antibody titer (IgG) were evaluated. Six months after immunization, a protective effect of immunized rabbits with detoxified venom was evaluated by injection of different lethal doses of native Aah venom and mortality was recorded.

During the immunization schedule, low levels of peripheral neutrophil, eosinophil cell count and peroxidase activities were observed in immunized animals with detoxified venom. However, immunological response showed high titers of IgG at one month after immunization followed by gradual decrease that persisted at six months. Results showed that detoxified venom can induce an immunoprotective effect six months after immunization against challenge with lethal doses until 6 DL50 of native venom.

It seems that detoxified venom associated to Alum adjuvant triggers a specific immune response with low inflammation. Despite antibody titer decreased with time, the protection remained higher. These results allowed to suggest a possible alternative to immunotherapy.

Key-words: detoxified venom, alum adjuvant, inflammatory and immune response, immunoprotection.

## Determination of eugenol and its derivative isoeugenol in *Globularia alypum* by solvent system extraction and comparative study of their antioxidant activities with various oxidation conditions.

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Globularia alypum leaves have been widely used for more years in gastrointestinal disorders as a traditional folk medicine. The aim of the present study was to determine the chemical composition of the petroleum ether extract and to evaluate its antioxidant activities in comparison with eugenol and its derivative isoeugenol. After phytochemical tests, a simple hydrodistillation was effectuated by Clevenger apparatus and the distillate was extracted with petroleum ether by decantation process. Gas chromatography mass spectroscopy was used to identify and quantify phenolic compounds in this extract. The antioxidant activity of petroleum ether extract from GA was measured in vitro by 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging method. However, total antioxidant capacity (Molybdate phosphate test), hydrogen peroxide scavenging and reducing power antioxidant ( $P^+ K^+$ ) were estimated. The petroleum ether extract demonstrated a low free radical scavenging capacity compared to eugenol and isoeugenol extracted from Syzygium aromaticum. Furthermore, the identification of this extract by CPG/ms led to the isolation of new known phenol named eugenol and also its derivative isoeugenol with considerable amounts (10.56%, 0.87%). The antioxidant capacities of the petroleum ether extract are probably associated with phenolic compounds detected and its principle compound indicates that this plant may be an important source of chemopreventive and chemotherapeutic natural products. The best of our knowledge is the combination of new detected compounds for the first time, eugenol and its derivative in this plant which has been tested separately as powerful antioxidant agents. However, further studies are required to determine if this is of clinical significance.

Key-words: *Globularia alypum*, petroleum ether extract, CPG/ms, eugenol, isoeugenol, antioxidant activities.

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