

# USE OF HET-CAM ASSAY FOR EVALUATE THE IRRITANT POTENTIAL OF NATURAL POLYMER HYDROGELS WITH AMINO ACID-DERIVED IONIC LIQUIDS

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**INTRODUCTION:** Hydrogels of natural polymers, such as alginate and chitosan, have been studied due to their potential use as dressings thanks to their greater biocompatibility and biodegradability. The incorporation of ionic liquids provides hydrogels improved physicochemical and mechanical properties, especially when using amino acids, allowing for reduced cytotoxicity. However, damage prediction is essential for their contact with biological tissues. **OBJECTIVE:** To evaluate the irritancy score (IS) of alginate and chitosan hydrogels incorporated with ionic liquids (ILs) derived from the amino acids glycine and lysine using the Hen's Egg Test chorioallantoic membrane assay (HET-CAM). **MATERIAL AND METHODS:** Hydrogels from chitosan and alginate polymers, incorporating with different concentrations of ILs were developed through physical crosslinking. To evaluate the irritant potential of the chorioallantoic membrane, fertilized chicken eggs with 10 days of incubation were used. For the assay, the outermost shell and the white membrane were removed and 0.3 g of the hydrogels, 600  $\mu$ L and 900  $\mu$ L for the pure IL of lysine and glycine, respectively, were applied to the chorioallantoic membrane ( $n = 6$ /formulation). In addition to negative control, consisting of saline solution. The positive control was performed with 0.1 N NaOH. The membrane was monitored for 300 s and the time for the occurrence of vasoconstriction, hemorrhage and coagulation phenomena were recorded to calculate the IS. Thus, the lesions were classified as: non-irritating (0–0.9), mildly (1–4.9), moderately (5–8.9) and severely irritating (9–21). Animal Use Ethics Committee approval UFSM: 1261190724. **RESULTS:** Seven hydrogels were obtained in different concentrations of lysine and glycine ILs, being: 0% (chitosan/pure alginate), LIS 5%, LIS 10%, LIS 15%, GLI 5%, GLI 10% and GLI 15%. The pure lysine and glycine ILs presented severely irritating scores (IS = 20.2

and 20.4 respectively), positive control (IS = 21). The hydrogels with LIs in different concentrations presented IS = 0, the same as the negative control. **CONCLUSION:** The hydrogels with LIs did not show toxicity, but further studies are needed for topical application.

Keywords: chorioallantoic membrane assay; hydrogels; ionic liquids

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