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Gelatin films crosslinking with glutaraldehyde for use in biosensors: influence of the concentration and the reticulation time

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Abstract – Biosensors based on the silicon technology require a protecting biocompatible coating for implanting in the human body. Among biomaterials, the gelatin is a promising coupling agent for biological entities with applications in several fields such as food, pharmaceutics and biomedicine. The obtained gelatin membranes were immersed in glutaraldehyde solutions with concentration of 0.5% to 2.0% for periods from 10m to 24h. The efficiency of the crosslinking agent was evaluated by swelling tests. Citotoxicity test were also performed. It has been observed that for 24h a glutaraldehyde concentration of 0.5% yields acceptable results.

Biosensors are able to provide quantitative or qualitative specific analytical information by using a recognition element of biological origin in close contact with a transducer. These devices are selective, versatile and relatively inexpensive, useful for real time measurements in, e.g., in bioprocesses, and in many industries such as food, health, oil, biomedicine and environment monitoring. Biosensors based on the silicon technology require a protecting biocompatible coating for implanting in the human body. Biomaterials like gelatin and chitosan are being increasingly used as coupling agents for biological entities. In order to reduce the enzymatic degradation and maintain the required mechanical properties during the implantation period a stabilization of these organic biomaterials is needed. Among the agents used to promote the crosslinking, the aldehyde and particularly the glutaraldehyde are mostly used, even when they are toxic at high concentrations. Thus, the aim of this work was to determine the lower acceptable concentration and crosslinking time for gelatin films as coatings for biosensors. The gelatin films were obtained and immersed for periods of 10m, 1h and 24h in solutions of glutaraldehyde with concentrations between 0.5% and 2.0%. The efficiency of the crosslinking agent was evaluated by swelling and toxicity tests. Fig. 1 shows the infrared spectrum of the gelatin films with the presence of the following principal bands: at 1650 cm^{-1}, characteristic of the I amide, due to the carbonyl stretching; at 1560 cm^{-1}, related to the II amide, due to the vibrations in the binding plane N-H and to the C-N stretching; at 1235 cm^{-1}, associated to the vibrations in the III amide plane, due to the C-N stretching and to the N-H deformation at 1450 cm^{-1} and near 3450 cm^{-1}, due to the O-H stretching [1]. By means of Scanning Electron Microscopy (SEM) the film morphology was analyzed. The gelatin showed a plane and rough surface (Fig. 2). The swelling process of the films diminished with the increase of the glutaraldehyde concentration as well as with the immersion times in the used concentrations, resulting in more stable films in aqueous solutions [2]. For a time of 24h it is possible to use a solution with a glutaraldehyde concentration of 0.5%.

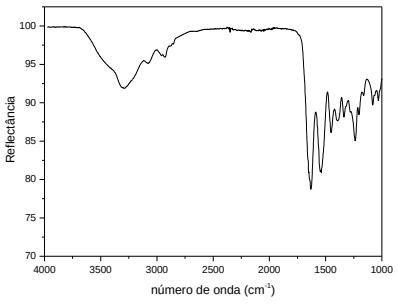

Figure 1: Fourier transformed infrared reflectance spectroscopy (FTIR) patterns of gelatin film.

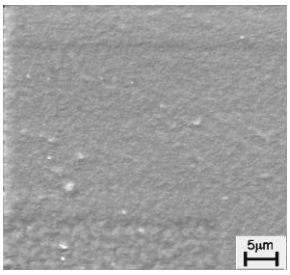

Figure 2: Scanning electron microscopy (SEM) of gelatin film.

[1] Chiono V.; Pulieri E.; Vozzi G.; et al. J Mater Sci: Mater Med, v.19, p.889-898 (2008).
[2] Bigi, A.; Cojazzi. G,; Panzavolta, S.; et al. Biomaterials, v.22, p.763-768 (2001).