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Synthesis, characterization and biological activity of pure hydroxyapatite and associated with gentamicin

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Abstract – This work studied the variation of a conventional route by adding gelatin in HA means of precipitation and then associating an antibiotic (gentamicin). The XRD analysis revealed the presence of single-phase HA. The micrographs obtained through TEM revealed the formation of crystal agglomerates of nanometric size with acicular form. Microbiological test was carried out to verify whether pure hydroxyapatite (without gentamicin) revealed antimicrobial properties and to evaluate differences between the pure material and the with gentamicin.

Phosphate ceramics have been largely investigated as bone implant material. Interest in a particular phase of calcium phosphates, the hydroxyapatite (HA), appeared because its chemical composition is similar to that of natural mineral, and it possess suitable biological properties such as biocompatibility, osteoconductivity and resorption^{1,2}. Nowadays bone infections still represent a problem in medicine and the main reason is the poor accessibility of the infected bone site by systemically administered antibiotics.³⁻⁵ The use of controlled drug release materials offers an important adjuntive route to administer antibiotcs for prevention or treatment of infections of bone.⁵ This work studied the variation of a conventional route by adding gelatin using concentrations of 1, 6, 10 and 60 g.L-1 in HA means of precipitation and then associating an antibiotic (gentamicin) in order to reduce contamination risk of the bone graft substitute^{3,5}. The HA was obtained by precipitation in water and gelatin, and then dried, sieved and calcined at 700°C. After prepared, samples received 1% in mass gentamicin sulfate and were then subject to a microbiological test to evaluate the incorporation efficacy. In material characterization, samples were analyzed through X-ray diffraction (XRD), granulometric, specific superficial area through BET method, transmission and scanning electronic microscopy and infrared spectroscopy (FTIR). The XRD analysis revealed the presence of single-phase HA. The micrographs obtained through TEM revealed the formation of crystal agglomerates of nanometric size with acicular form. Microbiological test was carried out to verify whether pure hydroxyapatite (without gentamicin) revealed antimicrobial properties and to evaluate differences between the pure material and the one with gentamicin, the test was conducted using human-saliva sample and biomaterials. These were inoculated in brain heart infusion (BHI) and incubated for 24 h. Thus, experimental suspensions were cultivated on the surface of Petri plates with BHI agar and were inoculated with the experimental suspensions, and incubated for 24 and 48 hours. Number of colony-forming units (CFU) was carried out after incubation periods for samples. CFUs were more evident 48 hours due to the increase of colonies size, rather than the number of colonies. It was possible to observe that pure hydroxyapatite presents slightly antimicrobial properties with significant reduction in the number of colony-forming units (CFU/mL) in comparison of the control sample human saliva. Hydroxyapatite samples associated to gentamicin showed the lowest number of CFUs or even did not present any proving association with the material obtained.



Figure 1: Photographs of the samples after 48 hours incubation. The sample A represents the control with human saliva and B, C e and D represent the samples with 1% in weight of gentamicin.

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