Mechanical Properties of a composite material made of HAp nanofibers

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ABSTRACT

Biomaterials based on hydroxyapatite (HAp) are often studied due to the favorable answer presented as a substitute of bone tissue. However, the mechanical properties of this material have not been studied a lot. In this paper, we synthesized nanofibers of HAp through the microwave assisted hydrothermal method using Ca(NO3)2, K2(HPO4) and KOH as precursors. The nanofibers presented a preferential crystalline orientation in the [300]. Structural studies were performed using X-Ray diffraction by powders, morphology and microstructure were analyzed by scanning electron microscopy and high resolution electron microscopy. Micrographs were processed in order to perform a deep analysis of HAp crystal structure.

After that, a porosity controlled ceramic was prepared using the HAp nanofibers through the Modified Gelcasting Process. This ceramic possesess meso and macroporosity, and both were relevant on the final mechanical properties.

Finally, the organic phase was added to the porous ceramic and the composite material was obtained. Then, compression tests were made to this composite. The obtained results showed a resistance under compression, in the composite material, quite similar to that observed in the trabecular bone.

Keywords: Modified Gel Casting Process, Oriented Crystal, Hydroxyapatite

1 INTRODUCTION

Different techniques have been used to improve strength and fracture toughness of HAp such as making composites and using different morphologies of HAp as raw material of the composite like nanopowders, whiskers or nanorods [1-3]. But, mechanical properties of the composite of HAp oriented fibers have not been studied a lot.

One factor that must be considered is the porosity which plays an essential role in cell migration and adhesion, tissue formation, mechanical properties and nutrient diffusion, among others [4]. Also, a key component in tissue engineering for bone regeneration is the scaffold generation, which should be used as a template for cell interactions and the formation of bone-extracellular matrix through its pores and thereby provides structural support to the newly formed tissue [5].

Moreover, other issue could enhance mechanical properties of HAp is decreasing the grain size of HAp [1]. With decreasing grain size, the fraction of the grain boundary phase is increased and thereby a greater amount of energy is absorbed during crack propagation through the grain boundaries of such fine grained microstructures (nanostructures) [6].

The aim of this research was the obtainning of a biomaterial with a mechanical resistance comparable to those observed in some bone tissue. The nanofibers samples were analyzed by XRD, SEM and TEM to verify the morphology and the size of the fibers of HAp. Then, those fibers were molded by the Modified Gelcasting process and a porous material was made. Afterward, the organic phase (Gelatin) was introduced inside the ceramic porosity to prepare composite materials and finally, compression tests were performed in the composite, and the results were compared to those mechanical resistences observed in several types of bone tissues.

2 METHODOLOGY

2.1 Synthesis of HAp

The synthesis of HAp was done by microwave assisted hydrothermal method. The reacting mixtures were prepared in two steps; first, one solution made of glutamic acid [C5H9NO4·H2O] (J.T. Baker FW 147.13) and calcium nitrate [Ca(NO3)2·4H2O] (Sigma Ultra C4955) was prepared in deionized water. The two components were dissolved by mechanical agitation during 2 h at 60° C.

In the second step, other solution of monobasic potassium phosphate [KH2PO4] (Mallinckrodt Chemicals FW 136.09) and potassium hydroxide [KOH] (SIGMA-ALDRICH 221473) was done in deionized water, and this solution was stirred by 20 minutes. Then, both solutions were mixed and the stirring was continued for another 10 minutes [7].

Finally this reacting mixture was spilled inside Teflon vessels which were closed immediately and placed inside a

microwave oven (Synthos 3000 by Anton Paar). All synthesis reactions were carried out at 170° C and 70 kPa and the synthesis reaction was performed within 45 minutes.

2.2 Modified gel casting process

Ethanol was used as a solvent and the required amount of Polyvinyl butyral (PVB) was added and then, the solution was mixed under magnetic stirring. Besides, Polyethylene glycol (PEG) was melted and mixed with Poly(acrylic acid) (APA) wich was incorporated with the mixture of Ethanol-PVB.

In the other hand, the HAp was mixed with oxalic acid and the necessary amount of Poly(methyl methacrylate) (PMMA). Finally, both dissolutions were carefully mixed to form a mixture which became a viscous blend. This blend was molded and dried at room temperature. Once the dry blend was removed from the mold, it was sintered by heating in a melting pot until a temperature of 1100°C for 2 h using a heating rate of 1°C/min.

Due to the fact that, the combination of polymers and hydroxyapatite to produce bone substitutes is a natural strategy, gelatin was used to generate a composite material. The ceramic scaffolds absorbed different solutions of gelatin in water, in concentrations of 5.7, 6, 6.6 % by weight related to HAp. Adsortion was made through capillarity, and in this way, the organic-inorganic composite materials were made.

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2.3 X-Ray Diffraction (XRD)

In order to identify the HAp crystal phase contained in the samples, all the nanostructures were analysed by X-Ray diffraction by powder using a D8 Advance diffractometer built by Bruker. The operation conditions in the diffractometer were 35 kV and 15 mA, using a CuKα radiation with wavelength of λ= 1.5406 Å. The measurements were done from 10° through 90° on a 2θ scale with a step size of 0.05°.

2.4 Scanning Electron Microscopy (SEM)

Observations of the morphology and microstructure of all materials were done using a scanning electron microscope Jeol JSM-6390 LV. The accelerating voltage used in the microscope was 20 kV and all the images were formed from secondary electrons. The sample was collected from a vessel by a pipette and separated from the solution at the end of the synthesis process. A small portion of the filtered solid was put on a sample holder and glued by carbon painting.

2.5 High Resolution Transmission Electron Microscopy (HRTEM)

Morphology and microstructure were observed through a JEOL JEM-2100F using an accelerating voltage of 120 kV. Conventional bright field and high resolution images were recorded. Moreover, most of the images were analysed using the software Digital Micrograph® designed by Gatan in order to analyse the HAp Crystal structure of nanofibers. Sample preparation was dried and then a little amount of material was put on a carbon-film copper grid of 3 mm of diameter.

2.6 Mechanical Testing

All the pieces were sanded up to reach a cubic shape with size of 5x5x5 mm^3. The mechanical tests of the composite materials were carried out in a Zwick/Roell Tensile and Compression Testing Machine model Z005 in compression mode, at room temperature and a deformation rate 1mm/min. For each type of material, 5 samples were tested and the average values were tabulated.

3 RESULTS AND DISCUSSION

3.1 X-Ray Diffraction

Figure 1 shows a typical diffractogram of HAp. The crystal phase identification was made by comparison using the International Centre for Diffraction Data ICDD powder diffraction file (PDF) bank, and the identified phases in the samples were the PDF´s 88-1199 and 86-120, both corresponding to HAp. A very important fact related to these diffractogram is that the intensity of the HAp exhibit the most intense reflexioan at 33º in 2θ, and it corresponds to the (300); this increment in the intensity of the reflexion indicates a cristalline preferential orientation.

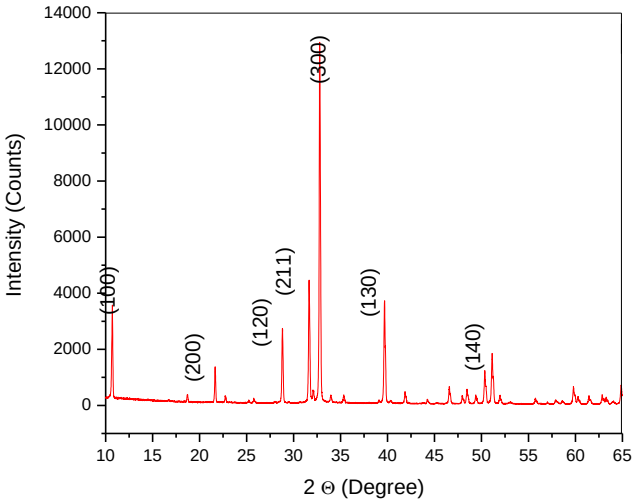

Figure 1. X-ray diffractogram recorder from a HAp sample obtained by the microwave assisted hydrothermal method.

Also, using the "EVA" program provided by Bruker, the average crystallite size of the HAp was determined and a value of 64.15 nm was found. It is important to notice that the signals in the diffractogram are well defined and have low noise, indicating a high crystallinity of HAp [8].

3.2 Scanning Electron Microscopy (SEM)

Observation made by the SEM allowed determining the morphology in the HAp samples, and the shape of microfiber was revealed as can be observed on Figure 2. It is necessary to remark that the morphology of fibers was mainly obtained and no nanoplates neither nanoparticles were observed, which is desirable for the purposes of this study.

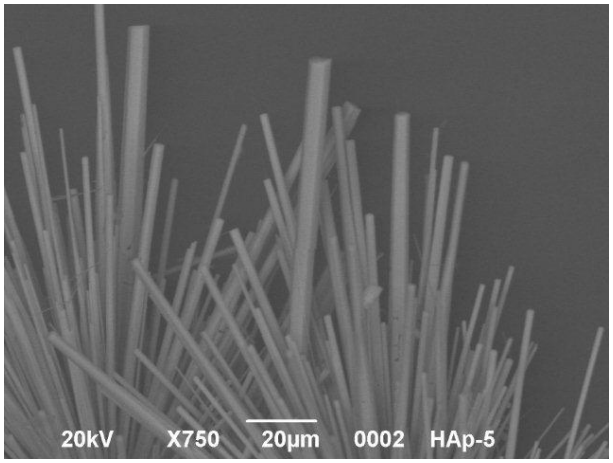

Figure 2. SEM Micrograph of HAp microfibers which have a hexagonal cross-section.

The microfiber surfaces appeared quite smooth and all fibers possess a hexagonal cross-section. The facets and edges that form the microfiber body look well-defined, and this is evidence of a high crystallinity.

3.3 High Resolution Transmission Electron Microscopy (HRTEM)

A magnified image obtained by HRTEM of one nanofiber edge is displayed on figure 3. An arrangement of parallel lines is evident in the image, which correspond to the crystal structure of HAp. This line arrangement was analyzed through the Digital Micrograph software, and some interplanar distances were determined. These distances were of 0.815 nm, which corresponds to the (100) planes, and 0.341 nm, which is related to the (002) planes of HAp crystal structure.

When a Fast Fourier Transform (FFT) was performed by the Digital Micrograph software to the image region within the white rectangle, the dot pattern on Figure 3(b) was produced. The dots array in this pattern is evidence of a high crystallinity. The symmetry 2 is clearly observed in this pattern and the most intense dots correspond to the (002) plane (labelled in the FFT). Other intense dots in the pattern correspond to the (300) and the (100) planes.

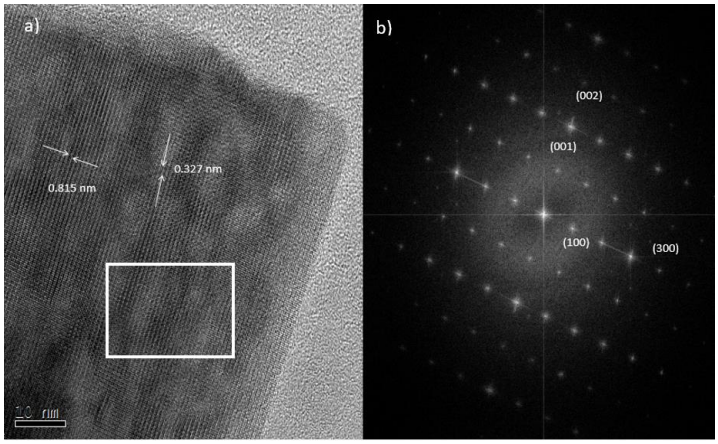

Figure 3. (a) HRTEM micrograph of one nanofiber edge, and (b) a FFT from the image region inside the white rectangle in (a).

This result also indicate that HAp fibers grew with a preferential crystalline orientation. According to this result and that from XRD and the morphology observed by SEM, it is possible to determine that the nanofibers grew along the "c"axis.

3.4 Mechanical Testing

The mechanical properties of a bone graft should be similar to those properties of bone receptors and consequently, to be adequate to support load. These mechanical properties vary depending on the type of bone. The maximum compressive strength effort is between 100 and 150 MPa for compact bone and between 2 and 12 MPa for trabecular bone.

On Figure 4 the average mechanical behavior under compression of molded HAp samples was observed. These samples contained 3 different concentrations of gelatin. Is evident how the ultimate strengh increase with the imcrement of gelatin concentration. The values of this ultime strengh were of 4.17, 4.82 and 13.14 MPa related to the concentrations of 5.7, 6, 6.6 % by weight respectively, these values correspond to those observed in the trabecular bone [9].

4 CONCLUSIONS

Through the microwave-assisted hydrothermal method and using the appropriate precursors, HAp nanofibers were synthesized. The fibers grew with a high crystallinity and with a preferential crystalline oriented in "c" direction, and the microwave-assisted hydrothermal method was proved to be a very efficient methodology to carry out the HAp synthesis with a significative saving of time and energy.

The composite material made of HAp nanofibers and gelatin were possible to obtain through the modified gel

casting method and these composite materials showed synergical mechanical properties.

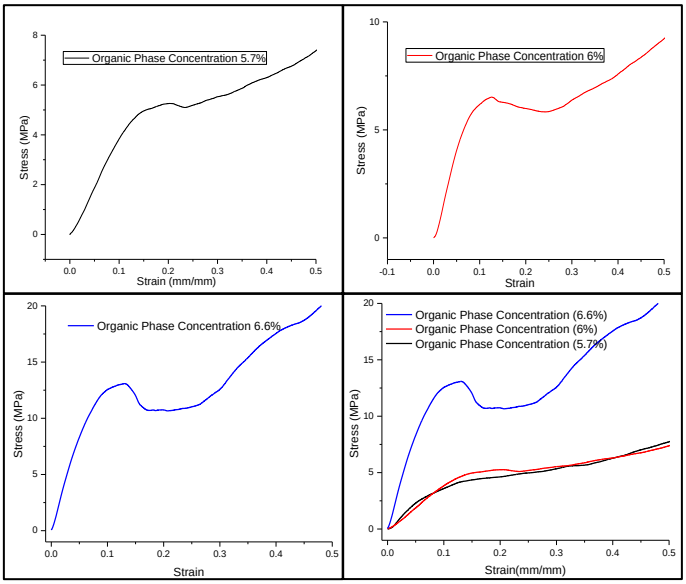

Figure 4. Stress-strain test graphic of the HAp composite

The maximum ultimate strength recorded for these composites was of 13.14 MPa at a low concentration of organic phase. This value is close similar to that observed in the trabecular bone, which is between 2 and 20 MPa. It is pssible that, a potential increment in the organic phase coud produce a higher ultimate strengh value, and this could be similar to that registered to the compact bone.

With the results obtained, it is possible to conclude that the nanofibers of HAp oriented in direction "C" has a favorable effect on structural, morphological and mechanical properties because

According with all results exposed above, it is also possible to supose that the composite material obtained here has the potential applicationa as a bone replacement material.

ACKNOWLEDGEMENT

The authors would like to thanks the financial support of CONACyT Fondo Salud 2013-01-201329 and the technical assistance of José Luis Cabrera Torres and Carmen Peza Ledesma for the microscopy observations.

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