SYNTHESIS AND CHARACTERIZATION OF BIOACTIVE CERAMIC IN $CaO\text{-}MgO\text{-}SiO_2$ SYSTEM

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Abstract

Na_2SiO_3 solution and chloride solutions of calcium and magnesium were used as raw materials. HCl was added to Na_2SiO_3 solution resulting in gel of silica. In order to obtain a mixture of Si gel, Ca^{2+} and Mg^{2+}, calcium and magnesium solutions were blended into the gel. After adding NaOH to the mixture, Ca^{2+} and Mg^{2+} ions precipitated as hydroxides. Subsequently, it was dried to attain the ceramic precursor powder, which was calcined at 900 ºC for 1h, compacted into a pellet form and sintered at 1200 ºC for 2h. The ceramic body was characterized by SEM, XRD and FTIR. *In vitro* test for bioactivity verification was performed in SBF. A surface deposition of hydroxyapatite on the ceramic body was observed after 3 days. The cytotoxicity test revealed that the material has cell viability above 70%, meaning that it is not toxic for the cells, according to ISO 10993-5 standard.

Keywords: Biomaterial, Glass-ceramic, $CaO\text{-}MgO\text{-}SiO_2$ system, Sol-gel synthesis.

Introduction

The use of ceramics[1] as biomaterials for the body is relatively recent and is due to its high chemical stability, better biocompatibility than metals and good tribological properties, being used for repair or replacement of connective tissues. These bioceramics[1] present a stable interface with the tissue, compatibility of the

implant mechanical behavior with the living tissue, and are classified as inert and bioactive.

The concept that the bioactive ceramics are able to bind to bone was first suggested by Larry Hench[2] in the 1970s through the Bioglass® synthesis. He observed that a hydroxyapatite layer was formed on the implant (Bioglass®) surface allowing interfacial bonding. Currently the ceramics[1], with bioactive characteristics studied are bioactive glasses, glass ceramics and calcium phosphate ceramics.

The glass-ceramics[3-4] are obtained from a heat treatment carried out on bioactive glass, with the aim of improving their mechanical resistance which is usually low and is composed of a crystalline phase and a residual glassy phase. They are based on Bioglass® where 45 mol% of the composition is SiO_2, and this concentration demonstrates a hasty interfacial bonding of material with the bone. According to Krishnan and Lakshmi[5] during interfacial bonding, genes that control growth factor and osteogenesis are activated and within 48h a hydroxyapatite superficial layer similar to bone tissue is obtained.

In this work, SiO_2, MgO and CaO were used in the mol concentration of 45.98%, 10.72% and 43.30% respectively, to obtain the CaO-MgO-SiO_2 system. According to Xianchun Chen[6] et al., this mol concentration has mechanical properties close to the cortical bone, good bioactivity and biocompatibility *in vitro* being used in bone regeneration.

In the system structure[7-9], SiO_2 is responsible for forming a network covalently attached to oxygen atoms at their vertices, which provides stability to the material and is called the network former. CaO is called the network modifier, because interrupt the covalent SiO_2 network by establishing ionic bonds with the oxygen atoms, which become unbound oxygen atoms. This modification improves the elastic modulus property and is essential for initiating the first stage of interfacial bonding by increasing the growth rate of the hydroxyapatite layer. MgO is also considered a network modifier by narrowing the SiO_2 covalent network, by altering mechanical properties such as micro hardness and fracture toughness, reduces the coefficient of thermal expansion and reduces the system degradation rate to favor bioactivity in the initial stages of hydroxyapatite formation.

Several techniques for prepare glass ceramics was studied, for example, methods of co-precipitation powder mixing[10], solid state reaction[11], pyrolysis

spray[12] and sol-gel[13]. The sol-gel method involves the synthesis of a sol, to obtain a gel which is thermally treated for the ceramic material crystallization[14]. Usually, powders that exhibit high purity, homogeneity[15] and high bioactivity[16], are produced from this process. Due to use of low temperature, favorable characteristics in obtaining the bioceramics related to the pores size and volume associated with the surface area[17] are reached by this method.

For this work, it was chosen the so-gel method combined with co-precipitation. Gel of silica was obtained from Na_2SiO_3 solution by adding HCl. Then chloride solutions of calcium and magnesium mixed to gel was co-precipitated as correspondent hydroxides with NaOH up to pH 10. The produced ceramic powders after sintering resulted in ceramic bodies that were characterized by the XRD, FTIR and SEM techniques. The diffractograms revealed the presence of diopside crystalline phase as majority. The ceramic body bioactivity was verified by immersion in SBF (simulated body fluid). The results were analyzed by FTIR and SEM. Cytotoxicity test confirmed cell viability above 70% and according to ISO 10993-5[18] is sanctioned as non-cytotoxic.

Materials and methods

Na_2SiO_3 solution and chloride solutions of calcium and magnesium were used as raw materials. In order to synthesize the bioactive CaO-MgO-SiO_2 ceramic powder was used sol-gel method combined with co-precipitation.

HCl was added slowly in Na_2SiO_3 solution in a beaker, under stirring to homogenize the mixture, and monolithic gel of silica was produced after 24h of rest. Then, it was broken with a stirrer and submitted to an ultrasound bath for 1h in order to result a suspension of gel. Subsequently this suspension was blended with Ca and Mg chloride solutions to obtain a gel with Ca^{2+} and Mg^{2+} ions homogeneously mixed. Afterward, NaOH was added up to pH 10, to co-precipitate Ca^{2+} and Mg^{2+} as hydroxides.

The resulted product was filtered and washed with deionized water until no Cl^- ion was detected, then it was dried at 70ºC to attain the ceramic precursor powder, which was calcined at 900ºC for 1h. The resulted white powder was characterized by XRD technique to investigate the crystalline phases. The powder was compacted into a pellet form (10 mm diameter and 3,5 mm thickness) and sintered at 1200ºC for 2h

to obtaining a ceramic body, that was also characterized by XRD technique and analyzed the changes in crystalline phases. Experimental procedure flow sheet is showed in Figure 1. *In vitro* tests were performed by soaking sintered samples in SBF (Simulated Body Fluid) at 37°C for 3, 7 and 21 days. The SBF solution was prepared according to the procedure described by Kokubo[19]. After the respective soaking time, the ceramic body samples were taken out, washed with deionized water and dried at 70ºC for 24 h and characterized by FTIR and SEM micrograph.

In vitro cytotoxicity was performed by the indirect method as recommended by ISO 10993-5 using Chinese hamster ovary cell line in micro plates. These micro plates were analyzed in a spectrophotometer reader at 495 nm. The results were compared with an extracts of negative control of alumina and a positive control of latex.

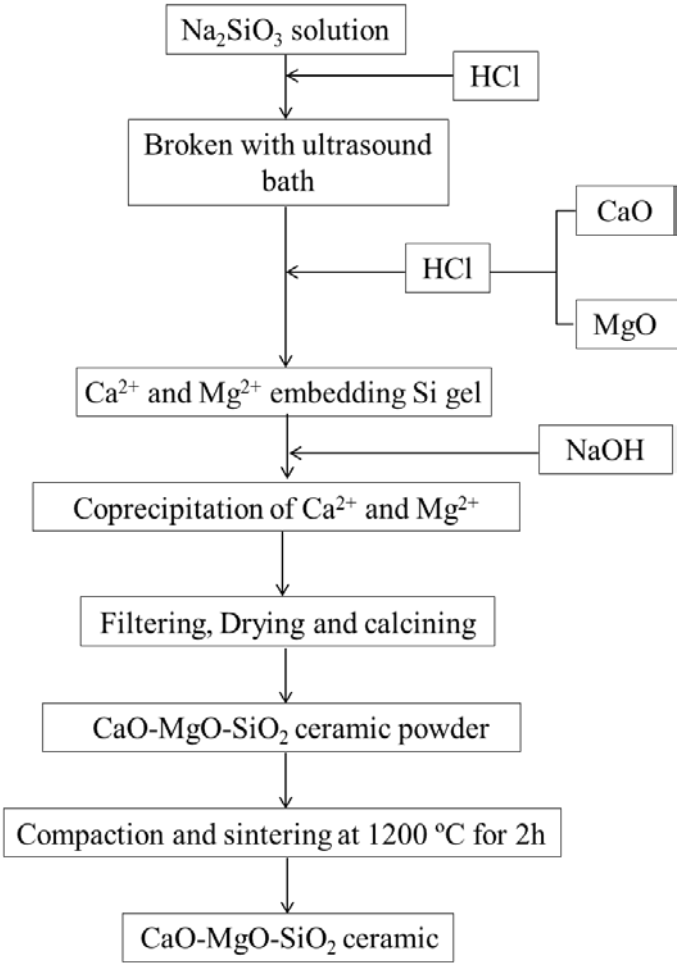

Figure 1: Experimental procedure flow sheet of CaO-MgO-SiO_2 ceramic preparation.

Results and discussion

The XRD pattern of CaO-MgO-SiO_2 ceramic powder calcined at 900ºC for 1h is showed in Figure 2.

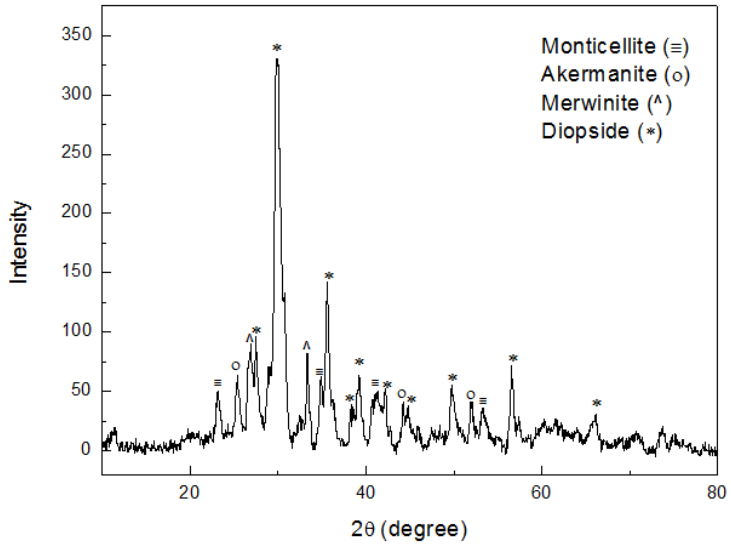

Figure 2: XRD pattern of CaO-MgO-SiO_2 ceramic powder calcined at 900ºC for 1h.

From Figure 2 it is observed a multiphase composite, with predominance of diopside phase, with other phases being: monticellite, akermanite and merwinite. The XRD pattern of sintered ceramic body at 1200ºC for 2h is presented in Figure 3. It is possible to observe a multiphase composite with diopside as main phase again, with other phases as monticellite, akermanite, merwinite, majorite and serendibite are present.

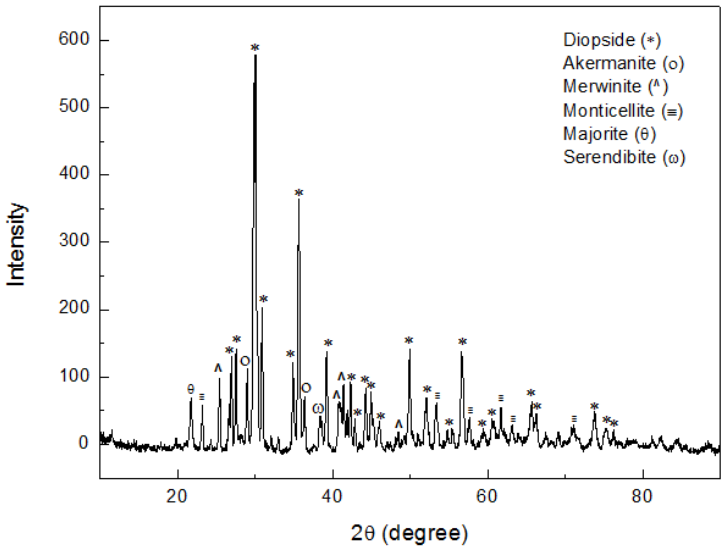

Figure 3: XRD pattern of sintered ceramic body of CaO-MgO-SiO_2 at 1200ºC for 2h.

The SEM micrograph on Figure 4 shows the CaO-MgO-SiO_2 ceramic body before (a) and after 3 days (b), 7 days (c) and 21 days (d) soaked in SBF solution. Figure 4(a) shows a flat surface with some reentrance in the sintered ceramic body. While in figure 4(b) to 4(d) it is observed the surface covered by spherical particles deposited.

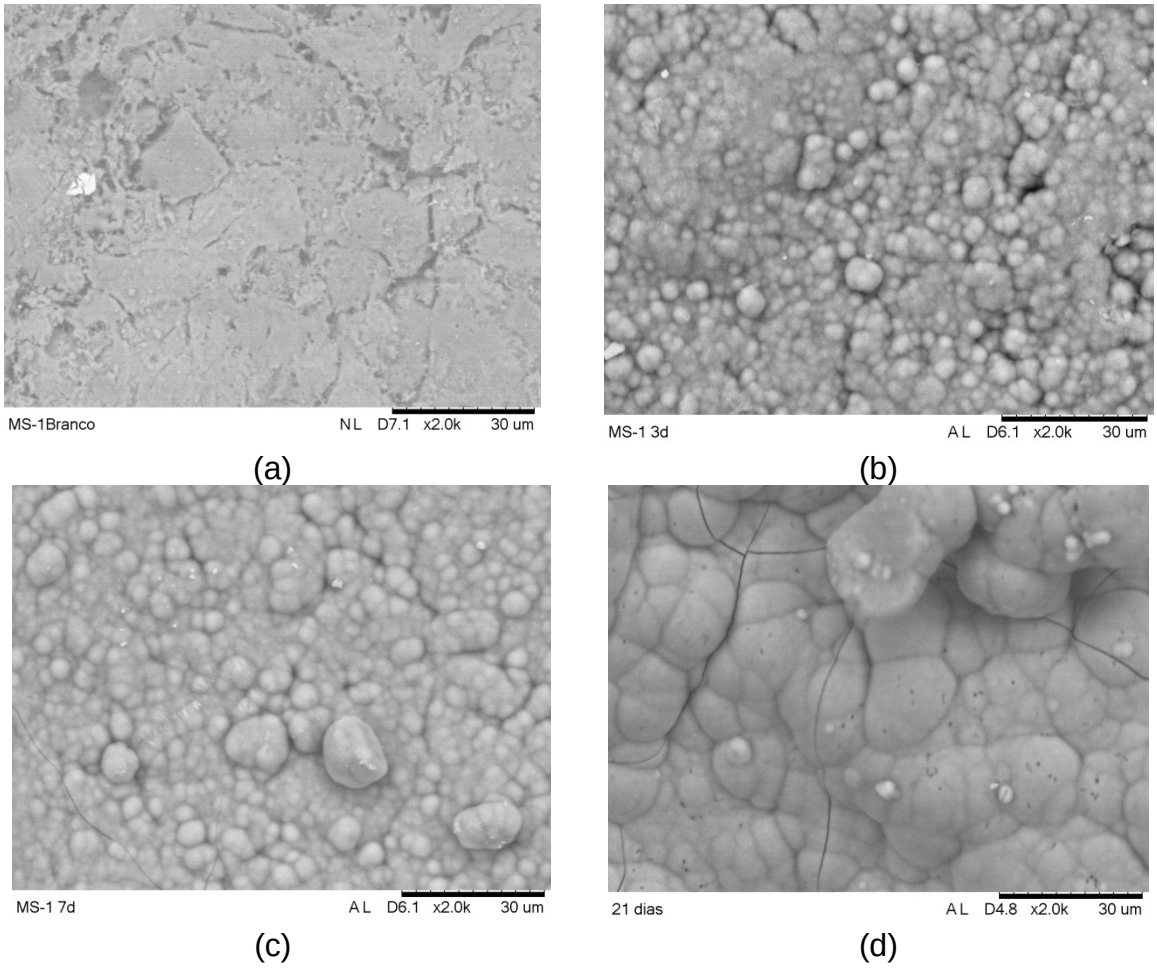

Figure 4: CaO-MgO-SiO_2 ceramic body before (a) and after 3 days (b), 7 days (c) and 21 days (d) soaked in SBF solution.

The FTIR spectra in Figure 5 display phosphate absorption band at 1020 cm^{-1} on spectra of the samples soaked for 3, 7 and 21 days, which indicates the hydroxyapatite layer on the sample surface of this ceramic pellets.

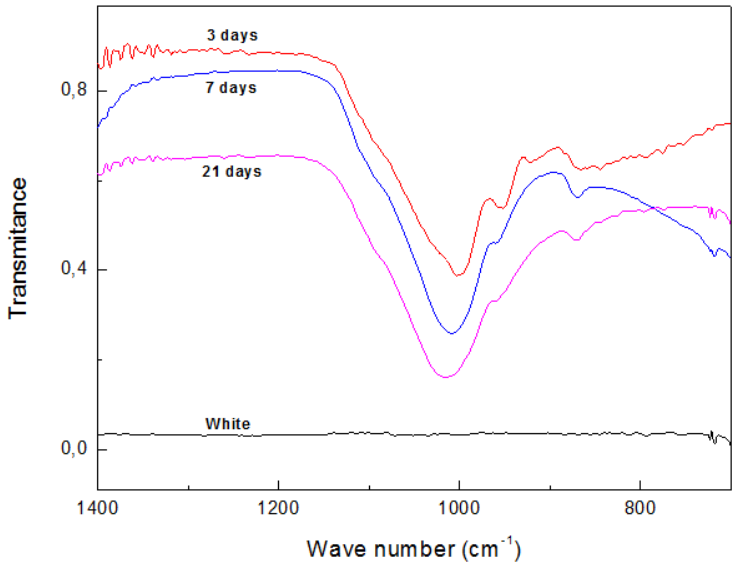

Figure 5: FTIR spectra showing phosphate absorption band at 1020 cm^{-1} on spectra of the samples soaked for 3, 7 and 21 days.

The ceramic body didn't show cytotoxicity as confirmed in Figure 6, because the cell viability of non-diluted extract (MS-1) is approximately 100% and for this reason was considered non cytotoxic.

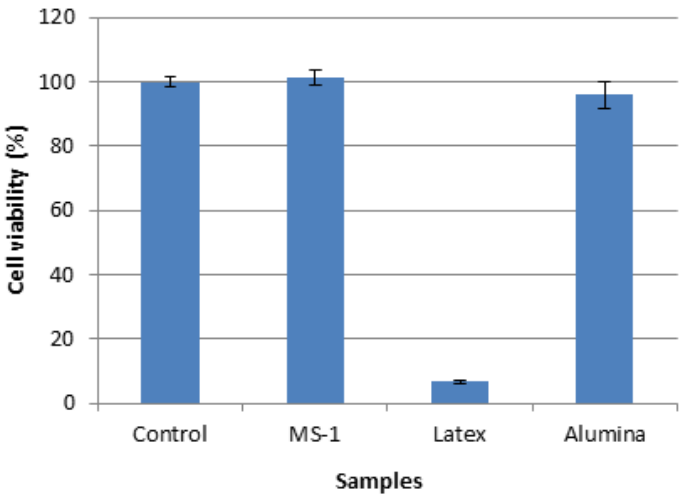

Figure 6: The cell viability of non-diluted extract (MS-1)

Conclusion

The results demonstrated that solution of Na_2SiO_3 and chloride solutions of calcium and magnesium produced a ceramic powder of good quality, when using the sol-gel method combined with co-precipitation for the synthesis. The ceramic body

calcined at 900°C for 1h and sintered at 1200°C for 2h presents a multiphase composition, with predominance of diopside phase.

The *in vitro* test performed on the SBF solution has demonstrated that a hydroxyapatite surface layer growth after three days, which reveals high ceramic bioactivity during interfacial bonding. The ISO 10993-5 reveals that cell viability of extract used in the indirect method should be higher than 70%, to be considered non-cytotoxic. The *in vitro* test demonstrated that ceramic body has a higher value and therefore can be considered non-cytotoxic.

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