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In vitro bioactivity assessment and mechanical properties of novel calcium titanate/borosilicate glass composites

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Abstract

Bioactive calcium titanate/borosilicate glass composites were developed. Powder mixtures of borosilicate glass and 10, 20 or 30 wt% of potassium polytitanate particles were uniaxially pressed and sintered at 850 °C for 1 h. After heat treatment the reaction between potassium polytitanate and borosilicate glass produced composites consisting of calcium titanate particles embedded in a B-rich amorphous phase. For the *in vitro* bioactivity assessment sintered samples were immersed in a simulated body fluid (SBF) for 21 days under physiological conditions of pH and temperature. The compressive strength of the composites was also evaluated. A homogeneous and thick apatite layer was formed on all the materials tested. Furthermore, an appropriate compressive strength was observed (68–85 MPa). These results indicate that these composites are potential materials for bone tissue replacement and regeneration.

Keywords: Borosilicate glasses; Potassium titanate; Calcium titanate; Apatite formation; Bioactive composites

1. Introduction

Since Bioglass® was discovered, a great quantity of bioactive materials (glasses, glass–ceramics, ceramics and composites) has been investigated [1–3]. As it is well known, bioactive systems exhibit the ability to bond the living bone through an apatite layer formed on their surfaces when they are in contact with real or simulated body fluid.

The main advantage of bioactive composites is that, as they are composed of two phases of different nature, each of the phases may provide different functional or structural properties to those of the starting materials [4]. Among such materials it is possible to note inorganic composites consisting of stainless steel fibers embedded in a Bioglass® matrix [1], bioactive glass ceramic toughened by zirconia particles [5], hydroxyapatite/diopside/alumina composite [6], bioactive glass (45S5)/titania composites [7], calcium titanate/titania coating composite [8], etc.

Using glass–ceramic technology, the effect of reinforcement is supported by different, preferably fiber-shaped, crystalline phases, which can be formed during the thermal treatment and partial crystallization of an original glass; whereas the same effect can be achieved with ceramic technology by sintering the compacted mixtures of powdered glass and reinforcement material.

On the other hand, it is necessary to note that high mechanical strength usually is typical for non-porous bulk materials, characterized with relatively small interface and, as a result, low bioactivity properties. Whereas, good bioactive properties are characteristic for the porous glass–ceramic composites representing relatively low mechanical strength [9,10].

The goal of this research is to establish a new binary system leading to the formation of glass–ceramic composites with both high mechanical and bioactive properties. The glass composition and reinforcement were selected taking into account the following reasons: the borosilicate glasses have attractive characteristics for biomaterial applications [11,12] and the total conversion of some borosilicate glasses into calcium phosphate without silica remaining into human body has been reported [13]. Furthermore, a rapid hydroxyapatite formation mechanism has

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Table 1
Ionic concentration of simulated body fluids and human blood plasma.

	Ion concentration (mM)							
	Na^+	K^+	Ca^{2+}	Mg^{2+}	Cl^-	HCO_3^-	HPO_4^{2-}	SO_4^{2-}
Human blood plasma	142.0	5.0	2.5	1.5	103.0	27.0	1.0	0.5
SBF	142.0	5.0	2.5	1.5	147.8	4.2	1.0	0.5

been observed when a special borosilicate glass is immersed in a simulated body fluid (SBF) [14].

The potassium polytitanate, obtained by molten salt method [15], was selected as another component of the system. This new precursor material of $K_2O\cdot nTiO_2$ chemical composition has semi-crystalline or amorphous structure; however, an additional heat treatment at temperatures higher of 800–850 °C promotes its crystallization obtaining fiber-shaped potassium tetratitanate or hexatitanate depending on the value of $n = TiO_2/K_2O$ in the original composition.

Before processing, the particles of the potassium polytitanate have spherical shape with a diameter of 20–30 μm and, unlike the fiber-shaped crystalline potassium titanates, can be easily compacted to produce green body of ceramic composite materials. Particles of $K_2Ti_4O_9$ and $K_2Ti_6O_{13}$ have demonstrated to possess high mechanical properties (an elasticity modulus close to that of the theoretical value, tensile strength about of 6 GPa [16]) and are very attractive as a reinforcement for different types of matrix materials [17]. In addition, it was shown in our previous research publication [18] that crystallization of the potassium polytitanate was accompanied with a formation of meso-porous structure which could be attractive for bone tissue regeneration. However, glass–ceramic composites based on potassium titanate have not been investigated for bone replacement applications.

2. Materials and methods

The borosilicate glass [13], used as a vitreous component to produce glass–ceramic composite, was prepared using $CaCO_3$, $NaCO_3$, $NH_4H_2PO_4$, H_3BO_3, Al_2O_3 and SiO_2 as raw materials. The homogenized raw materials mixture was melted in the platinum crucible at 1400 °C for 2 h and the obtained melt was quenched onto a stainless steel plate at room temperature. The glass was crushed and milled to obtain a powder (fraction of 100–325 μm). The obtained glass had the following chemical composition (wt%): SiO_2 (9.8), Al_2O_3 (1.9), P_2O_5 (3.6), CaO (44.0), Na_2O (3.0) and B_2O_3 (37.4).

The potassium polytitanate used as a precursor material to form crystalline fiber-shaped titanate was synthesized according to the stated in the literature [15], that is by heat treatment of the mixture, consisting of TiO_2 powder (anatase, Aldrich, 99%), KOH (Aldrich, 99%) and KNO_3 (Aldrich 98%) with a weight percent relation of 1:1:8, respectively. The heat treatment was performed at 500 °C for 2 h followed by washing with distilled water and drying at 60 °C for 4 h. The powdered product of the synthesis showed a molar ratio of $TiO_2/K_2O \sim 5.3$ according to energy dispersive spectroscopy (EDS) analysis.

The raw material mixtures used in the experiments contained 10, 20 and 30 (wt%) of potassium polytitanate. The products obtained by sintering of these mixtures were identified as 10HTK-90VBC1, 20HTK-80VBC1 and 30HTK-70VBC1. Green bodies of the composites were produced by uniaxial compressing to obtain the disks (weight of about 0.5 g), which were sintered at 850 °C for 1 h. The obtained specimens were investigated using X-ray diffraction (XRD, Philips PW3040), scanning electron microscopy (SEM, Philips XL30) and EDS.

The *in vitro* bioactivity assessment of the glass–ceramic composites was evaluated by immersing samples in 250 ml of simulated body fluid (SBF-K9) at 37 °C for 21 days. The SBF solution was prepared according to the procedure described by Kokubo et al. [19] by dissolving the following reagent-grade chemicals: NaCl, $NaHCO_3$, KCl, $K_2HPO_4\cdot 3H_2O$, $MgCl_2\cdot 6H_2O$, $CaCl_2\cdot 2H_2O$ and Na_2SO_4 into deionized water (1 dm^3) and buffered to pH 7.4 with tris(hydroxymethyl)-aminomethane and 1N HCl at 37 °C. The ion concentration of SBF is presented in Table 1.

The solubility of the samples was estimated by measuring the weight losses of the specimens (disks) after different times of immersion in SBF. The procedure was performed by taking out the samples from the flasks containing the SBF; then, the samples were dried in an oven for 1 h at 100 °C in order to eliminate the remaining SBF into the pores. Once the samples were dried, they were weighted. Three weight loss measurements were performed for each period of time and for each type of sample [14]. In addition, pH of SBF was also recorded for different periods of immersion and the content of boron, calcium and phosphate in the remaining SBF solutions (after 21 days of immersion of the samples) was evaluated by inductively coupled plasma spectrometry (ICP-IRIS Intrepid II XSP).

The compressive strength of sintered materials was determined according to the ASTM-C773 standard. The size of the samples was 8 mm in diameter and 11 mm in height. Six samples for each material were tested and the compressive load was applied at a cross-head speed of 0.5 mm/min.

3. Results and discussion

The XRD patterns of the samples after sintering at 850 °C for 1 h are shown in Fig. 1. Considerable changes were observed; these phase transformations indicate a high chemical interaction between the calcium and boron oxides of the glass and the potassium polytitanate particles. The change of phases was presented in all the cases, the reaction products of the chemical interaction were identified as tetragonal calcium

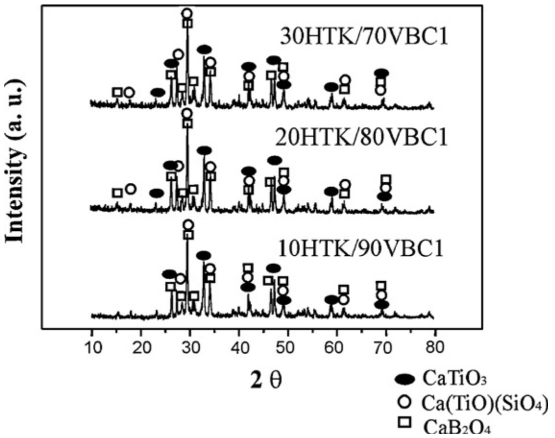

Fig. 1. XRD patterns of samples obtained after sintering at 850 °C for 1 h.

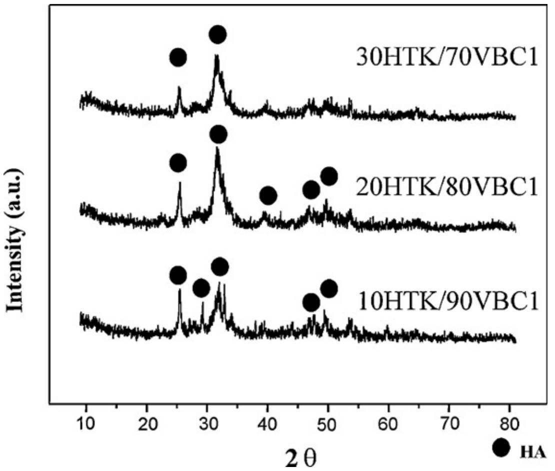

Fig. 2. XRD patterns of the 10HTK-90BVC1, 20HTK-80BVC1, 30HTK-70BVC1 composites after 21 days of immersion in (SBF).

titanate (main phase), which has been studied for bioactivity purposes [8], and titanite Ca(TiO)SiO$_4$ and calcium metaborate CaB$_2$O$_4$ in a lower content. The results suggest that an ionic exchange between two atoms of similar size Ca and K was produced into the crystalline structure during the sintering.

After sintering, the samples were immersed in SBF for 21 days in order to assess bioactivity. Fig. 2 shows the XRD patterns of the surfaces after this period of immersion in SBF. Hydroxyapatite (HA) was formed on all the composites (adding a quantity of 10–30 wt% of potassium polytitanate particles). These results indicate a high bioactivity, since no peaks corresponding to the substrate were detected by XRD.

Fig. 3 shows SEM images of the surface and cross-section of samples prepared by adding 20 wt% of potassium polytitanate after 21 days of immersion in SBF. A homogeneous and thick ceramic layer (Fig. 3A and B), which morphology closely resembles that formed on the highly bioactive existing systems, has been formed. The EDS spectrum (Fig. 3D), which corresponds to the sample shown in Fig. 3B, clearly indicates that this newly formed layer is rich in Ca and P. As observed in the cross-section analysis (Fig. 3C), an irregular in thickness, but yet a continuous layer was formed on the samples after immersion in SBF. The preferential sites for apatite nucleation were the calcium titanate particles, which leaded to the

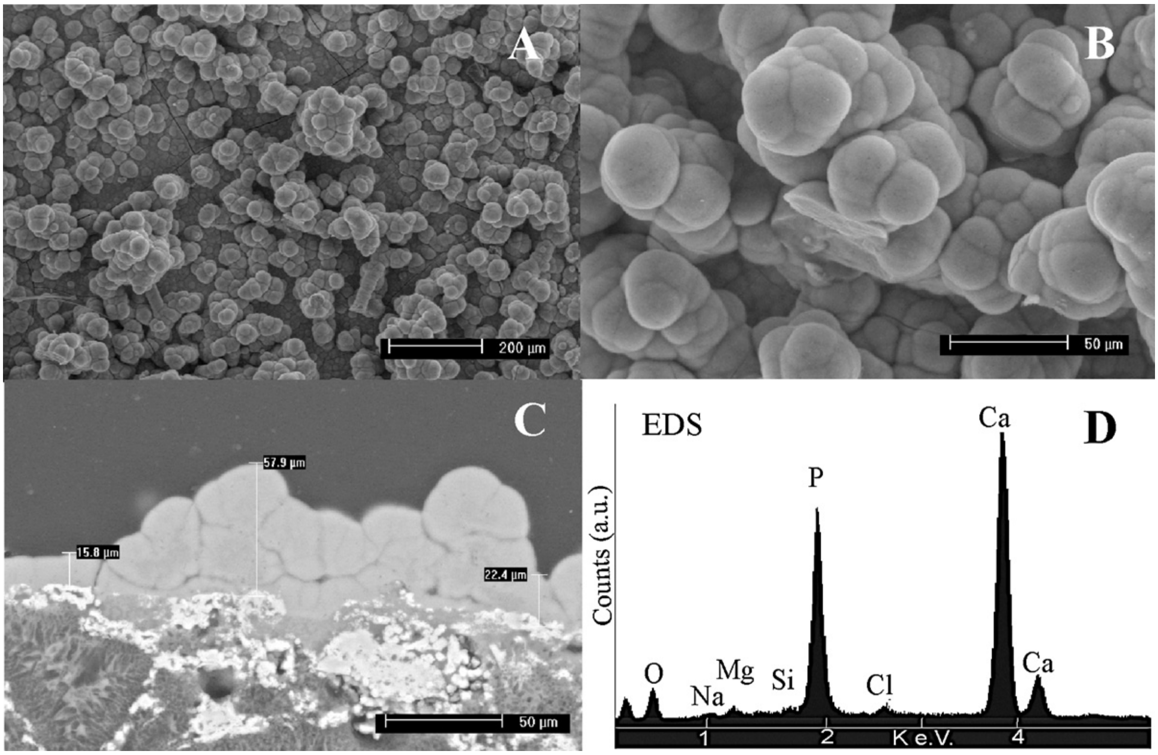

Fig. 3. SEM images of the 20HTK-80BVC1 composite after 21 days of immersion in SBF: (A, B) surface, (C) cross-section, (D) EDS spectrum corresponding the sample shown in (B).

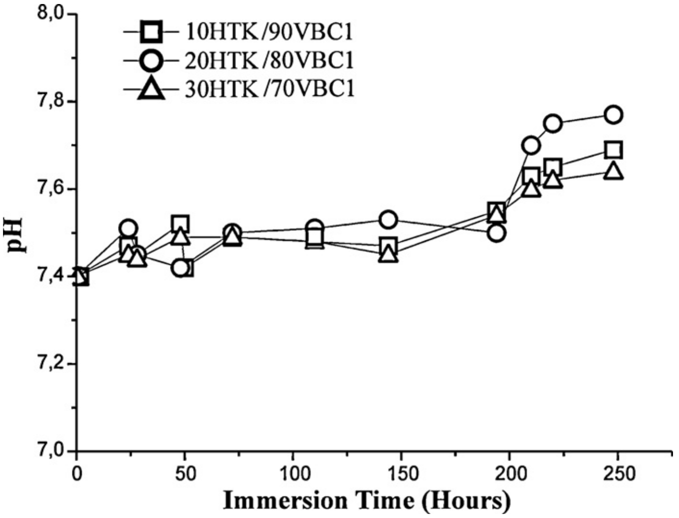

Fig. 4. Variation of pH in SBFs with time during the first 7 days.

formation of a dense and thick HA layer with a maximum thickness of around 60 μm. This layer consisted of apatite agglomerates of about 20 μm in diameter. The analysis of the cross-section shows also an intimate bond between the HA layer and the substrate.

Regarding the pH of the remaining SBF (Fig. 4), this value increases up to approximately 7.8 after 7 days in all the cases. A weight loss lower than 6% was observed (Fig. 5), this result is considerable lower than that presented in the literature for a specific borosilicate glass with no other components or phases in the microstructure (40% of weight loss) [14]. The behavior of pH of the remaining SBFs as a function of immersion time indicates an ionic exchange between the fluid and the glass which, according to the SEM analyses (Fig. 3) a partial and irregular dissolution of glassy phase takes place. However, after 7 days of immersion in SBF the weight loss of the composites become stable, this may be due to the formation of the bioactive layer, which decreases the dissolution of the glass since apatite acts as a protective coating. The results obtained by ICP analysis of the remaining SBFs show that the Ca and B concentration increases, mainly during the first 7 days of immersion, while that of P decreases in this period of time, Fig. 6. This may indicate that the first week of immersion is the main stage of the mechanism for apatite formation. According to the results described above, the mechanism for apatite formation on this particular kind of composites may take place due to the ionic exchange of Ca and B of the substrate with H^+ of the surrounding SBF, this ionic exchange increases the supersaturation degree of the fluid with respect to apatite. Meanwhile, as stated in the literature [20], the calcium titanate particles act as preferential sites for apatite nucleation due to the formation of Ti–OH groups on the particles surface.

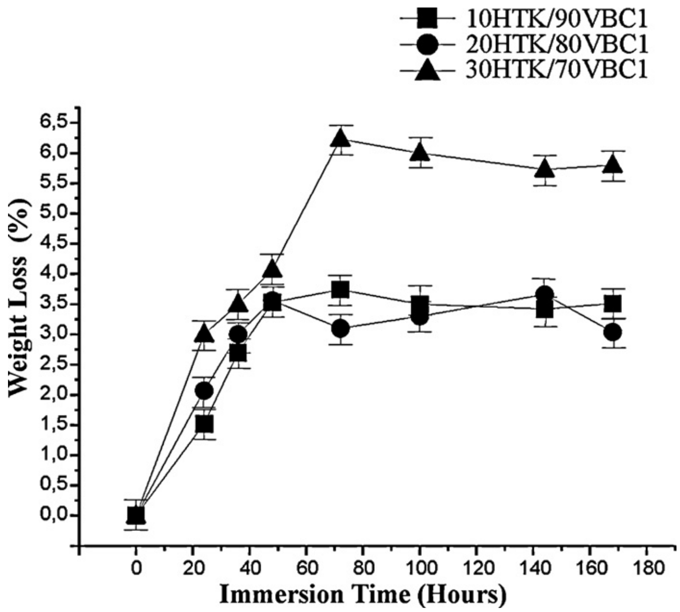

Fig. 5. Percentage weight loss of samples as a function of time.

The compressive strength of the sintered composites is presented in Fig. 7, where a slight decrease in this mechanical property is observed as the potassium titanate particles added is increased. This may be due to the fact that a higher porosity was observed as the quantity of crystalline particles was increased. In any case, an appropriate compressive strength was observed in all the cases (from 68 to 85 MPa). These results, including

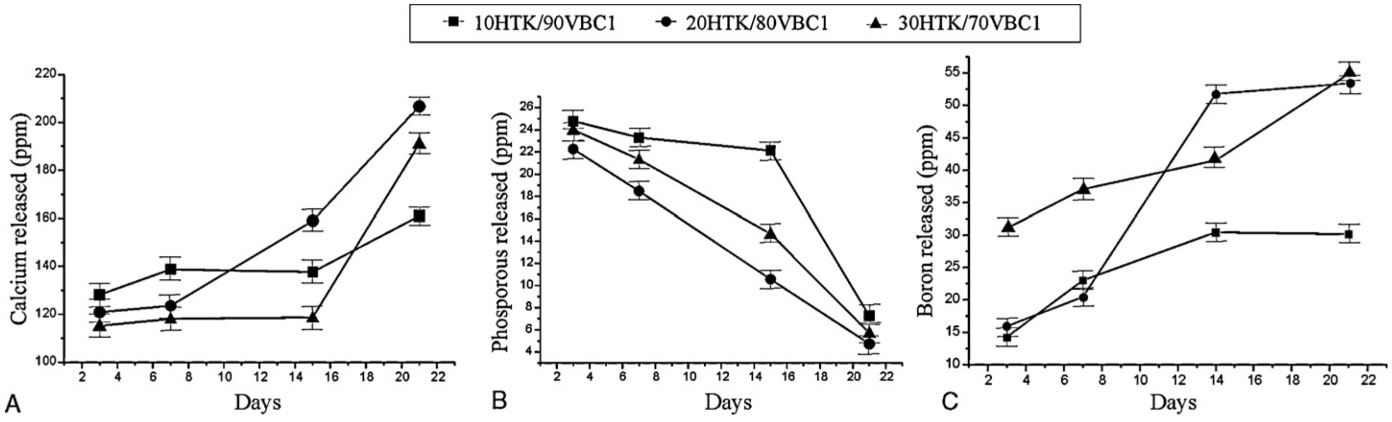

Fig. 6. Ions released of calcium (A), phosphorous (B) and boron (C) in SBFs remaining at 7, 14 and 21 days of immersion time.

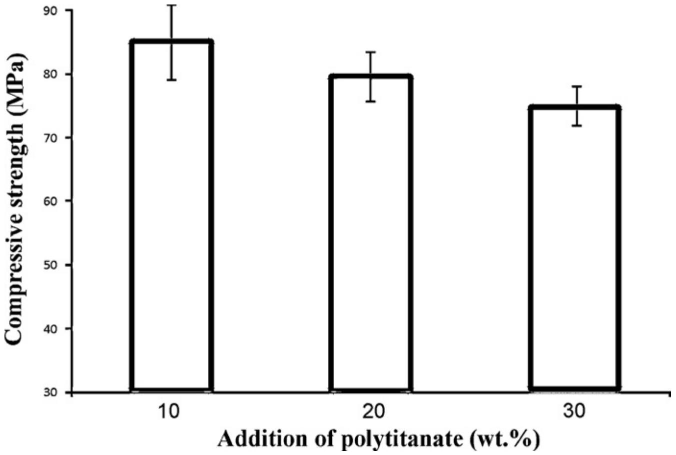

Fig. 7. Compressive strength of samples 10HTK-90BVC1, 20HTK-80BVC1, 30HTK-70BVC1.

those that demonstrate a high *in vitro* bioactivity of the materials tested, make of these composites potential and attractive materials for bone tissue regeneration.

4. Conclusions

Novel bioactive composites, prepared by mixing potassium titanate particles with a specific borosilicate glass, were developed. After sintering powder mixtures of 10, 20 and 30 wt% of potassium polytitanate particles with borosilicate glass at 850 °C for 1 h, different phases were formed, mainly calcium titanate and titanite which appeared to be embedded in a B-rich glassy phase. A continuous and thick apatite layer was formed on the sintered composites after immersing them in simulated body fluid for 21 days. In addition to their high *in vitro* bioactivity, the composites showed an appropriate compressive strength.

References

[1] L.L. Hench, Bioceramics, J. Am. Ceram. Soc. 82 (1998) 1705–1728.
[2] S.M. Best, A.E. Porter, E.S. Thian, J. Huang, Bioceramics: past, present and for the future, J. Eur. Ceram. Soc. 28 (2008) 1319–1327.
[3] S. Agathopoulus, D.U. Tulyaganov, J.M.G. Ventura, S. Kannan, M.A. Karakassides, J.M.G. Ferreira, Formation of hydroxyapatite onto glasses of the CaO–MgO–SiO_2 system with B_2O_3, Na_2O, CaF_2 and P_2O_5 additives, Biomaterials 27 (2006) 1832–1840.
[4] ASM International, "Ceramic and Glasses", Engineered Materials Handbook, ASM International, USA, 1991.
[5] E. Verne, R. Defilippi, G. Carl, C. Vitale Brovarone, P. Appendino, Viscous flow sintering of bioactive glass–ceramic composites toughened by zirconia particles, J. Eur. Ceram. Soc. 23 (2003) 675–683.
[6] Z. Xihua, L. Changxia, L. Musen, B. Yunqiang, S. Junlong, Fabrication of hydroxyapatite/diopside/alumina composites by hot-press sintering process, Ceram. Int. 35 (2009) 1969–1973.
[7] H.H. Beherei, K.R. Mohamed, G.T. El-Bassyouni, Fabrication and characterization of bioactive glass (45S5)/titania biocomposites, Ceram. Int. 35 (2009) 1991–1997.
[8] D. Wei, Y. Zhou, D. Jia, Y. Wang, Structure of calcium titanate/titania bioceramic composite coatings on titanium alloy and apatite deposition on their surfaces in a simulated body fluid, Surf. Coat. Technol. 201 (2007) 8715–8722.
[9] D.W. Hutmacher, J.T. Schantz, Ch.X.F. Lam, K.Ch. Tan, T.Ch. Lim, State of the art and future directions of scaffold-based bone engineering from biomaterials perspective, J. Tissue Eng. Regen. Med. 1 (2007) 245–260.
[10] R.C.C. Monteiro, M.M.R.A. Lima, Effect of compaction on the sintering of borosilicate glass/alumina composites, J. Eur. Ceram. Soc. 23 (2003) 1813–1818.
[11] W. Liang, M.N. Rahman, D.E. Day, N.W. Marion, G.C. Riley, Bioactive borate glass scaffold for bone tissue engineering, J. Non-Cryst. Solids 354 (2008) 1690–1696.
[12] A. Yao, D. Wang, W. Huang, Q. Fu, M.N. Rahaman, D.E. Day, *In vitro* bioactive characteristics of borate based glasses with controllable degradation behavior, J. Am. Ceram. Soc. 90 (2007) 303–306.
[13] Y. Li, M.N. Rahaman, Q. Fu, B.S. Bal, A. Yao, D.E. Day, Conversion of bioactive borosilicate glass to multilayered hydroxyapatite in dilute phosphate solution, J. Am. Ceram. Soc. 90 (2007) 3804–3810.
[14] M. Kulwant, S. Thind, G. Sharma, K. Singh, V. Rajendran, S. Aravindan, Soluble borate gasses: in vitro analysis, J. Am. Ceram. Soc. 90 (2007) 467–471.
[15] T. Sanchez-Monjaras, A. Gorokhovsky, J.I. Escalante-García, Molten salt synthesis and characterization of potassium polytitanate ceramic precursors with varied TiO_2/K_2O molar ratios, J. Am. Ceram. Soc. 91 (2008) 3058–3065.
[16] N. Bao, X. Feng, X. Lu, Z. Yang, Study on the formation and growth of potassium titanate particles, J. Mater. Sci. 37 (2007) 3035–3043.
[17] D.Y. Ding, J.N. Wang, Interfacial reactions in $K_2Ti_6O_{13}$ whisker/Al_2O_3/Al composite, J. Mater. Sci. Technol. 17 (2001) 833–836.
[18] M.A. Aguilar González, A. Gorokhovsky, A. Aguilar-Elquezabal, J.I. Escalante García, Síntesis y caracterización de adsorbentes cerámicos basados en polititanatos de potasio y vidrio SiO_2–B_2O_3–R_2O–Al_2O_3, Bol. Soc. Esp. Ceram. Vidr. 47 (2008) 29–34.
[19] T. Kokubo, H. Kuchitaniand, S. Sakka, Solution able to reproduce in vivo surface structure changes in bioactive glass ceramic A-W, J. Biomed. Mater. Res. 24 (1990) 721–734.
[20] Kokubo, T. Matsushita, H. Takadama, Titania-based bioactive materials, J. Eur. Ceram. Soc. 27 (2007) 1553–1558.