JOURNAL OF MATERIALS SCIENCE: MATERIALS IN MEDICINE 5 (1994) 529–532

In vitro formation of hydroxylapatite layer in a MgO-containing glass

J. S. MOYA
Instituto de Cerámica y Vidrio, CSIC, Arganda del Rey Madrid, Spain

A. P. TOMSIA
Department of Material Science and Mineral Engineering, L.B.L, University of California, Berkeley, USA

A. PAZO, C. SANTOS, F. GUITIÁN
Instituto de Cerámica, Universidad de Santiago, Santiago, Spain

In the present article, the behaviour of a biologically active glass (BAG) belonging to the system SiO_2–CaO–MgO–Na_2O–K_2O–P_2O_5, soaked in simulated body fluids (SBF_S), is studied. The amount of Ca, Mg and K leached from the glass, as well as variations in P concentration and pH of the solution were measured versus time. The formation of a hydroxylapatite (HA) layer on the glass surface was determined by XRD and SEM-EDX. The article reports the influence of pH variation of the medium on the crystallinity of the HA formed.

1. Introduction

Since the discovery of biologically active glasses (BAG) by Hench [1] in 1970, very active research work has been carried out in the last few years to understand the mechanism and parameters that control glass–bone bonding.

Nowadays it is generally accepted that the formation of a hydroxylapatite (HA) layer on the glass is a necessary condition to ensure a direct bone bonding [2].

Bioactive glasses and glass-ceramics belonging to the Na_2O–CaO–SiO_2–P_2O_5 system have been extensively studied by Hench [1]. Glass-ceramics containing apatite and wollastonite (A–W), in the SiO_2–CaO–MgO–P_2O_5 system have recently been investigated [3]. However, no available literature has been found directly related to glasses belonging to the SiO_2–CaO–MgO–Na_2O–K_2O–P_2O_5 system. Recently, the present authors [4] have shown that glasses belonging to the mentioned system can be used to coat Ti-based implants because of the formation of strong metal–glass bonding through a TiN layer formed at 1000 °C in N_2 atmosphere [5].

The aim of the present investigation is to study the *in vitro* formation of an HA layer on the surface of a glass belonging to the mentioned six-components system.

2. Materials and methods

A glass with the following composition (wt %): SiO_2 54.5, Na_2O 12.0, K_2O 4.0, CaO 15.0, MgO 8.5, P_2O_5 6.0, was used in the present investigation.

The glass was prepared starting from silica sand (SiO_2 99.9%) and reagent grade Na_2HPO_4, Na_2CO_3, K_2CO_3, and MgO. A homogeneous mixture was obtained by stirring the ingredients in isopropil alcohol. The powder mixture, after drying (110 °C, 48 h) and prefiring at 400 °C, 3 h, was fired in a Pt crucible at 1500 °C, for 4 h. The corresponding melt was poured into distilled water at room temperature. The glass fragments were then ball-milled to a particle size of < 5 μm.

Glass powder was melted at 1400 °C in a Pt crucible. After this, transparent and pore-free glass specimens with $10 \times 10 \times 10$ mm dimensions were obtained by casting the melt in a bronze mould, which had not been preheated.

Six glass plates of $10 \times 10 \times 1$ mm dimensions with polished surfaces obtained by finishing with alumina (0.5 μm) were soaked for different times in simulated body fluid (SBF) with ion concentration nearly equal to those of human blood. The ratio of solution to the surface area of the plates was 0.01 $cm^2\ ml^{-1}$. The plates were removed from the solution, washed with distilled water and air dried.

Glass powder with average particle size 20 μm and specific surface area 0.1 $m^2\ g^{-1}$ was obtained by grinding the original glass blocks in an agata mortar. 2 g of this powder were soaked in SBF with and without Mg (Table I). The temperature of the SBF was kept constant at 36.5 °C.

The concentration of Ca, Mg, and K leached from the glass were determined by atomic absorption spectroscopy (AAS). The pH variation of the solution was also measured versus time. After each determination, the suspensions were regenerated with SBF to the initial level; these corrections were taken into account for calculations.

The films formed on the glass plate after different soaking times were characterized by thin-film X-ray diffraction (XRD). Conventional XRD analyses were

TABLE I Chemical composition of simulated body fluids (SBF) I and II

	Na^+	K^+	Mg^{+2}	Ca^{+2}	Cl^-	CO_3H^-	PO_4H^{-2}
SBF I	142	5	1.5	2.5	103	13.4	1
SBF II	142	5	–	4	101.5	13.4	1

Ion Concentrations are expressed in mM

performed with the powder glass after different soaking times. The microstructure of the surface and the fracture surface of the reacted plate after 20 days soaking were observed by scanning electron microscopy with energy-dispersive X-ray spectrometer (SEM-EDX).

3. Results

Fig. 1 shows the leaching of Ca, Mg and K as a function of soaking time of glass plate and powder in SBF. As is shown, a similar trend is observed in both cases. In the first stage of soaking (3 days) an increase in concentration of the cations in the solution is detected. In the case of Ca, a depletion in the concentration occurs after these 3 days of soaking. Mg and K concentration becomes stabilized after 4–6 days soaking. As can be seen in Fig. 2, P content in the solution drastically decreases after 3 days of glass soaking.

In Fig. 3, pH versus soaking time plots corresponding to the glass powder in the two SBF fluids and those corresponding to the glass plate are shown. A pH value of 9 is reached after 4 days of soaking in the case of powder glass soaked in free Mg SBF. In contrast, the increase in pH value was less significant for powder and plate soaked in SBF containing Mg.

XRD patterns corresponding to the glass powder and the plate after 7 days of soaking in SBF I (with Mg), are shown in Fig. 4. For comparative purposes, XRD patterns corresponding to a well-crystallized synthetic HA powder and human bone HA are also included. As can be seen, HA formed on the surface of the glass powder exhibits higher crystallinity than HA developed on the glass plate surface. This pattern is quite similar to that corresponding to human bone HA.

The SEM micrograph of the fracture surface of a glass plate, together with different EDX microanalysis of selected areas, are shown in Fig. 5. A P–Ca-rich layer of 10 μm thickness is observed at the outer surface of the glass plate. This layer is separated from the bulk by a crack. An intermediate SiO_2-rich layer has been also detected.

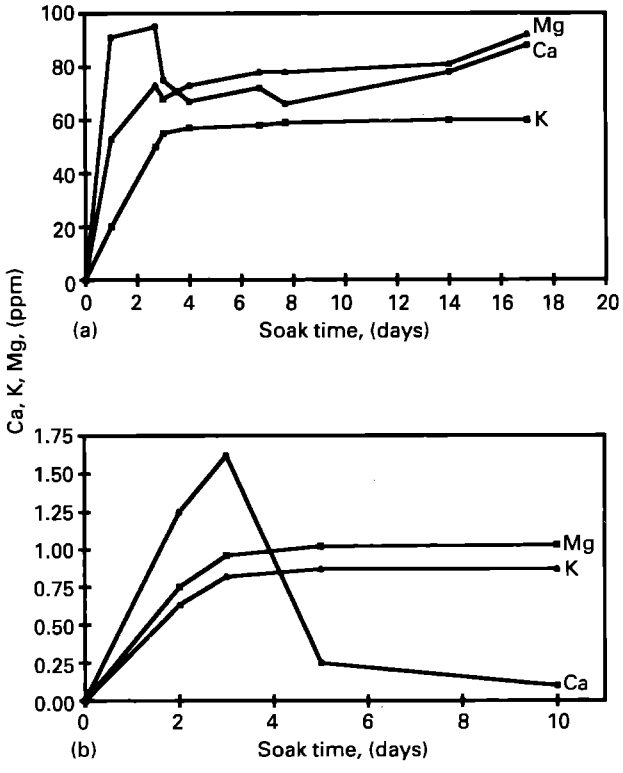

Figure 1 Variation of element concentration in SBF I versus glass soaking time. Rate glass/SBF = 1/100 (a) glass powder; (b) glass plate.

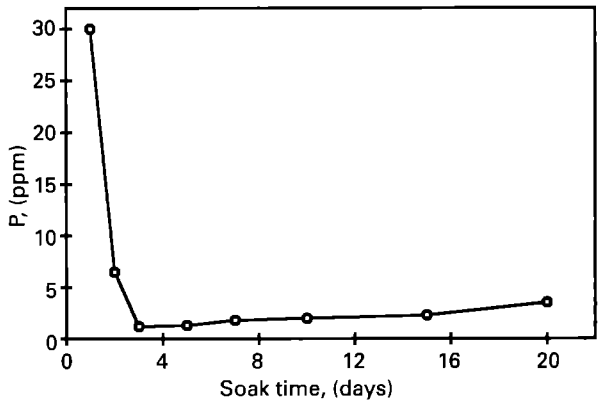

Figure 2 Evolution of P concentration in SBF I versus glass powder soaking time.

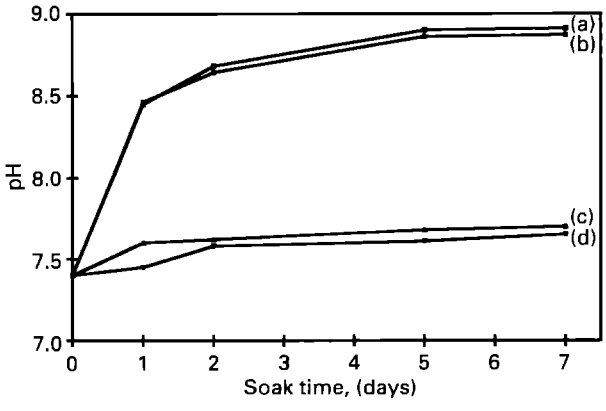

Figure 3 pH variation of SBFs versus glass soaking time: (a) glass powder/SBF II; (b) glass powder/SBF I; (c) glass plate/SBF II; (d) glass plate/SBF I.

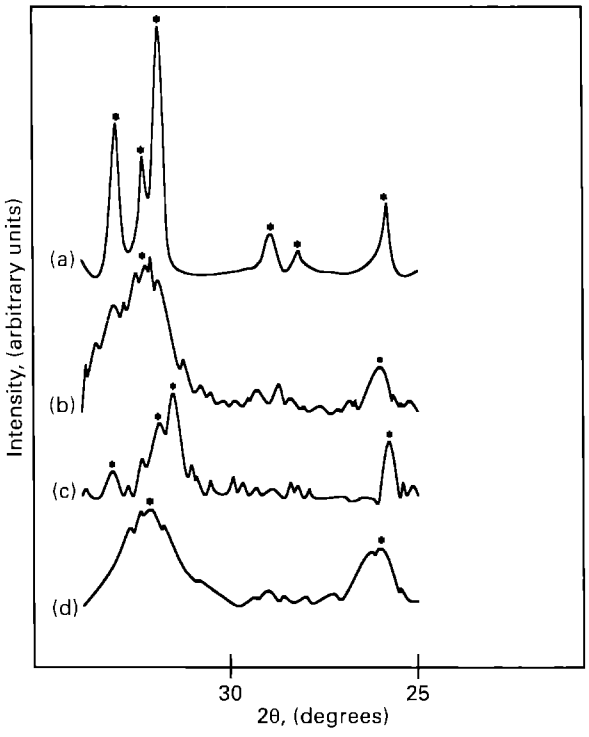

Figure 4 Comparison of XRD patterns corresponding to glass soaked in SBF I for 7 days and those corresponding to synthetic and human bone HA, *hydroxylapatite (a) synthetic HA; (b) human bone HA; (c) glass powder; (d) glass plate.

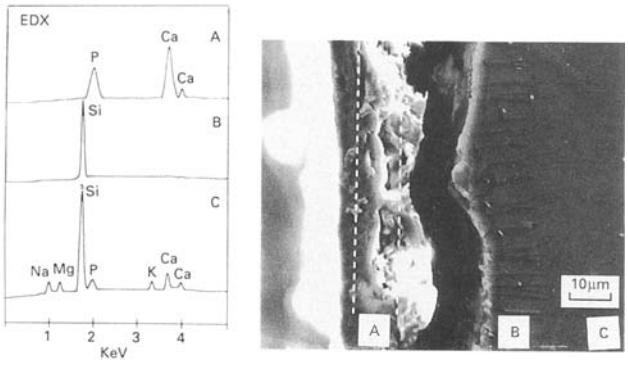

Figure 5 SEM micrographs and EDX spectra of selected areas of the fracture surface of the glass plate after 20 days soaking in SBF I.

The SEM-EDX study of the surface layer is presented in Fig. 6. From this study the following points can be deduced:

(i) The P–Ca-rich layer presents cracking, probably induced by differential shrinkage during drying.

(ii) The microstructure of this layer is in the form of rounded low-crystallinity particles, closely packed.

(iii) Immediatly below the P–Ca-rich layer, a silica-gel-like layer is located. In several areas of this layer some HA needle-like crystals (0.1 μm) were precipitated, adopting the morphology of spheroidal clusters.

4. Discussion

The *in vitro* formation of an HA layer on the surface of a SiO_2–MgO–CaO–P_2O_5–Na_2O–K_2O glass occurs by a mechanism similar to those operating in glasses belonging to the SiO_2–P_2O_5–CaO–Na_2O system [1]

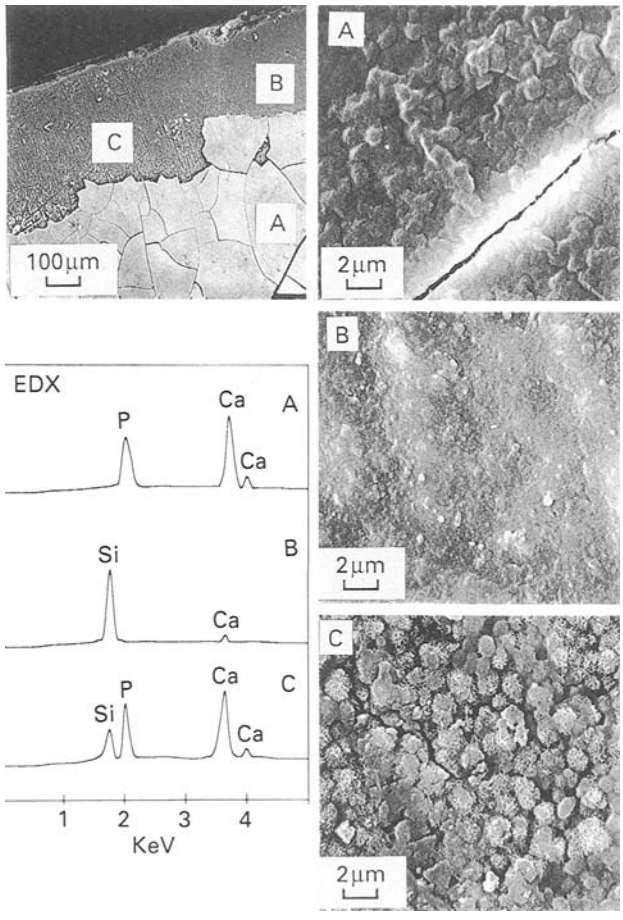

Figure 6 SEM micrographs and EDX spectra of selected areas corresponding to the surface of the glass plate after 20 days soaking in SBF I.

or in glass-ceramics with SiO_2–MgO–CaO–P_2O_5 composition [3]. The leaching of Ca, Mg and K (Fig.1) from the glass surface to the SBF produces the appearance of a silica-gel layer (Figs 5, 6) and the highly hydrated silica groups effectively induce heterogeneous nucleation of the HA, decreasing the concentration of Ca and P in the medium (Figs 1, 2).

In a recent investigation, Hisizawa *et al.* [6] claim that the formation of an HA layer on BAG is strongly influenced by the Mg^{++} content in SBF. These authors suggest that Mg^{++} condenses on the BAG surface and interferes with the ion exchange between H^+ and Na^+. As a consequence, the formation of an HA film is retarded or avoided due to an insufficient pH increase in the medium.

Our results clearly indicate that:

(i) The presence of Mg^{++} in SBF slightly depletes the pH of the medium (Fig. 3).

(ii) Even in the case of SBF with high Mg^{++} content (36 ppm) a well-developed HA layer was obtained.

According to our experiments, the pH value of the medium primarily affects the crystallinity of the HA layer, not its formation (Figs 3, 4).

The precipitation of needle-like crystals of HA on the silica-gel layer in the initial stage suggests that high pH values (> 10) were first reached near the silica layer. In a subsequent stage the crystallinity of

the HA layer decreases due to the lower average value of the pH in the medium.

(iii) No Mg^{++} condensation was detected on the glass surface after a long soaking time (20 days), as observed in the EDX spectra shown in Fig. 6.

In summary it can be stated that the MgO-containing glass studied in the present investigation behaves as a typical bioactive glass belonging to the SiO_2–CaO–Na_2O–P_2O_5 system. The role of Mg^{+2} in the formation of HA layer was found to be insignificant.

Finally, a new family of BAGs with a large range of thermal expansion coefficients can be tailored into the SiO_2–CaO–MgO–Na_2O–K_2O–P_2O_5 system. This fact gives rise to the possibility of selecting mechanically suitable glass–metal composities for future implant applications.

References

1. L. L. HENCH, T. K. GREENLEE JR. W. C. ALLEN and G. PIOTROWSKI, "An investigation of bonding mechanism at the interface of a prosthetic material", Report, No. 1, Contract No. DADA 17-70-C-0001, U.S. Army Research and Development Command; Clearinghouse for Federal Scientific and Technical Information, Washington, D.C., 1970.
2. L. L. HENCH, in "Handbook of bioactive ceramics, Vol. I, bioactive glasses and glass-ceramics", edited by T. Yamamuro, L. L. Hench and J. Wilson (CRC Press, Boca Raton, FL, 1990).
3. T. KOKUBO, "Handbook of bioactive ceramics, Vol. I, Bioactive glasses and glass-ceramics", edited by T. Yamamuro, L. L. Hench and J. Wilson (CRC Press, Boca Raton, FL, 1990).
4. A. P. TOMSIA, J. S. MOYA and F. GUITIAN, *Scrita Met. et Mater.* **31** (8), (1994) 995.
5. F. GUITIÁN, J. COUCEIRO, A. P. TOMSIA, J. S. MOYA, and S. DE AZA, Patent application 9200317 (14-2-92), Oficina Española de Patentes y Marcas.
6. H. ISHIZAWA, M. FUJINO and M. OGINO, "Handbook of bioactive ceramics, Vol. I, Bioactive glasses and glass-ceramics", edited by T. Yamamuro, L. L. Hench and J. Wilson (CRC Press, Boca Raton, FL, 1990).

Received 26 February and accepted 7 December 1993