Effects of ions dissolved from bioactive glass-ceramic on surface apatite formation

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Non-bioactive glass-ceramic A-W(Al) containing apatite and wollastonite in a $MgO–CaO–SiO_2–Al_2O_3$ glassy matrix did not form an apatite layer on its surface in a simulated body fluid with ion concentrations nearly equal to those of human blood plasma and also in the fluids with small amounts of the calcium and silicate ions added individually, but formed the apatite layer in the fluid with the calcium and silicate ions added simultaneously. This indicates that the calcium and silicate ions dissolved from bioactive glass-ceramic A-W containing the apatite and wollastonite in a $MgO–CaO–SiO_2$ glassy matrix play a cooperative and important role in forming an apatite layer on its surface in the body, to give the glass-ceramic bioactivity. The calcium ion might increase the degree of the supersaturation of the surrounding body fluid, and the silicate ion might provide favourable sites for nucleation of the apatite on the surfaces of glass-ceramic.

1. Introduction

Various kinds of bioactive glasses [1–4] and glass-ceramics [5–8] bond to living bone through an apatite layer which is formed on their surfaces in the body. This kind of apatite layer is not observed at the interface of non-bioactive glasses [3] and glass-ceramics [9] to the bone. This indicates that the essential condition for glasses and glass-ceramics to bond to living bone is formation of an apatite layer on their surfaces in the body.

The same kind of apatite layer can be formed on the surfaces of bioactive glasses and glass-ceramics even in an acellular simulated body fluid which has almost equal ion concentrations to those of the human blood plasma [10–14]. This means that the surface apatite layer is formed by a chemical reaction of the glasses and glass-ceramics with the surrounding body fluid. We have previously shown that glass-ceramic A-W containing crystalline apatite ($Ca_{10}(PO_4)_6(O,F_2)$) and β-wollastonite ($CaO \cdot SiO_2$) in a $MgO–CaO–SiO_2$ glassy matrix, glass-ceramic A containing only the apatite in a $MgO–CaO–SiO_2$ glassy matrix and a $MgO–CaO–SiO_2–P_2O_5$ glass G, all of which have the same nominal composition and are bioactive, dissolve appreciable amounts of the calcium and silicate ions into the simulated body fluid, whereas non-bioactive glass-ceramic A-W(Al) containing the apatite and β-wollastonite in a $MgO–CaO–SiO_2–Al_2O_3$ glassy matrix hardly dissolves these ions [15]. This suggests that the calcium and/or silicate ions dissolved from the bioactive glass and glass-ceramics play an important role in forming the surface apatite layer.

In the present study, to identify the important ion in forming the surface apatite layer, effects of ions added to the simulated body fluid on the formation of the surface apatite layer were investigated for some non-bioactive materials, including silica glass, glass-ceramic A-W(Al), sintered alumina and sintered zirconia, pure titanium and Ti6Al4V alloy. On the basis of the results, the mechanism of apatite formation of the surface of bioactive glass-ceramic A-W and its related glass and glass-ceramic is discussed.

2. Experimental

2.1 Samples

Seven kinds of non-bioactive materials of commercial fused silica glass (Central Glass Co.), glass-ceramic A-W(Al), commercial sintered alumina (Al_2O_3 99.5%, Murata Manufacturing Co. Ltd.), commercial sintered zirconia partially stabilized with 3 mol % Y_2O_3 (Murata Manufacturing Co. Ltd.), commercially pure titanium (Sumitomo Metal Industries Ltd.) and commercial Ti6Al4V alloy (Sumitomo Metal Industries Ltd.) were used in the present study. Glass-ceramic A-W(Al) contains 38 wt % of oxyapatite ($Ca_{10}(PO_4)_6O$) and 27 wt % of β-wollastonite ($CaO \cdot SiO_2$) in a 35 wt % of glassy matrix of the composition 10.3 MgO, 16.6 CaO, 55.0 SiO_2, 18.1 Al_2O_3 in wt % [15]. All of these materials neither bond to living bone nor form an apatite layer on their surfaces in the body.

They were cut into a rectangular piece $22 \times 4.0 \times 1$ mm^3 and polished with a diamond paste 1 μm in diameter.

2.2 Soaking in fluids

The specimens described above were washed with

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TABLE I Examined fluids

Fluid	Additive	(mM)				
SBF[a]	No					
SBF + Ca	Ca^{2+}	2.5				
SBF + P	P^{V}	1.0				
SBF + Si	Si^{IV}	1.8				
SBF + F	F^{-}	0.1				
SBF + Ca + Si	Ca^{2+}	2.5	Si^{IV}	1.8		
SBF + Ca + Si + Mg	Ca^{2+}	2.5	Si^{IV}	1.8	Mg^{2+}	3.0
SBF + Ca + Si + Al	Ca^{2+}	2.5	Si^{IV}	1.8	Al^{III}	0.19

[a] SBF Na^{+} 142.0, K^{+} 5.0, Mg^{2+} 1.5, Ca^{2+} 2.5, Cl^{-} 148.8, HPO_4^{2-} 1.0, HCO_3^{-} 4.2 mM.

pure acetone and ion-exchanged water, and then immersed into 200 ml of a simulated body fluid (SBF) which had almost equal ion concentrations to those of the human blood plasma and the fluids added with various extra ions, which are given in Table I. The simulated body fluid and the fluids added with excessive magnesium, calcium and/or phosphate ions were prepared by dissolving reagent grade chemicals of NaCl, $NaHCO_3$, KCl, $K_2HPO_4 \cdot 3H_2O$, $MgCl_2 \cdot 6H_2O$ and $CaCl_2$ into ion-exchanged water. The fluids added with silicate, aluminate and/or fluoride ions were prepared by dissolving reagent grade chemicals of $Na_2SiO_3 \cdot 9H_2O$, $AlCl_3 \cdot 5H_2O$ and/or NaF into the fluid prepared by the method described above. The amounts of the excess calcium and phosphate ions added were, respectively, equal to the amounts of these ions present in the original simulated body fluid. If these ions were added in the same amounts simultaneously, or in double amounts individually, some precipitations were formed in the fluid. The amount of the magnesium ion added was twice that of the same ion present in the original simulated body fluid. The amount of the silicate ion added was determined so that the sum of the silicate ion added and that dissolved from glass-ceramic A-W(Al) could be almost equal to the amount of the silicate ion to be dissolved from glass-ceramic A-W in the simulated body fluid within six days [15]. The amounts of the fluoride and aluminate ions added were, respectively, the maximum amounts of these ions which could be dissolved from glass-ceramic A-W or A-W(Al) [15]. All of the examined fluids were buffered at pH 7.25 with 50 mM trihydroxymethyl aminomethane ($(CH_2OH)_3(CNH_2)$) and 45 mM hydrochloric acid (HCl) in a polyethylene bottle and their temperatures were maintained at 36.5 °C.

2.3 Analysis of surface structure

The specimens were taken out from the fluids after seven days immersion in the fluids, and gently washed with pure acetone. Their surfaces were subjected to thin-film X-ray diffraction, Fourier transform infrared reflection spectroscopy and scanning electron microscopic observations. In the thin-film X-ray diffraction, an X-ray diffractometer with a thin-film attachment (Rigaku, Model 2651Al) was used and the specimen surface was fixed at an angle of 1° to the incident beam. In the infrared reflection spectroscopy, a Fourier transform infrared spectrometer (Japan Spectroscopic, FT/IR 5M) was used and the reflection angle was taken at 75°. These techniques enable the detection of a surface layer only 1 μm thick. In the electron microscopic observation, the surfaces were ion-coated with platinum and observed under a scanning electron microscope (Hitachi S-450).

3. Results and discussion

Except for glass-ceramic A-W, all the other examined materials, i.e. the silica glass, sintered alumina, sintered zirconia, pure titanium and Ti6Al4V alloy, showed no change in the thin-film X-ray diffraction patterns and the infrared reflection spectra of their surfaces even after soaking in any kind of the examined fluids for seven days.

The thin-film X-ray diffraction patterns and Fourier transform infrared reflection spectra of the surface of glass-ceramic A-W(Al), which was soaked in the fluids given in Table I for seven days, are shown in Figs 1 and 2, respectively, in comparison with those before soaking. In Figs 1 and 2, assignment of main peaks of the diffraction patterns and reflection spectra, which was made on the basis of the data previously reported [11], are also shown. It can be seen from Figs 1 and 2

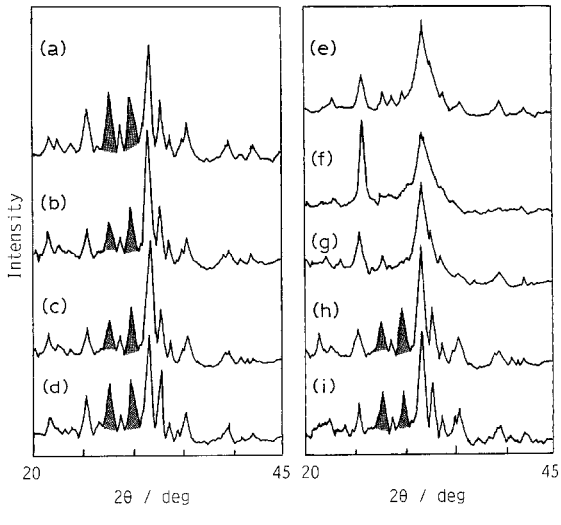

Figure 1 Thin-film X-ray diffraction patterns of glass-ceramic A-W(Al) soaked in various fluids for seven days: (a) SBF + P, (b) SBF + Ca, (c) SBF, (d) before soaking, (e) SBF + Ca + Si + Al, (f) SBF + Ca + Si + Mg, (g) SBF + Ca + Si, (h) SBF + F, (i) SBF + Si. Shaded peaks, wollastonite; unloaded peaks, apatite.

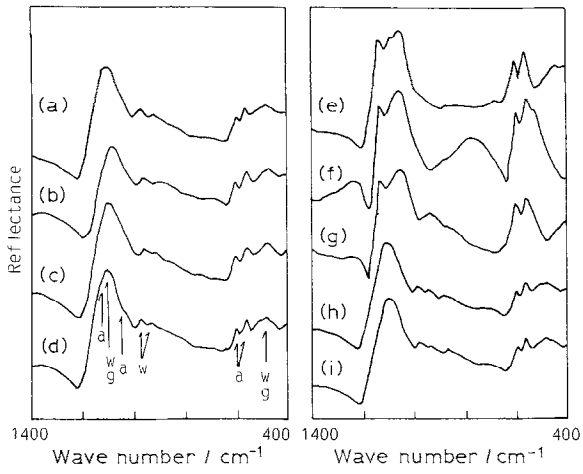

Figure 2 Infrared reflection spectra of glass-ceramic A-W(Al) soaked in various fluids for seven days: (a) SBF + P, (b) SBF + Ca, (c) SBF, (d) before soaking, (e) SBF + Ca + Si + Al, (f) SBF + Ca + Si + Mg, (g) SBF + Ca + Si, (h) SBF + F, (i) SBF + Si; (a) apatite, (w) wollastonite, (g) glassy phase.

that glass-ceramic A-W(Al) does not form an apatite layer on its surface in the fluids added with calcium, phosphate, silicate or fluoride ion individually, as well as in the original simulated body fluid, whereas it forms the apatite layer in the fluids added with the calcium and the silicate ions simultaneously. Even in the fluids added with the magnesium or aluminate ion, if the calcium and silicate ions are added simultaneously in addition to these ions, the apatite layer is formed on the surface of glass-ceramic A-W(Al). Fig. 3 shows scanning electron micrographs of the surface of glass-ceramic A-W(Al) soaked in three kinds of fluids added with the calcium and silicate ions simultaneously. It can be seen from Fig. 3 that the surface is completely covered with fragmental particles after the soaking in these fluids. The morphology of the particles is similar to that of the apatite formed on the surface of bioactive glass-ceramic A-W in the original simulated body fluid [11].

These results indicate that both the calcium and silicate ion dissolved from bioactive glass-ceramic A-W and its related bioactive glass and glass-ceramic cooperatively play an important role in forming the apatite layer on its surface in the body. The human body fluid is already supersaturated with respect to the apatite even under normal conditions [16]. The calcium ion dissolved from the glass and glass-ceramic could increase the degree of the supersaturation of the surrounding body fluid with respect to the apatite. The apatite formation on the surfaces of bioactive glass-ceramic A-W and its related bioactive glass and glass-ceramic, however, cannot be interpreted only in terms of the degree of the supersaturation [15]. This is apparent from the present observation that addition of the calcium ion alone to the simulated body fluid did not give apatite formation on the surface of glass-ceramic A-W(Al). The addition of the silicate ion in combination with the calcium ion was required for formation of the apatite layer. The silicate ion dissolved from the glass and glass-ceramics might provide favourable sites for nucleation of the apatite on the surface of the glass and glass-ceramics. This is

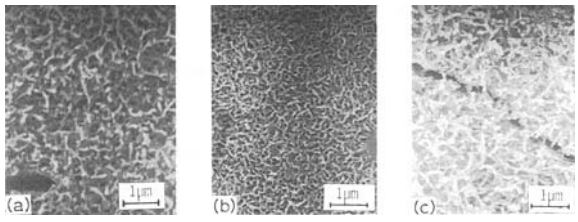

Figure 3 SEM photographs of surfaces of glass-ceramic A-W(Al) soaked in the fluids SBF + Ca + Si, SBF + Ca + Si + Mg, and SBF + Ca + Si + Al for seven days: (a) SBF + Ca + Si, (b) SBF + Ca + Si + Mg, (c) SBF + Ca + Si + Al.

supported by the present observation that the apatite layer was formed only on the surface of glass-ceramic A-W(Al) but not on the surfaces of other examined materials, including a silica glass, sintered alumina and pure titanium even in the fluid added with the calcium and silicate ions simultaneously. Glass-ceramic A-W(Al) could produce a considerable amount of silanol group on its surface when exposed to the simulated body fluid. Some of the silicate ion added to the simulated body fluid might combine with the silanol group to form a highly hydrated silicate structure on the surface of the glass-ceramic. Thus formed, highly hydrated silicate structures might provide favourable sites for nucleation of the apatite. In the case of bioactive glass-ceramic A-W and its related bioactive glass and glass-ceramic, the highly hydrate silicate structure might be spontaneously formed on their surfaces by dissoluton of appreciable amounts of the calcium and silicate ions when they are exposed to the simulated body fluid, and hence, the apatite layer might be formed on their surfaces even in the original simulated body fluid [10].

Hench *et al.* [17] and Anderson *et al.* [18] proposed that a silica hydrogel layer formed on the surfaces of Bioglass-type bioactive glasses might play an important role in forming an apatite layer on their surfaces. Their proposals are consistent with the present results, although the distinctive silica-rich layer could not be observed for bioactive glass-ceramic A-W [6, 7, 11, 19]. Carlisle *et al.* reported that the silicon also plays the essential role in forming the apatite in the living bone of mammals [20].

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