

In vitro dissolution behavior of $\text{SiO}_2\text{-MgO-Al}_2\text{O}_3\text{-K}_2\text{O-B}_2\text{O}_3\text{-F}$ glass-ceramic system

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Abstract Herein, we report the results of the in vitro dissolution tests, which were carried out by immersing the selected glass-ceramic samples in artificial saliva (AS) for various time periods of up to 42 days. In our experiments, the $\text{SiO}_2\text{-MgO-Al}_2\text{O}_3\text{-K}_2\text{O-B}_2\text{O}_3\text{-F}$ glass ceramics with different crystal morphology and crystal content were used and a comparison is also made with the baseline glass samples (without any crystals). The bioactivity of the samples was probed by measuring the changes in pH, ionic conductivity and ionic concentration of AS following in vitro dissolution experiments. High resistance of the selected glass-ceramic samples against in vitro leaching has been demonstrated by minimal weight loss (<1%) and insignificant density change, even after 6 weeks of dissolution in artificial saliva. While XRD analysis reveals the change in surface texture of the crystalline phase, FT-IR analysis weakly indicated the Ca-P compound formation on the leached surface. The experimental measurements further indicate that the leaching of F^- , Mg^{2+} ions from the sample surface commonly causes the change in the surface chemistry. Furthermore, the presence of (Ca, P, O)-rich mineralized deposits on the leached glass-ceramic surface as well as the decrease in Ca^{2+} ion concentrations in the leaching solutions (compared to that in the initial AS solution) provide evidences of the moderate bioactive or mild biomineralisation behaviour of investigated glass-ceramics.

1 Introduction

In biomaterials research, the in vitro studies involving dissolution experiments in solutions similar in composition to those present inside the human body (e.g. simulated body fluid, SBF, compositionally similar to human plasma or artificial saliva, AS, compositionally similar to human saliva) have now been recognized as preliminary/screening tests on new candidate implant materials. In an extensive study, Kim investigated and compared the bioactive properties of bioglass, hydroxyapatite, and A-W glass-ceramic [1]. It has been identified that the primary mechanism of bioactivity involves biomineralisation of Ca-phosphate nanocrystals on material surfaces, which subsequently encourages tissue integration. In a different study, Cluppera et al. [2] observed the crystalline hydroxyapatite layer formation on a commercial bioactive glass (Bioglass 45S5 grade, tape cast and sintered) after immersion in SBF solution for 20–24 h. Ebisawa et al. [3] reported the bone like apatite formation on a heat-treated glass, $40\text{FeO} \cdot \text{Fe}_2\text{O}_3\text{-}60\text{CaO} \cdot \text{SiO}_2$ (composition in wt. %) during immersion in SBF. Liu [4] fabricated a new bioactive glass-ceramic system ($\text{CaO-MgO-SiO}_2\text{-P}_2\text{O}_5\text{-F}$) and attributed the formation mechanism of Ca-P rich apatite layer to account for the excellent bioactivity. In one among many studies, Oliveria et al. [5] reported better adherence of the apatite surface layer on $\text{MgO-CaO-P}_2\text{O}_5\text{-SiO}_2$ glass-ceramic, when compared to the parent glass materials. Salinas et al. [6] reported that the chemical treatment by HCl can induce better apatite layer formation of a $\text{CaO} \cdot \text{P}_2\text{O}_5\text{-CaO} \cdot \text{SiO}_2\text{-CaO} \cdot \text{MgO} \cdot 2\text{SiO}_2$ glass system, after immersing for 4 days in SBF solution. The in vitro dissolution of $\text{SiO}_2\text{-Al}_2\text{O}_3\text{-B}_2\text{O}_3\text{-MgO-CaO-Na}_2\text{O-F}$ glass-ceramic showed evidence of mild biomineralisation after 7 days of immersion [7]. Also, submicron

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precipitates (calcium–phosphorus rich) were reported to cover the glass–ceramic surface. Padilla et al. [8] observed that phosphorus containing glasses can exhibit faster formation of apatite layer on a glass–ceramic surface ($\text{SiO}_2\text{--CaO--P}_2\text{O}_5$), when compared to $\text{SiO}_2\text{--CaO}$ glass–ceramic. In another recent work, Alemany et al. [9] observed the formation of a compact, continuous HA (Hydroxyapatite)-like layer within one week of SBF immersion of wollastonite glass–ceramic materials, irrespective of processing routes or condition. Xin et al. [10] reported the similar in vitro Ca–P formation on various sintered porous materials (SBF immersion for 1 day), including bioglass, glass–ceramic, HAp, α -TCP and β -TCP surfaces. Hench and co-workers [11] reported better bioactive property i.e. faster in vitro HCA (Hydroxycarbonate apatite) layer formation in $\text{Na}_2\text{O--CaO--SiO}_2$ glass–ceramic system, when compared with other bioactive materials, including hydroxyapatite ceramic, A–W glass–ceramic, ceravital[®] or Bioverit[®] material. Zhang et al. [12] reported the formation of apatite agglomerates in $\text{CaO--P}_2\text{O}_5\text{--TiO}_2\text{--MgO--Na}_2\text{O}$ glass–ceramic, characterized by needle-shaped crystals after immersion in SBF for a period of 8 weeks.

In case of mica based glass–ceramics, Vogel et al. [13] and Höland et al. [14] carried out some preliminary biological experiments on machinable bioactive glass–ceramics, consisting of both apatite and fluorophlogopite. They observed crystalline prominences i.e. a crystal rich layer with the absence of glassy matrix of about 0.5 μm thick at the surface of the glass–ceramics, after in vitro tests in Ringer's solution, which was attributed to the dissolution of glassy phase. They also reported the formation of a Ca-rich layer of 5–10 μm thick at the surface of the implant and strong bone-implant attachment in a mechanical push out test. In the light of the above mentioned experimental observations (in vitro dissolution) for different biomaterials in general and bio glass–ceramics in particular, it is quite understandable that the formation of calcium phosphate compounds (mainly apatite), either in the form of dispersed precipitate or as continuous layer covering the whole surface, is a true indicator of the bioactivity/biomineralization of the material. Such property evidently depends on the glass–ceramic composition as well as shape, size, distribution and chemistry of ceramic phase.

In the present study, two temperature variation batches e.g. sample heat treated at 1,000 and 1,080°C for 4 h and two time variation batches e.g. sample heat treated at 1,000°C for 12 h and 24 h, were immersed in artificial saliva for four different time periods (1, 2, 3, and 6 weeks). In accompanying work [15], we have reported the microstructural development and properties of the Macor glass–ceramics. This material is developed with the primary intention of their application for dental restoration purpose. Therefore, leaching behavior in artificial saliva was studied

in order to simulate the actual working environment inside the oral cavity. This was done by analyzing both the samples and the leaching solutions after different time periods.

2 Experimental

2.1 Materials

For our study, the $\text{SiO}_2\text{--MgO--Al}_2\text{O}_3\text{--K}_2\text{O--B}_2\text{O}_3\text{--F}$ glass–ceramic system was used and the composition of the base glass was closely related to the commercial composition available in that system (MACOR[®], Corning Glass Ltd.). The composition of the starting precursor materials was obtained from the previous work of Guedes et al. [16] and was rechecked from www.precisionceramics.co.uk. All the chemicals used for the preparation of the glass–ceramic material were of analytical reagent (AR) grade and more than 99% pure. Initially, the base glass was fabricated at 1,500°C in two steps i.e. first, the frit was prepared by quenching the melt in water and subsequently, by remelting the frit and quenching in air. The crystallization heat treatments were performed both by varying temperature from 1,000 to 1,120°C with 40°C temperature interval for 4 h constant time duration and by varying time from 4 to 24 h with 4 h time interval at a constant temperature of 1,000°C. During the heat treatment experiments, the heating rate remained constant at 6.5°C/min, for both the cases. The microstructural characterization of these heat-treated samples was carried out using optical microscope, SEM-EDS (FEI QUANTA 200, The Netherlands), and XRD (Isodebyeflex 2002, USA). The details of the material preparation procedure can be found elsewhere [15].

As far as the phase assemblage in investigated materials is concerned, XRD analysis of the heat-treated glass–ceramics showed the dominant presence of fluorophlogopite as the main crystalline phase in both the temperature variation (except 1,000°C, 4 h sample) and the time variation batches. In Table 1, the crystal volume fraction in different investigated ceramics is mentioned. As discussed in Ref 15, the temperature variation samples e.g. samples heat treated at 1,080°C exhibit the usual microstructure of 'straw' like interlocked mica flakes, randomly oriented in the glassy matrix. On the other hand, the morphology of the crystal phase in time variation samples is distinctly different. The important observation is that a typical crystalline morphology with a number of mica rods radiating from a central nucleus constitutes as the major microstructural phase. The microstructure is characterized by typical 'spherulitic-dendritic' shaped crystals with a number of mica rods, radiating from a central nucleus, which constitutes as the major microstructural phase. Such

Table 1 Different crystal shapes formed in different amounts by the different crystallization heat treatment experiments

Heat treatment condition	Crystal morphology	Crystal volume fraction (%)
1,000°C for 4 h	No detectable bulk crystallization	–
1,040°C for 4 h	Random, “straw” like	59.2
1,080°C for 4 h	Random, “straw” like	46.1
1,120°C for 4 h	Random, “straw” like	46.8
1,000°C for 8 h	Random, “butterfly” shaped	26.3
1,000°C for 12 h	Random, “butterfly” shaped	39.0
1,000°C for 16 h	Random, “butterfly” shaped	52.3
1,000°C for 20 h	Random, “butterfly” shaped	54.3
1,000°C for 24 h	Random, “butterfly” shaped	70.1

Crystal volume fraction corresponds to the fluorophlogopie crystals only

morphology of the crystal phases is common for all the time variation batches i.e. samples heat treated at 1,000°C for 8, 12, 16, 20 and 24 h. At the end of each of the mica rods, a characteristic ‘tree leave’ shaped structure is seen. The detailed microstructural analysis and explanations for the formation of different crystalline morphologies is already reported elsewhere [15].

2.2 In vitro dissolution tests

In order to study/simulate the dissolution behavior of the heat treated glass–ceramic material in physiological environment, the in vitro dissolution study was carried out by immersing four different batches of glass–ceramic samples in artificial saliva for four different time periods of 7, 14, 21 and 42 days. Each batch contains three identical samples. Amongst the four batches, three were from time variation batch i.e. heat treated at 1,000°C for different soaking time of 4, 12 and 24 h and one was from temperature variation batch i.e. sample heat treated at 1,080°C for 4 h. Such selection of samples has been made on the basis of our earlier observations, as reported in Ref. 15, that the sample, heat treated for 24 h at 1,000°C contains largest volume fraction (~70%) crystals among time variation batch; while a maximum crystal content (in case of temperature variation batch) is 54% in sample, heat treated at 1,040°C for 4 h. Also, one baseline glass sample, i.e. heat treated at 1,000°C for 4 h was selected, for the purpose of comparison.

Artificial saliva (AS) was prepared using the composition obtained from the work of earlier researcher [17, 18], and is given in Table 2. All the chemical reagents, used to prepare AS were of AR grade and almost 99% pure. The chemical reagents were mixed in distilled water using a magnetic stirrer at a temperature of 60°C and a speed of 40 rpm for 30 min. The solution was then cooled and kept in an airtight glass bottle at a temperature of 4°C for not more than one month. Three pieces (size 2.5 cm × 2 cm × 1 cm) with rectangular cross sections were cut from each of the four different heat treated batches for each set of experiments. All the samples were taken from a single bigger slab for each of the four heat treated batches in order to maintain the homogeneity in terms of crystal volume fraction, crystal morphology and surface features. Around 100 ml of AS was used to immerse each of the samples. The whole test assembly (sample immersed in solution) was kept in airtight glass bottles at 37°C (i.e. human body temperature). In the present study, the whole experimental setup was made to study the static leaching behavior of the materials. The solutions, therefore, neither stirred nor changed in the whole course of the present study. This condition is close to the actual operating environment present inside the oral cavity. After each time period, respective samples were taken out from the solution, cleaned with distilled water, dried for 3 h at 110°C, weighed in an electronic balance, and kept in airtight desiccators. The weight losses of the tested samples were calculated with respect to their respective initial weights. The electronic balance was calibrated to measure

Table 2 Composition of Artificial Saliva [17, 18]

Material	Amount (gm) per 1 liter of distilled water	Comment
NaCl	0.4	99.9% pure, AR grade
KCl	0.4	99.5% pure, AR grade
CaCl ₂ · 2H ₂ O	0.795	99.5% pure, AR grade
NaH ₂ PO ₄ · 2H ₂ O	0.78	99% pure, AR grade
Na ₂ S · 9H ₂ O	0.005	99.9% pure, AR grade
Urea	1	99.5% pure, AR grade

accurate weight up to fourth decimal place. All the solutions were kept inside the airtight glass bottles at 4°C, for not more than one month. The density of the glass–ceramic samples (heat treated at 1,000°C for 4 h, 1,000°C for 12 h, 1,000°C for 24 h and 1,080°C for 4 h) were measured following the Archimedis principle using the laboratory balance and density measurement setup, both at the beginning of the test and after 1, 2, 3, and 6 weeks of the dissolution.

2.3 Characterization

The detailed microstructural characterization of the leached surface was performed using XRD and SEM. Compositional analysis of the leached surface was carried out by using SEM-EDS. Additionally, IR spectroscopy (Perkin-Elmer System 2000 FT-IR spectrometer and PC data system, USA) of selected in-vitro tested samples was also performed in order to identify the change in surface bonding characteristics of the glass–ceramic samples, following in vitro test. Care has been taken during cleaning and handling of the samples in order to avoid any type of attachment of dirt or dust particles on the sample surface. The pH and ionic conductivity of the in vitro test solutions as well as the starting solution were measured using the dual channel ‘pH / Ion / Conductivity Meter’ (PC5500, Eutech Instruments), and fluoride concentrations in those solutions were measured using Orion portable meter (Model: 290A, Orion Research Inc., Boston, USA), having a single junction electrode. Atomic Absorption spectroscopy (AAS, model Spectra AA, 220FS, Varian, Australia) study was carried out in order to detect the concentration of metal ions in artificial saliva, both before and after in vitro tests.

3 Results and discussion

In the present study, the in vitro dissolution of the heat treated samples was studied in artificial saliva with the primary idea of understanding the material behavior in vitro as well as the basic mechanisms operating during dissolution of the material in saliva. All the characterizations were, therefore, set to more of a qualitative approach, rather than a quantitative one. The designation of different in-vitro tested samples and the corresponding solutions are provided in Table 3. The sample heat treated at 1,000°C for 4 h is designated as ‘Sample I’, at 1,000°C for 12 h as ‘Sample II’, at 1,000°C for 24 h as ‘Sample III’, and at 1,080°C for 4 h as ‘Sample IV’. The solution corresponding to the in-vitro test of Sample I for 1 week is designated as ‘Solution I1, for 2 weeks as I2 and so on.

The baseline glass sample (sample I, heat treated at 1,000°C, 4 h) experiences mostly weight gain during all the test intervals, except for the noticeable weight loss

Table 3 Sample and solution designation

Heat treatment condition	Sample designation	Time period (days)	Solution designation
1,000°C for 4 h	I	7	I1
		14	I2
		21	I3
		42	I6
1,000°C for 12 h	II	7	II1
		14	II2
		21	II3
		42	II6
1,000°C for 24 h	III	7	III1
		14	III2
		21	III3
		42	III6
1,080°C for 4 h	IV	7	IV1
		14	IV2
		21	IV3
		42	IV6

during the third week. For the other three samples, weight loss remains almost constant with a very little variation over the entire time period (up to six weeks). However, the weight loss or weight gain (in case of sample I) was very small (<1 %) for all the time periods, which seems to be good from the glass durability/clinical performance point of view. The density measurements of the samples, prior to and in course of dissolution for 1/2/3/6 weeks showed insignificant change, which implicates that the mechanism of dissolution process remains similar throughout the duration of the in-vitro experiment. However, it is possible during the course of in vitro test for extended time period, the dissolution mechanism changes from the leaching of glass phase alone to the simultaneous leaching of crystal and glass phase and it may be manifested in significant weight as well as density change.

3.1 Microstructural changes after dissolution study

Figure 1 plots the XRD spectra acquired from the selected sample surface, before and after the in vitro dissolution test. The recorded changes in XRD peak evolution, as compared to untreated bulk samples, provide interesting information. In case of sample I, similar broad X-ray peak, indicative of the presence of glass on leached surface, like in virgin surface, was recorded (not shown). For sample III (heat treated at 1,000°C for 24 h), it is important to observe that in vitro leaching actually reduces (compared to unleached surface) the intensity of the strongest peak from the (200) plane and some new peaks are evolved (see Fig. 1c). These peaks are identified to be the characteristic fluorophlogopite

peaks from two other crystallographic planes (113) and ($\bar{1}$ 13). This suggests that the surface texture of the glass–ceramic material has been changed, possibly due to leaching of different ions to the solution. X-ray peaks at higher 2θ values possibly signify for the amorphous layer formation on the surface of the tested samples and corresponding broadening of the existing peaks. Another possibility is that since the intensity of the strong peaks (evolved at lower 2θ values) decrease due to the leaching of the crystal phases from the surface, the overall background becomes stronger. It can be recalled that sample III contains highest crystallinity ($\sim 70\%$), so that the effect of leaching and corresponding surface texture change is more pronounced for this sample. However, no characteristic peaks, corresponding to any CaP-compounds, like TCP (Tri Calcium Phosphate) or HA (Hydroxyapatite) was detected for any of the samples, within the detection limit of XRD.

In discussing the *in vitro* leaching induced microstructural changes, the reference will be made to the corresponding untreated/virgin surface microstructure. Figure 2 shows the SEM images of the baseline glass (sample I) surface, tested for six weeks along with the corresponding EDS analysis. For sample I (heat treated at $1,000^\circ\text{C}$ for 4 h), some amorphous like material can be observed on the surface, which has different topographical features from the original sample surface (see Fig. 2a and

b). EDS analysis shows that the relative intensity of Mg, Al as well as O-peak increases, compared to that of the virgin sample. The increased oxygen content of the leached surface indicates the potential occurrence of polycondensation of Si–OH to form Si–O–Si bonds during *in vitro* leaching, although more experimental proof is awaited to support this conclusion as EDS is not the most appropriate technique to measure the presence of oxygen. Also, the surface topographical features do indicate the leaching of metal ions from the baseline glass surface.

Some important observations were made after *in vitro* testing of the glass–ceramic samples, containing ‘spherulitic-dendritic’ crystals (see Fig. 3). A comparison of Fig. 3a with that of Fig. 4a indicates that the ‘spherulitic-dendritic’ crystals are much larger in size than ‘straw’ like crystals. Overall, the ‘spherulitic-dendritic’ crystal shape is retained upon *in vitro* dissolution experiments (see Fig. 3b). A closer look at Fig. 3 reveals the dissolution of glass phase, which otherwise was retained in the inter-mica platelet region. The deposition of brighter contrast phase is critically noticed at various places on leached surface. As recorded with other investigated samples, the stronger presence of P can be seen in the inset of Fig. 3b. Also, the relative intensity of O-peak, compared to that from virgin surface increases after leaching. A comparison of Fig. 4a and b clearly reveals the absence of any change in crystal

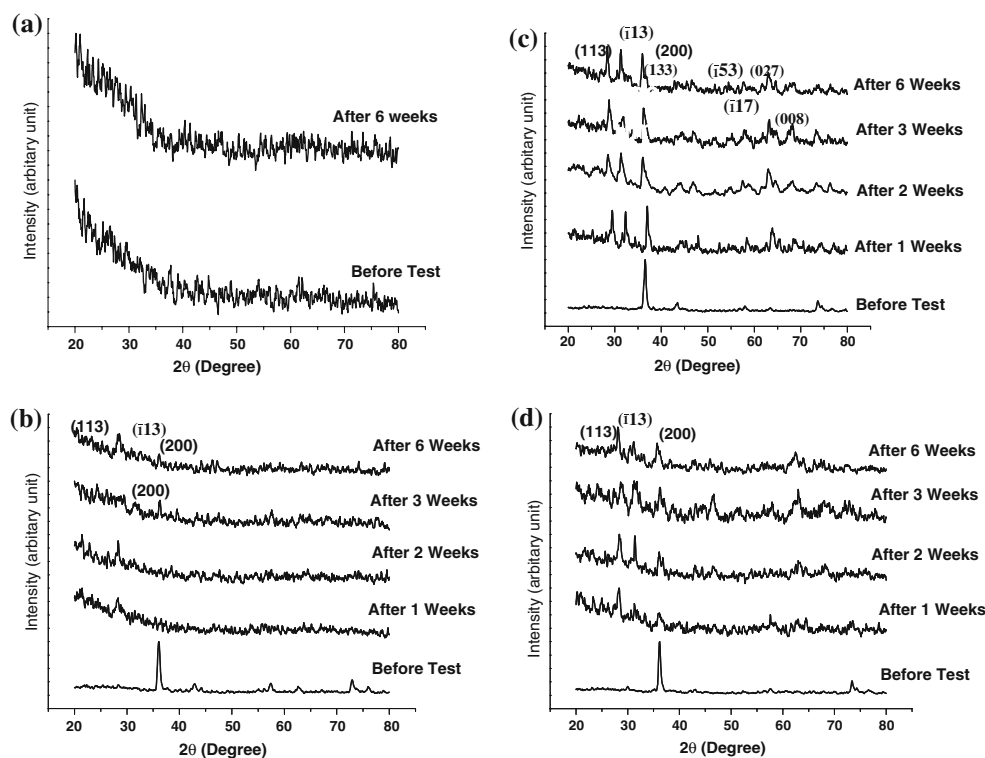


Fig. 1 Phase analysis (XRD spectra) of the heat treated samples, before and after *in-vitro* dissolution study for samples heat treated under various conditions: (a) $1,000^\circ\text{C}$ for 4 h (Sample I), (b) $1,000^\circ\text{C}$

for 12 h (Sample II), (c) $1,000^\circ\text{C}$ for 24 h (Sample III), and (d) $1,080^\circ\text{C}$ for 4 h (Sample IV)

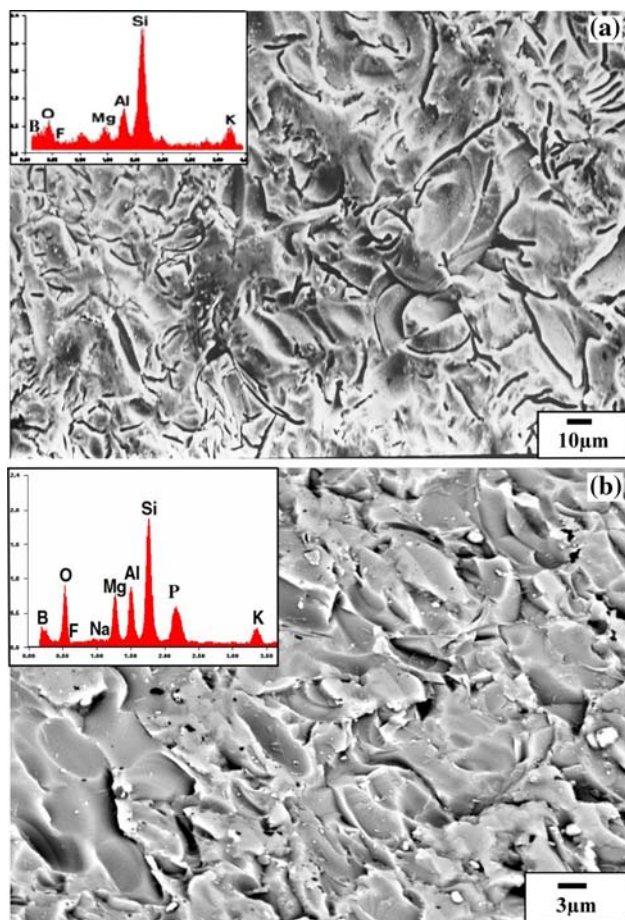


Fig. 2 SEM images revealing the surface topographical features of baseline glass sample (Sample I, heat treated at 1,000°C for 4 h): (a) virgin surface i.e. before in vitro dissolution, and (b) after 6 weeks of dissolution in artificial saliva. Representative EDS analysis of the surface composition are shown in insets

shape for the glass-ceramic containing the largest amount of crystals. Also, the EDS analysis of the brighter contrast phase, appearing to be the dissolution product' shows strong peak of P, but not of Ca. The changes in microstructure of the sample IV, containing straw like mica crystals, due to in vitro dissolution can be noticed in Fig. 5. The as-leached surface exhibits signs of surface dissolution as well as deposition of bright contrasting particles of irregular shapes and varying sizes (1–5 μm). EDS analysis (see Fig. 5b inset) further confirms that these particles are rich in (Ca, P). This observation undoubtedly confirms the formation of CaP-based compounds on the investigated glass-ceramic surface, when immersed in an artificial saliva solution. Similar formation of Ca-P rich dispersed precipitate is commonly reported to form during in vitro leaching treatment of various glass/glass-ceramics materials [12, 19].

Furthermore, in order to provide complementary evidences of the dissolution and leaching of the glass-ceramics, FT-IR spectroscopy was performed on both the

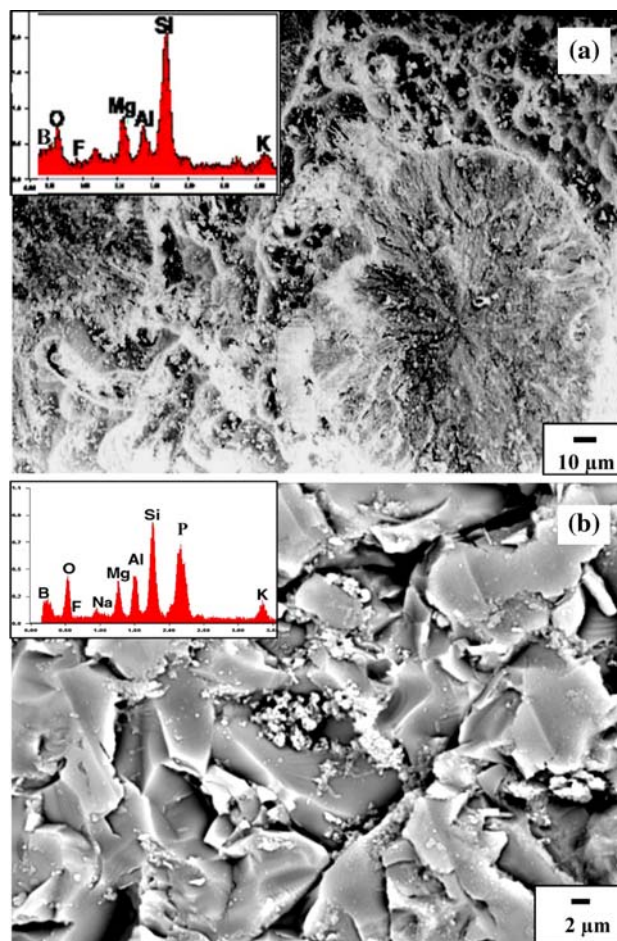


Fig. 3 (a) SEM image revealing the 'spherulitic-dendritic' shaped crystals on the virgin surface (before in vitro dissolution) of the glass ceramic sample, heat treated at 1,000°C for 12 h (Sample II), and (b) SEM image illustrating the surface topographical features of the same sample after in vitro dissolution for 6 weeks in AS solution. EDS analysis of the crystals in (a) and from the brighter particles on leached surface in (b) are shown in insets

virgin surface as well as with the leached glass-ceramic surface, after 6 weeks of dissolution. The representative results are provided in Fig. 6. In case of virgin surface, the observation of a broad IR band, spreading over a large region of 750–1,500 cm^{-1} with a peak at around 1,034 cm^{-1} indicates the characteristic Si–O–Si stretch band. In contrast, the observation of narrow IR bands, located at around 1,024 cm^{-1} , near 1,200–1,243 cm^{-1} , 1,600 cm^{-1} as well as those located at $\sim 2,940 \text{ cm}^{-1}$ and $\sim 3,265\text{--}3,282 \text{ cm}^{-1}$ on the virgin surface of sample III are comparable with that of the standard phlogopite (Phlogopite in KBr, Canada, NMNH 124158) spectrum obtained from FDM electronic handbook database. The evolution of IR bands is different after in vitro leaching experiments for both the samples. After 6 weeks dissolution, the IR spectra of the sample I (heat treated at 1,000°C for 4 h, without any crystallinity) appear to be subtly different with the higher

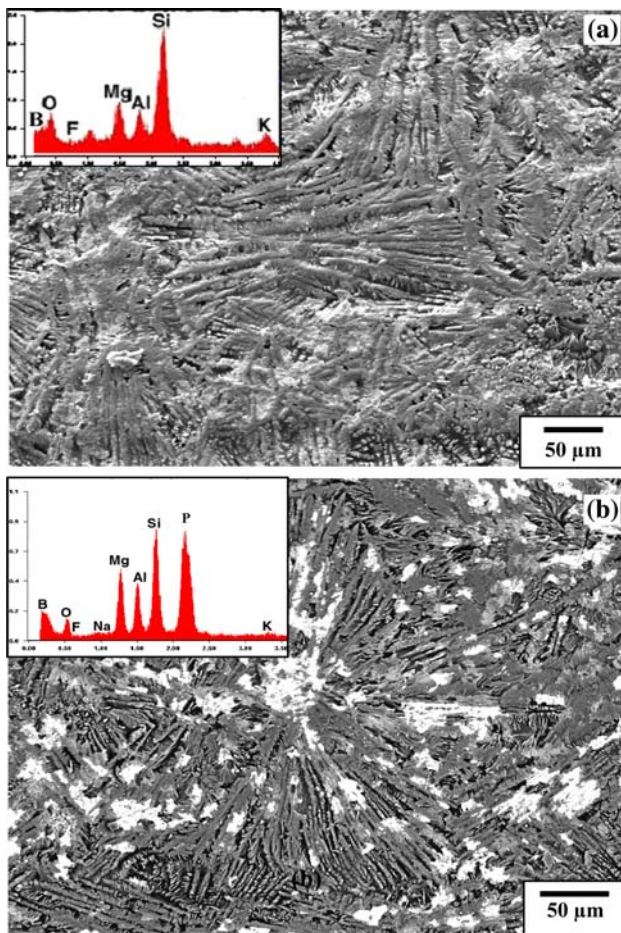


Fig. 4 (a) SEM images of glass–ceramic sample revealing the large amount of ‘spherulitic-dendritic’ shaped crystals on the virgin surface (before in vitro dissolution) of the glass ceramic sample, heat treated at 1,000°C for 24 h (Sample III), and (b) SEM image showing the as-leached surface, in particular the appearance of the leached ‘butterfly’ crystal of the same sample after in vitro dissolution for 6 weeks in AS solution. EDS analysis of the crystals in (a) and leached crystal in (b) are shown in insets

order bands disappeared and the lower order bands, corresponding to Si–O–Si bonding, being shifted to lower wave number value (see Fig. 6a). This observation possibly signifies for the severe leaching of glass phase in AS solution and corresponding change in the surface chemistry of the leached sample. Similar such modification in the IR spectrum (see Fig. 6b) is observed for the leached surfaces of sample III (heat treated at 1,000°C for 24 h). For the later, only higher order FT-IR bands tend to disappear and the shifting of lower order bands is negligible. This confirms that the presence of crystals actually suppress the leaching phenomena to a noticeable extent than the glass phase. Another important observation in the IR spectra is the presence of a small band at around 1,370–1,425 cm^{-1} , which actually corresponds to the carbonated Ca–P bond [19].

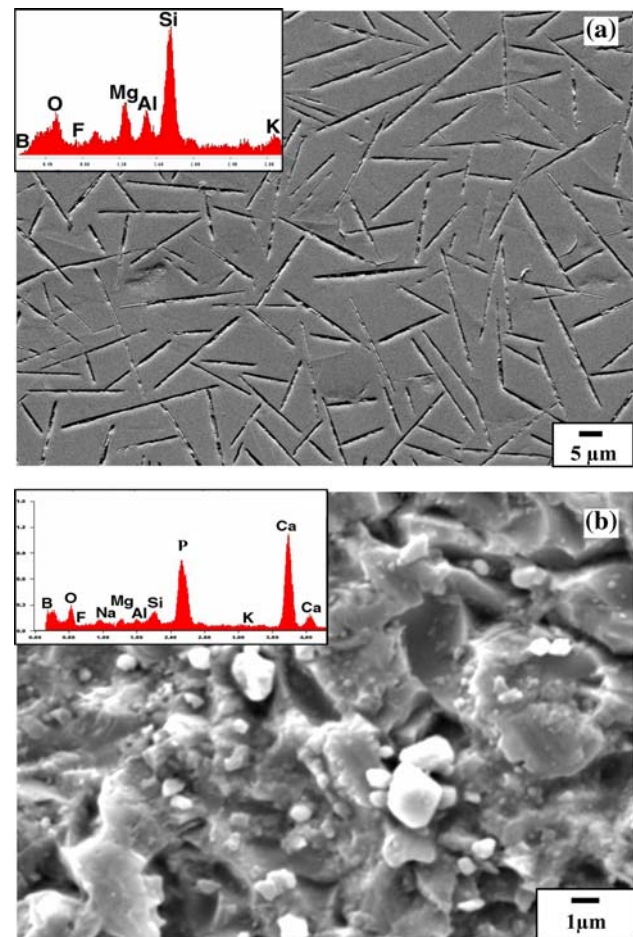


Fig. 5 (a) SEM image illustrating the presence of a large amount of ‘straw’ like crystals on the virgin surface (before in vitro dissolution) in the glass ceramic sample, heat treated at 1,080°C for 4 h (Sample IV), and (b) SEM image revealing the deposition of Ca–P-rich brighter contrast particles after in vitro dissolution of the same sample for 6 weeks in artificial saliva solution. EDS analysis of crystalline phase in (a) and bright contrast particles in (b) are provided in insets

3.2 pH and ion concentration changes of artificial saliva

In order to understand the details of leaching/dissolution behavior, the changes in pH and ionic conductivity of the solutions are systematically measured. Figure 7a shows the ionic conductivity of all the leaching solutions. It can be observed that the ionic conductivity of the leaching solutions (after first week) increases vis-a-vis initial artificial saliva solution, which means that the total ion concentration in the solution has increased due to leaching. The increase in ionic conductivity with prolonged time duration of in vitro dissolution experiments is however insignificant. This probably implicates that the leaching, once started, is a dynamic process i.e. ions move from sample to solution and solution to sample and consequently, the total number of ions remains constant in the solutions.

Fig. 6 IR spectra of the heat treated samples, before and after in-vitro dissolution study for samples heat treated under various conditions: (a) 1,000°C for 4 h (Sample I), (b) 1,000°C for 12 h (Sample II), (c) 1,000°C for 24 h (Sample III), and (d) 1,080°C for 4 h (Sample IV)

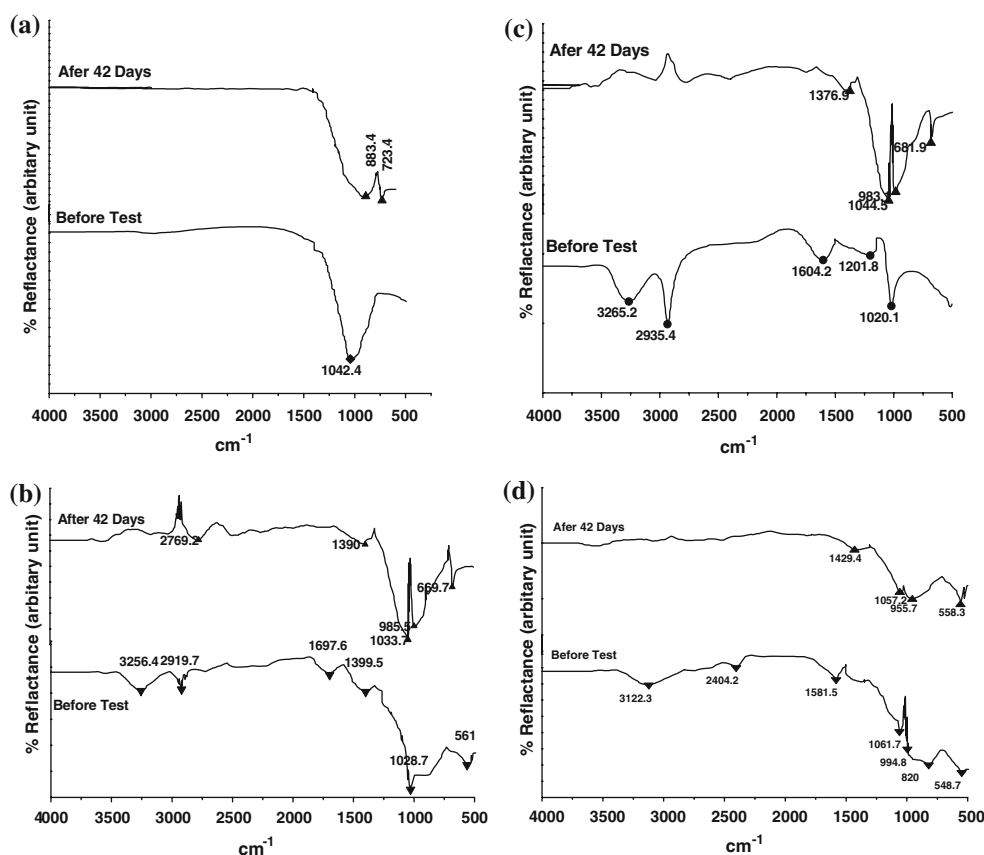


Figure 7b shows the changes in pH after various time periods of in vitro dissolution. It can be clearly noticed that pH, in all cases increases initially within first 2 weeks, when compared to the pH of the initial solution. Subsequently, pH of solution decreases noticeably and the final pH, in all cases, remains in acidic regime. Such dynamic changes in pH again support the occurrence of metal ion leaching i.e. chemical changes of material surfaces at different time periods. The increase in pH actually signifies for the reduction in the concentration of H^+ due to the replacement of metal ions in the glass and subsequent production of OH^- ions, due to breaking of siloxane bond. The interesting observation is that for the solutions II3, III3, and IV3, pH decreases to a lower value, than the initial solutions II1 and II2, III1 and III2, and IV1 and IV2. The reason may be attributed to considerable leaching of the glassy matrix from the surface, while the crystals still have to get leached. When the surface crystals get leached, pH again increases for solutions II4, III4, and IV4. Another observation is the decrease in pH of the solution I4 compared to that of solution I2 and I3. This is understandable as on the sixth week, leached layer is removed and fresh layer is exposed, so that leaching is just started in the sixth week and therefore, demand for hydrogen ions is comparatively less. Similar change in pH i.e. with decrease in pH to acidic region after 6 weeks of in vitro dissolution is

also measured for $45CaO-45P_2O_5-5K_2O-5MgO$ (composition in mol%) [20].

In order to gain further insight into leaching process during in vitro dissolution, the amount of metal ions and F^- ion in the solution were experimentally determined. Table 4 lists the concentration of different ions in the leaching solutions after various time periods. The initial solution only contains Na^+ , K^+ , Ca^{2+} etc., but not Mg^{2+} or F^- . The presence of fluoride ions in the leaching solutions suggests that fluorine is leached out from the baseline glass/glass-ceramic samples. Leaching of fluorine is particularly important for the biomaterials, as fluorine can be partially incorporated in the hydroxyapatite structure to form fluorohydroxyapatite. Additionally, fluorine being toxic to human health, leaching of fluorine is not desirable during in-vivo working condition. However, the amount of the leached fluorine is below the critical amount. Similar to F^- ions, the presence of Mg^{2+} ions is also observed in the leaching solutions, while the initial solution does not contain any Mg^{2+} ions. The presence of F^- and Mg^{2+} in the leaching solution definitely indicates a change in the glass/glass-ceramic surface chemistry, which was earlier mentioned. The amount of K^+ ion is found to be almost constant in all the leaching solutions, with the concentration being almost the same as the initial solution. This observation suggests that leaching of K^+ ions from the

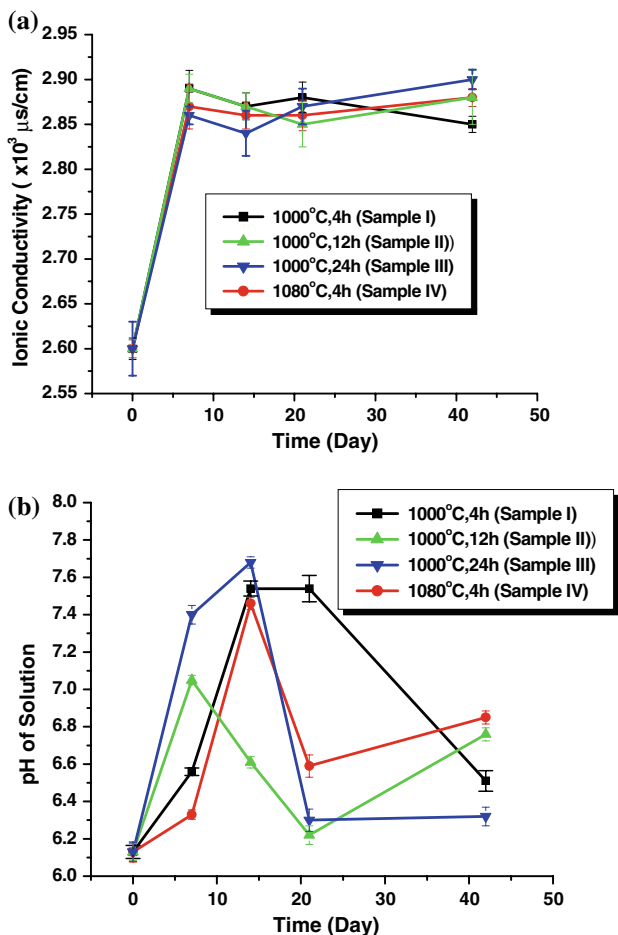


Fig. 7 (a) Ionic conductivity and (b) pH of all the leaching solutions in comparison with the starting SBF solution

sample is actually compensated by the absorption of K^+ on the sample surface. This, however, is a good signature, as far as the strength and machinability of the glass–ceramic material is concerned because of the fact that K^+ ions form the loosely bound (001) plane, which are the preferred cleavage plane in the mica crystals during crack

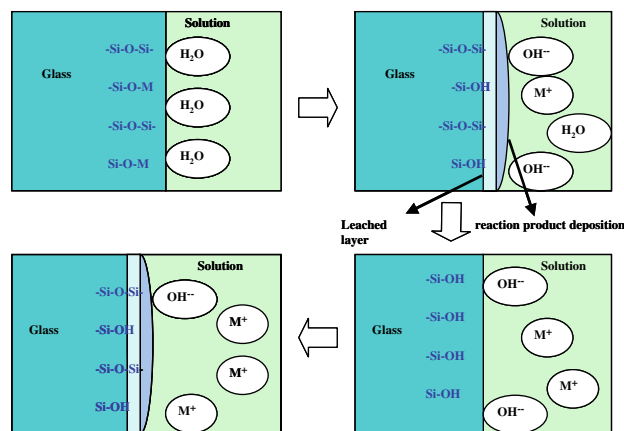


Fig. 8 Schematic representation of the dissolution process of baseline glass sample (Sample I, heat treated at 1,000°C, 4 h). Various ions and bond formation are also shown [M^{n+} = alkali ion, incorporated in glass network]

propagation. Importantly, the amount of Ca^{2+} ions in solutions is found to decrease for all investigated cases. It is possible that Ca^{2+} ions get deposited at the sample surface, as has been earlier observed [1–12] and helps in the biocompatibility and bioactivity of the glass–ceramic materials during operational condition inside the oral cavity. Furthermore, the decrease in Ca^{2+} ion concentration can be correlated with the presence of Ca–P rich phase deposition on leached surface. It should be noted here that in the present study, although the weight of the immersed sample to the volume of the solution ratio were kept almost constant for each set of experiments, the non-homogeneity in the surface geometry and surface chemistry may lead to difference in leaching behavior for different samples. Consequently, there will be variation in the concentration of different ions without showing any detectable trend. However, the primary aim here is to identify the leaching behavior of the heat treated samples and accordingly, the focus has been to study the qualitative presence of the ions and not on their absolute amounts.

Table 4 Changes in concentration of various ions in the leaching solution after different time periods of in-vitro dissolution

Average concentration of ions (mg/l)	Duration of test (days)	Heat treatment condition			
		1,000°C for 4 h	1,000°C for 12 h	1,000°C for 24 h	1,080°C for 4 h
Ca^{2+} (At starting = 396.598)	7	259.8	253.7	239.3	323.7
	42	207.7	186.01	176.09	218.2
K^+ (At starting = 97.004)	7	94.1	97.3	96.01	103.7
	42	95	111.6	97.31	86.3
Mg^{2+} (At starting = 0)	7	1.3	1.9	1.34	0.5
	42	0.6	0.87	0.5	0.61
F^- (At starting = 0)	7	0.09	0.06	0.05	0.06
	42	0.04	0.007	0.02	0.007

It should be noted here that the initial solution (before start of the in-vitro dissolution experiment) contains neither F^- nor Mg^{2+}

Summarizing, the schematic of the leaching of base line glass sample is shown in Fig. 8. The formation of Ca–P rich deposit on leached surface layer as well as cross-transport of several metal ions and F^-/H^+ , in case of glass–ceramics are illustrated. It is believed that similar dissolution process is operational in case of ‘spherulitic-dendritic’ crystals containing glass–ceramics. In case of ‘spherulitic-dendritic’ crystals, the characteristic crystal shape is retained, following the in vitro test. Also, the large ‘spherulitic-dendritic’ crystals with lesser surface area, present in the samples II and III may hinder the leaching of metal ions from the crystals more effectively than the small ‘straw’ like crystals present in sample IV.

4 Conclusions

Based on the experimental results obtained with the in vitro dissolution tests with SiO_2 – MgO – Al_2O_3 – K_2O – B_2O_3 – F glass ceramics, the following conclusions can be drawn:

- In our experiments, very minimal/insignificant weight loss as well as density variation are measured with the selected glass–ceramic samples, after 6 weeks of in vitro dissolution in artificial saliva solution. This indicates better durability of the investigated glass/glass–ceramics against leaching for extended time period of up to 6 weeks.
- Systematic XRD analysis importantly indicates the changes in surface texture of the glass–ceramic samples during in vitro leaching. This is reflected in the appearance of stronger (113), ($\bar{1}$ 13) as well as additional X-ray peaks of Fluorophlogopite phase on leached surface, as compared to virgin glass–ceramic material.
- The dynamic changes in pH with the final pH in acidic regime were recorded during in vitro dissolution experiments. The ionic conductivity of the leaching solution changes insignificantly during prolonged leaching, which implicates the continuous ion exchange process or deposition of various ionic species at the glass–ceramic/solution interface. The experimental measurements further indicate that the leaching of different ions (F^- , Mg^{2+}) from the sample surface causes the change in the surface chemistry of all the glass–ceramic materials.
- For the investigated glass–ceramics, a moderate bioactive behaviour is observed. The presence of (Ca, P, O)- rich deposits on the leached surface as well as the Ca^{2+} ion concentration profiles of leaching solution provide evidences for mild biomineralisation of both ‘straw’ and ‘spherulitic-dendritic’ crystals

containing glass–ceramics. Both FT-IR and SEM-EDS analysis provide complementary evidences to support this observation. The mineralized deposit is observed to contain individual micron sized crystals or aggregates of submicron crystals. No observable change in shape of ‘spherulitic-dendritic’ crystals is observed after in vitro dissolution.

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