Studies on preparation and characterizations of CaO-Na2O-SiO2-P2O5 bioglass ceramics substituted with Li2O, K2O, ZnO, MgO, and B2O3

M.R. Majhi, Ram Pyare and S.P.Singh

Abstract--The bioactive glass 45S5 (Hench glass), having composition 45 SiO2 - 24.5 Na2O - 24.5 CaO -6 P2O5 (wt %) were prepared with substituted Li2O, K2O, ZnO, MgO, and B2O3 by conventional melting process in an electric globar furnace at 1400±10 OC. The Controlled crystallization were carried out to convert the bioglasses to their corresponding bioglass ceramics. Nucleation and crystallization regimes were carried out by differential thermal analysis. The X-ray diffraction patterns of the bioactive glass-ceramics were show the presence of two main crystalline phases of sodium calcium silicate (Na2CaSi3O8, Na2CaSi3O9). The effect of introduction of B2O3 in place of SiO2, to the bioactive glass (45S5) lead to the formation of a new crystalline phase of calcium sodium borate (Na2CaB5O10) and the effect of introduction of ZnO and MgO in place of CaO, to the bioactive glass 45S5 there is no additional crystalline phases were developed other than two main crystalline phases of sodium calcium silicate (Na2CaSi3O8, Na2CaSi3O9). The bioactivity of the prepared glass and glass ceramics were done by infrared absorption and reflection spectrometry before and after immersion in the simulated body fluid for different periods of time at 37.8 OC. The Chemical durability of bioglass and bioglass ceramics were determined by pH measurement methods and it was found that pH of the solution varies with change in compositions after immersed in SBF solution from 1 to 30 days.

Keywords - Bioactive glass; pH Measurement; SBF; Chemical durability; Crystallization

1. INTRODUCTION

Bio- materials implanted into bone defects are generally encapsulated by fibrous tissue isolating them from the surrounding bone [1] .This is the normal response of the body towards inert materials. However, some ceramics, such as Bio glass [2], glass–ceramic A–W [3] and sintered hydroxyapatite [4] form bone-like apatite on their surfaces in the living body and bond to living bone through this apatite layer. This bone-bonding ability is called bioactivity. These bioactive ceramics are already used clinically as important bone-repairing materials. Their bone-bonding ability is achieved by the formation of a biologically active apatite layer after reaction of the ceramics with the simulated body fluid[5,6] .A controlled surface reaction of the ceramic is an important factor governing its bioactivity, as well as its biodegradability[7,8].In the present study, the introduction of MgO in the bioglass composition improves the chemical durability because of their presence in network forming sites by forming structural units such as MgO_4 [9,10] ,which shows that MgO causes obvious phase separation in silicate melts and glasses .The addition of zinc oxide to the bioactive glass–ceramic would control the reaction between the glass–ceramic and the surrounding body fluid. Zinc oxide was selected to control the reactivity since zinc is an essential trace element that has stimulatory effects on bone formation [12]. The zinc ions released from the glass–ceramic may enhance bone regeneration. With regards to ceramics designed to release zinc ions, [12, 13, 14] recently developed calcium phosphate ceramics containing zinc. However, they used polycrystalline ceramics, so the range where zinc can be incorporated is limited and the behavior of their materials is expected to be different from the glass-based materials. The bio-glass (45S5) of 5–10% B_2O_3 for SiO_2 substituted the various bioactive glass compositions to form glass–ceramics has minor effect on the final ability of the material to form a bone bond[11].The objective of this work is to study the effect of the addition of Li_2O, K_2O, ZnO, MgO, and B_2O_3 to CaO-Na_2O-SiO_2-P_2O_5 glass–ceramics on their bioactivity, density ,compressive strength, XRD, DTA and FTIR analysis were done during present investigation.

2. Experimental Methods

2.1. Glass preparation

The bioactive glass 45S5 (Hench glass), having composition 45 SiO_2 - 24.5 Na_2O - 24.5 CaO - 6 P_2O_5 (wt %) were prepared by substituted with Li_2O, K_2O, ZnO, MgO, and B_2O_3. Different compositions of bioactive glasses were prepared as shown in Table 1. For preparation of 100 grams of bioactive glasses fine-grained quartz is used as the source of silica, sodium carbonate (Na_2CO_3):, calcium carbonate ($CaCO_3$):, ammonium dihydrogen orthophosphate ($NH_4H_2PO_4$):,aluminium oxide (Al_2O_3) ,boric acid (H_3BO_3):, potassium carbonate (K_2CO_3):,Zinc Oxide (ZnO) and magnesium carbonate ($MgCO_3$) are used as a the source of sodium oxide (Na_2O), calcium oxide (CaO), phosphorus pentaoxide (P_2O_5), boron trioxide (B_2O_3), potassium

M.RMajhi is an Assistant Professor, Dept. of Ceramic Engg, IT.BHU -Varanasi (India) email: mrmajhi.cer@itbhu.ac.in
Ram Pyare is a Professor, Dept. of Ceramic Engg, IT.BHU -Varanasi (India) email:pyare_ram55@yahoo.co.in
S.P.Singh is a Professor, Dept. of Ceramic Engg, IT.BHU -Varanasi (India) email:spsinghceram@gmail.com

oxide (K_2O) ,zinc oxide (ZnO) and magnesium oxide (MgO) respectively. Bioactive glasses were prepared by conventional method. For preparing bioactive glasses amounts of materials are weighed using an electronic balance and mixed homogeneously with an agate pestle-mortar. Premixed batch is melted in platinum crucibles, at 1400±10°C, for 4 hours using globar furnace. After 4 hours of melting the glass was taken out from the furnace and, it was poured on an aluminum sheet and then cooled to room temperature .After crushing the glass, it was remelted in the furnace for another two hours to ensure complete homogeneity of glass. After homogenization and remelting, it was taken out from the furnace and poured on a hot rectangular mould kept on aluminum sheet and annealed at 480 ^oC to remove the thermal stresses and strains from the glass. Further, the glass was cooled to room temperature with controlled rate of cooling and the annealed glass samples were preserved for their properties and structural determinations such as density, compressive strength, DTA, XRD and FTIR spectrometry.

2.2. Density and Compressive strength of Bioactive Glasses

The density were studied by the Arcmedic's principle method and compressive strength of the sintered bioglass ceramic samples having size of 3x2x1 cm^3 dimension were subjected to compression test. The test was performed using Instron Universal Testing Machine at room temperature (cross speed of 0.05 cm/min and full scale of 5000 kgf)).

2.3. Differential thermal analysis

Differential Thermal Analysis measurement was carried out on powdered bioglass samples which were examined up to 1000 ^OC using a powdered alumina as a reference material (SETARAM Instrumentation, France) and the heating rate was $10^oC\ min^{-1.}$ The DTA data were used to obtain the proper heat treatment temperatures to obtain the corresponding glass–ceramic derivatives with high crystallinity. Briefly, these results showed that the values obtained were for glass nucleation temperature and the crystallization temperatures.

2.4. Preparation of glass to glass-ceramic samples (heat treatment process

The bioglass samples were thermally heated in two-step regime, at the deduced temperatures .Each bio glass sample was heated slowly to the first nucleation temperature for the formation of sufficient nuclei sites and after holding for 4 hrs, it was then further heated to reach the second chosen crystal growth temperature for performing the perfect crystal growth. After a second hold for 6 hrs time, the specimen was left to cool inside the muffle furnace to room temperature at a rate of 20^oC per hour.

2.5. X-ray diffraction measurement

The crystalline phase was identified by using X-ray diffraction analysis, the heat-treated bioglass ceramic samples were ground to 75 microns and the fine powder was subjected to XRD test using Cu-Kα radiation (λ = 1.5405A°) in a 2θ range between 20^o and 80^o. Step size and measuring speed were set to 0.02° and 1^o /min; respectively, with a tube voltage of 40 kV and current of 35mA. The JCPDS-International Centre for diffraction Data Cards were used as a reference.

2.6.In vitro bioactivity study of bioglass and bioglass–ceramics by FTIR – Reflectance and Transmittance spectra

The in vitro bioactivity of treated samples were assessed by evaluating the formation of calcium phosphate layer on the surface of the samples after immersion in SBF solution. The sample was immersed in 40ml of SBF solution in a small plastic container at 37.8 ^oC with pH= 7.4 in an incubator at static condition for time period 1 to 30 days. The SBF solution was prepared as described by Kokubo et al. [7]. The samples were prepared in to a disc shape by mixed with KBr powder in the ratio 1:100 (sample: KBr respectively) and the mixtures were subjected to a load of 10 tons/cm^2 in an evocable die to produce clear homogeneous discs. The Infrared transmittance and reflectance spectra of the bio glasses and their ceramic derivatives were measured at room temperature in the frequency range of 4000–400 cm−1 using a Fourier transform infrared spectrometer, (VARIAN scimitar 1000, USA) and (Is10 Nicolet, USA). The test results were compared with functional groups [49, 50].

2.7.pH measurement of SBF for different time periods for determine the chemical durability of glasses and their glass ceramic derivatives

The glass and glass-ceramic samples in the form of palate in the size range 1cm diameter, were subjected to the action of simulated body fluid , prepared by Kokubo et al[7] at 37.8 ^oC and pH 7.4 for different time periods (1 to 30 days). The pH of solution were measurement at different time periods by the help of pH meter to find out their chemical durability.

3. Results and Discussion

3.1.Density and Compressive strength of Bioactive Glasses

Results presented in figure 1 show that the density and the compressive strength increases with increasing the concentration of Li_2O and K_2O in the place of Na_2O in bio-glass composition No.(2) and (3) respectively. Similarly density and the compressive strength of the bio glass composition No.(4) and (5) increases with increasing ZnO and MgO in the place of CaO. Similar trend was observed while B_2O_3 is substituted for SiO_2 in bio-glass composition No.(6) and (7) respectively.

3.2. Differential Thermal Analysis

The differential thermal analysis (DTA) curve reveals information about the transformations that have occurred, such as glass transitions and crystallization temperature. The differential thermal analysis (DTA) curves of bioactive glasses has been shown in figure.2.

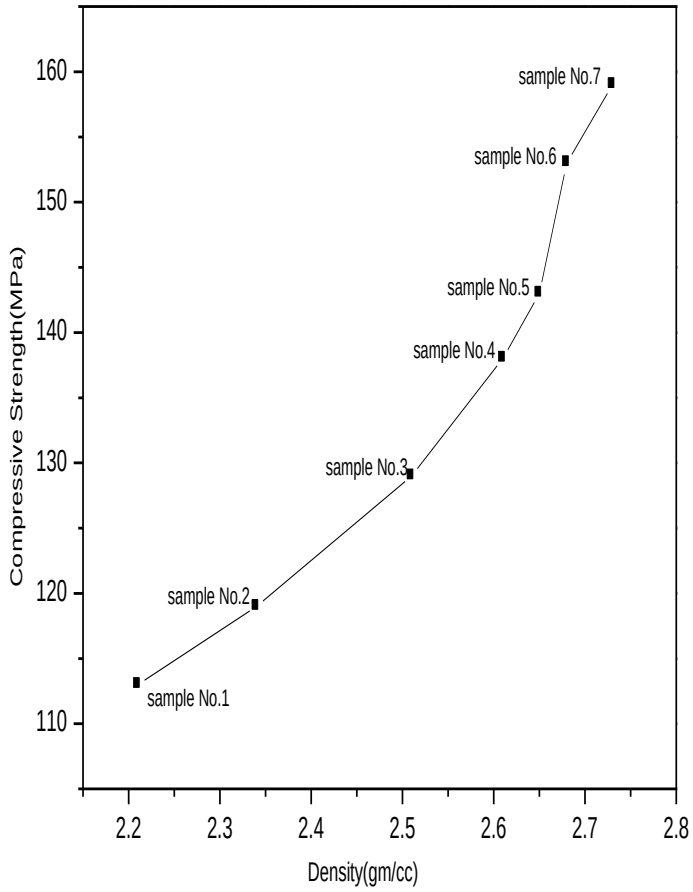


Fig.1 .Variation of Density and compressive strength of bio-glass ceramics Samples from Sl.No. (1-7)

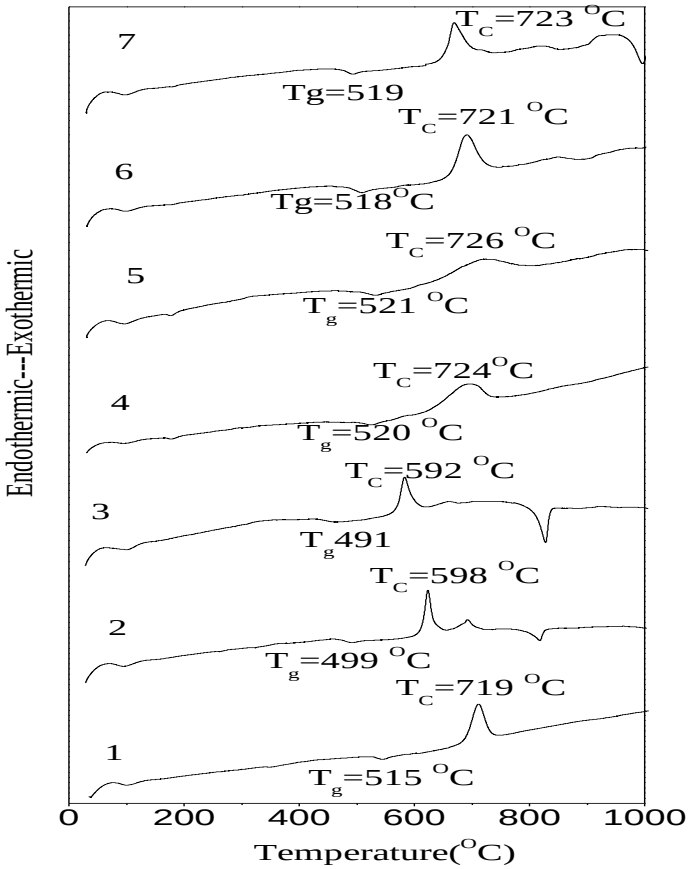
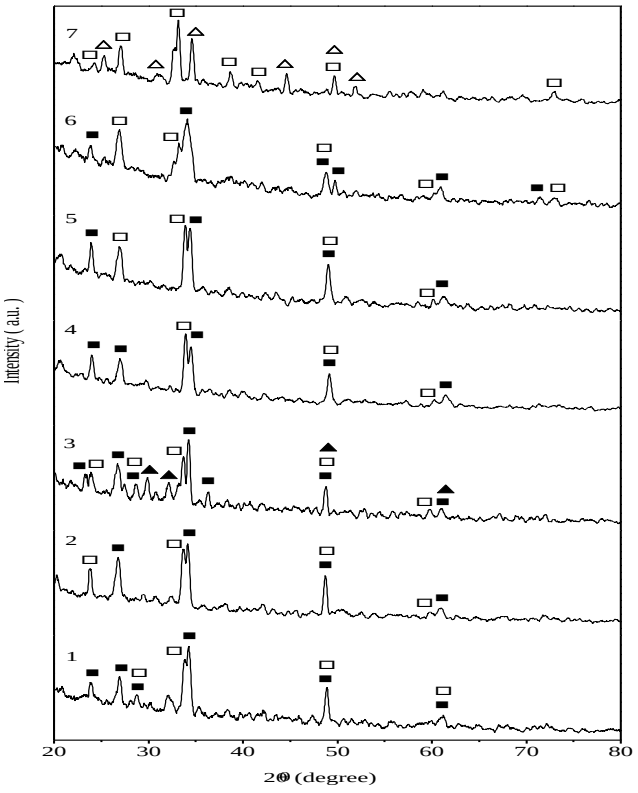


Fig .2. Differential Thermal Analysis (DTA) Curves of Bioactive Glasses Ceramics samples from Sl.No (1-7)



[□-(Na_2CaSi_3O8,)],[■-($Na_2CaSi_3O_9$)],[△-($Na_2CaB_5O_{10}$)],[▲- ($Li_2CaMg(PO_4)_2$]

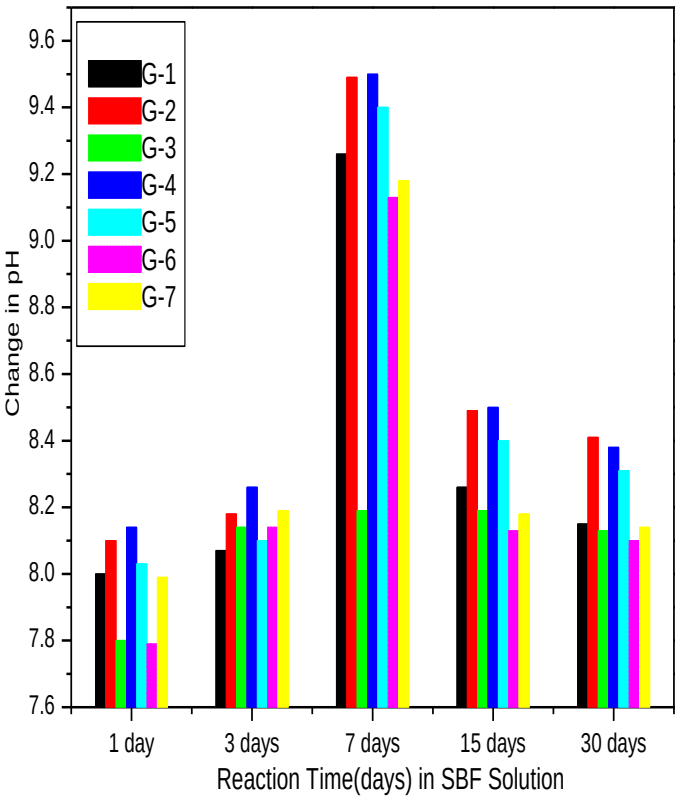


Fig. 4. Variation of pH with different time periods of bio glass samples at initial pH =7.4

Fig.3.X-ray Diffraction (XRD) Patterns of Ceramic Derivatives of Bioactive Glasses from Sl.No(1-7)

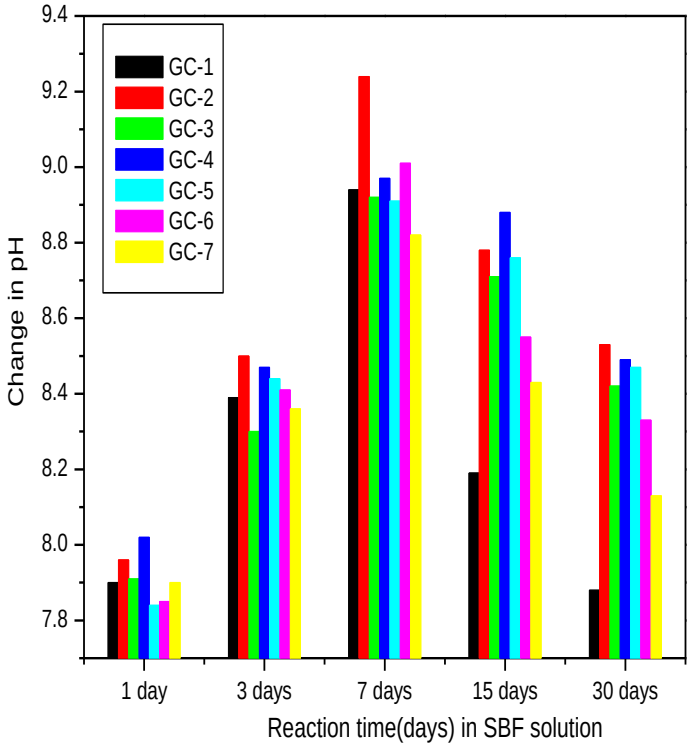


Fig. 5. Variation of pH with different time periods of bio glass ceramic samples at initial pH = 7.4

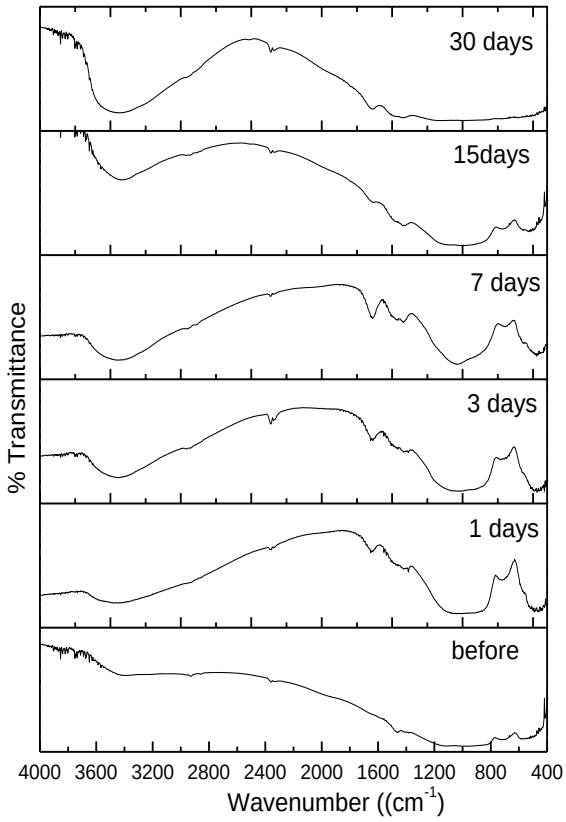


Fig.6.FTIR transmittance Spectra of Bio glass before and after SBF treatment (G-2)

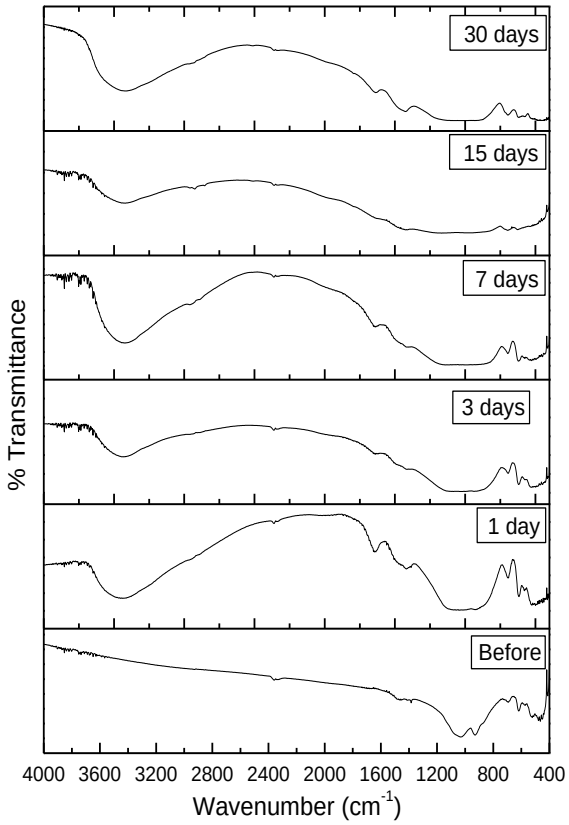


Fig..7: FTIR transmittance Spectra of Bio glass ceramics before and after SBF treatment (GC-2)

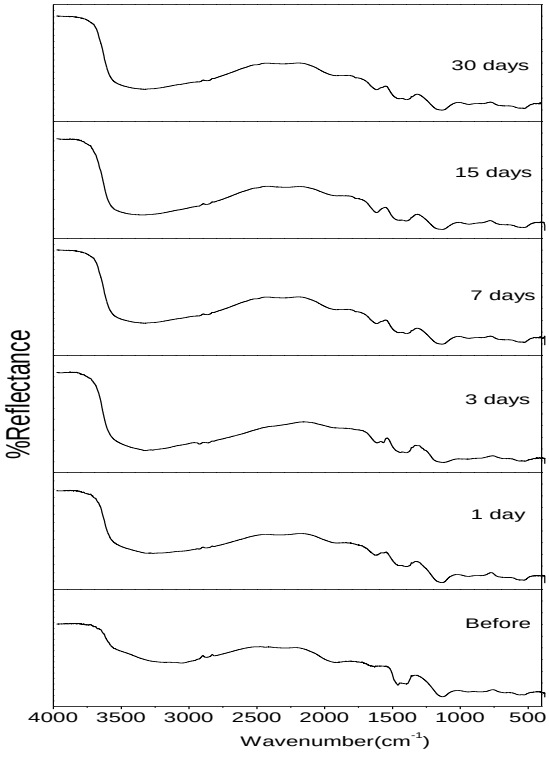


Fig.8.FTIR reflectance Spectra of Bio glass before and after SBF treatment (G-2)

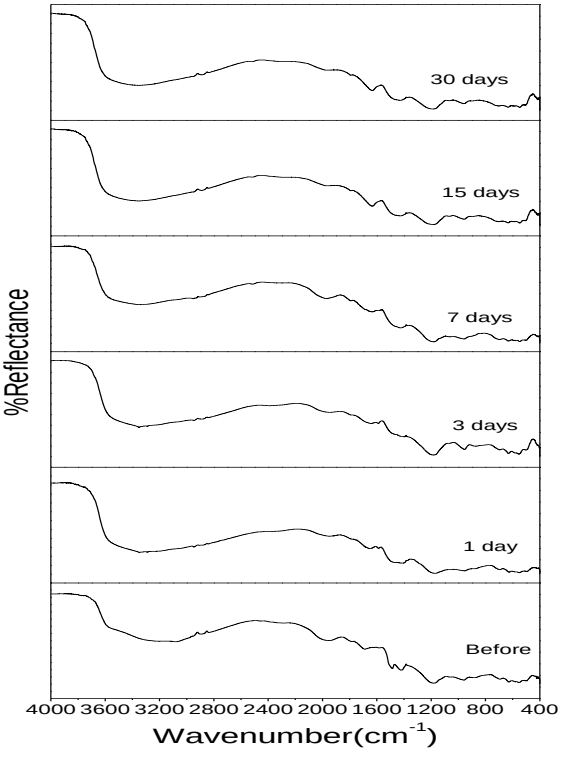


Fig..9: FTIR reflectance Spectra of Bio glass ceramics before and after SBF treatment (GC-2)

Table.1: Composition of Bioactive Glasses

Sl.No	SiO_2 (wt%)	Na_2O (wt%)	CaO (wt%)	P_2O_5 (wt%)	Li_2O (wt%)	K_2O (wt%)	ZnO (wt%)	MgO (wt%)	B_2O_3 (wt%)
1	45	24.5	24.5	6	-	-	-	-	-
2	45	19.5	24.5	6	2.5	2.5	-	-	-
3	45	14.5	24.5	6	5	5	-	-	-
4	45	24.5	19.5	6	-	-	2.5	2.5	-
5	45	24.5	14.5	6	-	-	5.0	5.0	-
6	40	24.5	24.5	6	-	-	-	-	5
7	35	24.5	24.5	6	-	-	-	-	10

Table. 2. Correlation between Wave number at which Transmittance Bands emitted and Functional Groups in Bioactive Glasses and their Ceramic Derivatives after immersing in Simulated Body Fluid (SBF) [44]

Wavenumber(cm^{-1})	Functional groups
400-500	Si-O-Si (bend)
500 - 560	P-O (Bend) (Crystalline)
560 - 600	P-O (Bend) (Amorphous)
720 - 840	Si-O-Si (Tetrahedral)
860 - 940	Si-O (Stretch)
1000 - 1100	Si-O-Si (Stretch)
1100 - 1200	P-O (Stretch)
1400 - 1530	C-O (Stretch)

Table. 3. Correlation between Spectral Frequencies and Functional Groups in a Bioactive Glass and their Ceramic Derivative and the Steps of Surface Changes by immersing in Simulated Body Fluid (SBF) [43]

Wavenumber(cm^{-1})	Vibrational mode	Surface reaction stages
860 - 940	Si-O (Stretch)	Stage 1 and 2
720 - 840	Si-O-Si (Tetrahedral)	Stage 3
560 - 600	P-O(Bend) (Amorphous)	Stage 4
500 - 560	P-O(Bend) (Crystalline)	Stage 5

The differential thermal analysis (DTA) curves of bioactive glasses show the glass transition temperature (endothermic effects) in the range of 491-525 ^{0}C and crystallization temperature (exothermic effects) in the range of 592-726^{0}C. The substitution of Na_2O with Li_2O and K_2O (bioactive glass 2 and 3) decreases of glass transition and crystallization temperature, with d the substitution of CaO with ZnO and MgO (bioactive glass 4 and 5) cause increase of glass transition and crystallization temperature, while the substitution of SiO_2 with B_2O_3 (bioactive glass 6and 7) cause increases of glass transition temperature and crystallization temperature [27].

3.3. X-ray diffraction results for bio glass–ceramics

The X-ray diffraction (XRD) pattern reveals information about the different crystalline phases. The X-ray diffraction (XRD) patterns of Ceramic Derivatives of Bioactive Glasses has been shown in Fig. 3. The X-ray diffraction (XRD) patterns of ceramic derivatives of all the bioactive glasses show the presence of two crystalline phases of sodium calcium silicate of the formula (Na_2CaSi_3O8, card number: PDF# 12-0684) and ($Na_2CaSi_3O_9$, card number: PDF# 45-0550) (except ceramic derivative of bioactive glass 3 and 7).The X-ray diffraction (XRD) pattern of ceramic derivative of bioactive glass 3 shows an additional crystalline phase, which is attributed to the formula calcium lithium silicate phase ($Li_2CaSi_3O_8$), while the X-ray diffraction (XRD) pattern of ceramic derivative of bioactive glass 7 shows an additional crystalline phase which is attributed to sodium calcium borate of the formula (($Na_2CaB_5O_{10}$)], card number: PDF# 38-0827).X-ray diffraction (XRD) patterns of ceramic derivatives of all the bioactive glasses indicate the formation of crystalline phases after the specified heat treatment. Each ceramic derivative of bioactive glass reflects specific crystalline phases depending on the chemical composition of the bioactive glasses and heat treatment conditions. The reason of the ease of crystallization of the bioactive glasses can be correlated with the presence of phosphate and silicate network, and the possible crystalline phase separation even in micro scale of the two crystalline phases depending on heat treatment conditions. It is well known that the addition of P_2O_5 (a few percent) to silicate glasses promotes volume nucleation and glass-ceramic formation as indicated by Elbatal[46]. have shown that the addition of B_2O_3 promotes volume crystallization in bioactive glasses but they showed that nucleation rates for CaO - P_2O_5 - B_2O_3 glasses were too low for practical glass–ceramic formation[36]. Hench reviewed the nature of crystalline phase separation in oxide glasses and showed that Li_2O, K_2O ,ZnO and MgO increases the tendency towards crystalline phase separation .ElBatal et al [46] has shown that the heat treatment of the parent bioactive glass 45S5 (bioglass or Hench glass) at a glass transition temperature of 525^{0}C and followed by heating at a crystallization temperature of 726^{0}C produced a ceramic derivative of bioactive glass containing the sodium calcium silicate crystalline phase ($Na_2Ca_2Si_3O_9$)[43,48]. Hench et al. showed that there is a relationship between the local structure of the modifier cations in silicate glasses of the systems (SiO_2 – CaO – Na_2O and SiO2 – CaO) and the nucleation ability [48]. These glasses can easily nucleate in the volume because they have similar local structures to their crystalline phases. They detected the crystalline phase $Na_2Ca_2Si_3O_9$ in the system SiO_2 – CaO - Na_2O after heat treatment at a crystallization temperature of 730 ^{o}C. It is obvious that the system SiO_2 – CaO - Na_2O - P_2O_5 has the tendency to form the sodium calcium silicate crystalline phase of the formula $Na_2Ca_2Si_3O_9$ as the main crystalline phase [36]. This tendency is confirmed in all the bioactive glasses (except bioactive glass 3 and7) where the $Na_2Ca_2Si_3O_9$ crystalline phase is detected by X-ray diffraction (XRD). In addition, there is another secondary crystalline phase ($Na_2CaSi_3O_8$) [40]..The effect of introduction of 5 and 10 % B_2O_3 to the bioactive glass 45S5 (bioactive glass 6,7) is shown to lead to the formation of a new crystalline phase of calcium sodium borate of the formula ($CaNa_3$ B_5O_{10}) [48]. The effect of introduction of ZnO and MgO to the bioactive glass 45S5 composition (bioactive glass 4 and 5) there is no additional crystalline phase occur.

3.4:Fourier Transform Infrared (FTIR) Absorption spectroscopy of Bio-glass and Bio-glass ceramic samples

Figs. 6–7 show the corresponding FTIR transmittance spectra of the bio glass and ceramic derivatives before and after treatment with SBF solution for different time period from 1 to 30 days. The FTIR transmittance bands of bioglasses and their ceramic derivatives were correlated before and after immersion in simulated body fluid (SBF) and were compared with vibrational mode according to Kim et al. [49] and Filgueira et al.[50] .The functional group was shown in table 2. Fig .6 show the bioglass (G-2) FTIR transmittance spectra peaks were noticed before SBF treatment at 413, 453, 498, and 713 cm-1. After SBF treatment the FTIR transmittance spectra peaks were noticed at 418, 432, 507, 526, 570, 686, 727, 1386, 1643 and 3454 cm-1. After SBF treatment new peaks were developed around at 507 and 526 cm-1 (P-O Bend- Crystalline), 570 (P-O Bend- Amorphous), 1643 (C-O stretch) and 3454 cm-1 (O-H stretching). The hydroxy carbonated apatite layer (HCA) formed on the surface of the bioglass and there was a slight change in the peaks intensity as the soaking time increases. The stages of formation of hydroxyl carbonate apatite were also mentioned earlier by Hench [1], ElBatal [46] and Oscar peitl [51]. Fig .7 show the bioglass-ceramic of GC-2 FTIR transmittance spectra peaks were noticed before SBF treatment at 410, 441, 534, 621, 692 and 731 cm-1. After SBF treatment the FTIR transmittance spectra peaks were noticed around at 420, 526, 570, 619, 686, 727, 1386, 1643 and 3454 cm-1. After SBF treatment new peaks developed at 507, 526 (P-O Bend-Crystalline), 570 (P-O Bend- Amorphous), 1643 (C-O stretch) and 3454 cm-1 (O-H stretching). The hydroxy carbonated apatite layer (HCA) formed on the surface of the bioglass-ceramic.

3.5. Chemical durability of bioglass (Changes in pH)

Fig.4 and Fig.5 show the change in pH with different time period for all the bioglasses and glass-ceramics. It was clearly observed from Figs 4 & 5 that the all samples the pH varies with change in compositions within 1 to 30 days compared to the initial pH of the solution (pH= 7.4) which is due to the fast release of Na^+ and Ca^{2+} ions through exchange with H+ or H_3O^+ ions into the solution[21]. The H^+ ions being replaced by cations, thereby increase in hydroxyl concentration of the solution, which leads to attack in silica glass network and formation of silanols to decrease in pH. The changes in pH are due to ion leaching i.e. chemical changes of material surfaces at different time periods. The increase in pH shows that the reduction in the concentration of H+ ion due to the replacement of cation ions in the glass and subsequent production of OH^- ions. It is also observed that the decrease in pH of the solution after 15days due to breaking of glass network. The reason may be considerable leaching of the glassy matrix from the surface. It can be understood that after 15 days the leached layer is removed and fresh layer is exposed, and therefore, demand for hydrogen ions is comparatively less. Similar change in pH, i.e. with decrease in pH to acidic region after 15 and 30 days respectively.

3.6.FTIR reflection spectra of bio glasses and their ceramic derivatives

Fig.8 and 9 show the FTIR reflectance spectra of the bioglas and their ceramic derivatives before and after immersion in SBF form 1to 30 days. The reflectance bands observed in FTIR spectra of bioglass and their ceramic derivatives were correlated wavenumbers corresponding to functional groups with (table 2) before and after immersion in simulated body fluid (SBF) as shown by Kim et al. [49] and Filgueira et al.[50].Fig.8 show the corresponding band frequencies of bioglass 448 (P-O Bend-Crystalline), 799 (P-O-P Stretch), 1558 (C-O Stretch) and 1605 cm-1 (C-O Stretch) and the frequencies of bioglass ceramics 519 (P-O Bend-Crystalline), 1558 (C-O Stretch) and 1604 (C-OStretch) as shown in fig .9 The formation of hydroxyl carbonate apatite on the surface was explained earlier by Hench [1], ElBatal [46] and Oscar peitl [51]. Fig .9show the reflectance spectra of bioglass samples then the reflectance bands were observed within the frequency range 442, 631, 1384 and 1470 cm-1 showing the presence of functional groups. These frequencies are due to various modes of vibrations 442 (Si-O-Si bend), 631 (P-O bend), 1384 (C-O-Stretch), and 1470 cm-1 (C-O-C Stretch). Bioglass treated with SBF solution with increasing duration of soaking time the reaction occurred as explained above and correlated with table 2 and 3. When the bioglass was converted to bioglass ceramic in fig 9, after then the FTIR reflectance spectra was recorded. The presence of various functional groups were identified with reference to corresponding frequencies 457 Si-O-Si (bend), 525, 613 (P-O Bend-Crystalline), 671 (P-O Bend), 722 (Si-O-Si -Tetrahedral), 921 (Si-O Stretch), 1031 ,1098 (Si-O-Si Stretch), 1384 cm-1 and 1469 cm-1 (C-O Stretch). Bioglass-ceramic treated with SBF.

4. Conclusion:

The density and the compressive strength increases with increasing the concentration of Li_2O and K_2O in the place of Na_2O in bio-glass composition No. (2) and (3) respectively. Similarly density and the compressive strength of the bio glass composition No.(4) and (5) increases with increasing ZnO and MgO in the place of CaO. Similar trend was observed while B_2O_3 is substituted for SiO_2 in bio-glass composition No. (6) and (7) respectively.The differential thermal analysis (DTA) curves of bioactive glasses reveals that the effect of introduction of Al_2O_3 and B_2O_3 in place of SiO_2 both nucleation and glass transition temperature increases ,an introduction ZnO and MgO in place of CaO respectively, in the bioactive glass 45S5 cause increase of both glass transition and crystallization temperatures, while the effect of introduction of K_2O in place of Na_2O, in the bioactive glass 45S5 cause decrease of both glass transition and crystallization temperatures. The X-ray diffraction (XRD) patterns of the bioactive glass-ceramics show the presence of two main crystalline phases of sodium calcium silicate ($Na_2CaSi_3O_8$, $Na_2CaSi_3O_9$). The effect of introduction of B_2O_3 in place of SiO_2, to the bioactive glass 45S5 is shown to lead to the formation of a new crystalline phase of calcium sodium borate of the formula ($Na_2CaB_5O_{10}$) and the effect of introduction of ZnO and MgO in place of CaO, to the bioactive glass 45S5 is shown that no additional crystalline phase developed other than two main crystalline phases of sodium calcium silicate ($Na_2CaSi_3O_8$, $Na_2CaSi_3O_9$) . The pH of the solution has varies from different glass samples No.(1-7) with substitution of different oxides after immersed in SBF solution. The fourier transform infrared (FTIR) absorption spectroscopy reveals that the effect of introduction of B_2O_3 in place of SiO_2, in the bioactive glass 45S5 has decreasing of bioactivity, a minor change of bioactivity in an introduction of Li_2O and K_2O in place of Na_2O, while the effect of introduction of MgO and ZnO in place of CaO, in the bioactive glass 45S5 cause a decrease in bioactivity. The fourier transform infrared (FTIR) reflectance spectra of the bioglass and bioglass-ceramic samples have also revealed that an increase in the number of bonds formed with increasing soaking time in SBF.

References:

1. L.L. Hench, R.J. Splinter W.C. Allen, T.K. Greenlee Jr. Bonding mechanism at the interface of ceramics prosthetic materials. J Biomed Mater Res Symp 2(1971) 117–41.
2. S.F. Hulbert. The use of alumina and zirconia in surgical implants. In:Hench LL, Wilson J, editors. An introduction to bioceramics. Singapore: World Scientific; (1993) 25–40.
3. T.Kokubo , Shigematsu M, Nagashima Y, Tashiro M, Nakamura T,Yamamuro T, et al. Apatite- and wollastonite-containing glass ceramics for prosthetic application. Bull Inst Chem Res Kyoto Univ. 60(1982) 260–68.
4. J. M.archo, J.L Kay, Gumaer RH, Drobeck HP. Tissue, cellular and subcellular events at bone-ceramic hydroxyapatite interface. J Bioeng 1(1977)79–92.
5. L.L. Hench Bioceramics: from concept to clinic. J Am Ceram Soc 74(1991)1487–510.
6. M. Neo, T .Nakamura, C. Ohtsuki,T. Kokubo ,T.Yamamuro . Apatite formation on three kinds of bioactive material at an early stage in vivo: a comparative study by transmission electron microscopy. JBiomed Mater Res 27(1993) 999–1006.
7. T. Kokubo A/W glass-ceramic: processing and properties. In: Hench LL, Wilson J, editors. An introduction to bioceramics. Singapore:World Scientific; (1993) 75–88.
8. S.D. Stookey, Ind. Eng. Chem. 51 (1959) 805.
9. W. Vogel, Chemistry of Glass, American Ceramic Society, Columbus, OH,(1985)
10. P.F. James, in: M.H. Lewis (Ed.), Glasses and Glass–Ceramics, Chapman and Hall,London, (1989) 59.
11. A.A. Ahmed, H.A. ElBatal, N.A. Ghoneim, F.A. Khalifa, J. Non-Cryst. Solids 41(1980) 57
12. M.Yamaguchi,H Oishi , Y Suketa . Stimulatory effect of zinc on bone formation in tissue culture. Biochem Pharmacol 36(1987)4007–12.
13. A Ito, K.Ojima , H.Naito , N Ichinose , T .Tateishi . Preparation,solubility and cytocompatibility of zinc-releasing calcium phosphate ceramics. J Biomed Mater Res 50(2000)178–83.

14.Y. Sogo ,T. Sakurai , K. Onuma ,A. Ito . The most appropriate (Ca + Zn)/P molar ratio to minimize the zinc content of ZnTCP/HAP ceramic used in the promotion of bone formation. J Biomed Mater Res .62 (2002) 457–63.
15. L.L. Hench, R.J. Splinter, W.C. Allen, T.K. Greenlee Jr., J. Biomed. Mater. Res. Symp. 2 (1971) 117–141.
16.L.Larry . Hench & June Wilson, "An Introduction to Bioceramics", World Scientific
Publishing Company: (1993).
17.N. Mohan Kumar, L. Szpiro et al., "Bioceramics: Properties, Characterizations and Applications", Springer: (2008).
18.L.Larry . Hench, "Bioceramics: From concept to clinic", Journal of American Ceramic Society: (1991) 1487 – 1510.
19.J.J.Vidueau ,V. Dupuis ., "Phosphates and biomaterials", European Journal of Solid State Inorganic Chemistry 28 (1991)303 – 343
20.L. L. Hench, "Bioceramics: Research and development opportunities", Brazilian Journal of Physics 22 (1992) 70 - 84
21.J.P.Zhong .,G.P. La Torre et al., "The kinetics of bioactive ceramics part VII: Binding of collagen to hydroxyapatite and bioactive glass", In Bioceramics: 7 (1994) 61 – 66.
22D.C..Greenspan .,J.P. Zhong et al., "Effect of surface area to volume ratio on in vitro surface reactions of bioactive glass particulates", In Bioceramics: 7(1994)28 – 33.
23.J.J.Mazer , J.V.Walther ., "Dissolution kinetics of silica glass as a function of pH between 40 and 85C", J Non-Crystalline Solids: 170 (1994) 32 – 45.
24.M.M.Pereira , A.E Clark. et al., "Effect of texture on the rate of hydroxyapatite formation on gel-silica surface", J American Ceramic Society: 78 (1995) 2463 – 2468.
25. O.P. Filho, G.P. La Torre, L.L. Hench, J. Biomed. Mater. Res. 30(1996) 509–514.
26.Zhong J.P., Greenspan D.C., "Bioglass surface reactivity: From in-vitro to in-vivo", In
Biomaterials: 10 (1997)391 – 394.
27.M. Brink, Tia Turunen et al., "Compositional dependence of bioactivity of glasses in the system Na_2O-K_2O-MgO-CaO-B_2O_3-P_2O_5-SiO_2", J Biomed Mater Res: 37 (1997) 114 –121.
28.D.C.Greenspa., J.P.Zhong . et al., "The evaluation of degradability of melt and sol–gel derived Bioglass in vitro", In Bioceramics: 10 (1997)391 – 394.
29.D.C.Greenspan , J.P.Zhong et al., "Bioactivity and biodegradability: Melt vs. sol-gel derived bioglass in vitro and in vivo", In Bioceramics: 11 (1998)345 – 348.
30.L.L..Hench "Biomaterials: A forecast for the future", Biomaterials: 19(1998)1419 –1423.
31.I. Rehman, J. C. Knowles et al., "Analysis of in vitro reaction layers formed on bioglass using thin-film X-ray diffraction and ATR-FTIR microspectroscopy", J Biomed Mater
Res: 41(1998) 162 – 166.
32.A. Stoch, W. Jastrzebski et al., "FTIR monitoring of the growth of the carbonate containing apatite layers from simulated and natural body fluids", Journal of Molecular Structure: 511 (1999) 287 – 294.
33. J.D. Rinehart Taylor T.D. et al., "Real-time dissolution measurement of sized and unsized calcium phosphate glass fibres", J Biomedical Materials Research: 48 (1999) 833 – 840.
34.Oscar Peitl, Edgar Dutra Zanotto, et al., "Highly bioactive P_2O_5-Na_2O-CaO-SiO_2 glass- ceramics", Journal of Non-Crystalline Solids: 292(2001) 115 - 126.
35.Mathai Mathew and Shozo Takagi, "Structures of Biological Minerals in Dental Research", Journal of Research of the National Institute of Standards and Technology: 106,(2001)1035 – 1044.
36.P. Sepulveda, J.R. Jones et al., "In vitro dissolution of melt-derived 45S5 and sol-gel derived 58S bioactive glasses", J Biomed Mater Res: 61 (2002)301 – 311.
37.E. Kontonasaki, T. Zorba et al., "Hydroxy carbonate apatite formation on particulate bioglass in vitro as a function of time", Crystal Research and Technology: 37 (2002) 1165 – 1171.
38.H.A. ElBatal, M.A. Azooz et al., "Characterization of some bioglass–ceramics", Materials Chemistry and Physics: 80 (2003)599 – 609.
39.R. Xin, Yang Leng et al., "A comparative study of calcium phosphate formation on bioceramics in vitro and in vivo" Biomaterials: 26 (2005) 6477 - 6486.
40.Irene Barrios de Arenas, Carol Schattner et al., "Bioactivity and mechanical properties of Na_2O-CaO-SiO_2-P_2O_5 modified glasses", Ceramics International: 32 (2006) 515 – 520.
41.Qizhi Z. Chen, Ian D. et al., "45S5 Bioglasss-derived glass-ceramic scaffolds for bone tissue engineering", Biomaterials 27(2006)2414-2425.
42.Aldo R. Boccaccini, Qizhi Chen et al., "Sintering, crystallization and biodegradation behaviour of bioglass-derived glass–ceramics", The Royal Society of Chemistry: 136 (2007) 27 – 44.
43.G.A. Stanciu, I. Sandulescu et al., "Investigation of the hydroxyapatite growth on bioactive glass surface", Journal of Biomedical & Pharmaceutical Engineering: 1 (2007) 34 - 39.
44. O.P. Filho, G.P. La Torre, L.L. Hench, J. Biomed. Mater. Res. 30 (1996) 509–514
45.S.M. Best, A.E. Porter et al., "Bioceramics: Past, present and for the future", Journal of the European Ceramic Society: 28 (2008)1319–1327.
46. H. ElBatal, Amany ElKheshen, "Preparation and characterization of some substituted bioglasses and their ceramic derivatives from the system SiO_2–Na_2O–CaO– P_2O_5 and effect of gamma irradiation", Materials Chemistry and Physics: 110, (2008), 352 – 362.
47M.H. Fathi, A. Doost Mohammadi, "Preparation and characterization of sol–gel bioactive glass coating for improvement of biocompatibility of human body implant", Materials Science and Engineering: 474 (2008) 128–133.
48. Hench L.L., Splinter R.J. , Allen W.C., Greenlee Jr T.K.., J. Biomed.Mater. Res. Symp. 2 (1971) 117–141.
49. C.Y. Kim, A.E. Clark, L.L. Hench, J. Non-Cryst. Solids 113 (1989)195–202.
50. Filgueiras M.R.T., La Torre G.P., Hench, L.L. J. Biomed. Mater. Res. 27 (1993) 1485– 1493.
51. Oscar peitl, E.D.Zanotoo, L.LHench : " Highly bioactive P2O5-Na2O-CaO-SiO2 glass- ceramics", Journal of non-crystalline solids 292(2001)115-126.