PREPARATION AND CHARACTERIZATION OF SILICON FREE BIOACTIVE GLASS BY QUECHING

PROJECT REPORT SUBMITTED IN PARTIAL FULFILMENT OF

REQUIREMENT FOR THE AWARD OF THE DEGREE OF
MASTER OF SCIENCE

IN

PHYSICS

BY

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ACKNOWLEDGEMENT

I would like to express my deep sense of regards to my supervisor, Prof. B.K. Choudhuri, Ph.D, D.Sc, Department of Physics, NIT, Rourkela for his helpful discussions, valuable suggestion, constant encouragement and support throughout the course of my research work.

I am also thankful to Prof. D.K Bisoyi Head of department of National Institute of technology, Rourkela for his profound support, without which this work would have been impossible.

I express my deep sense of gratitude towards all the faculty members & research staff of department of NIT Rourkela, for their continuous support and cooperation.

I also want to express my deep sense of gratitude towards Mr Udaya Kumar Department of Metallurgy and Material science for helping me in making glass which is necessary for the study of my project.

Especially I am grateful to our senior research fellow for their relentless help, support and heartily cooperation throughout my research work.

Date: Madhusmita Behera

CERTIFICATE

This is to certify that the thesis entitled, **"Preparation and characterization of Silicon free bioactive glass by Quick quenching"** being submitted as 1 year training report for fulfilling the requirements for the award of the degree of Masters of Science in physics of the **National Institute of Technology, Rourkela**, is a faithful record of bona fide research work carried out by **Ms. Madhusmita Behera** under the supervision of **Prof B.K Choudhuri, Visiting Professor,** NIT, Rourkela and that no part of this thesis has been submitted for any other degree or diploma or published in any form. It is further certified that the assistance and help availed of during the course of the study have been duly acknowledged.

(Prof. D.K Bisoyi) **(Prof. B.K choudhuri)**

DECLARATION

I hereby declare that the thesis entitled***, "Preparation and characterization of Silicon free bioactive glass by Quick quenching"*** submitted to Department of Physics, National Institute of Technology, Rourkela, in partial fulfilment of the requirements for the award of degree of Masters of Science in physics is an authentic record of the work carried out by me and no part of this thesis has been submitted for any other degree or publication in any form.

Date: Madhusmita Behera

ABSTRACT

Synthesis of bioactive glass that is a silicon free bioactive borate glass is achieved by the traditional method means by melt-casting. The glass was prepared by taking (40.9%) CaO-(34.9%) B_2O_3-(24.2%)BaO (BBCO) then by quick air quenching from 1250C to room temperature. XRD, DTA, and TGA studies are made to characterize the glass. Hydroxyapatite (HAp) or $Ca_{10}(PO_4)_6(HO)_2$ formation on the glass surface was tested by soaking the glass in K_2HPO_4 (0.25M with pH = 8.5) at 37^0C. Nanorods or bubbles of HAP are formed on the glass surface by soaking the glass for longer time. Fourier transform infrared spectroscopy (FTIR), scanning and election and high resolution transmission electron microscopic studies confirmed HAP formation.

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CHAPTER-1

1.1 INTRODUCTION:

There are many recent advancement and development of Tissue engineering and applications. The current researches in the field of "tissue engineering and application" mostly focus on the use of bioactive glass in tissue culture. Though its inherent brittleness, bioactive glass has several alluring characteristics as a scaffold material for bone tissue engineering. New bioactive glasses made up of borate and borosilicate compositions and also showing the ability to stimulate new bone formation when compared to bioactive glass made up of silicate composition. Borate based bioactive glass also have reliable degradation rates, so the degradation of bioactive glass implant can be more precisely matched to new bone formation [1]. Before going towards materials and methods of synthesis and characterization of silicon free bioactive glass in details one should have the basic understanding about historical background of bioactive glass, glass, type of glass and its structure and properties, bioactive glass and finally about the material and methods of synthesizing and characterizing the bioactive glass.

1.2 GLASS:

Glass is an amorphous material which means non-crystalline solid material showing glass transition. Glass is commonly brittle and can be optically transparent. There are many feasible definitions of glassy phase. An interesting one is that glass is both solid and liquid at the same time. Which means the glass is solid, morphologically but its internal structure is similar to that of liquid. Somehow this indicates the reasonable process necessary to obtain a glass. A glass is formed when a liquid is over cooled in absence of enough nucleation sites, where they can attain minimum energy configuration for their stability [2]. Therefore glass is also called as super cooled liquid. The resulting structure of the glass is not completely organized, quite of the crystalline structure typical of all other solids. For this reason glass structure is called amorphous which means without a definite shape. For a better understanding of glass structure we must understand glass transition.

1.3 GLASS TRANSITION:

The glass transition is described to be the reversible change in amorphous materials between a hard, brittle state into a rubbery molten state. Materials which are capable of going through a glass transition are named as glass.

Before considering particular glass forming techniques, some of the temperature sensitive properties of glass materials must be introduced. Glassy or non-crystalline materials do not solidify in the same manner as the crystalline materials do. While cooling a glass becomes more and more viscous in a continuous manner with decreasing temperature; there is no exactly specific temperature at which a liquid transforms to a solid as the crystalline materials usually transform its phase. In fact, one of the differences between crystalline and non-crystalline materials exists in the dependence of specific volume (or volume per unit mass, the reciprocal of density) on temperature. More clearly in crystalline materials there is a discontinuous decrease in volume at the melting temperature Tm. However for glassy material, volume decreases continuously with decrease in temperature; but there is a slight decrease in slope of the curve occurs at a point what is called the glass transition temperature, or fictive temperature, Tg. Glass transition temperature is a metastable transition. Below this temperature, the material is specified to be a glass it means is typically hard and brittle; above, it is first a super cooled liquid ie a rubber like viscous liquid, and finally liquid [3].

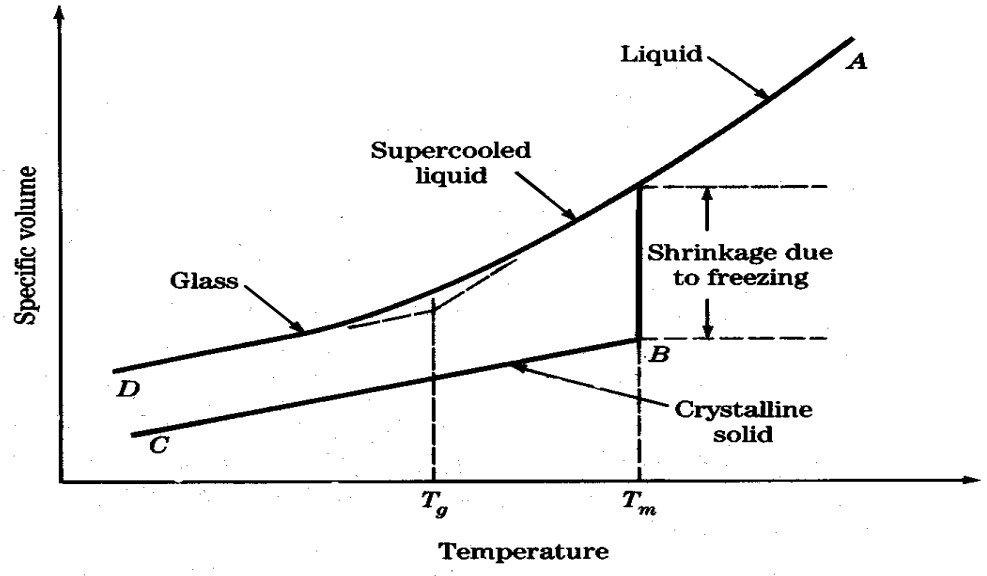

Figure 1.3.1 [4]

Contrast of specific volume – versus temperature behaviour of crystalline/glassy and non-crystalline materials. Crystalline materials solidify at the melting temperature Tm. Characteristics of the non-crystalline/glassy state is the glass transition temperature Tg.

1.4 GLASS ITS STRUCTURE AND PROPERTIES:

Most of glasses are silicate glass. In a crystalline solid made of silica (eg quartz), SiO_4 tetrahedral are organized in a well-defined network i.e, in a proper periodic way giving rise to long range ordering. But in amorphous silica SiO_4 tetrahedral are still linked together but the angle between them is not constant and as a result structure is not organized. The SiO_2 is called glass former because the glass structure is mostly kept together by SiO_4 tetrahedral. Other oxides can be used as B_2O_3 and P_2O_5. Alkaline and alkaline-earth oxides are added together to glass as glass modifiers, in that they interrupt the network created by glass former. The resulting structure is called random network [5].

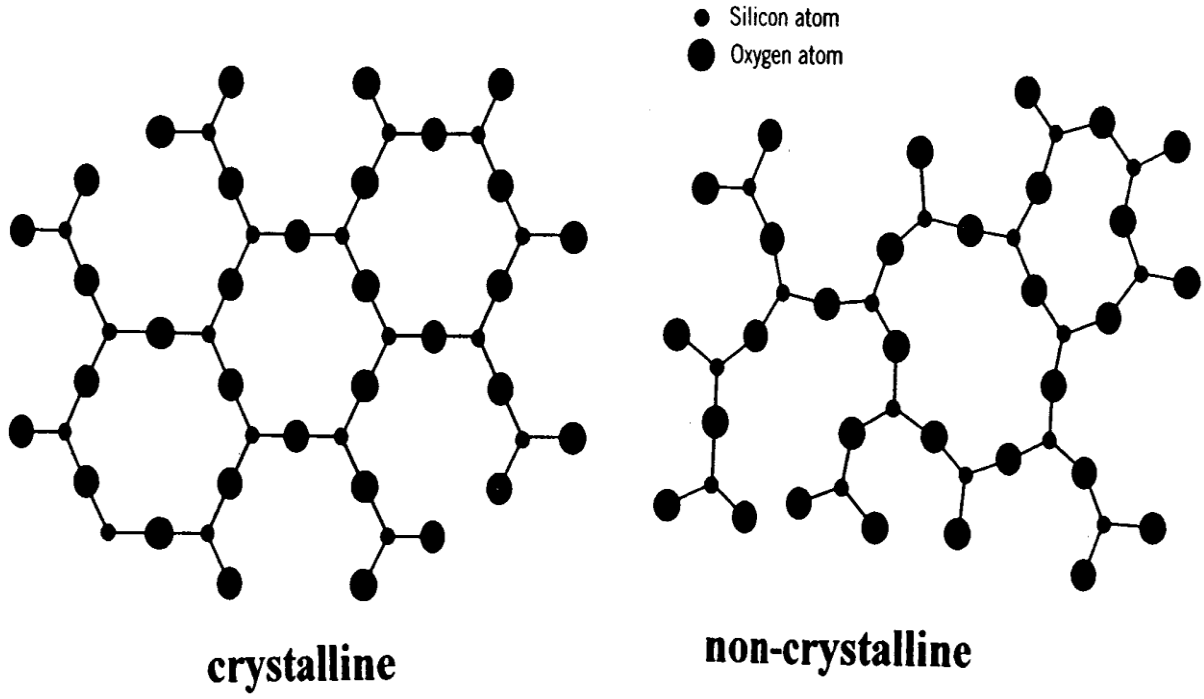

Figure-1.4.1

1.5 TYPES OF GLASS: There are different types of glass namely Optical glass, Soda-lime glass, Photo-chromatic glass, Pyrex glass, Borosilicate glass, Lead glass, Insulating glass, Metallic glass, Semiconducting glass and Bioactive glass etc [6]. Here our main concern is the study of Bio-active glass. So one has to understand what is bioactive glass?

1.6 BIOACTIVE GLASS (bio glass 45S5)

These bioactive glass or bio glasses were generally used for bone replacement therapy. Usually these bio glasses dissolved in the body fluid or blood plasma present in our body and takes the place of bones, because of its composition stimulates bone and cell regeneration. It was believed that Hydroxyapatite to be the best biocompatible replacement material, after Larry Hench experiment, he developed a material using silica (glass) as the host material, assimilate with calcium and phosphorous to bind broken bones. This material resembles bone material and stimulates the re-growth and regeneration of new bone material. Thus, due to its biocompatibility and osteogenic capacity it came to be known as "bioactive glass- bio glass" .The original bio glass (45S5) composition is as follows: 45% silica (SiO_2), 24.5% calcium oxide (CaO), 24.5%sodium oxide (Na_2O), and 6% phosphorous pentoxide (P_2O_5) in weight percentage. Bioglass material is composed of minerals that are naturally occurring in the body (SiO_2, Ca, Na_2O, H, and P), and the molecular proportions of the calcium and phosphorous oxides are similar to those in the bones. After the implantation of bioglass in the body or when it is subjected to an aqueous solution, or body fluids, converts to a silica-CaO/P_2O_5-rich gel layer that subsequently mineralizes into hydroxycarbonate in a matter of hours.[7,8,9] More the dissolution, better the bone tissue growth.[10] This gel layer resembles hydroxyapatite matrix so much that osteoblasts were differentiated and new bone was deposited.[11]

$Ca_5(PO_4)_3(OH)$ is the chemical formula for hydroxyapatite, a natural mineral form of calcium apatite and usually written as $Ca_{10}(PO)_6(OH)_2$ [12]

1.8 OBJECTIVE OF SYNTHESIZING SILICON FREE BIOACTIVE GLASS:

Since there is a huge demand for a stable biomaterial which is chemically inert, biocompatible as well as long lasting material, therefore different research materials have been put forward in this competition of stable biomaterial. The Bio ceramic and Bio active glass has been proved themselves stable enough in different field of tissue engineering and application Hence before moving to synthesis and characterization of bioactive glass one has to understand the glass and its structure and properties.

Different types of bioactive materials have been developed over last 3 decades. Among these the main bioactive materials used clinically are: bioactive glasses in the SiO_2-Na_2O-CaO-P_2O_5 system, commonly known as 45S5 system [1]. The Biocompatibility (capability of cells

adhesion, proliferation and differentiation) of 45S5 system and some oxide glasses having silicon has lead them used as implant materials in human body to repair, and replace diseased or damaged bones or tissues [13].These surface reactive oxides glasses induce bioactivity forming strong bond with living bone tissues. The specialty of a bioactive glass is that hydroxyapatite (HAp) or $Ca_{10}(PO_4)_6(OH)_{2(}$,(i.e, the mineral phase of bone or) the main component of bone, is formed on the glass surface when comes in contact with the body fluid *in vivo* [14] or simulated body fluid (SBF) *in vitro* [15-16]. Due to this important property, different bioactive glasses with SiO_2 were prepared and characterized for tissue engineering applicants [16-20]. The high temperature (more than 1200^0C) melting SiO_2 based bioactive glasses viz. 45S5 (46.1 mole% SiO_2, 26.9 mole% CaO, 24.4 mole% Na_2O and 2.5 mole% P_2O_5), 45 (60 mole% SiO_2, 36 mole% CaO and 4 mole% P_2O_5) , 70S30C(70 mole% SiO_2, 30 mole% CaO) have been elaborately studied by different research groups [21-26]. In some cases silica present in these glasses remains in human body, after being implanted for a long time. This might create health problems [27-29//31-33].Some Bioactive borate glasses also showed HAP formation on their surface by soaking them in KH_2PO_4 solution [x]. Therefore different SiO_2 free borate glasses (like Na_2O-CaO-B_2O_3 , Na_2O–BaO–B_2O_3,CaO-B_2O_3 etc. [18,19,28-30///,27-30]) have been thoroughly investigated showing faster and almost complete conversion into HAp **[**18,19,28].Upon immersion of these glasses or corresponding glass powders **[**18,19,31**]** in an aqueous phosphate solution, the metal ions like Na and B are dissolved into the solution, whereas the Ca ions react with the phosphate the Ca ions react with the phosphate ions to form a calcium phosphate (typically HAp with Ca/P~1.67) with proper pH values. The interface reaction and HAp formation processes start at the surface of the glass and move uniformly inward, until the glass is completely converted to HAp (if sufficient phosphate ions are available). Most of these high melting and multi-component bioactive glass powder are used for making porous scaffolds [28-31//32,34] for tissue engineering applications. Some high melting glasses like Al_2O_3-P_2O_5-ZnO have also been made in the form of fibres **[**32//35**]** for growing human skeletal muscle cells.

CHAPTER-2

LITERATURE SURVEY

2.1 Synthesis, characterization and in vitro bioactivity of SiO_2-CaO-P_2O_5 sol- gel glasses highlighted by XRD technique.

Ungureanu D.N., Angelescu N., Avram D., Catangiu A., Bratu V., Stoian E.V

The scientific bulletin of VALAHLA University- MATERIALS and MECHANICS-Nr. 6(year 9) 2011.

In vitro bioactive test means test of the sample in a simulated bio fluid solution whose ion and mineral concentration is likely to be equally with blood plasma. The XRD Technique can evaluate the deposition of Hydroxy apatite which is the main component of bone. But the method of preparation of glass is through a sol-gel method, which is one of the chemical approaches.

2.2 Characterization of bioactive glasses: "Effect of the immersion in solutions that stimulates body fluids".

Marta Giulia Cerruti

University of Turin Department of chemistry IFM Ph.D. thesis

The history of bioactive material leads to the innovation in preparation of such a bio glass which is inert and non toxic to the human body. Glass former can be boron oxide and phosphorous pentoxide and the structure of glass is dependent on the glass modifier. Index of bioactivity can be calculated by using some mathematical formulae. Different techniques used to analyse the bioactive glass.

2.3 Bioactive Glass and Glass-Ceramic Scaffolds for Bone Tissue Engineering

Lutz-Christian Gerhardt 1 and Aldo R. Boccaccini 1,2,*

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2 Institutes of Biomaterials, University of Erlangen-Nuremberg, 91058 Erlangen,

Bioactive glass-ceramic composite is showing very good cell adhesion with the living tissue for tissue culturing technique. Porous scaffolds showing the good adhesion with the bioactive glass-ceramic materials and the formation of Hydroxyapatite increases in this type of composites.

2.4 Preparation and in vitro characterization of novel bioactive glass ceramic nanoparticles Zhongkui Hong,1,2 Rui L. Reis,1,2 Joa˜o F. Mano1,2

13B's Research Group – Biomaterials, Biodegradables and Biomimetics, Department of Polymer Engineering,University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal

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Received 3 September 2007; revised 5 October 2007; accepted 18 October 2007

Bioactivity increases with adding certain amount of nano crystals. Also annealing the glass up to certain temperature gives rise the formation of nano crystals as a result bioactivity increases.

CHAPTER-3

3.1. MATERIALS AND METHODS:

Since we are concerned about silicon free bioactive glass so we must stick to borate glasses like Na_2O-CaO-B_2O_3, Na_2O–BaO–B_2O_3,CaO-B_2O_3 etc. Therefore we require different chemicals for the preparation of glass. Here one can prepare two types of bioactive glasses

i- Melt –derived bioactive glass (physical approach)

ii- Sol-gel bioactive glass (chemical approach)

Here we were concerned with physical approach i.e, melt derived method of bioactive glass preparation. In this method the compounds of different compositions of chemicals is melted at a higher temperature and suddenly quenched to room temperature within seconds in atmospheric air hence also known as air quenching. Here in this chapter will discuss about methods of preparation and method of characterization of bioactive glass in details.

3.2. MATERIALS:

Analytical grade $CaCO_3$, H_3BO_4, $BaCO_3$ and H_2HPO_4 all of purity 99% were purchased from E Merck, India. Dulbecco's modified eagle medium, DMEM (Gibco, USA), Ficoll Hypaque (Histopaque-1077; Sigma, MO, USA), phosphate buffer solution (PBS) and (FBS) were purchased from Gibco,(USA) .

3.3. GLASS PREPARATION:

The bioactive BBCO glass with composition (40.9%) CaO-(34.9%) B_2O_3-(24.2%) BaO was prepared by using $CaCO_3$, H_3BO_4, and $BaCO_3$ utilizing using standard splat cooling method **[38]**.This typical glass composition was chosen as its glass quenching temperature (~1150C) lies within 1200K. Most of the other glasses with varying compositions, liquid quenching temperatures exceed 1200C. The appropriate amounts of the well mixed powders were melted in a platinum crucible around 1150^oC for one hour and then quickly quenched between to copper block kept at room temperature. Thin (1-2mm thickness) transparent glass plates were thus obtained. Fine thin glass fibres (0.05-0.1mm diameter) were also prepared by pulling the same in molten condition.

3.4. SOAKING THE DIFFERENT BORATE GLASSES IN KDP SOLUTION AND SBF:

Transparent glass plates (~1-1.2mm thick) and thin fibres' (0.05mm diameter) were immersed in K_2HPO_4 solution (0.25M with pH =9.5) in different 100ml glass beakers kept at 37^oC for (7 to 30 days). Phosphate ions present in the solution react with the BBCO glass forming hydroxyapatite. A glass plate (~10mm in diameter and ~ 5 mm thickness) or a glass fibre was immersed in 100 cm^3 of the phosphate solution. After a given immersion time (maximum 30days), the glass/fibre was washed well with distilled water and dried in vacuum and used for surface morphology studies using Field emission scanning electron microscope (FESM).

3.5. METHODS OF CHARACTERIZATION:

Glass samples and HAp were characterized by XDR (Bruker, Model no. D8 Germany), FTIR (Perkin–Elmer spectrum 100 FTIR spectrometer with a 4 cm^{-1} resolution), DTA/TGA analysis (TA Instruments: SDQ600). Field emission scanning electron microscope (FESEM: JEOL JSM-6700F), and high resolution transmission electron microscope (HRTEM: Model JEM-2010, JEOL) studies. Prior to SEM analysis, cell-seeded scaffolds were rinsed in Dulbecco's Phosphate Buffered saline DPBS buffer, and fixed with 2.5% gutaraldehyde in DPBS overnight at 4^0C. The construct were dehydrated by exposure to a gradient series of alcohol followed by aseptic critical point drying, and coated with platinum before observing under JEOL JSM-840A scanning electron microscope.

3.5.1. X- RAY DIFFRACTION:

X-ray diffraction is the non-destructive technique used to characterize the sample. This technique is used for analysing the internal structure of the sample ie whether it is crystalline, amorphous, and liquid or glass. XRD technique is generally used for the identification of phases. In case of crystalline solid it also provides information about the crystal orientation as well as the unit cell dimensions of the crystal sample.

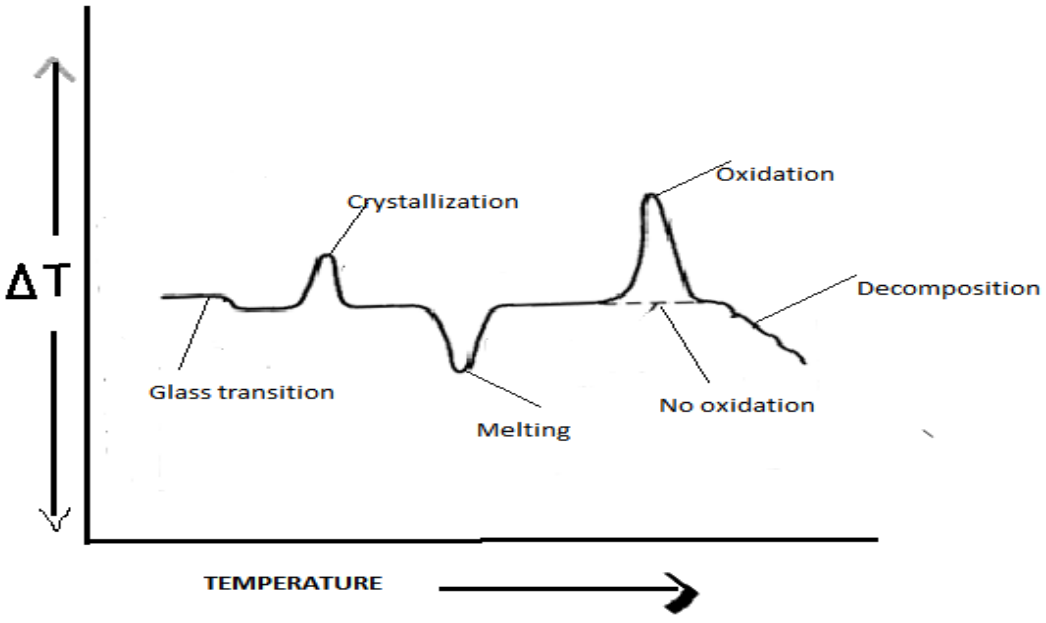
In this work we used this technique to evaluate the glassy phase after preparation of glass by quenching, and also identify the phase formation after being soaked in the SBF solution. X-ray diffraction patterns were recorded with a PAN analytical system X'pert

HIGH SCORE with Ni Filtered Cu Kα (λ=1.53A°). From this we found that X-ray diffraction has become a significant and a powerful tool for the structure and characterization in solid state physics and material science. The analyzed data is given below [3].

3.5.2. DTA: DIFFRENTIAL THERMAL ANALYSIS:

Differential thermal analysis (DTA) is the most extensively used thermal analysis techniques. This technique used to examine thermal events in a sample ie the crystallization, melting, solidification, decomposition, oxidation etc by heating or cooling without mass exchange with its surroundings. In this technique sample is examined with increasing and decreasing the temperature. The word differential is coming in to picture because in this technique two materials are taken in to consideration while heating or cooling the sample. One material is taken as reference material which is inert and other one is the sample. Therefore one thermo couple is used such that the change in temperature can be measured during heating and cooling process. During the process of heating or cooling the thermal events in the sample takes place with respect to the inert reference material and it will exactly measure time and temperature at which the thermal event took place as well as the change in temperature of the sample with respect to the reference material [37].

In this work we want to analyse the glass transition temperature which is a thermal event so for measuring the glass transition temperature we used this technique.



3.5.3. TGA: THERMO GRAVIMETRIC ANALYSIS:

Thermogravimetry (TG) is a technique which is used for measuring change in mass of a sample with temperature. It measures the change in weight of the sample while heating and cooling the sample. The sample which is to be measured is placed within a furnace and its mass change is observed. A TGA consists of a sample pan that is supported by a precision balance. That pan lies within the furnace and is heated or cooled during the experiment. The mass of the sample is observed during the experiment [37].

In our study this technique is used for the confirmation of glass transition temperature of the prepared glass.

3.5.4. SEM:

The scanning electron microscope used a focused high energy electron beam to produce a range of signals of the surface of the solid specimens. These signals are derived from electron sample interaction and giving information about the sample including external and surface morphology, crystal structure and orientation, chemical composition and deposition or formation of any phase on the surface of material. It can generate two dimensional scanned images over a selected area of the sample surface. Areas of the sample surface ranging from approximately 1cm to 5 micron in width can be imaged and scanned thoroughly in a scanning mode using conventional SEM.

Here in our case we used this microscopy technique to analyse the deposition or formation of phase over the glass surface.

3.5.4. FE-SEM: (Field Emission Scanning Electron Microscope) :

FESEM is microscope the electrons beams are generated by a field emission source. The object is scanned by electrons in a zig-zag pattern to visualize very small topographic details on the surface or entire or fractioned sample. Researchers apply this technique to observe structures that may be as small as 1 nanometer (= billion of a millimeter). In this technique one can able to see the nanoparticles, nanorods, nanowires, nano powders, nanofibres etc [37].

3.5.5. FTIR SPECTROSCOPY:

Fourier transform infrared spectroscopy is the non destructive technique used for the compositional analysis of the sample. In this spectroscopy method different wave numbers are corresponding to different functional group. These wave numbers are also due to the mode of vibration of bonds within the sample. Each sample has unique characterization peak and this uniqueness in the peaks will identify the composition and elements present within the sample. [37]

In our work we required to evaluate the material composition of the deposited material on the soaked glass surface.

3.5.6. HR-TEM (High resolution transmission electron microscope):

In TEM is the ability to obtain full morphological (grain size, grain boundary and interface, secondary phase and distribution, defects and their nature, etc.), atomic crystallographic structural and microanalytical such as chemical composition specially at nm scale, bonding (distance and angle), electronic structure, coordination number) data from the sample makes it unique.TEM is the most systematic and flexible technique for the characterization of materials. The HRTEM allows direct imaging of the atomic structure of the sample.

In our study we use this technique for direct imaging of nano crystals.

3.5.7. SAED (Selected Area Electron Diffraction):

A diffraction pattern is formed on the back-focal plane of the objective lens of Transmission electron microscope when an electron beam passes through a crystalline specimen. In the diffraction mode, the nano crystalline structure can be analysed similar to that of XRD. In this mode pattern of selected area diffraction (SAD) can be further enlarged on the screen and also recorded by a camera. SAED in a TEM, however, shows its special characteristics compared with X-ray diffraction. Constructive diffraction from a lattice plane (hkl) generates a range of intensity spot on the screen arranged in the form of rings in SAED mode [37].

This technique is used in our study to find the interplannar spacing of the nano crystallites.

CHAPTER-4

4.1. RESULT AND DISCUSSION:

The Differential thermal analysis of the bioactive glass sample was carried out and the glass transition temperature of the prepared BBCO Glass was found out through DTA-TGA analysis. X-ray diffraction analysis (XRD) is performed by using a X-ray diffractometer adopting Ni filter and Cu target of wavelength Cu Kα- 1.53 A°. The XRD patterns of the sample were recorded in a 2θ range of 20-70°. The JCPDS – International centre for diffraction data cards was used as a reference data for the XRD patterns interpretation in the present work. The bioactive glass samples were investigated by Fourier transform infra-red reflectance FTIR spectroscopy. The FTIR reflectance spectra were obtained between wavenumber 4000-400 cm^{-1} resolutions.

In vitro bioactivity test is performed in simulated body fluid. Whenever bioactivity test is carried out in actual living body then it is called as in vivo bioactivity test. And when this bioactive test is being carried out in simulated body fluid then it is called in vitro bioactive test. The concentration of ions in simulated body fluid is nearly equal to that of human blood plasma. This simulated body fluid (SBF) solution was prepared by dissolving the required amount of reagent grade chemicals, the Sodium chloride (Nacl,) sodium bicarbonate ($NaHCO_3$), potassium chloride (KCl), di-potassium hydrogen phosphate (K_2HPO_4:$3H_@O$) magnesium chloride hexahydrate ($MgCl_2$:$6H_2O$), calcium chloride dehydrate($CaCl_2$: $2H_2O$) and sodium sulphate (Na_2SO_4) in distilled water and was buffered with pH value of 7.40. In this SBF prepared the bioactive glass of some measured thickness and size soaked for around 15-20 days. Then its surface morphology was studied with SEM, FE-SEM, HRTEM, SAED analysis and interpreted with previous literature available regarding deposition of HAp over the glass surface.

Figure 3.2.1

Figure 3.2.2

Figures-3.2.1 and 3.2.2 showing, respectively, the transparent glass plate and a glass plate soaked in K2HPO4 solution for 30 days.

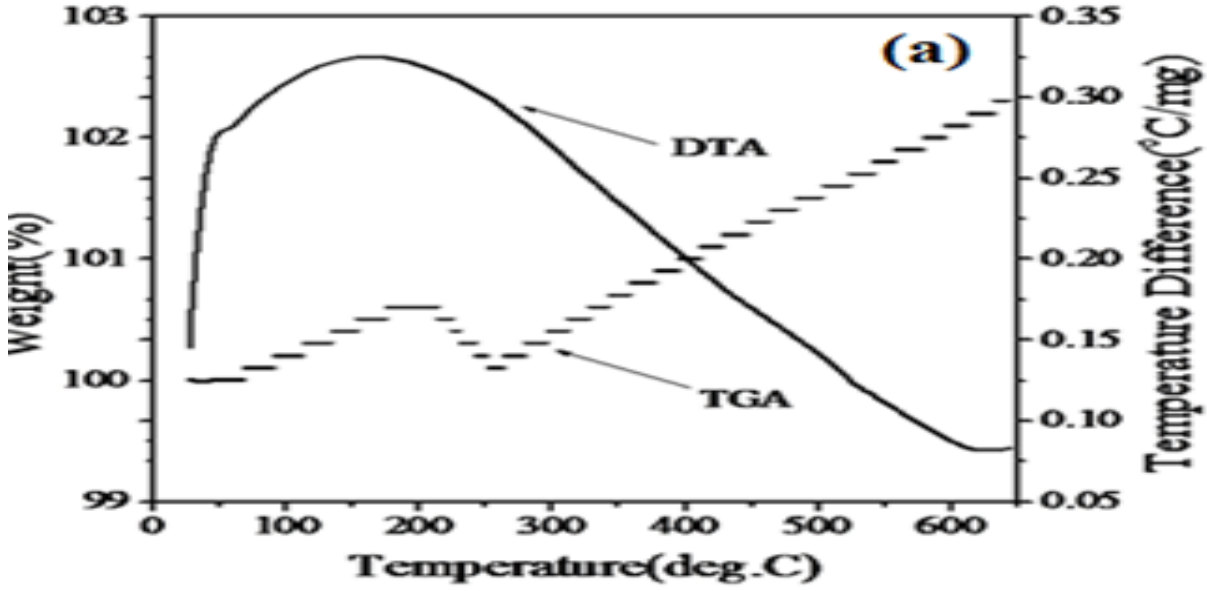


Figure-3.2.3

DTA and TGA of the Glass are shown in figure-3.2.3. The glass transition appears around 195^{o}C from both the curves. The glass transition temperature of BBCO is relatively lower compared to the SiO_2 based bioactive glasses [42//x41].

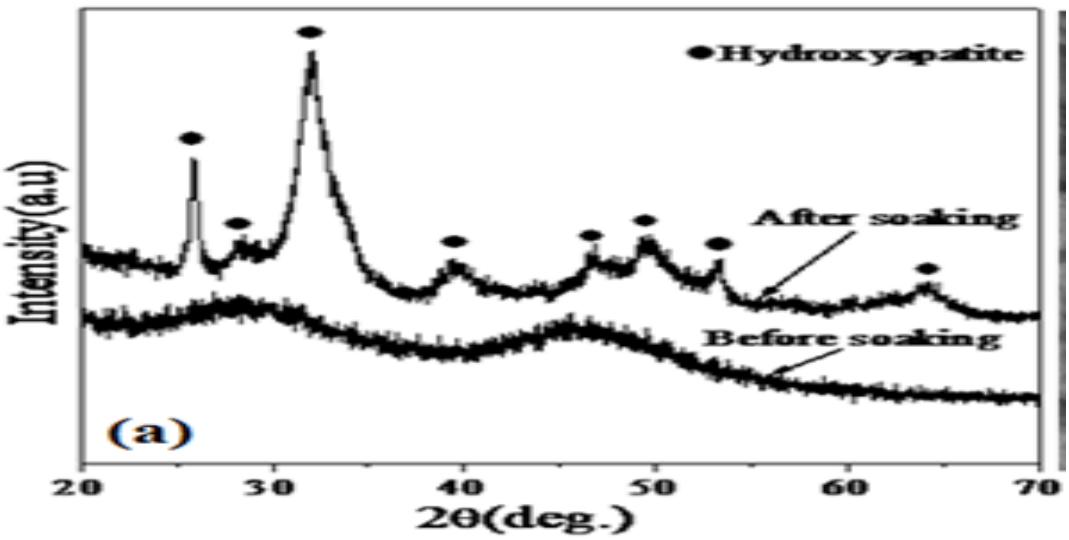


Figure-3.2.4

XRD of the as quenched glass is shown in Fig-3.2.4 i.e., the lower one humps like peaks showing before soaking indicates amorphous character of the glass. And so shows the corresponding XRD of the soaked glass surface for 15 days. Most of the XRD peaks (figure 3.2.3) were identified (JCPD, 9-0432) with the standard HAp phases.

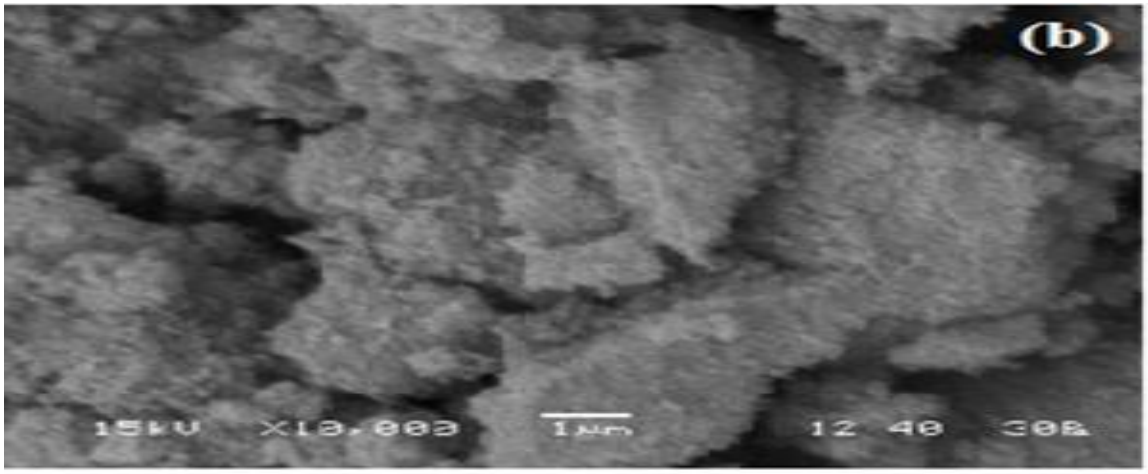


Figure- 3.2.5

Fig-3.2.5 shows the SEM micrograph of the same HAp powder. From this image it has been seen the powder like deposition over the glass surface.

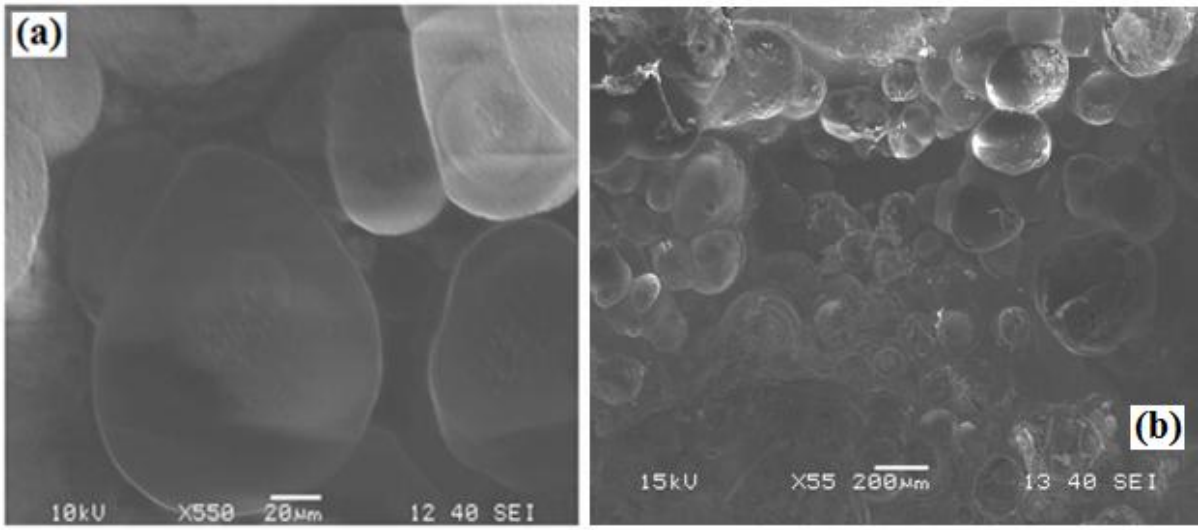


Figure-3.2.5

Figure-3.2.5FESEM micrographs of the HAp formed on the BBCO glass wire (a) and plate (b) surfaces showing nano-rods and bubble like structures.
FE-SEM micrographs of the glass surface after soaking for 30days showing rod and bubble like surface morphology of the soaked glass fibre and plate, respectively. Similar HAp bubbles were formed on the porous borate glass powder scaffold [45//43].

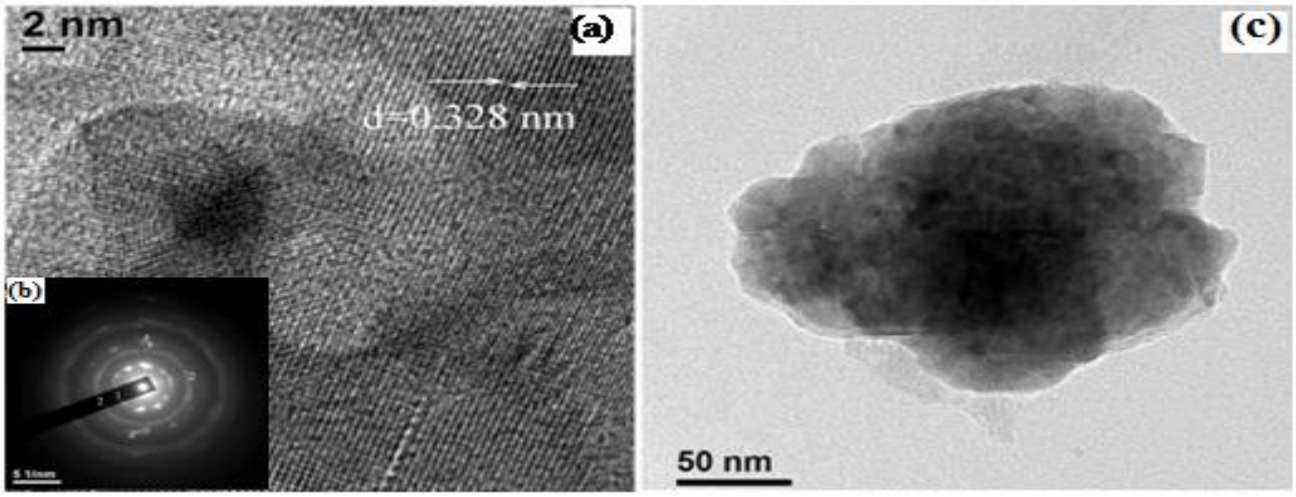


Figure-3.2.6

Figure-3.2.6 (a)HRTEM and (b)SAED images representing the morphology and crystal pattern of the nano HAp with diffraction pattern.(C)- TEM of single HAp grain.

This figure-3.2.6 shows the TEM of a HAp grain and the corresponding HRTEM and SAED micrographs indicating the crystalline nature of the HAp nanoparticles are shown in figures-3.2.6 (b and c)

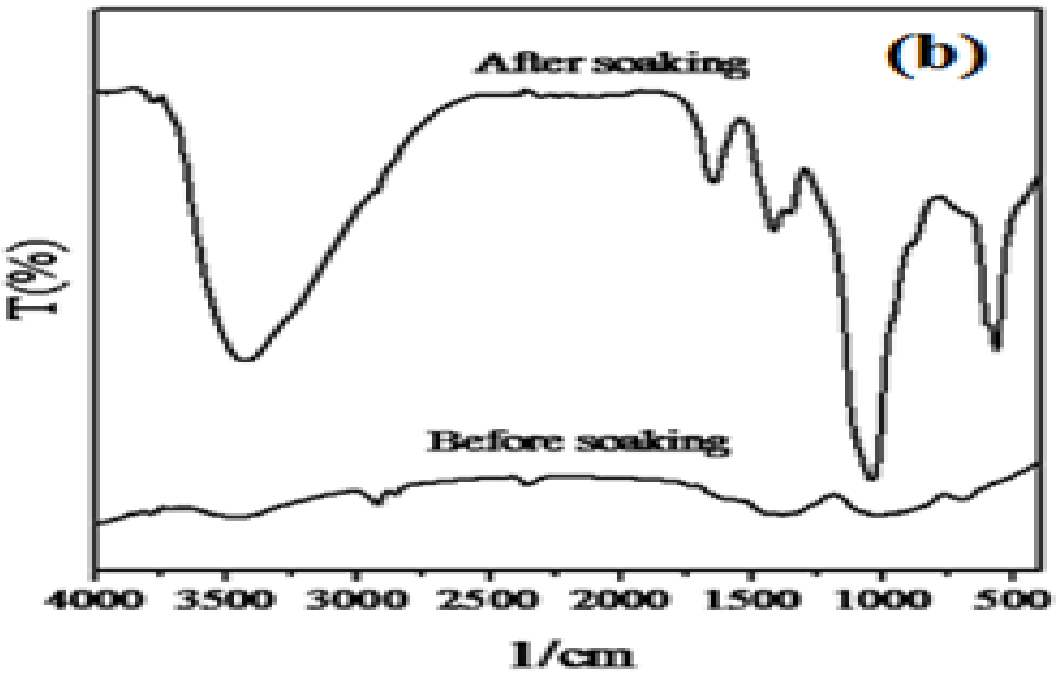


Figure-3.2.7

FTIR spectra of the quenched glass and HAp nanopowder are shown in figure-3.2.7. The FTIR spectrum of the quenched BBCO glass shown in figure-3.2.7a i.e, before soaking.. Figure-3.2.7-b shows the FTIR spectrum of the soaked glass surface. The band at 3429.2 cm^{-1} belongs to the vibration of hydroxyl group. The band at 1037.63 cm^{-1} is the characteristic band of phosphate stretching vibration, while the bands at 864 and 565 cm^{-1} are due to phosphate bending vibrations. The bonds at 601.75 and 565cm are due to the phosphate binding vibration. This FTIR spectral behaviour agrees well with that of the reported nano HAp sample [41-42//x39-4].

CHAPTER-5

5.1 CONCLUSION:

In conclusion, nano HAp layers formed on the BBCO glass surface soaked in K_2HPO_4 solution due to reasons explained earlier [[xx] conversion of glass into HAp. Depending on the thickness of the plate, HAp conversion rate can be varied. The speciality of the BBCO glass is that it can be made in form of fine fibres or powder (by by grinding).

5.2 SCOPE AND FUTURE DIRECTION:

- Better bioactive material can be prepared by adding silver and gold nano-particle and this could be area research in the future
- It was also found that graphene-oxide is showing bioactivity hence graphene oxide glass composite can be prepared and could be the research area for future study.

REFERENCES

[1]- [1] Will J, Gerhardt LC and Boccaccini AR "Bioactive glass based scaffolds for bone tissue engineering", 2012.

[2]- Corno M, Dissertion for bachelor degree, turin Italy, 2004.

[3]- Willy publication, Callister " Material science and engineering",2008.

[4]-"Cooling to glass transition temperature''[NATURE 410:259-267 (8 March 2001)]

[5]- Marta Giulia Cerruti "Characterization of bioactive glasses. Effect of the immersion in solutions that stimulates body fluids." ,University of Turin Department of chemistry IFM Ph.D. thesis, Phd in chemical science 2001/2004

[6]- www. Preservearticle.com *201012291928/glass-types.html*

[7]- Andersson OH, Karlsson KH, Kangasniemi K. Calcium phosphate formation at the surface of bioactive glasses *in vivo*. J Non-Cryst Solids. 1990;119:290–6.

[8]- Hench LL, Wilson J. Surface-active biomaterials. Science. 1984;226:630–6.

[9]- Wallace KE, Hill RG, Pembroke JT, Brown CJ, Hatton PV. Influence of sodium oxide content on bioactive glass properties. J Mater Sci Mater Med. 1999;10:697–701.

[10]- Ducheyne P, Qui Q. Bioactive ceramics: The effect of surface reactivity on bone formation and bone cell function. Biomaterials. 1999;20:2287–303.

[11]- Developments in Biocompatible Glass Compositions. Medical Device and Diagnostic Industry Magazine MDDI Article Index. An MD and DI, Column Special Section, 1999 Mar

[12]- Anjuvan Singh "Hydroxyapatite, a biomaterial: Its chemical synthesis, characterizationand study of biocompatibility prepared from shell of garden snail, Helix aspersa" Bull. Mater. Sci., Vol. 35, No. 6, November 2012, pp. 1031–1038.c Indian Academy of Sciences.

[13]- L. L. Hench, J. Mater. Sci. Mater . Med. 17(2006), 967-78.

[14]-L. L. Hench, J. Mater. Sci. Mater . Med. 17(2006), 967-78.

[15]-O.H.Anderson, I. Kangasniemi;J. Biomed. Mat. Res. 25;1991, 1019-30.

[16]- L.L.Hench, J.M. Polak, Science, 295 (2002) 1014-1017.

[17]- T. Kokubo, Biomaterials, 12 (1991) 155-163.

[18] W. Huang, M. Rahman, D.E. Dey, X. Liu, J Am. Cerm. Soc. 91(2008) 2105-2111.

[19] L.L. Hench, R.J. Splinter, W.C.Allen, T.K. Greenlee, J. Biomed. Mater. Res. 2(1971) 11.
[20] N. Ozawa, T.J. Yao, Biomed. Mater. Res. 62 (2002) 579−586.
[21] J. Shi, N.M. Alves, J.F.Mano, Adv. Funct. Mater.17 (2007) 3312−3318.
[22] A. Pedone, T.Charpentier; G. Malavasi; M.C. Menziani, (2010)Chem. Mater.22(2010) (19): 5644–5652.
[23] M.W.G. Lockyer, D.Holland; R.Dupree ,188(1995).J. Non-Crys. Sol.188(1995) 207–219.
[24] E,E Meling, E M A Hamzawy, A M El-Kady, A Salama, A El-Rashedi, J. Mat. Sci. Mat. Med. (2012) 23; 2069-2080]
[25] L.L. Hench, R.J. Splinter, W.C.Allen, T.K. Greenlee, J. Biomed. Mater. Res. 2(1971) 11.
[26] J. Liu, K.Li, H. Wang, M. Zhu, H. Yan, Chem. Phys. Letts. 396(2004) 429-432.
[27] J. R. Jones, L. M. Ehrenfried, L. L. Hench, Biomaterials, 27 (2006) 964–973.
[28] X. Liu, H. Pan, H .Fu, Q. Fu, M. N. Rahaman, W. Huang. Biomed Mater. 5 (2010) 015005-10.
[29] M. N. Rahaman, D. E. Day, B. S. Bal, Q. Fu, S. B. Jung,L. F. Bonewald, A. P. Tomsia, Acta Biomater 7(2011) 2355–2373.
[30] W. Liang, M. N. Rahaman, D. E. Day, N. W. Marion,G. C. Riley, J. J. Mao,.354(2008)1690–1696]
[31] W. Huang, D. E. Day, M.N. Rahaman, J. Am. Ceram. Soc., 90(2007)838–44.
[32] M. Hamadouche, A. Meunier, D.C. Greenspan , C. Planchat , J.P. Zhong , G.P.L.Torre, L. Sedel , J. Biomed. Res. 54 (2001)560-6.
[33] M. Manadouch, A. Meunier, D.C. Greenshpan, C. Blanchat, J.P. Zhong, G.P.L. Torre, J. Biomed. Mat. Res. 45(2001) 560.
[34] J. Ning, A. Yao, D. Wang, W. Huang, H. Fu, X. Liu, X. Jiang, X. Zhang, Mater. Lett. 61 (2007) 5223-5226]
[35] Q. Chen, J.A. Roether, A.R Boccaccini, in Topics in Tissue Engineering: Ed. By N R. Ashammaki, R.Reis , R.Chiellini : (2008), Vol4 (Chapter-6) pp 1-27.
[36]- E. A. Saleh, M. Whyte and P. G. Genever, Eur. Cells. Mater. 22 (2011) 242-257.
[37]- Yang leng " Material characterization: introduction to microscopic and spectroscopic methods", mar 2009.
[38] S.K.Swain, S.V.Dorozhkin, D. Sarkar, Mater. Sci. Eng. C 32 (2012) 1237-1240.
[39] H. Sakata, K Sega, and B K Chaudhuri:Phys. Rev. B60(1991)3230-3236
[40x] D. Bhadra, Md. G. Masud, S. Sarkar, J. Sannigrahi, S. K. De and B. K. Chaudhuri, J. Appl. Polm. Sci. Part b: Polym Phys, 50, 572 (2012)

[41] E. E Meling, E M A Hamzawy, A M El-Kady, A Salama, A El-Rashedi, J. Mat. Sci. Mat. Med. (2012) 23; 2069-2080]

[42] K.P.Santosh , A. Balakrishnan, T.N. Kim , S.J.Cho , Bull. Mater. Sci.32(2009)465-70.

[43] S.K.Swain, S.V.Dorozhkin, D. Sarkar, Mater. Sci. Eng. C 32 (2012) 1237-1240.

[44] W Liang, M N Rahaman, D E dey, N W Marion, G C Riley, J M Mao,J. Non cryst Solid. 354(2008)1690-96]

[45] J.P. Zhong, G.P. La Torre et al , Bioceramics 7(1994)28-33.].