CHAPTER-1

Introduction and literature review

1.1 Introduction

Bioactive glasses are of great importance for biomedical applications due to their ability to chemically bond to bone and stimulate new bone growth. Under physiological conditions, bioactive glasses dissolve in a controlled manner releasing calcium and phosphorous into solution. Ca and P form an amorphous calcium phosphate layer (ACP) which then crystallizes to form hydroxyapatite (HA) / Hydroxyl carbonate apatite (HCA): the naturally occurring mineral present in both teeth and bones. Recent development in tissue engineering in the field of orthopedic implants look forward to developing the regeneration capabilities of the host tissues using advanced designing methods for the preparation of implants to match the structure of the host tissues to accelerate the rejuvenation of the damaged tissues.

This requires the preparation of implants which are similar to that of the host tissue structure both in terms of structure as well as mechanical and biological properties. In reference to the above requirements, bioactive glasses have shown promising prospects. Due to their good bioactivity confirming both osteoconduction and osteoproduction, have become the material of major interest. Since the revolutionizing paper by Hench on bioactive glass in the year 1970, the composition has been optimized several times for better results than the last one. Glasses are composed of glass network former, modifier, and intermediate oxides. They present an amorphous character reflecting a structural disorder. Metal elements introduced in the glass matrix may involve specific changes in thermal behavior. In the glassy form, if there are not enough alkaline ions, the intermediate oxides will be a network modifier by creating two oxygen bridges. Conversely, if there are enough alkaline ions, the intermediate oxides will be a network former by creating two bridging oxygen. Studies have shown the impact of titanium on the thermal properties.

Biomaterials normally described as a combination of substances originating from natural, inorganic, or organic materials that are biocompatible in exactly or partially with the physiological fluid over a healing period. They entail complete or part of a living organism or biomedical devices which perform augmentation and replacements of any natural function [Boretos et al. 1984]. The biomaterial is a nonviable substance used in medical devices for the intention of the interaction with biological systems. Their usage within the physiological medium must need characteristic features such as efficiency and reliability. These distinguishing features have provided with a suitable combination of chemical, mechanical, physical, and biological properties [Williams et al. 1987]. Recently, biomaterials are widely used in various medical devices and systems; drug delivery systems; tissue engineering; screws, plates, wires, and pins for bone treatments; total artificial joint implants; partial or total hip replacement, skull repair or reconstruction; dental and maxillofacial applications [Binnaz et al. 2012]. In other words, a biomaterial is a non-toxic material that can be used to construct artificial organs, rehabilitation, and augmentation of medical devices or prostheses, and to replace biological tissues. Hench classified the application of biomaterials in tissue engineering into three-time frames [Hench 1998]:

The past: removal of tissues;

The present: replacement of tissues;

The future: regeneration of tissues;

In the past years, the aim of developing biomaterials was to create much strong and chemically inert biomaterial for the augmentation of mechanical strength of the bones or other physiological parts. The first skeletal repairing materials used were metals, which were considered purely as bio-inert. Since then, plenty of successful applications in orthopedics are carried out with metallic implants annually. However, no material implanted in living tissues is completely inert: all materials elicit a response

from the host tissue. Although inert metal implants can provide high strength and corrosion resistance, relative movement, called micromotion, can occur due to a lack of chemical or biological bonding at the interface [Cao et al. 1996]. Also, the inert metal implants have the risk of releasing some ions from the surface texture to cause immunological effects in the body. These led to a search for materials that can repair and regenerate tissues rather than replace them. These types of materials are called bioactive materials.

In a general sense, a bioactive material has been defined as a material that has been designed to induce specific biological activity. In a more narrow sense, a bioactive material has been defined as a material that undergoes specific surface reactions, when implanted into the body, leading to the formation of a hydroxyl - carbonate apatite (HCA) layer which is responsible for the formation of a firm bond with tissues [Kokubo 1991]. The ability of a bioactive material to form an HCA layer when immersed in body fluid is often taken as an indication of its bioactivity [Kokubo et al. 1990].

Bioactive glasses were invented by [Hench et al. 2006], which helped in interfacial bonding with the surrounding or the damaged tissue regarded as the second generation bio-materials. Since then, various kinds of bioactive materials have been developed over the last three decades. Among these, the main bioactive materials used clinically are:

silica-based bioactive glasses [Hench et al.1971],
hydroxyapatite (HA) [$Ca_{10}(PO_4)_6(OH)_2$] [Jarcho et al. 1977],
β - tricalcium phosphate (TCP) [$Ca_3(PO_4)_2$] [Rejda et al. 1977],
HA/TCP bi-phases ceramic and bioactive glass-ceramic,
A-W containing crystalline oxyfluoroapatite [$Ca_{10}(PO_4)_6(O,F)_2$] and
β-wollastonite [$CaO.SiO_2$] in $MgO\text{-}CaO\text{-}SiO_2$ glassy matrix [Kokubo et al. 1982]. Figure 1.1 shows the in vitro and in vivo performance of bioactive materials.

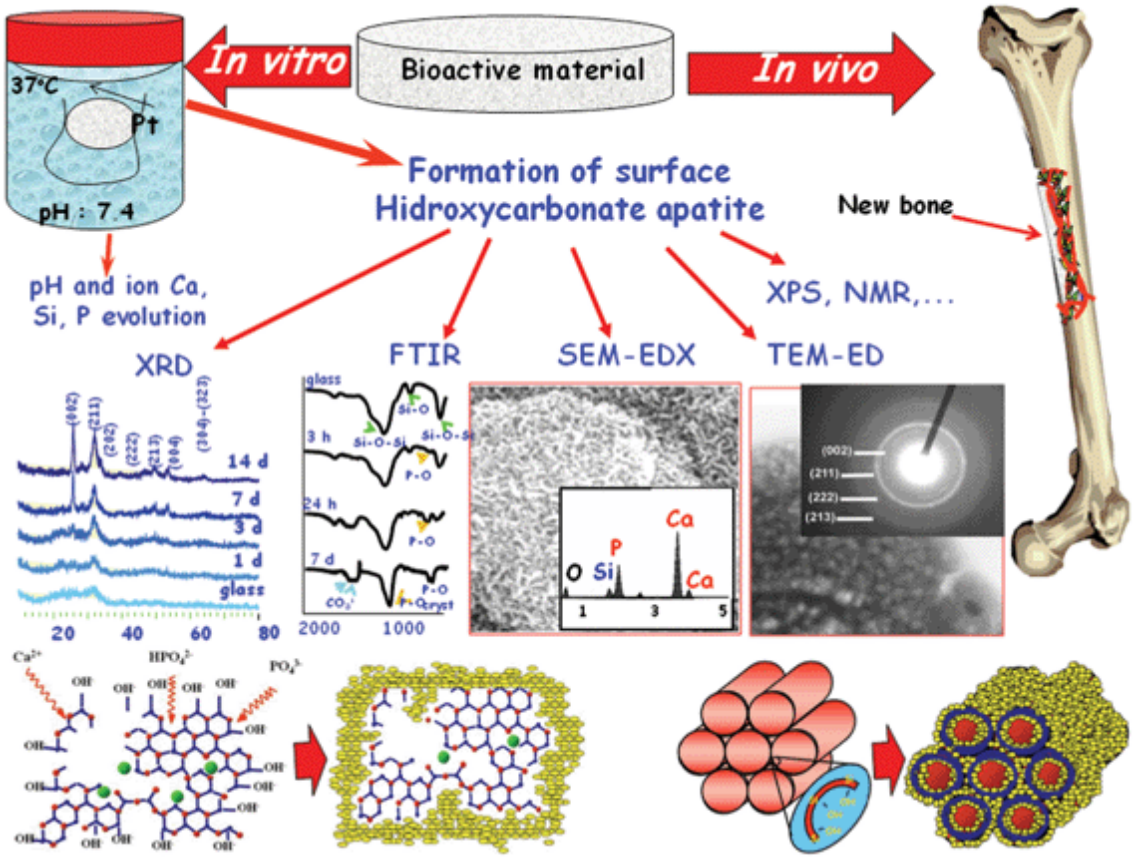

Figure 1.1- In vitro and In vivo performance [Antonio et al. 2013]

In 1991 Kokubo developed simulated body fluid (SBF), SBF has become the most widely used solution for in vitro investigation of material bioactivity by providing conditions very close to those found in in vivo, which is carried out in the living body. A fast, economical, and reliable bioactivity test of any material can thus be carried out in SBF solution.

By adjusting the oxide composition of glass, its properties, and rate of bonding to tissues can be controlled. These aspects make bioactive glasses different from other bioactive materials [Hench 1998]. The 45S5 bioactive glass and glass-ceramic have been used widely because of bonding capability with hard & soft tissues. One of the significant applications of bioactive glass and glass-ceramic is as an artificial bone graft. Therefore, it is a promising material in the field of biomedical application. It has inferior mechanical properties in comparison to cortical bone. Generally, the purpose of

biomaterials is to substitute for a damaged or diseased part of human body bones. In the 45S5 bioactive glass [$45SiO_2$-$24.5Na_2O$-$24.5CaO$-$6P_2O_5$ (wt %)]

S denotes the network former SiO_2 followed by a specific Ca/P molar ratio of 5:1 [Best et al.2008]. The key compositional features that are responsible for the bioactivity of 45S5 bioactive glass are its low SiO_2, high Na_2O, and CaO contents as well as high CaO/P_2O_5 ratio [Rahaman et al.2011 and Srivastava et al. 2012]. However, it suffers from a mechanical weakness, and low fracture toughness due to the amorphous nature of glass, and it may not be suitable for load-bearing applications [Shi et al. 2004]. The network former in the bioactive glass holds the three-dimensional non-periodic glass structure during the selective dissolution of cations (Na^+, Ca^{2+}, etc.,) by suppressing the detachment of some other ions [Hench et al. 1971]. The presence of SiO_2 also helps in the precipitation or surface reconstruction of the loose silica-rich layer and hence enhances the formation of hydroxylapatite layer [Hench et al. 1991, Paschall et al.1974 and Hench et al. 1997]. The interactions between the bone tissues and the bioactive implants, in particular, the interfacial reaction kinetics and the sequence of responses have been critically reviewed by earlier workers [Hench et al. 1991, Hench et al. 1998 and Lim al el.2005].

The present investigation aims to improve our understanding in vitro bioactivity and physical-mechanical properties of CoO, TiO_2, and ZrO_2 substituted 1393 bioactive glasses. The motivation for the selection of Co, Ti, and Zr is due to the following observations:

Titanium has excellent biological property.

The major components of the oxygen-carrying part of blood cells and significantly improve the blood vessel formation.

Titanium has the highest strength to density ratio of any metallic property.

Titanium in bone marrow mesenchymal stem cells enhances cell migration.

Titanium has a paramagnetic and low electrical and thermal conductivity.

TiO_2 addition in the ternary system Na_2O-TiO_2-P_2O_5, (from 0 to 2.5 mole% TiO_2) resulted in a nonlinear increase of glass transition temperature and dilatation softening temperature.

The increase of TiO_2 content involves the increase of the contribution of the surface crystallization mechanism.

The increase of TiO_2 content, it has been proved that there is an improvement in the mechanical properties of bioactive glass.

Co has excellent biological property.

The major components of the oxygen-carrying part of blood cells and significantly improve the blood vessel formation.

Co also helps protect our cells from being damaged by certain chemicals.

Co, along with vitamin B12, is essential for keeping blood vessels and skin elastic and flexible.

[Hoppe et al. 2014] investigated that cobalt oxide-releasing from 1393 bioactive glass derivative scaffolds for bone tissue engineering applications as cobalt was known as angiogenesis agent.

The authors prepared a melt derived 1393 glass having wt% composition ($53SiO_2$-$6Na_2O$-$12K_2O$-5MgO-20CaO-$4P_2O_5$) substituted with CoO for CaO in the glass which was further used to produce a three-dimensional (3D) porous scaffolds by the foam replica technique. Structural properties of bioactive glasses by FTIR spectrometry and their thermal behavior as well as scaffold macrostructure, compressive strength, cellular bioactivity and Co-release in simulated body fluid (SBF) were investigated. The addition of CoO for CaO in 1393 bioactive glass was done from 1.0 to 5% by weight has shown to act in a

concentration-dependent manner as both network former as well as a modifier in bio-glass. The SBF investigation done by authors for all glass scaffolds containing 1 to 5% cobalt oxide had shown the formation Ca-P layer incorporated with cobalt on the surface of scaffold samples. The maximum concentration of Co^{2+} ions around 12 ppm released in SBF after 21 days of the reaction was found to be within the therapeutic range of divalent cobalt. So, considering the impact of surface chemistry on cell attachment proliferation, the resulting formation of Ca-P layer incorporated by Co^{2+} ions at the scaffolds-SBF interface would be very important for improving the understanding of mineralization behavior and cell response to bioactive scaffolds. The authors mentioned that these Co^{2+} releasing scaffolds could be used as hypoxia-mimicking novel biomaterials with a high degree of mechanical integrity, making them interesting candidates for bone tissue engineering applications [Hoppe et al. 2014].

Zr has excellent biological property.

The major components of the oxygen-carrying part of blood cells and significantly improve the blood vessel formation.

Zirconia was used as a bone substitute since it has excellent mechanical properties.

The ZrO_2 substituted is widely used as a substrate in hard tissue applications due to its excellent strength and fracture toughness [Hulbert, S.F et al. 1993]. It was discovered from the reaction product obtained after heating gems by the German chemist Martin Heinrich Klaproth in 1789 [Julian et al. 2009].

Several research articles have suggested that zirconia has good chemical and dimensional stability, mechanical strength, and toughness, and it is also

biologically inert [Miao et al. 2007].

Many studies have shown that the compressive strength of ZrO_2 is relatively higher than porous HA and ZA20 (20 wt.% Al2O3 added TZP) ceramics.

In vitro evaluation has also shown that ZrO_2 is not cytotoxic [Sarkar, R et al. 2010; Zhang, S et al. 2013, Ducheyne et al. 1980]

1.2 Biomaterials

Any natural or synthetic material that is implanted in the living body may be referred to a biomaterial. According to Williams "a biomaterial is a non - toxic material of natural or man-made origin, which is intended to interface with a biological system to treat, augment or replace any tissue, organ, or function of the body and that evokes a minimal biological response" [Ratner et al. 1996]. In simple words, a biomaterial is a non - toxic material that can be used to construct artificial organs, rehabilitation devices or prostheses, and to replace natural tissues. The mechanism of tissue attachment of an implant is directly related to the tissue response at the implant interface [Hench et al. 1991]. No material implanted in a living body is inert: all materials elicit a response from the host tissue. According to the different types of implant-tissue attachment, biomaterials are classified into four types, which are summarized in Table 1.1.

Table 1.1- Types of tissue attachment of biomaterials [Hench et al. 1996]

Type of implant	Type of attachment	Example
Nearly inert	Mechanical interlock (morphological fixation)	Metals, Alumina, Zirconia, Polyethylene
Porous	In growth of tissues into pores (biological fixation)	Hydroxyapatite , Hydroxyapatite coated porous metals
Bioactive	Interfacial bonding with tissues (bioactive fixation)	Bioactive glasses, Bioactive glass - ceramics, Hydroxyapatite
Resorbable	Replacement with tissues	Tricalcium phosphate, Polylactic acid

The tissue response to a biologically inactive, nearly inert implant, in the formation of a non-adherent fibrous capsule. This attachment is called “morphological fixation.” The thickness of the fibrous layer depends on many factors, such as the conditions of the implant, the conditions of the host tissue, the conditions of motion and fit at the interface and the mechanical load. A chemically stable material like alumina elicits a fragile capsule under an optimal mechanical fit. More chemically reactive metallic implants elicit thicker interfacial fibrous layer. Because the interface is not chemically or biologically bonded, relative movement can occur, called micromotion. This movement results in the progressive development of the non-adherent fibrous capsule and eventually leads to deterioration in the function of the implant or the host tissue at the interface or both.

Porous biomaterials provide interfacial fixation by ingrowths of tissue into pores on the surface or throughout the implant. This attachment is called “Biological Fixation.” It is capable of withstanding more complex stress than dense nearly inert implants which achieve only “morphological fixation.”

Resorbable implants are designed to degrade gradually with time and be replaced with natural host tissues. For example, resorbable sutures composed of poly (lactic acid)-poly (glycolic acid) are metabolized to carbon dioxide and water. Tricalcium phosphate ceramics degrade to calcium and phosphate salts. Because large quantities of materials must be handled by cells, the constituents of a resorbable implant must be metabolically acceptable. Another requirement for a resorbable implant is that the resorption rate must be matched to the repair rates of tissues.

Bioactive implants offer another approach to achieve interfacial attachment. When a bioactive material is implanted in the body, a series of biophysical and biochemical reactions occur at the implant-tissue interface. These reactions eventually result in a mechanically strong chemical interfacial bonding. This attachment is called “Bioactive Fixation.”

1.3 Bioactive materials

In a general sense, a bioactive material is one that elicits a specific biological response at the interface of the material which results in the formation of a bond between the tissues and the material [Hench et al. 1994]. In a more narrow sense, a bioactive material has been defined as a material that undergoes specific surface reactions, when implanted into the body, leading to the formation of a hydroxyl - carbonate apatite (HCA) like layer that is responsible for the formation of a firm bond with hard and soft tissues [Kokubo et al. 2006)].

The level of bioactivity of a specific bioactive material can be related to the time for more than 50% of the interface to be bonded. An index of bioactivity (I_b) introduced by Hench [Hench et al. 1993] as:

$$I_b = 100/t_{0.5bb}$$

Where $t_{0.5bb}$ is the time for more than 50% of the implant interface to be bonded to tissues

Bond strength and the time needed for bonding depend on the type of bioactive material and its bonding mechanism, as well as the thickness of the bonding zone. However, the critical character of bioactive material is its ability to undergo chemical/biological bonding in the interface. Based on the type of biochemical bonding at the interface, bioactive materials have been classified into two types:

Class A (osteoproductive materials) and

Class B (osteoconductive materials) [Hench et al. 1994].

Osteoproduction has been defined [Wilson et al. 1994] as “the whereby a bioactive surface is colonized by osteogenic stem cells free in the defect environment as a result of surgical intervention.” Class A bioactivity occurs when a material elicits both an intracellular and an extracellular response at its interface. However, the materials of

Class B, the osteoconductive materials, elicit only an extracellular response at their interface [Cao et al. 1996]. Bioactive material includes a wide range of materials such as: bioactive glasses, bioactive glass - ceramics, hydroxyapatite, bioactive composites, and bioactive coatings.

1.4 Simulated body fluid

In 1991, Kokubo proposed that the essential requirement for an artificial material to bond to living tissues is the formation of hydroxyl-carbonate apatite (HCA) on its surface when implanted in the living body, and that this in vivo HCA formation can be reproduced in a simulated body fluid (SBF) with ion concentrations nearly equal to those of human blood plasma. SBF developed by Kokubo differs in some ions compared to human blood plasma [Kokubo et al.2006]. Some other researchers have tried to correct this difference by preparing SBF with alternative compositions. [Kokubo et al. 2006] made a revised SBF (r-SBF) in which the concentrations of Cl- and HCO3- ions were adjusted to the levels in human blood plasma. However, calcium carbonate showed a strong tendency to precipitate from r-SBF. Takadama et al. [Kokubo et al. 2006] also proposed a modified SBF (n-SBF) in which only the Cl- ion concentration was increased. This n-SBF does not differ from the SBF by Kokubo instability and reproducibility.

1.5 Bioactive glasses

Most of the published works on bioactive glasses are concentrated on silica-based materials. Silica-based bioactive glasses have supplied successful solutions to different bone defects and soft tissue treatments during the last decades [Hench et al.1993]. The high biocompatibility and the positive biological effects of their reaction products (both leached or formed at the surface) after implantation have made silica-based bioactive

glasses one of the most interesting bioactive materials during the last 40 years [Arcos et al. 2010]. In contrast, the poor mechanical properties of these bioactive glasses have severely limited the range of clinical applications. These bioactive glasses in different forms are needed for various clinical applications. Some clinical applications of silica-based bioactive glasses are given bellow

Table 1.2- Clinical applications of silica - based bioactive glasses [Arcos et al. 2010]

Material form	Clinical application
Solid shapes	– Ossicle replacement in the middle ear
	– Cone shaped devices for jaw defects filling
	– Curved plates for restoring eye orbit floor
	– Soft tissue sealing for transdermal implants
Particulates	– Bone tissue replacement in periodontal diseases
	– Soft tissue augmentation in paralysis of vocal cords
Particulates and autologous bone	– Maxillofacial reconstructions
	– Spine
Particulates by injection	– Urological tissue augmentation

Silica-based glasses have an amorphous network structure based on the SiO_4^{4-} tetrahedron as the structural unit. The tetrahedra are linked to each other only at the oxygen ions at the corners. In crystalline silica, the tetrahedra are regularly arranged, as shown in Figure 1.2. However, a silica-based glass has a more open structure due to the existence of non-bridging oxygen ions. The open structure of the silica-based glass is formed by the disruption of the network structure by the presence of network modifiers, e.g., Na^+, K^+, Ca^{2+}, Figure 1.3. Strand [Starnd et al. 1992] suggested that the bioactivity of glass is based on the mean number of non - bridging oxygen ions in the silica tetrahedron. Instead of sharing a corner with another tetrahedron, the charge of the oxygen ion in the corner is balanced by a network modifier cation, e.g., Na^+, K^+, Ca^{2+}, in Figure 1.3. In the silica-based glass, each silicon is bonded to four oxygen atoms, and thus the number of non - bridging oxygen ions in the tetrahedron can take any value between 0-4. The number 0 represents a crystalline SiO_2 structure or quartz glass; the

number 4 means a dissolved SiO_4^{4-} ion. To be bioactive for a silica-based glass, the number of non-bridging oxygen ions per tetrahedron must be greater than 2.6 [Ylanen et al. 2000]. Traditional silica-based glasses consist of more than 65% SiO_2 by weight, less than 15% Na_2O by weight and about 10% CaO by weight. The composition of silica-based bioactive glasses is different from traditional silica-based glasses, though bioactive glasses resemble them. Bioactive glasses typically contain less than 60% SiO_2 by weight and large amounts of alkali and/or alkaline earth oxides.

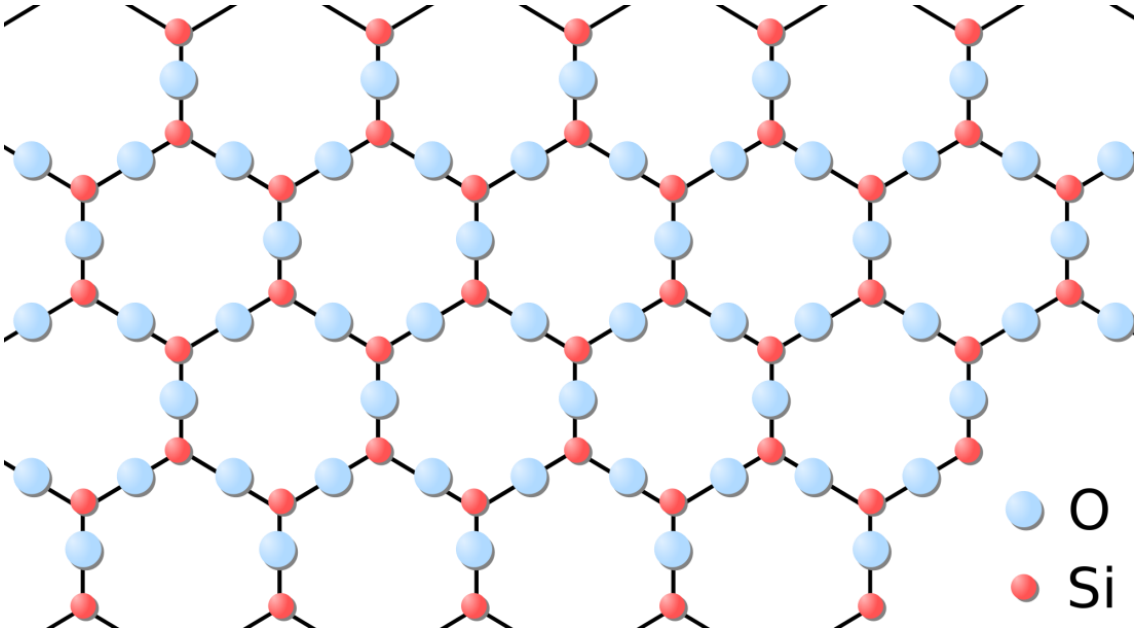

Figure 1.2- Two - dimensional presentation of the structure of crystalline SiO_2

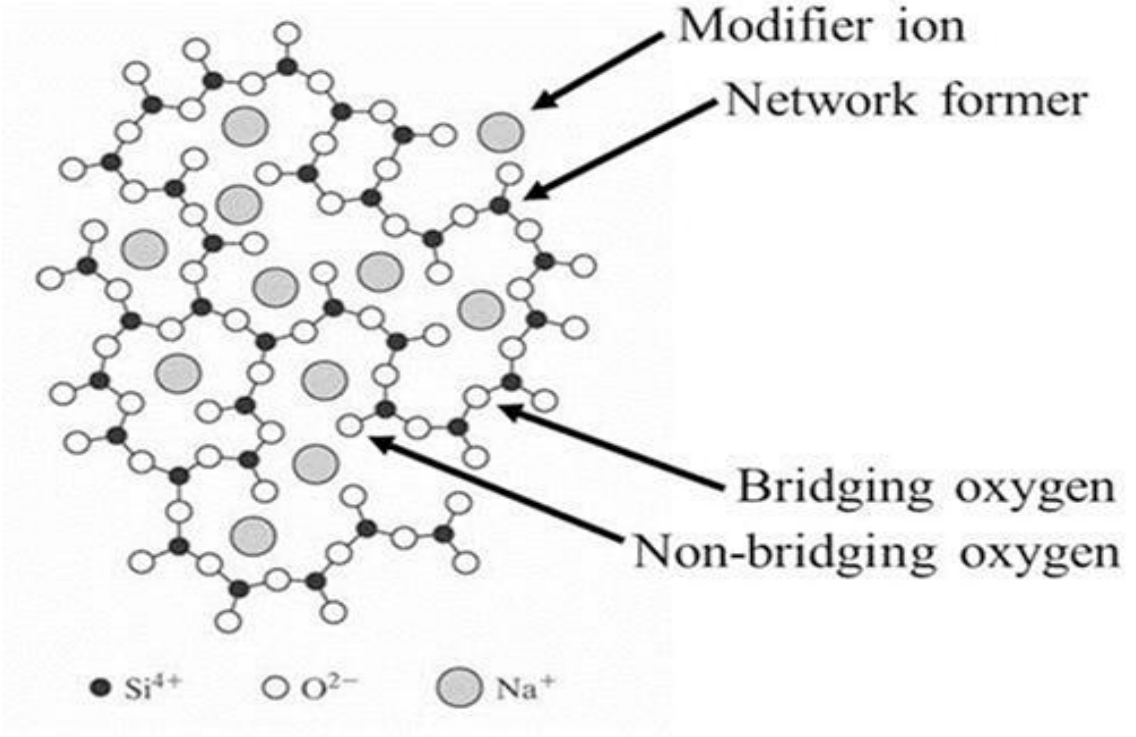

Figure 1.3- Two dimensional presentation of a random glass network composed of network modifiers and network formers (SiO_4^{4-} units) [Starnd et al. 1992]

According to Hench et al. [Hench et al. 1991], three key compositional features distinguish bioactive glasses from traditional SiO_2-Na_2O-CaO glasses:

1. Amount of SiO_2 is 40 - 60% by weight;
2. High Na_2O and high CaO content;
3. High CaO/P_2O_5 ratio.

If the content of SiO_2 > 60% by weight, the number of bridging oxygen ions is so large that it will dramatically reduce the network dissolution rate of the glass, thus leading to loss of bioactivity. However, the content of SiO_2 < 40% by weight, will give dissolved monomeric SiO_4^{4-} units. It is questionable whether obtaining a glass phase of this composition is possible [Ducheyne, 1987]. Thus, to show bioactivity, the SiO2 content of the glass should be between 40 and 60% by weight. The base components in most silica-based bioactive glasses are SiO_2, Na_2O, CaO, and P_2O_5. Previously, it was

assumed that P_2O_5 was required for a glass to be bioactive. However, phosphate in the glass was later found only to aid in the nucleation of the calcium phosphate phase on the surface. Phosphate is not a critical constituent because the surface can adsorb phosphate ions from solution [Hench et al. 1993].

The mechanisms of tissue bonding of silica-based bioactive glass have been attributed to the formation of a hydroxyl - carbonate apatite (HCA) layer on the glass surface when it is in contact with the body fluid. While some details of the chemical and structural changes are not clear, the HCA layer is generally believed to form as a result of a sequence of reactions on the surface of the bioactive glass implant, as described by Hench [Hench 1998b]:

1. Rapid exchange of cations such as Na^+ or Ca^{2+} with H^+ or H_3O^+ ions from the solution, leads to creation of silanol (Si - OH) groups on the glass surface:

$$Si - O - Na^+ + H^+ + OH^- \rightarrow Si - OH + Na^+ (solution) + OH^-$$

2. Loss of soluble silica in the form of silicic acid, $Si(OH)_4$, to the solution, resulting from breaking of Si - O - Si and the continued formation of Si - OH groups on the glass surface:

$$Si - O - Si + H_2O \rightarrow Si - OH + OH - Si$$

3. Condensation and polymerization of amorphous silica (SiO_2) - rich layer on the surface of the glass depleted in Na^+ and Ca^{2+}:

$$OH - Si - OH + OH - Si - OH \rightarrow OH - Si - O - Si - OH + H_2O$$

4. Migration of Ca^{2+} and PO_4^{3-} ions to the surface through the amorphous SiO_2 - rich layer, leading to the formation of an amorphous calcium phosphate ($CaO-P_2O_5$) layer on the surface of the amorphous SiO_2 - rich layer, followed by growth of the amorphous $CaO-P_2O_5$ by incorporation of soluble Ca^{2+} and PO_4^{3-} ions from solution.

5. Crystallization of amorphous $CaO\text{-}P_2O_5$ layer by incorporation of OH^-, CO_3^{2-} anions from the solution to form crystalline HCA layer. The HCA layer on the silica based bioactive glass surface is similar to biological apatite having the chemical formula given below [Hench et al.1993]:

$[(Ca, M)_{10} (PO_4, CO_3, Y)_6 (OH, F, Cl)_2]$

Where M = Na^+, K^+, Mg^{2+}, Sr^{2+}, Pb^{2+}, Ba^{2+} etc. and Y = HPO_4^{2-}, SO_4^{2-}, BO_3^{2-} etc.

With the initial formation of an HCA layer a sequence of events that appear to be associated with the formation of a bond with tissues are [Hench 1998b]:

1. Adsorption of biological moieties in the amorphous SiO_2 - rich and HCA layer.
2. Action of macrophages.
3. Attachments of stem cells.
4. Differentiation of stem cells.
5. Generation of matrix.
6. Mineralization of matrix.

The biocompatibility of silica-based bioactive glass has long been established [Wilson et al. 1994]. After implantation, silica-based bioactive glass undergoes degradation, releasing alkali ions such as Na^+ and Ca^{2+}. Si, presumably in the form of $Si(OH)_4$, is also released during the degradation by dissolution mechanisms. The release of Si from silica-based bioactive glass implanted in the living body has been studied to determine the pathway of released Si [Lai et al. 2002]. By measuring the Si released in urine and blood samples for up to 7 months post-implantation, and using chemical and histopathological analyses of bone and several tissues, it was found that the Si resulting from the 45S5 bioactive glass degradation was harmlessly excreted in soluble form through the urine. Formed amorphous SiO_2 - rich layer eaten by phagocytes and excreted out.

The first bioactive glass developed by Hench et al. [Hench et al. 1993] and named 45S5 bioactive glass [composition (weight %) $45SiO_2$-$24.5Na_2O$-$24.5CaO$-$6P_2O_5$], which exhibits a high bioactivity and can join readily even to soft tissues, is a silica-based bioactive glass. A silica-based bioactive glass designated 1393 [composition (weight %) $53SiO_2$-$6Na_2O$-$12K_2O$-$20CaO$-$5MgO$-$4P_2O_5$] is based on the 45S5 bioactive glass composition, but it has a comparatively higher SiO_2 content and additional network modifiers, such as K_2O and MgO, when compared to 45S5 bioactive glass, is also used clinically [Rahaman et al. 2011]. However, 1393 bioactive glass degrades and converts to an HCA material more slowly than 45S5 bioactive glass. Other invented silica-based bioactive glasses are:

45S5.4F[Composition (weight %) $45SiO_2$-$24.5Na_2O$-$14.7CaO$-$9.8CaF_2$-$6P_2O_5$],

52S4.6 bioactive glass [Composition (weight %) $52SiO_2$-$21Na_2O$-$21CaO$-$6P_2O_5$], 55S4.3 bioactive glass [Composition (weight %) $55SiO_2$-$19.5Na_2O$- $19.5CaO$-$6P_2O_5$], 55S4.3 bioactive glass [Composition (weight %) $55SiO_2$-$19.5Na_2O$-$19.5CaO$-$6\ P_2O_5$], 6P53S bioactive glass [composition (weight %) $52.7SiO_2$-$10.3Na_2O$-$2.8K_2O$-$18.0CaO$-$10.2\ MgO$-$6P_2O_5$] and 58S bioactive glass [Composition (weight %) $58.2SiO_2$-$32.6CaO$- $9.2P_2O_5$], [Hench et al. 1993]. More recent works have shown that certain borate-based glasses such as

[composition (weight %) $53B_2O_3$-$10.3Na_2O$-$2.8K_2O$-$18CaO$-$10.2\ MgO$ - $6\ P_2O_5$] and [Composition (weight %) $56.6B_2O_3$-$5.5Na_2O$-$11.1\ K_2O$-$18.5CaO$-$4.6\ MgO$-$3.7P_2O_5$] are also bioactive [Rahaman et al., 2011]. Because of their lower chemical durability, borate-based bioactive glasses degrade faster and convert more completely to an HCA like material, when compared to silica-based bioactive glasses. Borate based bioactive glasses have been shown to support cell proliferation and differentiation in vitro as well as tissue infiltration in vivo. Borate based bioactive glasses have also been shown to

serve as a substrate for drug release in the treatment of bone infection. A concern associated with borate-based bioactive glass is the toxicity of boron released into the solution as borate ions $(BO_3)^{3-}$. Recent work has shown the ability to control the degradation rate of silica-based bioactive glasses by manipulating its composition. For example, by partially replacing the SiO_2 in silica-based bioactive glasses with B_2O_3 (yielding a borosilicate bioactive glass), such as:

45B15S5 bioactive glass [Composition (wt %) $30SiO_2$-$15B_2O_3$-$24.5Na_2O$-24.5CaO-6 P_2O_5] and 1393B1 [Composition (wt %) $34.4SiO_2$-$19.9B_2O_3$-$5.8Na_2O$-$11.7K_2O$-19.5CaO-4.9MgO-$3.8P_2O_5$], the degradation rate can be varied over a wide range. The ease of manufacture and the ability to control the degradation rate of silica-based bioactive glasses make them particularly useful for promoting the regeneration of tissue. By controlling the glass composition, it should be possible to match the degradation rate of silica-based bioactive glass with the tissue regeneration rate. Some phosphate-based glasses such as: [composition (weight %) $9.3Na_2O$-19.7CaO-$71P_2O_5$] are also bioactive. As their constituent ions are present in the organic mineral phase of bone, these glasses have a chemical affinity with bone. The solubility of these glasses can be controlled by modifying their composition; therefore, these glasses may have additional clinical potential as resorbable materials. Depending on the manufacturing process, bioactive glass can be divided mainly into two groups: sol-gel bioactive glasses and melt - derived bioactive glasses. Sol-gel bioactive glasses are made by a chemically based process at much lower temperatures than the traditional processing methods. Sol-gel bioactive glasses have been investigated by many research groups [Balamurugan et al. 2007; Li et al. 2005; Liu et al. 2004; Xia et al. 2006]. Li and co-workers in 1991 have shown that sol-gel bioactive glasses in the system of SiO_2-Na_2O-CaO are bioactive even up to 85 mol % SiO_2. The wide range of bioactive oxide compositions makes it possible to tailor

the reactivity of the glasses to various applications. Also, sol-gel processing offers the potential advantages of ease of powder production, high purity of the material, and better control of bioactivity through changes in processing parameters [Li et al. 1991]. Compared with the sol-gel process, melting requires much higher working temperatures. However, melting is a simple and low-cost technique and is much less time consuming than sol-gel processing. For the production of a large amount of bioactive glasses, the melting process is very suitable and reliable. Because of these benefits, melting is a dominant process for producing bioactive glasses.

1.6 Hydroxyapatite

Hydroxyapatite is a crystalline form of calcium phosphate similar to the mineral present in bone. It is a compound with a definite crystallographic structure and of a definite composition, $Ca_{10}(PO_4)_6(OH)_2$. The mineral component in the living bone is also hydroxyapatite, the so-called biological apatite. The amount of biological apatite in bone is approximately 70% by weight. It was believed that hydroxyapatite used for a bone replacement would be entirely compatible with the body. When exposed to body fluids, hydroxyapatite will bond to the bone by forming indistinguishable unions. The bonding starts by formation of hydroxyl - carbonate apatite (HCA) crystals on the bone, thus promoting the adhesion of matrix - producing cells and organic molecules as a result of surface chemistry and surface charges [Hench et al. 1993]. Biological apatite, which comprises the mineral phase of human bone, is usually referred to as hydroxyapatite. Biological apatite differs from pure hydroxyapatite, and it is more appropriate to refer to as carbonate apatite [Hench et al. 1993]. Biological apatite contains ions such as Na^+, K^+, Mg^{2+}, F^- and Cl^- in solid solution. Some of the PO_4^{3-} may also be replaced by CO_3^{2-}. Thus, the ideal Ca/P molar ratio of pure hydroxyapatite (1.67) differs slightly from that of biological apatite (1.72-1.80) [Ylanen 2000]. The

possibility of improving apatite by changing its composition has inspired researchers to deliberately substitute ions in order to modify its properties and behavior [Hayakawa et al. 2008; Osaka et al. 2007]. The main goal in these studies has been to enhance bone bonding between the implant and tissue.

Hydroxyapatite can be prepared in either dense or macroporous forms. The surface chemistry of porous sintered and cemented hydroxyapatite is the same as that of the dense forms [Hench et al. 1993]. However, tissue response to porous hydroxyapatite implants is inherently different from its response to dense hydroxyapatite because of the opportunity for in growth. Thus, porous hydroxyapatite has now replaced dense hydroxyapatite form. When porous hydroxyapatite has been placed into bone defects, bone growth into the pores has ranged from 18% to 74% [Holmes et al. 1988; Martin et al. 1993]. The entire porous space of the implant is probably never completely filled with bone [Hulshoff et al. 1997; Rosen et al. 1990]. Porous hydroxyapatite is osteoconductive, biocompatible, and practically inert; it resorbs with time, but the degradation rate is slow [Ikeda et al. 1999]. Due to the brittle nature of porous hydroxyapatite, it can be used only in non - loading sites. In general, synthetic hydroxyapatite is widely used in dental, craniofacial and orthopedic surgery, mainly as granules, and as a bioactive coating on load-bearing implants, etc.

1.7 Bioactive composites

Although bioactive materials can form a strong biochemical bond with the bone and soft tissues, the mechanical properties of the materials themselves are usually unsuited for load-bearing applications. One approach to solve this problem is to combine them with a fracture tough material to produce a composite.[Cao et al. 1996] divided bioactive composites into two groups based on the goal of the implant. The first group consists of the compositions in which a fracture-resistant phase (metal fibers or

tough ceramic particles) is used to reinforce the bioactive material [Cao et al. 1996]. In other words, the matrix of the composite is a bioactive material, and the reinforcing phase is a tough material (matrix/reinforce = bioactive material/tough material). In 1993, Ducheyne et al. introduced the first bioceramic composites of metal fibers and glasses, i.e., stainless steel fiber/bioactive glass and titanium fiber/bioactive glass composites. These discontinuous metal fiber/ceramic composites were shown to maintain the bioactivity of the ceramics, but to have enhanced fracture resistance and strength compared to the ceramic alone. One year later, the metal fiber-reinforced bioactive glass composites were patented by Ducheyne [Hench et al. 1993]. Other types of composites also have been developed. For example, Boccaccini et al. [Boccaccini et al. 1997] described a processing route for the fabrication of metallic fiber mat reinforced bioactive glass matrix composites. However, the disadvantage of these composites is that they have elastic moduli greater than bone and thus give rise to stress shielding of bone. The second group of composites uses powders, particles or fibers of bioactive materials to reinforce an elastically compliant and biocompatible polymer matrix, for example, poly(D,L Lactice)/45S5 bioactive glass composite [Helen et al. 2006, Roether 2002], Poly(ether ether ketone)/hydroxyapatite composite and poly(DL - Lactide - coglycolide)/ bioactive glass composite foams [Orava et al. 2007]. Polymer - biomaterial composites solve the problem of stress shielding of bone. Bonfield demonstrated that an increase in the volume fraction of particulate hydroxyapatite from 0 to 0.5 (50% volume) produced an increase in the Young's Modulus of the hydroxyapatite reinforced polyethylene composite, thus approaching the lower range of values associated with the bone itself. Rich et al. studied in vitro bioactivity of poly (ε-caprolactone-co-DL-Lactide) reinforced by different amounts (40, 60 and 70 % by weight) of the bioactive glass S53P4 [Rich et al. 2002]. They found that the in vitro

bioactivity was dependent on the weight fraction and granule size range of the bioactive glass used. The in vivo studies of the glass-fiber-reinforced composite has been done by [Tuusa et al. 2005]. They also developed glass-fiber-reinforced composite with bioactive glass granule coating, and the in vivo tests showed that the coated composite implant provided an alternative for bone defect reconstruction, especially in head and neck area [Tuusa et al. 2007]. The bioactive part of the composite can also be a mixture of two biomaterials [Juhasz et al. 2004]. For applications without load-bearing requirements, composites derived from two bioactive materials have also been developed. Bioactive glass-reinforced hydroxyapatite composites have higher fracture resistance and greater bone-bonding ability than that of commercial hydroxyapatite. Miao et al. [Miao et al. 2007a] recently developed a porous calcium phosphate ceramic modified with poly (lactic-co-glycolic acid) bioactive glass. This composite showed enhanced strength due to infiltration of the PLGA (poly (lactic-co-glycolic acid)) into the inner pores of the porous ceramic. An additional coating achieved bioactivity with the bioactive glass PLGA.

1.8 Bioactive coatings

To resolve problems of the mechanical limitations of bioactive materials in load-bearing applications, it is to apply the material as a coating on a mechanically tough substrate. Bioactive coatings can modify the surface of implants and create an entirely new surface, thus giving the bioactive implant properties which are quite different from those of the uncoated implant. The bioactive coating materials successfully combine the bioactivity of bioactive materials and the good mechanical properties of tough materials. The bone-bonding capacity of these coatings may help to provide cementless fixation of orthopedic prostheses, especially for short term stabilization of the implants [Cao et al. 1996]. But in long term implantation, the bioactive coated materials suffer from a lack

of stability of the coating/implant interface. Because of its similarity to the inorganic component of bone and tooth, hydroxyapatite was one of the first materials considered for coating metallic implants [Greenspan et al. 1999]. A group in Japan developed a carbonate hydroxyapatite coating to titanium, for use in bone bonding implants [Salinas et al. 2000; Yan et al. 1997]. The coating significantly increased the bone-bonding strength by providing a bioactive surface. Similar research was carried out by [Kumar et al. 2002] by producing coating materials with different ratios of TiO_2/hydroxyapatite. Hydroxyapatite a coating, on the other hand, is clinically important implants such as porous zirconia have also been studied [Miao et al. 2007b]. Osaka et al. [Osaka et al. 2007] developed a bioactive composite coating consisting of one layer of titania and one layer of apatite on a titanium substrate. Thermal spraying, in particular, plasma spraying, is the most common method for applying hydroxyapatite coatings. Other techniques have also been investigated for commercial applications, including electrophoretic deposition processes, hot isostatic pressing, ion beam sputtering, radio frequency sputtering, and thermal spray techniques other than plasma spraying, such as the high velocity oxy-fuel technique [Arcos et al. 2002; Arcos et al. 2003; Ebisawa et al. 1997; Jiang et al. 2009].