**Head Start:**

**Graduate Level Resources**

**in Materials Engineering**

**Issue 4: Calcium Phosphates for Medical Applications**

**Sharon Kehoe**

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**Head Start: Graduate Level Resources in Materials Engineering**

This serial publication includes technology and science topics relevant to research in materials engineering. It is intended to supplement discipline specific undergraduate education and also provide detailed information on specific processing systems.

**Forward**

Issue 4: Calcium Phosphates for Medical Applications, September 2008.

Graduate level research has traditionally involved an initial period of intensive reading. Obvious resources include peer reviewed papers, conference papers and theses. However, for most students considerable time is also spent on technical descriptions of equipment and procedures, and on reading textbooks on relevant topics not covered by their own undergraduate training. The latter can be particularly pertinent to engineers working on a cross discipline project extensively involving materials chemistry or biological systems. While there is no doubt as to the value of a broad understanding of the context of a project, nor the need to understand relevant equipment, chemical processes and biological systems, this information is not generally at research level.

Nonetheless it can be a very time consuming exercise to come ‘up to speed’. Add to this the wealth of research being made accessible through electronic databases, and the rapidly growing volume of research being published, and the initial phase of reading oneself into a project can be very daunting.

Having observed many graduate students of the Materials Processing Research Centre at Dublin City University struggle through this process, I noted that we were not building effectively on this type of understanding gained by each student. There has also been confusion as to what is appropriately included in theses, with examiners taking different views on the amount of content to be dedicated to explaining concepts, terminology and systems from the complementary discipline.

From these observations came the idea of a series of publications dedicated to giving graduate students a ‘head start’. These are written by graduate students, largely based on their reading of text books in science and engineering disciplines relevant to their project. Where it is appropriate, students have collaborated on a theme. It is intended that the texts consolidate existing MPRC background knowledge on a topic, thus providing a ‘fast track’ for new researchers to start on their own work on a related project. It is intended they be an open ended serial, with new students and all MPRC members being welcome to submit new titles.

Lisa Looney

Series Editor

Director, Materials Processing Research Centre.

Dublin City University

Ireland

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Elsevier & C.S. Elliot (Figure 3-1: Solubility isotherms of CaP phases in the system Ca(OH)2 – H3PO4 – H2O at 37ºC. The solubility is expressed in the total amount of calcium ions in solution)

Dr. Shozo Takagi, National Institute of Standards Technology (Figure 3.2: Crystal structure of hexagonal HAp projected down the *c* –axis, Figure 3.4: Crystal structure of octacalcium phosphate projected down the c -axis. The region with shaded atoms displays similarity to Hap. Hydrogen atoms are omitted for reasons of clarity, Figure 3.5: A projection of the structure of α -Ca3(PO4)2 on the (001) plane, Figure 3.7: Crystal structure of DCPD is shown, as viewed down the b-axis)

Prof. Nora de Leeuw, University College London (Figure 3.3: View onto the (0001) plane of the FAp structure, showing hexagonal symmetry and the relationship between a hexagonal unit cell (pink) and a monoclinic unit cell (blue) (Ca=blue; O=red; P=yellow; F=green))

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**CONTENTS**

[1 Bioceramics 1](#_Toc200810396)

[1.1 Introduction 1](#_Toc200810397)

[1.2 Classification of Bioceramics with Natural Tissues 4](#_Toc200810398)

[1.2.1 Bioinert Ceramics 4](#_Toc200810399)

[1.2.2 Bioactive Ceramics 4](#_Toc200810400)

[1.2.3 Biodegradable Ceramics 5](#_Toc200810401)

[1.3 Compositional Classification of Bioceramics 6](#_Toc200810402)

[1.3.1 Oxide Ceramics 6](#_Toc200810403)

[1.3.2 Carbons 7](#_Toc200810404)

[1.3.3 Glasses 7](#_Toc200810405)

[1.3.4 Calcium Phosphate Ceramics 8](#_Toc200810406)

[1.3.5 Composites 9](#_Toc200810407)

[2 Biological Apatite 10](#_Toc200810408)

[2.1 Introduction 11](#_Toc200810409)

[2.2 Types of Natural Bone 12](#_Toc200810410)

[2.3 Chemical Composition of Natural Bone 12](#_Toc200810411)

[2.4 Physical Properties of Natural Bone 13](#_Toc200810412)

[2.5 Natural Bone Cells 14](#_Toc200810413)

[2.5.1 Osteocytes 14](#_Toc200810414)

[2.5.2 Osteoblasts 14](#_Toc200810415)

[2.5.3 Osteoclasts 15](#_Toc200810416)

[2.6 Natural Bone Remodelling by Osteoblasts and Osteoclasts 15](#_Toc200810417)

[3 Calcium Apatites 16](#_Toc200810418)

[3.1 Introduction 16](#_Toc200810419)

[3.2 Calcium Phosphate Compounds 17](#_Toc200810420)

[3.2.1 Low Temperature Calcium Phosphates 18](#_Toc200810421)

[3.2.2 High Temperature Calcium Phosphates 21](#_Toc200810422)

[3.3 Crystal Structures of Calcium Phosphates 23](#_Toc200810423)

[3.3.1 Hydroxyapatite 23](#_Toc200810424)

[3.3.2 Fluorapatite 25](#_Toc200810425)

[3.3.3 Chlorapatite 26](#_Toc200810426)

[3.3.4 Octacalcium Phosphate 27](#_Toc200810427)

[3.3.5 Tricalcium Phosphate (α and β) 28](#_Toc200810428)

[3.3.6 Tetracalcium Phosphate 28](#_Toc200810429)

[3.3.7 Amorphous Calcium Phosphate 29](#_Toc200810430)

[3.3.8 Dicalcium Phosphate Dihydrous 29](#_Toc200810431)

[3.3.9 Dicalcium Phosphate Anhydrous 30](#_Toc200810432)

[3.4 Substitutions in Calcium Phosphates 30](#_Toc200810433)

[4 Hydroxyapatite (HAp) 32](#_Toc200810434)

[4.1 Introduction 32](#_Toc200810435)

[4.2 Physico-chemical properties of HAp 32](#_Toc200810436)

[4.2.1 Mechanical Properties of HAp 32](#_Toc200810437)

[4.2.2 Chemical Composition of HAp 33](#_Toc200810438)

[4.2.3 Crystal Structure of HAp 34](#_Toc200810439)

[4.2.4 Impurity Contents of HAp 34](#_Toc200810440)

[4.3 Techniques for HAp Powder Synthesis 36](#_Toc200810441)

[4.3.1 Precipitation Route 37](#_Toc200810442)

[4.3.2 Hydrothermal Route 40](#_Toc200810443)

[4.3.3 Hydroylsis Route 42](#_Toc200810444)

[4.3.4 Sol-Gel Route 43](#_Toc200810445)

[4.3.5 Solid-State Reaction Route 48](#_Toc200810446)

[4.3.6 Spray Drying Reaction 50](#_Toc200810447)

[4.3.7 Freeze Drying Reaction 51](#_Toc200810448)

[4.3.8 Novel Routes for Synthesising HAp 53](#_Toc200810449)

[4.3.9 Use of Novel Materials for HAp Synthesis 54](#_Toc200810450)

[4.4 Precipitation Kinetics in HAp Synthesis 56](#_Toc200810451)

[4.4.1 Creation of Supersaturation 56](#_Toc200810452)

[4.4.2 Kinetics of Nucleation 59](#_Toc200810453)

[4.4.3 Kinetics of Crystal Growth 60](#_Toc200810454)

[4.5 Powder Processing of HAp 63](#_Toc200810455)

[4.5.1 Thermal Behaviour of HAp 63](#_Toc200810456)

[4.5.2 Grinding 67](#_Toc200810457)

[4.5.3 Sieving 68](#_Toc200810458)

[4.6 Calcium Phosphate Coating Application onto HAp 68](#_Toc200810459)

[4.7 Biocompatibilty and Toxicity 70](#_Toc200810460)

[4.8 *In vivo* Interfacial Reactions with Bone 70](#_Toc200810461)

[5 SUMMARY 71](#_Toc200810462)

[6 References 72](#_Toc200810463)

**Figures**

[Figure 2‑1: Composition of natural bone 11](#_Toc200810366)

[Figure 3‑1 Solubility isotherms of CaP phases in the system Ca(OH)2-H3PO4-H2O at 37°C. The solubility is expressed in the total amount of calcium ions in solution [33] 17](#_Toc200810367)

[Figure 3‑2 Crystal structure of hexagonal HAp projected down the *c* –axis [29] 24](#_Toc200810368)

[Figure 3‑3 View onto the (0001) plane of the FAp structure, showing hexagonal symmetry and the relationship between a hexagonal unit cell (pink) and a monoclinic unit cell (blue) (Ca=blue; O=red; P=yellow; F=green) [55] 25](#_Toc200810369)

[Figure 3‑4 Crystal structure of octacalcium phosphate projected down the c -axis. The region with shaded atoms displays similarity to Hap. Hydrogen atoms are omitted for reasons of clarity [29] 27](#_Toc200810370)

[Figure 3‑5 A projection of the structure of α -Ca3(PO4)2 on the (001) plane. Adapted from [29] 28](#_Toc200810371)

[Figure 3‑6 Schematic illustration of the TTCP structures projected on the (1 0 0) plane [60] 29](#_Toc200810372)

[Figure 3‑7 Crystal structure of DCPD is shown, as viewed down the b-axis [29] 30](#_Toc200810373)

[Figure 4‑1 A flow chart for the synthesis of HAp powders via the precipitation route for Reaction 1 (left hand side) and Reaction 2 (right hand side) 39](#_Toc200810374)

[Figure 4‑2: A flow chart for the sol-gel synthesis of HAp powder 44](#_Toc200810375)

[Figure 4‑3: Schematic of spray dry apparatus for direct HAp powder synthesis 50](#_Toc200810376)

[Figure 4‑4: Overall view of the freeze-drying chamber 52](#_Toc200810377)

**Tables**

[Table 1‑1 Biomedical applications of Bioceramics materials (Adapted from [1]) 2](#_Toc200810378)

[Table 1‑2 Mechanical Properties of bioceramics used in medical applications (Adapted from [1]) 3](#_Toc200810379)

[Table 1‑3 Biological responses for various bioceramics 5](#_Toc200810380)

[Table 2‑1: Comparative composition of enamel and human bone (Adapted from [16, 20, 21]) 12](#_Toc200810381)

[Table 2‑2: Mechanical properties of human bone (Adapted from [16, 20]) 14](#_Toc200810382)

[Table 3‑1 Calcium phosphate compounds: chemical formulae, Ca/P molar ratios, dissolution rates and their acronyms 20](#_Toc200810383)

[Table 3‑2 Unit-cell positions of the HAp lattice [16] 24](#_Toc200810384)

[Table 3‑3 Unit-cell positions of the FAp lattice [54] 25](#_Toc200810385)

[Table 3‑4 Summary of unit cell Information for HAp, FAp and ClAp 26](#_Toc200810386)

[Table 3‑5 Summary of possible apatite structures 31](#_Toc200810387)

[Table 4‑1: Mechanical Properties for HAp powder [16, 79, 84] 33](#_Toc200810388)

[Table 4‑2: Composition of synthetic HAp (Adapted from [16, 21]) 33](#_Toc200810389)

[Table 4‑3 Impurity content in HAp. Adapted from [16] 35](#_Toc200810390)

[Table 4‑4: Analytical overview of the wet chemical methods 47](#_Toc200810391)

[Table 4‑5: Analytical overview of the dry chemical methods 49](#_Toc200810392)

[Table 4‑6: Analytical overview of the vapour phase reactions 52](#_Toc200810393)

[Table 4‑4: Solubility Products, Ksp, for various precipitation temperatures. Adapted from [16] 58](#_Toc200810394)

[Table 4‑5: Summary of HAp coating techniques for application onto orthopaedic implants 69](#_Toc200810395)

# Bioceramics

## Introduction

Bioceramic materials are a class of ceramics used for repair and replacement of diseased and damaged parts of musculoskeletal systems. Bioceramics have a wide array of medical applications; such as, structural functions in joint or tissue replacements. They can be used as coatings to improve the biocompatibility of metal implants, and can function as resorbable lattices which provide temporary structures and a framework that is dissolved and replaced as the body rebuilds tissue. Some ceramics even feature drug-delivery capability. Materials for successful surgical implantation and use within medical devices must therefore be non-toxic. Table 1-1 examines the biomedical applications of bioceramics.

The perfect material for medical applications would not only be biocompatible, but also have physical properties similar to those of the tissue being replaced or repaired. Ceramics, though they include good chemical and corrosion-resistant properties, are notoriously brittle. Researchers therefore have sought ways of combining desirable ceramics with other materials to tailor properties such as strength and elasticity to meet the desired requirements. Composites, functionally gradient materials, and coatings have been studied to optimize material choices.

Much work has been devoted to the interfacial reactions of biological systems with Hydroxyapatite (HAp) is used as a coating for metal surgical implants (most often made of titanium and its alloys, or stainless steels), and recent studies have examined the possibility of its use in composite form, in materials that combine polymers with ceramic or metal/ceramic combinations. Considerable research has been performed on methods of coating application and in-situ synthesis of apatites, and the implications for ceramic properties and microstructure.

Bioceramics in a number of forms and compositions are currently in use or under consideration, with more in continuous development. Alumina and zirconia are among the bioinert ceramics used for prosthetic devices. Bioactive glasses and machinable glass-ceramics are available under a number of trade names. Porous ceramics such as calcium phosphate-based materials are used for filling bone defects. The ability to control porosity and solubility of some ceramic materials offers the possibility of use as drug delivery systems. Glass microspheres have been employed as delivery systems for radioactive therapeutic agents, for example.

Material selection must also include consideration of the ease of forming into sometimes complex shapes, and with strict dimensional tolerances. Devices for use within the body must be able to withstand corrosion in a biological environment and endure use for years without undue wear (and without causing damage to surrounding tissues). Before insertion, they should be unchanged during storage, and must be sterilized without damage. Materials scientists must keep in mind a multitude of properties and capabilities as they seek to develop the materials that will serve to improve the lives of patients. The thermal and chemical stability of ceramics, high strength, wear resistance and durability all contribute to making ceramics good candidate materials for surgical implants.

Table ‑ Biomedical applications of Bioceramics materials (Adapted from [1])

|  |  |  |
| --- | --- | --- |
| ***Devices***  | ***Function***  | ***Bioceramic Material***  |
| Artificial total hip, knee, shoulder, elbow, wrist  | Reconstruct arthritic or fractured joints  | High-density alumina, metal bioglass coatings  |
| Bone plates, screws, wires  | Repair fractures  | Bioglass-metal fiber composite, Polysulfone-carbon fiber composite  |
| Intramedullary nails  | Align fractures  | Bioglass-metal fiber composite, Polysulfone-carbon fiber composite  |
| Harrington rods  | Correct chronic spinal curvature  | Bioglass-metal fiber composite, Polysulfone-carbon fiber composite  |
| Permanently implanted artificial limbs  | Replace missing extremities  | Bioglass-metal fiber composite, Polysulfone-carbon fiber composite  |
| Vertebrae Spacers and extensors  | Correct congenital deformity  | Al2O3  |
| Spinal fusion  | Immobilize vertebrae to protect spinal cord  | Bioglass  |
| Alveolar bone replacements, mandibular reconstruction  | Restore the alveolar ridge to improve denture fit  | Polytetra fluro ethylene (PTFE) - carbon composite, Porous Al2O3, Bioglass, dense-apatite  |
| End osseous tooth replacement implants  | Replace diseased, damaged or loosened teeth  | Al2O3, Bioglass, dense hydroxyapatite, vitreous carbon  |
| Orthodontic anchors  | Provide posts for stress application required to change deformities  | Bioglass-coated Al2O3, Bioglass coated  |

There exists, a markedly strong quantitative difference of the mechanical properties between natural bone and bioceramics, as depicted in Table 1-2. These differences lead to strong gradients of the modulus of elasticity (Young´s modulus) that ultimately give rise to so-called “stress shielding”. This means that load put on the implant during movement will not be transmitted by the bone itself, but instead through the stiff ceramic femoral ball into the likewise very stiff titanium alloy stem. The absence of regular tensile loads will eventually lead to atrophic loss of cortical bone matter, as these loads are required for living bone to stay healthy.

Table ‑ Mechanical Properties of bioceramics used in medical applications (Adapted from [1])

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Material**  | **Young’s Modulus (GPa)**  | **Compressive Strength (MPa)**  | **Bond strength (GPa)**  | **Hardness**  | **Density** **(g/cm3)**  | **Fracture Toughness,** **(MPam1/2)**  |
| AluminaAl2O3 | 380  | 4000  | 300-400  | 2000-3000 (HV)  | >3.9  | 5.0-6.0  |
| ZrO2 (PS)  | 150-200  | 2000  | 200-500  | 1000-3000 (HV)  | ≈6.0  | 4.0-12.0  |
| Graphite  | 20-25  | 138  | NA  | NA  | 1.5-1.9  | NA  |
| (LTI) Pyrolitic Carbon  | 17-28  | 900  | 270-500  | NA  | 1.7-2.2  | NA  |
| Vitreous Carbon  | 24-31  | 172  | 70-207  | 150-200 (DPH)  | 1.4-1.6  | NA  |
| HAp  | 73-117  | 600  | 120  | 350  | 3.1  | <1  |
| Bioglass  | ≈75  | 1000  | 50  | NA  | 2.5  | 0.7  |
| AW Glass Ceramic  | 118  | 1080  | 215  | 680  | 2.8  | ≈2  |
| Bone  | 3-30  | 130-180  | 60-160  | NA  | NA  | NA  |

*The variation in Young's Modulus noted for some of the materials listed is due to variation in density of test specimens. PS - Partially Stabilized; HA - Hydroxyapatite; NA - Not Available; AW - Apatite-Wallastonite; HV - Vickers Hardness; DPH - Diamond Pyramid Hardness*

While alumina is relatively stiffer, with a higher compressive strength in comparison to zirconia, the latter performs mechanically better in terms of tensile and flexural strengths and also, fracture toughness.

## Classification of Bioceramics with Natural Tissues

Insertion of a synthetic material into the human body, elicits a biological response, in which the biological tissue reacts towards the implant in a variety of ways, depending on the composition of the material. The mechanism of tissue interaction (if any) depends on the tissue response to the implant surface. Bioceramics are classified in accordance to their biological response in contact with living tissues, to which they illicit (refer to Table 1-3). The three main classes for Bioceramics are as follows: (1) Bioinert, (2) Bioactive and (3) Biodegradable.

### Bioinert Ceramics

Bioinert ceramics do not release any toxic constituents, but also do not show positive interaction with biologica tissue. The term bioinert refers to a material that once placed in the human body has minimal interaction with its surrounding tissue. As a response of the body to these materials; a non-adherent capsule of connective tissue is usually formed around the bioinert material that in the case of bone remodelling manifests itself by a shape-mediated contact osteogenesis. Through the bone-materials interface, only compressive forces will be transmitted (“bony on-growth”).

Examples of these include alumina, partially stabilised zirconia, and calcium alumina. Generally a fibrous capsule might form around bioinert implants hence its biofunctionality relies on tissue integration through the implant.

### Bioactive Ceramics

Bioactive ceramics are durable materials that can undergo interfacial interactions with surrounding biological tissues. This occurs through a time-dependent kinetic modification of the surface, triggered by their implantation within the living bone. An ion-exchange reaction between the bioactive implant and surrounding body fluids, results in the formation of a biologically active apatite (such as, HAp) layer on the implant that is chemically and crystallographically equivalent to the mineral phase in bone. In contrast to bioinert ceramics, there is chemical bonding to the bone along the interface, believed to be triggered by the adsorption of bone growth-mediating proteins at the biomaterials surface. As a result of this, there will be a biochemically-mediated strong bonding osteogenesis [2]. To a certain degree, tensile and shear forces can be transmitted through the interface (“bony ingrowth”) in addition to compressive forces.

Examples of these materials include synthetic HAp [Ca10(PO4)6(OH)2], glass ceramic A-W and bioglass® [3]. A particular advantage of Bioglasss (45S5 Bioglasss) is its ability to bond to both hard and soft tissues [4]. The primary shortcoming of Bioglasss is mechanical weakness and low fracture toughness due to an amorphous two-dimensional glass network. The bending strength of most Bioglasss compositions is in the range of 40–60 MPa, which is not suitable for major load-bearing applications.

Table ‑ Biological responses for various bioceramics

|  |  |  |
| --- | --- | --- |
| ***Bioceramic Material*** | ***Medical Application*** | ***Biological Behaviour***  |
| Al2O3 | Femoral balls, inserts of acetabularcups | bioinert |
| ZrO2  | Femoral balls | bioinert |
| HAp | Bone cavity fillings, coatings, ear implants, vertebrae replacement | bioactive |
| Tricalcium phosphate, TCP | Bone replacement | bioactive |
| Tetracalcium phosphate, TTCP | Dental cement | bioactive |
| Bioglass | Bone replacement | bioactive |

### Biodegradable Ceramics

Biodegradable ceramics are otherwise termed; soluble or bioresorbable (that is, it eventually dissolves and is replaced or incorporated into biological tissue). The dissolution rate depends upon the chemical structure and composition of the bioceramic. Examples of bioresorbable materials include tricalcium phosphate (TCP, Ca3(PO4)2), calcium oxide (CaO) and calcium carbonate (CaCO3).

## Compositional Classification of Bioceramics

### Oxide Ceramics

The primary oxide ceramics of interest include alumina (Al2O3)-based ceramics and zirconia (ZrO2)-based ceramics. An overview of both of these ceramics is detailed in the following descriptions for the valuable physical and chemical characteristics which favour its use for certain medical applications.

#### Alumina (Al2O3)-based ceramics

Alumina bioceramics have a proven bioinertness, as established since 1975. The mechanical properties for alumina ceramics are presented in Table 1-2 demonstrating high levels of hardness and abrasion resistance. The surface energy and surface smoothness of this ceramic attribute to the excellent wear and friction behaviour of Al2O3, although, there exists only one thermodynamically stable phase (that is, Al2O3 that has a hexagonal structure with aluminium ions at the octahedral interstitial sites). The abrasion resistance, strength and chemical inertness of alumina have resulted in its use as a ceramic for dental and bone implant applications.

The biocompatibility of alumina ceramic has been tested by many researchers. Recent experimental studies conducted by Standard et al. [5] investigated the bone ingrowth into a porous bioinert alumina ceramic, using a single-pore in vivo model. They implanted alumina tubes (1.3 mm outside diameter, 0.6 mm inside diameter, and 15 mm length) into the femoral medullary canal of female rats for up to 16 weeks. The tissues formed in the tubes were identified by histological analysis and were quantified by image analysis. A tissue front consisting of fibrovascular tissue, osteoid, woven bone, and some lamellar bone advanced into the tubes with increasing time of implantation. Behind this front, lamellar bone lined interior surfaces of the ceramic tubes and enclosed a central lumen of marrow tissue. The progression of tissue into the tubes was considered to represent the cascade of tissue differentiation within a bioinert porous structure.

#### Zirconia-based ceramics

Zirconia is a biomaterial that possesses high mechanical strength and fracture toughness. Zirconia ceramics have several advantages over other ceramic materials due to the transformation toughening mechanisms operating within their microstructure. Its mechanical properties are outlined in Table 1-2. The research on the use of zirconia ceramics as biomaterials commenced about twenty years ago and now zirconia is in clinical use in total hip replacement (THR), but developments are in progress for application in other medical devices. Today's main application of zirconia ceramics is in THR ball heads. Fini et al. [6] have investigated the osteointegration of zirconia in normal and osteopenic rats by means of histomorphometry. They found the tested material was biocompatible in vitro and confirmed that bone mineral density is a strong predictor of the osteointegration of an orthopaedic implant and that the use of pathological animal models is necessary to completely characterize biomaterials.

### Carbons

Carbon is an extremely versatile bioceramic and exists in a variety of forms. Table 1-2 outlines the physical characteristic with respect to the mechanical properties of various types of carbons used in medical applications. Bokras et al. [7] emphasised that the good compatibility of carbonaceous materials with bone and other tissue and the similarity of the mechanical properties of carbon to those of bone; indicate that carbon is indeed an exciting candidate for orthopedic implantation. As opposed to metals, polymers and other ceramics, these carbonaceous materials do not suffer from cyclic fatigue. However, their intrinsic brittleness and low tensile strength limits their use in major load bearing applications. It is used as biomaterial particularly in contact with blood. Hence it is important to evaluate its blood compatibility.

Shi et al. [8] studied the thromboembolic rates in the mitral and aortic positions of omni-carbon valve constructed entirely of pyrolytic carbon. They found a total of 569 aortic omnicarbon valves had thromboembolic events of 0.5% and a total of 298 mitral omnicarbon valves had a thromboembolic rate of 1.6%. Other studies conducted by Zimmerman et al. [9], studied the compatibility of filamentous carbon fibre. It was speculated that it does not corrode and does not elicit any significant foreign body response *in vivo*.

*In vitro* tests were conducted through work carried out by Evans et al. [10] on diamond like carbon films on germanium and various metals and oxides that has been extended to coating plastics used in medical and bioengineering applications. The test proved its biocompatibility and allowed cells to grow without inflammatory response and with cell integrity maintenance.

### Glasses

Bioactive glasses were discovered in 1969 and provided for the first time an alternative; interfacial bonding of an implant with host tissues [11]. Bioglasses are an important class of biomaterials, which are prepared from a mixture of silica, alumina, magnesia, calcium oxide, sodium oxide, and phosphorous oxide. Hench et al. [11] have extensively studied ternary phases of these oxides containing some P2O5 and evaluated the biocompatibility of the quenched glasses of various compositions.

Structurally, all silica-based glasses have the same basic building block –SiO44-. Glasses of various compositions can be obtained to yield varied physical properties. Bioglasses have also found a place in the use of prosthetics as a medical application. These bioglasses are embedded in a biomaterial support to form prosthetics for hard tissues. Such prosthetics are biocompatible, show excellent mechanical properties and are useful for orthopaedic and dental prosthetics [11].

Evidence of a lack of toxicity of various bioglass formulations has been reported from many studies carried out both *in vivo* and *in vitro* [12]. The bioactivity of bioglass results from the formation of a HAp layer on the surface.

Recent studies have demonstrated how a controlled release of the ionic dissolution products of bioactive glasses can lead to regeneration of surrounding tissues. The mechanism for *in situ* tissue regeneration involves the upregulation of seven families of genes that control the osteoblast cell cycle, cell division and cell differentiation. It is necessary to have the critical concentrations of Si and Ca ions present within 48 hours, so that osteoblasts are capable of differentiation into a mature osteocyte phenotype, and can ultimately begin to proliferate and succeed in the regeneration of newly formed bone. Osteoblasts that are not in the correct phase of the cell cycle and unable to proceed towards differentiation are switched into apoptosis by the ionic dissolution products. Gene activation by controlled ion release provides the conceptual basis for molecular design of a third generation of biomaterials optimised for in situ tissue regeneration. [3]

### Calcium Phosphate Ceramics

An increasing number of bioceramics are specifically prepared to impart a biological activity promoting the integration of the implant into biological tissues, thus favouring their repair. Implanted bioceramics interact mechanically and chemically with the host tissue and with cells of the biological environment.

The primary focus is on calcium phosphate based bioceramics and their medical application in the form of porous ceramics, cements, coatings, composites and their use in tissue engineering. Independent of the calcium phosphate form selected, the bioceramic generally fulfils only part of the tissue functions. Currently, biomimetism appears as an appealing concept for biomaterials used as tissue or organ substitutes [13].

### Composites

Biocomposites offer many important and exciting possibilities for medical applications, and are a rapidly expanding area of research. Through the combination of a ductile polymer matrix with hard, bioactive particulate ceramic filler, optimal materials can be designed. The desirable mechanical properties of the matrix component compensate for the poor mechanical behaviour of the ceramic, while in turn the desirable bioactive properties of the filler improve those of the polymer, expanding the possible uses of each material within the body. With optimised matrix selection, filler content, particle size and morphology, and implant surface topography, improved biocomposites can be produced and will continue to advance the medical and orthopaedic field.

# Biological Apatite

The mineral in bones and teeth is an impure form of HAp with carbonate as the major impurity, and for this reason HAp is extensively used in the manufacture of prostheses and implants. It has been demonstrated through experimental and theoretical studies, that biological and synthetic HAp’s display crystal structure similarities and differences that affect greatly the bioactivity of the synthetic materials. Recent developments in structure research of bioapatites report experimental values of the hydroxyl concentration in biological apatites in the range of ~20% of the amount in synthetic HAp.

Previous attempts in studying the hydroxyl content of HAp indicate that the mineral phase in bone contains less hydroxyl than that of stoichiometric HAp. However, experimental results calculated were questionable due to specimen pretreatment that was used to eliminate interference from the bone organic matrix. Cho et al. [14] estimated that the hydroxyl content in human cortical bone is only about 20% of that in stoichiometric HAp, via a method which avoided the need for any such pre-treatment.

In addition to the difference in hydroxyl content, there is also a size difference between the nano-crystallites in bone HAp and the micro-crystallites in the synthetic material.

## Introduction

Bone is a natural composite material (see Figure 2-1), which by weight contains about 62 wt. % inorganic phase (that is, bone mineral), 25 wt. % organic phase (that is, the matrix), 10 wt. % water and 3wt. % residual [15, 16]. Bone is also a living tissue, with about 15% of its weight being due to the cellular content [17].

The matrix of bone is comprised primarily of Type I collagen that is highly aligned, yielding a very anisotropic structure. This organic component of bone is predominantly responsible for its tensile strength. The collagen is laid down in the form of fibres, which aid in the provision of flexibility to bone.

The mineral component of bone is rarely stoichiometric, containing many substitutions such as magnesium, sodium, potassium, fluorine, chlorine, and carbonate ions [18]. This inorganic phase consists of submicroscopic crystals of an apatite of calcium and phosphate, whose crystal structure closely resembles that of HAp. The apatitic mineral in bone is closely associated with the collagen fibers and is made up of long, flat, plate-like nanocrystals that are approximately 40 nm long, 10 nm wide and 1-3 nm thick. This mineral component gives rise to the compressive strength of bone. In the body, bone serves a number of functions, such as providing the cells, found in the marrow, that differentiate into blood cells, and also acting as a calcium reservoir. Nevertheless, its primary purpose is to provide mechanical support for soft tissues and serve as an anchor for the muscles that generate motion. [19].



Figure 2‑1: Composition of natural bone

## Types of Natural Bone

There are two types of bone, compact or cortical, and cancellous or trabecular (also known as spongy) bone. Compact bone is very dense, consisting of parallel cylindrical units (osteons), and is found in the shafts of the long bones as well as on the outer surface of the smaller bones in the body. Trabecular bone is less dense and is made up of an array of rods and struts that form an open-cell foam, the pores of which are filled in by marrow. This type of bone is found at the ends of the long bones and inside the smaller bones (ribs, spine).The anisotropic structure of bone leads to mechanical properties that exhibit directionality. This directionality results from the fact that bone has evolved to be both tough and stiff, two competing properties which are optimised in bone but with an inherent loss in isotropy. Nevertheless, bone exhibits extraordinary mechanical properties, displaying both viscoelastic and semi-brittle behaviour

.

## Chemical Composition of Natural Bone

The constituent elements within bone mineral are given in Table 2-1. Mg, Na, K and carbonate ions are also present in bone salts, although they are not organised into specific crystals. Fluid Ca-P compounds in the matrix are transformed to HAp.

Table ‑: Comparative composition of enamel and human bone (Adapted from [16, 20, 21])

|  |  |  |  |
| --- | --- | --- | --- |
| **Mineral Component** | **Chemical Formula** | **Enamel** | **Bone** |
| **Constituents (wt. %)** |  |  |  |
| Calcium | *Ca2+* | 36.0 | 24.5 |
| Phosphorous | *P* | 17.7 | 11.5 |
| Ca/P molar ratio |  | 1.62 | 1.65 |
| Carbonate | *CO32-* | 3.2 | 5.8 |
| Sodium  | *Na+* | 0.5 | 0.7 |
| Magnesium | *Mg2+* | 0.44 | 0.55 |
| Potassium | *K+* | 0.08 | 0.03 |
| Chloride | *Cl-* | 0.30 | 0.10 |
| Fluoride | *F-* | 0.01 | 0.02 |
| **Total Inorganic (mineral)** |  | **97.0** | **65.0** |
| **Total Organic** |  | **1.0** | **25.0** |
| **Absorbed H2O** |  | **1.5** | **9.7** |
| **Trace elements** | ***Sr2+, Pb2+, Zn2+, Cu2+, Fe3+*** |  |  |

The main mineral components forming human bone are calcium (36.7 wt. %) and phosphorous (16.0 wt. %) with trace amounts of other minerals. A significant amount of carbonate (7.4 wt. %), however, also appears to be present within the biological apatite, bone. Slosarczyk et al. [22] confirm that biological apatites present in bone: dentin: enamel contain different amounts of carbonate 7.4: 5.6: 3.5 wt% respectively. The presence of CO32- in their structure is of paramount importance; as it is the main source of lattice distortion, creating microstresses and crystalline defects in its vicinity which, in turn, play a fundamental role in the solubility. As a consequence, synthetic apatites aimed at emulating the biological scenario should exhibit small particle sizes and the presence of CO32-. Among the hetero-ions substituting in biological HAp, CO32- is the most abundant (2–8 wt. %, and partially substitutes both in the PO43- site (B-type CHAp) and the OH- site (A-type CHAp) of the HAp structure. B-type carbonated HAp demonstrates improved solubility, collagen deposition *in vitro* and reabsorption *in vivo*, compared with stoichiometric HAp and/or to A-type CHAp [23].

The type and amount of ionic substitutions in the apatite phase of bone vary from the wt% level (such as, 5wt% CO32-) to the ppm - ppb level (such as, Sr2+ or Ba2+) [24]. Although these levels of substitution are small, the replacement ions play a major role in the biochemistry of hard tissues. Certain metals, such as aluminium, iron, cadmium and lead, are known to cause bone pathologies in humans and animals. Studies have been reported concerning the effects of aluminium in bone disorders in individuals with chronic renal disease subjected to dialysis [25]. Levin and Goldberg [26] concludes that lead poisoning, otherwise termed, ‘plumbism’’, is caused by an inhalation of lead in the form of dust or its absorption through the skin, the mechanism involved being attributed to isomorphous substitution of Ca2+ by Pb2+ in bone, leading to formation of solid solutions of HAp and lead HAp. Also, the interaction of cadmium with biological apatites is responsible for an osseous disease with effects similar to osteoporosis [27].

## Physical Properties of Natural Bone

Compact bone has a compressive strength in the longitudinal direction (parallel to the long axis) ranging from 131-224 MPa, and a Young’s modulus between 17-20 GPa. It also exhibits good fracture toughness, which is much higher in the transverse direction than in the longitudinal one. The mechanical properties of trabecular bone are highly dependent on its density. Compressive strength varies with the second power of density, whereas Young’s modulus scales as the second or third power, with values ranging between 5-10 MPa and 50-100 MPa for strength and modulus, respectively [16]. An overview of the mechanical properties for natural bone is shown in Table 2-2.

Table ‑: Mechanical properties of human bone (Adapted from [16, 20])

|  |  |
| --- | --- |
| **Mechanical Property** | **Test direction related to bone axis** |
| **Parallel** | **Normal** |
| Tensile Strength (MPa) | 124 - 174 | 49 |
| Compressive Strength (MPa) | 170 - 193 | 133 |
| Young’s Modulus (GPa) | 17 – 18.920 – 27 (random) | 11.5 |
| Micro Hardness (VPN) | 30-60 | - |
| Fracture Toughness (MPa m1/2) | 2-12 | 8 |
| Bending Strength (Mpa) | 160 |  |
| Shear Strength (Mpa) | 54 |  |
| Ultimate Tensile Strain (UTS) | 0.014 – 0.031 | 0.007 |
| Ultimate Compressive Strain | 0.0185 – 0.026 | 0.028 |
| Yield Tensile Strain | 0.007 | 0.004 |
| Yield Compressive Strain | 0.010 | 0.011 |

## Natural Bone Cells

In any specimen of bone, three functionally distinct cell types are present; osteocytes, osteoblasts, and osteoclasts [28]. An overview for each cell type is described in this section.

### Osteocytes

Osteocytes are small, inactive cells, seemingly isolated from one another in individual lacunae. Inconspicuous cell processes extend out through tiny canaliculi and provide gap-junctional contact among neighboring osteocytes as a means of communication and nutrient supply.

### Osteoblasts

Osteoblasts(bone-forming cells) are small cuboidal cells, usually found lying adjacent to one another upon lamellae they have just secreted. Osteoblasts lay down new bone lamellae.  They are active in bone development and also in bone remodelling. Populations of osteocytes and osteoblasts may be interchangeable, reflecting different stages in the activity of individual cells.  That is, active osteoblasts that become enclosed in bone may adopt the resting osteocyte form, while osteocytes which are released from their bony matrix (by osteoclast activity) may become active osteoblasts.

### Osteoclasts

Osteoclasts (bone-removing cells) are large cells with multiple nuclei, each one typically sitting alone within a small hollow (Howship's lacuna) that it has just made by "eating" away the adjacent bone matrix. Osteoclasts remove preexisting bone.  They are active in bone development and also in bone remodelling. Osteoclasts are more closely related to macrophages than to osteocytes or osteoblasts.  They form a distinct cell population derived from the same precursor cells as macrophages.

## Natural Bone Remodelling by Osteoblasts and Osteoclasts

The modelling and remodelling processes of human bone are not indifferent at cellular level. They are based on the separate actions of bone resorbing cells, called osteoclasts, and bone forming cells, called osteoblasts [28]. The remodelling process begins at a quiescent bone surface with the appearance of osteoclasts. These are large multinucleated cells that form by fusion of mononuclear precursors of haemotopoetic origin. They attach to the bone tissue matrix and form a ruffled border at the bone/osteoclast interface that is completely surrounded by a “sealing” zone. Thus the osteoclast creates an isolated microenvironment. Subsequently, the osteoclast acidifies the microenvironment and dissolves the organic and inorganic matrices of the bone. For a brief period after this resorptive process stops, osteoblasts appear at the same surface site. The osteoblasts derive from mesenchymal stem cells found in the bone marrow, periosteum and soft tissues. They deposit osteoid and mineralise it, so actually forming new bone. Some of the osteoblasts are encapsulated in the osteoid matrix and differentiate to osteocytes. Remaining osteoblasts continue to synthesize bone until they eventually stop and transform to quiescent lining cells that completely cover the newly formed bone surface. These lining cells are highly interconnected with the osteocytes in the bone matrix through a network of canaliculi. It appears that osteoclasts and osteoblasts closely collaborate in the remodelling process in what is called a Basic Multicellular Unit (BMU).

# Calcium Apatites

## Introduction

Calcium phosphates with clinical applications are used in reconstructive surgery. When considering the repair of a skeletal section, two diverse routes are considered: (i) to replace the damaged part or (ii) to substitute it, thus regenerating the bone. This second route constitutes the role played by calcium phosphates, which as previously discussed, are classified among the bioactive ceramics group. Bioceramics, and therefore, calcium phosphates, exhibit very good biocompatibility and bone integration qualities, and display materials showing closest similarity to the mineral component of bone; this fact, together with their bioactivity, make them very good candidates for bone regeneration.

Other various orthopaedic and dental medical applications of calcium phosphates, include repair of bone defects, repair of periodontal defects, alveolar ridge augmentation, ear implants, eye implants, maxillofacial reconstruction, spine fusion, bone space fillers, bone cement additives, composites and implant coatings.

It is only in the past 20–30 years that interest in the use of dense HAp for implantation has developed. Significant research work was reported in the 1980s and 1990s by the groups led by DeGroot, Jarcho, Driessens, Bonfield and Zhang [29]. Calcium phosphates are now used for a variety of different applications covering all areas of the skeleton including spinal fusion, craniomaxillofacial reconstruction, and treatment of bone defects, fracture treatment, total joint replacement (bone augmentation) and revision surgery. Only certain compounds are useful for implantation in the body, compounds with a Ca/P ratio less than 1 are not suitable for biological implantation due to their high solubility. The known pure calcium phosphates have been classified into three major structural types [29]:

**Apatite type, **

These include derivatives of Hap, (X = OH) and fluorapatite, FHAp (X = F) as well as those related to apatite-type structures; such as octacalcium phosphate (OCP), [Octacalcium bis(hydrogenphosphate) tetrakis(phosphate) pentahydrate],  and tetracalcium phosphate (TTCP), .

**Glaserite type,**

These can be considered to include all polymorphs of tricalcium phosphates (TCP), .

**Ca-PO4 sheet-containing compounds**

These include dicalcium phosphate dihydrate (DCPD),, dicalcium phosphate anhydrous (DCPA), , and monocalcium phosphates, and .

## Calcium Phosphate Compounds

Various calcium phosphate compounds have been used in the field of biomedical engineering owing to the range of properties that they offer, from TCP being resorbable to HAp being bioactive [30], while microscale HAp is typical highly phase stable and thus less bioresorbable [31].

Calcium phosphates (CaPs) contain the phosphate group. Two different categories of CaP can be categorised as follows: (1) CaP obtained by precipitation from an aqueous solution at or around room temperature (low-temperature CaP) and (2) CaP obtained by thermal reaction (high-temperature CaP). The most important property of CaP is its solubility in water, since the *in vivo* behaviour of CaPs can be predicted to a large extent by their solubility [32]. If the solubility of a CaP, (such as, HAp) is less than the mineral part of bone, it degrades extremely slowly if at all (see Figure 3-1).



Figure ‑ Solubility isotherms of CaP phases in the system Ca(OH)2-H3PO4-H2O at 37°C. The solubility is expressed in the total amount of calcium ions in solution [33]

If the solubility of a CaP is greater than that of the mineral part of bone, it is degraded. Therefore, using the different solubility isotherms of CaP, the in vivo degradation rate of CaP can be predicted to be in the order of (at pH 7.0):

MCPM > TTCP ≈ α-TCP > DCPD > DCPA > OCP > β-TCP > Ca-dHAp > HAp

However, the surface of a highly-soluble CaP can be reactive and may become covered with a poorly soluble CaP, hence reducing its degradation rate.

### Low Temperature Calcium Phosphates

The most common low-temperature CaPs are dicalcium phosphate dihydrate (DCPD), octocalcium phosphate (OCP) and precipitated hydroxyapatite (PHA). An overview of each of these compounds is described in this section.

#### 3.2.1.1 Monocalcium Phosphate Monohydrate

Monocalcium phosphate monohydrate (MCPM; Ca(H2PO4)2.H20) is the most acidic CaP and the most soluble CaP at almost all pH values. Consequently, MCPM is not biocompatible and cannot be used alone as a bone substitute. However, it can be used in combination with a basic CaP compounds, such as α-TCP or β-TCP [34].

#### 3.2.1.2 Dicalcium Phosphate

Dicalcium phosphate (DCP; CaHPO4) powders have proven to be both biocompatible and biodegradable [35, 36]*.* The compound has previously been reported to be present in bone [37]. DCP results normally from the recrystallization of DCPD and is the most stable CaP at low pH. The conversion is faster in water at higher temperature and acidity [37].

#### 3.2.1.3 Dicalcium Phosphate Dihydrate

Dicalcium phosphate dihydrate (DCPD; CaHPO4.2H2O) has been detected in callus, bone and kidney stones. It is also considered as a precursor of HAp in bone [38]. DCPD is the simplest CaP to synthesise. Moreover, it is biocompatible, biodegradable and osteoconductive. DCPD is metastable and can be converted into DCP, OCP or TCP. *In vivo,* DCPD is converted into HAp or biodegraded and replaced by newly formed bone. When a large amount of DCPD is transformed *in vivo* into HAp, inflammatory reactions can occur due to the release of large amount of acid. DCPD is the end product of brushite CPC.

#### 3.2.1.4 Octacalcium Phosphate

Octacalcium phosphate (OCP; Ca8H2(PO4)6.5H2O) has been shown to enhance bone regeneration accompanied by the conversion into hydrolysed apatetic product in situ and the biodegradation. OCP has been advocated to be a precursor of biological apatite [36]. Previous research studies have demonstrated evidence supporting the involvement of OCP as the initial crystal formation in dentin, enamel and bone minerals and has been examined as an alternative to HAp on Ti6A14V, as a biomimetic coating [36]. It has also been used to fill osseous defects. OCP is known to be physicochemically resorbable more so than HAp or β-TCP under physiological conditions [39] OCP can be prepared by hydrolysis of DCPD or α-TCP[40]. It also can be precipitated starting from saturated CaP solutions [36, 41].

#### 3.2.1.5 Precipitated Hydroxyapatite

Precipitated hydroxyapatite (PHAp; Ca10-x(HPO4)x(PO4)6-x(OH)2-x) exhibits a complex chemistry, since PHAp can possess a Ca/P molar ratio ranging from 1.33 to 1.67 (refer to Table 3-1), and sometimes even outside this range*.* PHAp is obtained by precipitation in an aqueous solution above a pH of 7. PHAp crystals are normally poorly crystalline and of submicron dimensions. It has a very large specific surface area, typically 25-1000 m2/g and chemically similar to the apatite present in bone. The main difference is the absence of impurities in the structure, mainly carbonate and magnesium ions. The solubility of PHAp increases with a decrease of the Ca/P molar ratio, crystallinity, and crystal size. The solubility of TCP (PHAp with a Ca/P molar ratio of 1.50) is estimated to be close to that of β-TCP. The solubility of stoichiometric PHAp (Ca:P = 1.67) showed a 20% difference in solubility, depending on the method of preparation [42].

#### 3.2.1.6 Amorphous Calcium Phosphate

An approximation for the chemical formula for Amorphous Calcium Phosphate (ACP) is outlined in Table 3-1. The amorphous nature of ACP is evidenced by the absence of peaks in an X-ray diffraction spectrum. ACP is reported to be more soluble than DCPD [33]. Bohner [37] reported on a highly reactive ACP in combination DCPD with the final composition displaying a poorly crystalline HAp material and resembling natural bone.

Table ‑ Calcium phosphate compounds: chemical formulae, Ca/P molar ratios, dissolution rates and their acronyms

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Ca/P** **molar ratio** | **Calcium Phosphate** | **Mineral Phase** | **Chemical Formula** | **Acronym** |
| 2.0 | Tetracalcium phosphate | Hilgenstockite | Ca4O(PO4)2 | TTCP |
| 1.67 | Hydroxyapatite | Hydroxyapatite | Ca10(PO4)6(OH)2 | HAp |
| 1.67 | Oxyapatite |  | Ca10(PO4)6O | OXA |
| 1.33 – 1.67 | Calcium deficient hydroxyapatite |  | Ca10-xHx(PO4)6(OH)2-x | *Ca-d*HAp |
| 1.5 | β – Tricalcium phosphate |  | β – Ca3(PO4)2 | β – TCP |
| 1.5 | α - Tricalcium phosphate |  | α - Ca3(PO4)2 | α - TCP |
| 1.5 | γ – Tricalcium phosphate |  | γ - Ca3(PO4)2 | γ – TCP |
| 1.33 | Octacalcium phosphate |  | Ca4H(PO4)3 3H2O | OCP |
| 1.00 | Dicalcium phosphate anyhdrous | Monetite | CaHPO4 | DCPA |
| 1.00 | Dicalcium phosphate dihydrate | Brushite | CaHPO4.2H2O | DCPD |
| 1.00 | Calcium pyrophosphate (α, β, γ) |  | α - Ca2P2O7β - Ca2P2O7γ - Ca2P2O7 | CPP |
| 1.00 | Calcium pyrophosphate dihydrate |  | Ca2P2O7.2H2O | CPPD |
| 0.7 | Heptacalcium phosphate |  | Ca7(P5O16)2 | HCP |
| 0.65 | Tetracalcium dihydrogen phosphate |  | Ca4H2P6O20 | TDHP |
| 0.5 | Monocalcium hydrate monohydrate |  | Ca(H2PO4)2.H2O | MCPM |
| 0.5 | Calcium metaphophate (α, β, γ) |  | Ca(PO3)2 | CMP |
|  | Amorphous calcium phosphate |  | Ca3(PO4)2⋅3H2Oa | ACP |

***a = an approximation [43]***

### High Temperature Calcium Phosphates

Traditionally, all CaPs used in medical applications are high-temperature CaPs, in particular: β-TCP, HAp, and β-TCP/HA composites, otherwise known as called bicalcium phosphates (BCP). An introductory overview of some of these compounds is outlined in this section.

#### 3.2.2.1 Monocalcium Phosphate

Monocalcium phosphate (MCP; Ca(H2PO4)2) is obtained by heating or precipitation, at a temperature range of 100-110°C. De Waal [44] reports a method of producing MCP from CaP, which includes reacting the CaP with sulphuric acid to produce MCP.

#### 3.2.2.1 β – Tricalcium Phosphate

β – Tricalcium Phosphate (β-TCP; β-Ca3(PO4)2) can only be obtained at a high temperature (above about 700ºC). This has two consequences: (1) the fabrication of round and monodisperse particles is very difficult due to sintering and (2) β-TCP can never be in true equilibrium in aqeous solution, since it cannot precipitate like other calcium phosphates. Several routes, however, for its preparation are possible and include: (1) A mixture of an equimolar amount of DCPD and HAp (Ca/P ratio = 1.67) and subsequent calcination; (2) calcination of HAp (Ca/P ratio = 1.50). β-TCP has extensively been used as bone substitute, since it is degradable by osteoclastic activity [45].

#### 3.2.2.2 α – Tricalcium Phosphate

(α-TCP, α-Ca3(PO4)2) has exactly the same chemical composition as β-TCP but posses a different crystallographic structure. This difference makes α-TCP highly soluble in comparison to that of β-TCP. It is obtained by heating β-TCP above temperatures of 1125°C or 1166°C, and quenching it to prevent a reverse transformation [46]. α-TCP is readily transformed into HAp in an aqueous solution. It is biocompatible and more biodegradable than β-TCP.

#### 3.2.2.3 Hydroxyapatite

HAp (Ca5(PO4)3OH) defined here as the high temperature form of a stoichiometric PHAp, is highly crystalline, the most stable CaP in an aqueous solution, and the most biocompatible CaP [37]. At temperatures higher than ≈900°C, partial decomposition of HAp may take place resulting in oxyapatite (OXA). HA, partially dehydrated HA, or OXA decompose above 1300°C into α-tricalcium phosphate (α-TCP) and tetracalcium phosphate (TTCP)

#### 3.2.2.4 Oxyapatite

OXA (Ca10(PO4)6O) is obtained at temperatures higher than ≈900°C. It results from the partial decomposition of HAp. Otherwise, OXA is poorly understood due to the difficulties of detecting its phase [37].

#### 3.2.2.5 Tetracalcium Phosphate

Phase pure TTCP (Ca4(PO4)2O) is directly obtained by a solid state-reaction at high temperatures (~1500°C), through mixing of equimolar quantities of DCPD and CaCO3 [47]. TTCP preparation can be divided into two categories, that is, solid-state reaction and a wet encapsulating deposition. While the latter has advantages of low cost, short duration and simple process, it is often difficult to control the purity and the molar ratio of Ca/P. TTCP product is frequently mixed with impurities such as HAp, CaO and α and/or β-TCP, when the starting materials are heated to the temperature range of 1000ºC - 1400ºC.

It presents the most soluble CaP at a pH less than 5. Furthermore, it is biocompatible but poorly biodegradable. Brown and Chow [48] have indicated that mixing and reaction of TTCP with DCPA in a diluted phosphate-containing solution can lead to the formation of near stoichiometric HAp, when excess TTCP is present.

## Crystal Structures of Calcium Phosphates

Apatites are a structural type for compounds of the general formula M10(XO4)6Y2 rather than specific compounds. In general, they are known to be capable of accommodating a wide variety of modifications and combinations of substitutions of ions and groups within the apatitic lattice. However, the term “apatite” has been extensively and synonymously used to represent the calcium phosphates, Ca10(PO4)6X2, where X = F- OH-, or Cl- and this concept will be discussed further in the following section. Apatites are thermodynamically the most stable phases among the calcium phosphates and, therefore, can be considered as the probable end product in many reactions.

This section considers the crystallographic features of various calcium phosphates of biological interest. The phosphates containing both  and, generally constitute biologically relevant calcium phosphates. The structure of HAp is considered as the prototype for the inorganic component of bones and teeth and hence is discussed with respect to the types and locations of ionic substitutions. Octacalcium phosphate (OCP) is a known precursor in biological mineralization. OCP has a layered type structure, with one layer quite similar to that of HAp and the other, a hydrated layer consisting of a more widely spaced Ca, and PO4 ions and the water molecules. The closeness of fit in the apatetic layers of OCP and HAp accounts for in-situ conversion of OCP to HAp.

### Hydroxyapatite

It is generally accepted that slightly nonstoichiometric HAp has the hexagonal space group P63/m structure, with *a =* 9.421 and *c* = 6.884 nm, while in the case of stoichiometric HAp the structure becomes monoclinic P21/b [49]. This is characterised by ordering within OH ion columns to form a sequence OHOHOHOH, with an ordered arrangement of these columns, so that the *b*-axis is doubled giving lattice parameters *a* = 9.421(8), *b =* 2*a*, *c =* 6.8814(7) nm, γ = 1203 [50]. Normally, only those preparations that have a final high-temperature stage have the possibility of yielding monoclinic HAp. Other preparations are normally hexagonal, presumably because sufficient OH ions are missing, replaced by H2O or impurity ions, so that the ordering is disturbed.

Recent theoretical studies by Calderin et al. [51] however, find the hexagonal and monoclinic structures both energetically acceptable for HAp, but Haverty et al. [52] finds that the P63/m or the P63 hexagonal structural models are energetically unfavorable in comparison with a model based on the P21/b symmetry or the newly proposed monoclinic P21 structural model. They also conclude from Rietveld analysis of X-ray diffraction patterns from a NIST standard reference specimen that HAp (SRM2910) crystallizes in a mixture of 23% P21/b and 77% P21 monoclinic phases [53].



Figure ‑ Crystal structure of hexagonal HAp projected down the *c* –axis [29]

The corners of the unit cell depicted in Figure 3-2 (as identified by shaded circles) are occupied by OH- for HAp and F- for FAp. The unit cell identified as *a'* and *b'* presents an alternative arrangement for the hexagonal HAp structure. Table 3-2 outlines the lattice coordinates for its unit cell.

Table ‑ Unit-cell positions of the HAp lattice [16]

α = 90º, β=90º, γ =120º

|  |  |  |  |
| --- | --- | --- | --- |
| **Atom**  | **x/a**  | **y/b**  | **z/c**  |
| Ca(I)  | 1/3 | 2/3 | 0.0010 |
| Ca(II)  | 0.2464 | 0.9938 | 1/4 |
| P  | 0.3999 | 0.3698 | 1/4 |
| O(I)  | 0.3272 | 0.4837 | 1/4 |
| O(II)  | 0.5899 | 0.4666 | 1/4 |
| O(III)  | 0.3457 | 0.2595 | 0.0736 |
| Hydroxyl | 0.0000 | 0.0000 | 0.1930 |

### Fluorapatite

Fluorapatite (FAp), Ca10(PO4)6F2, is the most stable among the CaPs and possesses a hexagonal crystal structure with a P63/m space group. The lattice parameters for the structure are as follows; *a* = *b* = 9.367(1) Å and *c* = 6.884(1) Å, *Z* = 1. The positions of the two sets of Ca2+ ions and the PO43- ions are nearly identical to those of OHAp. However, the F- ions occupy the center of the CaII triangles (6*h* positions), on the mirror planes at *z* = 1/4 and 3/4.

Figure 3-3 and Table 3-3 outlines the lattice coordinates for its unit cell. There are two non-equivalent, symmetrically and crystallographically different Ca2+ ions in the apatite structure. Out of the ten Ca ions in a unit cell, four Ca ions occupy four-fold (*4f*) sites and the other six Ca ions occupy the six-fold (*6h*) sites. These two types of Ca ion positions have been defined in previous studies as CaI and CaII respectively.The CaI site is the larger of the two sites, and has a trigonal symmetry surrounded by nine polyhedra of oxygen atoms. The six CaII sites are smaller in size and lower in symmetry because they are surrounded by six oxygen atoms and one fluorine atom. [54]



Figure ‑ View onto the (0001) plane of the FAp structure, showing hexagonal symmetry and the relationship between a hexagonal unit cell (pink) and a monoclinic unit cell (blue) (Ca=blue; O=red; P=yellow; F=green) [55]

Table ‑ Unit-cell positions of the FAp lattice [54]

α = 90º, β=90º, γ =120º

|  |  |  |  |
| --- | --- | --- | --- |
| **Atom**  | **x/a**  | **y/b**  | **z/c**  |
| Ca(I)  | 0.6667  | 0.3333  | 0.0010  |
| Ca(II)  | -0.0071  | 0.2423  | 0.2500  |
| P  | 0.3690  | 0.3985  | 0.2500  |
| O(I)  | 0.4849  | 0.3237  | 0.2500  |
| O(II)  | 0.4667  | 0.5875  | 0.2500  |
| O(III)  | 0.2575  | 0.1342  | 0.0705  |
| F  | 0.0000  | 0.0000  | 0.2500  |

### Chlorapatite

Chlorapatite (ClAp), Ca10(PO4)6(Cl)2, has been described in the hexagonal space group *P*63/*m*, with cell parameters, *a* = *b* = 9.598(2) Å, *c* = 6.776(4) Å, *Z* = 1 [29]. It is chemically similar to HAp, in that the Cl- is disordered like the OH- in HAp, and displaced from the midpoint of the CaII triangles, and located at positions 1.2 Å above and below the mirror planes. The Cl- is so far removed from the mirror plane towards the midway point between the two CaII triangles, which results in an additional weak bond developing between the CaII and a second Cl- ion. Stoichiometric ClAp has also been reported to crystallize in the monoclinic space group with space group *P*21/*b* having cell parameters *a* = 9.628(5) Å, *b* = 2*a*, *c* = 6.764(5) Å, γ = 120º, *Z* = 2 [56]. The structure is very similar to the hexagonal one, but the Cl- ions are ordered in two columns on pseudohexagonal axes as in the case of the monoclinic HAp. Table 3-4 outlines the similarity between the crystal structures for HAp, FAp and ClAp

Table ‑ Summary of unit cell Information for HAp, FAp and ClAp

|  |  |  |  |
| --- | --- | --- | --- |
|  | **HAp** | **FAp** | **ClAp** |
| **Formula** | Ca5(PO4)3OH  | Ca5(PO4)3F  | Ca5(PO4)3Cl |
| **Form Wt.** | 502.322  | 504.313  | 520.767 |
| **Density** | 3.153  | 3.201  | 3.185 |
| **Mol Volume** | 159.334  | 157.527  | 163.527 |
| **Z** | 2 | 2 | 2 |
| **Crystal System** | Hexagonal | Hexagonal | Hexagonal |
| **Crystal Class** | 6/*m* | 6/*m* | 6/*m* |
| **Space Group** | *P*63/*m* | *P*63/*m* | *P*63/*m* |
| **Cell Parameters** |  |  |  |
| **a** | 9.424  | 9.367  | 9.628 |
| **c** | 6.879  | 6.884  | 6.764 |
| **Vol** | 529.09  | 523.09  | 543.01 |

### Octacalcium Phosphate

The crystal structure of octacalcium phosphate (OCP), Ca8(HPO4)2(PO4)4.5H2O, was initially determined in 1962 [57]. The crystals are triclinic, space group *P*, with cell parameters *a* = 19.692(4) Å, *b* = 9.523(2) Å, *c* = 6.835(2) Å, α = 90.15(2) º, β = 92.54(2) º, γ = 108.65(1) º and *Z* = 2. The structure of OCP is illustrated in Fig. 3-4. The positions of all atoms in the region *x* = 0 to ≈ 1/4 in OCP corresponds very closely to that of HAp. This portion consists of two Ca2+ and two PO43- groups, corresponding to each triangular set and two Ca positions in one column in apatite, thus accounting for the Ca6 (PO4)4 unit.

****

Figure ‑ Crystal structure of octacalcium phosphate projected down the c -axis. The region with shaded atoms displays similarity to Hap. Hydrogen atoms are omitted for reasons of clarity [29]

### Tricalcium Phosphate (α and β)

α - TCP crystallizes in the monoclinic space group *P*21/*a* with *a* = 12.887(2) Å, *b* = 27.280(4) Å, *c* = 15.219(2) Å, β = 126.20(1) º, *Z* = 24 [58]. The Ca2+ and PO43- ions are packed in two kinds of columns along the *c* -axis, one containing only Ca2+ and the other both Ca2+ and PO43- ions in the ratio 1:2. The crystal structure arrangement for α-TCP illustrated in Figure 3-4, graphically demonstrates its columnar arrangement and distortion from linearity. Oxygen atoms of the PO4 groups have been omitted for purposes pertaining to clarity. The arrangement of these columns conform to a pseudohexagonal form.

β-TCP crystallizes in the rhombohedral space group *R*3*c* with unit cell parameters *a* = 10.439(1) Å, *c* = 37.375(6) Å, *Z* = 21 (hexagonal setting) [59]. The main difference associated between the structures of α- and β-TCP is that there exists, no cation - cation columns in the β phase.

****

Figure ‑ A projection of the structure of α -Ca3(PO4)2 on the (001) plane. Adapted from [29]

### Tetracalcium Phosphate

Tetracalcium phosphate (TTCP), Ca4(PO4)2O, is part of the monoclinic, space group *P*21, with unit cell parameters *a* = 7.023(1) Å, *b* = 11.986(4) Å, *c* = 9.473(2) Å and γ = 90.90(1)º [59]. The Ca2+ and PO43- ions in TTCP are located in four sheets perpendicular to the *b*-axis. Each sheet contains two Ca-PO4 columns and one Ca-Ca column. The arrangement of these columns is similar to those in glaserite where the oxide ions are extra. However, two adjacent sheets in TTCP form a layer that is closely related to that of apatite.



Figure ‑ Schematic illustration of the TTCP structures projected on the (1 0 0) plane [60]

### Amorphous Calcium Phosphate

Amorphous calcium phosphate (ACP) can be based on the approximated molecular formula, Ca3(PO4)2⋅3H2O, as proposed by Eanes [43] and is generally accepted as a TCP. At present, there is no conclusive evidence that ACP is an integral mineral component in hard tissues. However, it plays a special role as a transient phase in biomineralisation.

### Dicalcium Phosphate Dihydrous

Dicalcium phosphate dihydrate (DCPD), CaHPO4.2H2O, occur as the mineral phase of brushite to crystallize in the monoclinic space group *Ia* with unit cell parameters *a* = 5.812(2) Å, *b* = 15.180(3) Å, *c* = 6.239(2) Å and β = 116.42(3) º, *Z* = 4 [61]. The opposite edges of HPO42- ions are linked to Ca2+ ions to form linear chains that are stacked in a zig - zag fashion to form corrugated sheets parallel to the (010) face.

****

Figure ‑ Crystal structure of DCPD is shown, as viewed down the b-axis [29]

The Ca2+ and HPO42- ions are linked together to form linear chains along the plane. The linkages between chains are indicated by dashed lines. The Ca2+ - HPO42- chains are stacked in a zig-zag fashion, forming corrugated sheets parallel to (010). There are two sheets per unit cell, but only one is illustrated here for clarity.

### Dicalcium Phosphate Anhydrous

Dicalcium phosphate anhydrous (DCPA), CaHPO4, crystallizes in the triclinic space group,with *a* = 6.910(1) Å, *b* = 6.627(2) Å, *c* = 6.998(2) Å, α = 96.34(2)º, β = 103.82(2)º and γ = 88.33(2)º, *Z* = 4 [29]. The structures of DCPD and DCPA are closely related with similar Ca-PO4 chains arranged in corrugated sheets.

## Substitutions in Calcium Phosphates

Apatite is capable of accommodating several substituents, while still maintaining its basic structure. Ca2+ can be substituted by various cations, such as, monovalent (Na+, K+) [62], divalent (Mg2+, Sr2+, Ba2+, Pb2+) [63] and trivalent (Y3+) cations [63]. Significant anionic substitutions, include the replacement of OH by CO32-, F- , Cl-, and PO43- by CO32- [65, 66], AsO43- and VO43- [67]. Some substitutions are coupled with others to maintain the charge balance in the apatite, such as, CO32- for PO43- coupled with Na+ for Ca2+. The trivalent anionic phosphate sites cannot accept vacancies, as the trivalent anions are quite large and vacancies are believed to destabilise the lattice [68]. Adversely, the cationic sites can accept vacancies, up to a maximum of 2 sites out of the 10 existing in stoichiometric apatites [69]. The possible substitutions for each sublattice are indicated in Table 3-5.

Table ‑ Summary of possible apatite structures

|  |
| --- |
|  |
| M = | Ca, Sr, Ba, Cd, Pb, Mg, Na, K, H, D, …  |
| Z = | P, CO3, V, As, S, Si, Ge, Cr, B, … |
| X =  | OH, OD, CO3, O, BO2, F, Cl, Br, vacancies |

Due to its interesting chemical and physical properties, synthetic apatites find numerous medical applications. In particular, what concerns the applicability of the apatite structure within the biomedical field is the capacity of its lattice to act as a host for different chemical species. It is known that the bone regeneration rate depends on several factors, such as; porosity; composition; solubility and the presence of certain elements that, when released during the resorption of the ceramic material, facilitate the bone regeneration carried out by the osteoblasts. Therefore, small amounts of strontium, zinc or silicates are believed to stimulate the action of these osteoblasts and, in consequence, the new bone formation. Carbonate and strontium favour the dissolution, and therefore the resorption of the implant. Silicates, however are found to increase the mechanical strength, a very important factor in particular for porous ceramics, and also accelerate the bioactivity of apatite [70].

In recent years, substitution of magnesium (Mg) in the apatite structure has received much attention due to its impending role on bone metabolism, reducing cardiovascular disease, promoting catalytic reaction and controlling biological functions [71].

# Hydroxyapatite (HAp)

## Introduction

HAp has a similar chemical and phase composition to living natural bone. The bioactive behaviour of this ceramic has proved to accelerate the integration of a prosthesis *in vivo*. The first x-ray diffraction study of bone was published by De Jong in 1926, in which apatite was identified as the only recognizable mineral phase [72]. He also reported marked broadening of the diffraction lines of bone apatite, which he attributed to small crystal size. It was not until the 1970’s that synthetic HAp was accepted as a potential biomaterial that forms a strong chemical bond with bone *in vivo*, while remaining stable, under the harsh conditions encountered in the physiologic environment [73].

Stoichiometric HAp is biocompatible, osteoconductive, nontoxic, noninflammatory, nonimmunogenic agent and bioactive (that is, it possesses the ability to form a direct chemical bond with living tissues) [74, 75].

## Physico-chemical properties of HAp

### Mechanical Properties of HAp

HAp has been clinically applied in many areas of dentistry and orthopaedics due to its excellent osteoconductive and bioactive properties, which is due to its chemical similarity with the mineral portion of hard tissues [75 - 77].

Poor mechanical properties (in particular, fatigue properties) relating to HAp powders, however, mean that HAp cannot be used in bulk form for load bearing applications such as orthopaedics. Instead, coatings of HAp have good potential as they can exploit the biocompatible and bone bonding properties of the apatitic bioceramic, while utilising the mechanical properties of substrates such as Ti6Al4V and other biocompatible alloys.

One of the major concern with HAp is the low fracture toughness (that is, <1MPam1/2 of the sintered body [78, 79]. As a result various studies have been carried out to improve the mechanical properties of sintered HAp [80 - 82]. These include the development of new powder processing/synthesis techniques and composition modification. The ultimate goal is to identify the most appropriate synthesis method and conditions to produce well-defined particle morphology [78 - 83] that is sinter active at low temperatures to produce HAp that exhibits enhanced mechanical properties. The mechanical properties for HAp are shown in Table 4-1.Ultimately, the final mechanical properties obtained are dependent upon the synthesis route taken.

Table 4‑1: Mechanical Properties for HAp powder [16, 79, 84]

|  |  |
| --- | --- |
| **Mechanical Properties** | **Metric Units** |
| Ultimate Tensile Strength (UTS) | 38 - 48 MPa |
| Modulus of Elasticity | 7 - 13 GPa |
| Flexural Strength | 100 - 120 MPa |
| Compressive Strength | 350 - 450 MPa |
| Fracture Toughness, KIC | <1.0 Mpa.m1/2 |
| Poisson's Ratio | 0.27 |

### Chemical Composition of HAp

The constituent elements within synthetic HAp are given in Table 4-2. Biological apatite differs from that of pure Hap in stoichiometry, composition, crystallinity and mechanical properties. Biological apatites are usually calcium deficient and are always carbonate substituted. It is therefore more appropriate that biological apatites are referred to as carbonate apatite and not as stoichiometric HAp. A disadvantage of carbonate substitution in synthetic HAp, so as to replicate biological apatite, however, is the capacity to increase its solubility and slow down the transformation process from ACP > HAp.

 Table ‑: Composition of synthetic HAp (Adapted from [16, 21])

|  |  |  |
| --- | --- | --- |
| **Mineral Component** | **Chemical Formula** | **HAp** |
| **Constituents (wt. %)** |  |  |
| Calcium | *Ca2+* | 39.6 |
| Phosphorous | *P* | 18.5 |
| Ca/P molar ratio |  | 1.67 |
| Carbonate | *CO32-* | - |
| Sodium  | *Na+* | tr |
| Magnesium | *Mg2+* | tr |
| Potassium | *K+* | tr |
| Chloride | *Cl-* | - |
| Fluoride | *F-* | - |
| **Total Inorganic (mineral)** |  | **100.0** |
| **Total Organic** |  | **-** |
| **Absorbed H2O** |  | **-** |
| **Trace elements** | ***Sr2+, Pb2+, Zn2+, Cu2+, Fe3+*** |  |

### Crystal Structure of HAp

As previously discussed in Section 3.3.1, It is widely accepted that slightly nonstoichiometric HAp is part of the hexagonal space group P63/m structure, with *a =* 9.421 and *c* = 6.884 nm, while in the case of stoichiometric HAp the structure becomes monoclinic P21/b [49]. Much research has also led to the proposal of other crystallographic structures for HAp [51, 52].

### Impurity Contents of HAp

An important consideration in the production and processing of HAp is been the control and reduction in resultant impurity levels. These impurities may be classified as either substitutional or additional phases.

The most common example is the preparation of CO3-substituted HAp and fluoride-substituted HAp [65]. Synthetic carbonated HAp (CHAp), CO3, has been classified as either type A or B depending on the mode of carbonate substitution: CO32- for OH- (type A) or CO32- for PO43- (type B). Biological apatites are principally type B [22]. In synthetic powders prepared by wet methods some fractions of PO43- as well as OH- groups are replaced by CO32- groups (type AB). Among the variety of HAp-based bioceramics, CHAp seems to be a more promising material for bioresorbable bone substitution rather than bioactive coatings in orthopaedic application. Sintering time, temperature and the atmosphere are important parameters to control the level and type of carbonate substitution. The presence of CO32- in HAp structure influences the decomposition, sinterability, solubility and biological reactivity of CHAp implantation materials [22]. The presence of CO32- in the HAp structure influences decomposition, sinterability, solubility and biological reactivity of implantation material. Slosarczyk. et al. [22] observed that the decomposition of HAp synthesised without any CO32- additive did not occur. CO32- ions, hence, are found to lower the thermal stability of HAp powders and produces CaO as a secondary phase, whose amount grows as treatment temperatures are increased to 1250ºC. CO32- replacing PO43- increase the Ca/P ratio of 1.667, resulting in non-stoichiometric solutions which are thermally less stable that those containing no substitutions. These results indicate that although carbonate is found in natural bone, it is an undesirable impurity in synthetic HAp.

Magnesium content in raw materials used as precipitation precursors has also been found to have an adverse affect on final HAp powder properties achieved [71]. The presence of magnesium can alter the conditions of calcium phosphate precipitation and more precisely of HAp precipitation: it delays nucleation, growth and HAp maturation. It is also found to lead to the production of non-stoichiometric HAp decomposing upon subsequent calcinations [71].

Foreign elements obtained in HAp in the range of parts per million will not alter the overall biocompatibility response of the material. Impurity levels found in HAp powders are shown in Table 4-3.

Table 4‑3 Impurity content in HAp. Adapted from [16]

|  |  |
| --- | --- |
| **Trace element** | **HAp powder** |
| **(ppm)** | **Laboratory (ppm)** | **Commercial (ppm)** |
| Al | - | 100 - 1000 |
| Cu | 8 | 1 - 10 |
| Fe | 30 - 40 | 100 - 1000 |
| Ge | - | 10 - 100 |
| Mg | 50 | 1000 - 2000 |
| Mn | 3 - 8 | 100 - 1000 |
| Na | 400 | 1000 - 3000 |
| Pb | 7 | 1 - 10 |
| Si | 200 | 100 - 1000 |
| Ti | - | 30 - 1000 |
| Zn | 20 | 10 - 100 |

Another potential method for improving the biological activity of HAp is the incorporation of silicon (or silicate groups) into its lattice. Several methods for the synthesis of silicon-substituted HAp’s have been reported. The hydrothermal method was implemented by Tanizawa et al. [85] in order to obtain materials with a Ca/(P+Si) ratio higher than that of pure HAp. Boyer et al. [86] examined the synthesis of silicon-substituted HAp through the solid-state reaction. However, this study also involved the incorporation of a secondary ion, such as La3+ or SO42-. Gibson et al. [87] synthesised silicon-containing HAp using the wet method, and its in vitro bioactivity studies demonstrated favourable results. A similar method was also utilized in work conducted by Marques et al. [88] synthesized by wet method; HAp with silicon content up to 0.15 wt%, obtaining stable materials at 1300ºC and noting that the unit cell volume and the a parameter length of the HAp decreased as the silicon content increased. It is not clearly known whether the silicon present in the material substitutes completely the phosphorus in the HAp structure, or whether the replacement is partial, or even if in any of the described synthesis the silicon species remain as an independent phase.

Sr and CO3 co-substituted HAp nanopowders have been synthesised by neutralization in a recent research study carried out by Landi et al. [89] for medical applications, such as, a resorbable bone filler or bone substitute scaffold.

## Techniques for HAp Powder Synthesis

The various methodologies employed to produce synthetic HAp are discussed in this section to provide the necessary background information with respect to HAp powder synthesis techniques, used to date. It is important to note that each method is greatly dependent upon the precursor materials. Bernard et al. [90] has investigated this, by examining the effect of the purity of the starter raw materials on the resultant purity of the HAp product. It was established that the stoichiometry (that is, optimum concentration) of the HAp precipitate and its purity post thermal treatment, depends on the quality of the raw materials used. The production of ceramic HAp powders may be classified under four main headings:

1. Wet Chemical Synthesis (such as, Precipitation, Hydrothermal, Hydrolysis and Sol-Gel Techniques)
2. Dry Chemical Synthesis (such as, solid-state reactions, mechanochemical synthesis)
3. Vapour Phase Reactions (such as, spray and freeze-drying)
4. Novel Techniques

The advantage of the wet process is that the by-product consists primarily of water and the probability of contamination during processing is very low. Low processing costs has also been reported. Its disadvantage is that the resulting product can be greatly affected by even a slight difference in the reaction conditions. The dry process, on the other hand, demonstrates high reproducibility in spite of a high risk of contamination during stages of milling and long heat treatment times [20, 31, 91]. An overview of these reaction routes are discussed, in the following sections.

### Precipitation Route

In the precipitation route, HAp powders are obtained from a chemical reaction of inorganic oxide solutions [20]. The two most popular ways in precipitating HAp are described in the two reactions below [20, 92, 93]:

#### Reaction 1

The precipitation method for the reaction of orthophosphoric acid with calcium hydroxide:

  Equation 1

#### Reaction 2

The precipitation method for the reaction of diammonium hydrogen phosphate with calcium nitrate:



Equation 2

Reaction 1 is simple, cheap, suitable for large scale industrial production and non-polluting by nature, but often leads to the production of non-stoichiometric HAp, which creates subsequent problems during sintering (thermal decomposition of non-stoichiometric HAp leads to TCP when the Ca/P molar ratio < 1.67), or to CaO when the Ca/P molar ratio > 1.67. In contrast, Reaction 2 is expensive to produce and polluting by nature, in that it’s by products must be removed. The quality of the initial reagents also play an important role in obtained high quality phase pure HAp. Synthesis, based on Reaction 1 has demonstrated that the purity and the properties of the chemicals and process parameters, such as, reagent addition flow rate, stirring rate, pH and reaction temperature all have a significant influence on the final quality of the HAp in relation to its crystallinity, crystallite size, morphology, particle size distribution, density and surface area. Another study has shown the effect of ripening time on crystallinity and sinterability and mechanical properties for dense HAp can be related to the initial reagent properties.

HAp synthesised by work conducted by Kweh et al. [92] using Reaction 2, have found that the reaction temperature, the reactant concentrations, rate of mixing the reactants and the residence time can affect the overall characteristics of the HA produced. The research also found that other phases can be produced with different reaction pH values.

HAp with a Ca/P molar ratio, ranging from 1.5 to 1.667 (0 ≤ x ≤ 1) using the conventional aqueous precipitation method from the addition of a diammonium phosphate solution ((NH4)2HPO4) into a reactor containing a calcium nitrate solution was prepared by Raynaud et al. [94]. The device implemented was fully automated with the reactor placed in an argon atmosphere under dynamic flow in order to prevent any presence of CO2, which could result in carbonate apatite formation. In this study, pH and temperature of synthesis were the preponderant parameters for the control of the precipitate composition.

A preparation method used to a lesser extent; to obtain HAp has been patented by Barsa, et al. [95]. It involved a reaction of anhydrous trisodium phosphate with calcium chloride in a formamide/water, (HCONH2/H2O), solution. The calcium phosphates at 37°C were precipitated from solutions mixing calcium chloride with sodium chloride and anhydrous trisodium phosphate with sodium hydroxide. The research also indicated that brushite was formed at a pH < 6.5, while amorphous calcium phosphate (ACP) was formed at a pH > 8.0. HAp was not detected in the first precipitate, but occurred after a few days.

Overall, precipitation techniques can produce small particle size and high-purity HAp powders, when the precipitation system and conditions are well controlled to obtain a reproducible precipitate. The important precipitation process variables include solution concentration (Ca/P ratio), pH, acid addition rate, stirrer speed, temperature, reaction time and atmospheric condition. [92]

The overall production routes for both precipitation reactions are outlined in Figure 4-1.

 

Figure 4‑1 A flow chart for the synthesis of HAp powders via the precipitation route for Reaction 1 (left hand side) and Reaction 2 (right hand side)

In an investigation carried out by Xu et al. [96], nano-sized HAp powder particles were synthesised using the precipitation approach of method (2). This was achieved by reacting 0.6 mol of orthophosphoric acid (H3PO4) with 1 mol of calcium hydroxide (Ca(OH)2). The precipitation reactants were carried out at 40 ±5 °C in house and terminated when the pH reached 9 through the addition of H3PO4. After the complete mixing of the reactants, the precipitate was stirred for two more hours and left overnight to settle. The so-formed HAp suspension was then transported into a spray dryer, where the atomization and drying processes took place. The spray-dried HAp powders were collected and kept in an oven at 60 °C for 12 h to remove any absorbed moisture. The spray-dried HAp powders were then sieved and particles with a size less than 20 μm were heat-treated at 1000 °C for 5 h in order to increase the thermal stability of the HAp feedstock. A similar approach was taken by Kumar et al. [97] who also produced HAp powders using the precipitation reaction of method (2) by reacting orthophosphoric acid (H3PO4)with calcium hydroxide. H3PO4 was added drop-wise at a rate of 1.5L/h into a bath of calcium hydroxide maintained at 40°C until reaching a pH of about 8. The suspension was stirred for 2h and left to settle overnight. Centrifugation was used, as opposed to the spray drying method employed by Xu et al. [96]. Polymetametylcrylate (PMMA) was added as a deflocculant to reduce the HAp slurry viscosity.

Fluoride-substituted apatites were prepared by Jha et al. [98] by performing reactions in aqueous medium using a precipitation reaction between diammonium orthophosphate and calcium nitrate 4-hydrate and ammonium fluoride. This research suggested that the incorporation of the fluoride ions into the apatite structure increased the crystallinity and the Ca/P ratio.

Overall, precipitation techniques can produce small particle size and high-purity HAp powders, when the precipitation system and conditions are well controlled to obtain a reproducible precipitate. The important precipitation process variables include the initial calcium concentration, acid addition rate, stirrer speed, reaction temperature and control of the atmospheric environment. [16, 92]

### Hydrothermal Route

The hydrothermal route employs solutions of various calcium and phosphoric compounds, reacted hydrothermally, at moderate temperatures to form HAp, such as [20]:

 Equation 3

 Equation 4

Pure HAp is produced by reaction of a Ca(OH)2 suspension, at 95ºC under stirring, with H3PO4 at a pH = 10.6 +/-1, and ripening under reflux. The phase composition remained unchanged at 900ºC for 2h. Needle like, 58nm length HAp was synthesised using aqueous solutions of (CH3COO)2Ca and K2HPO4, under stirring, heating to 100ºC reflux, ageing, filtering and then freeze-drying. Also, needle like high quality 150nm length (surface area; 31 – 43 m2/g) HAp has been economically synthesised by ball-milling mixing Ca(OH)2 and Ca(H2PO4)2.H2O in ethanol for 1h, drying, and heating at 109ºC with water, under pressure for 1 – 3h [20]. Specimens after sintering at 1100 - 1300ºC (without decomposition) yielded mechanical properties of δf = 120MPa at 3-point bending, Hv = 5.1GPa and KIC = 1.2 MPa.m1/2. The hydrothermal route has also been used in the production of highly crystalline and homogeneous HAp via a transformation of coral aragonite (CaCO3) based on the following reaction [99]:



Equation 5

A combination of the precipitation route (reaction 2: equation 2) and the hydrothermal route is also reported, producing rod-like nanosized HAp in the prescenc of polyacrylic acid. In this study [100], it was found that both routes facilitated the stabilization of HAp synthesis.

Young et al. [101] synthesised HAp using the solid-decomposition reaction to produce their precursor starting materials. The mixed oxides were reacted in distilled water at 1000°C by the hydrothermal method. The reactions were as follows:

**Solid-decomposition reaction(s):**

  Equation 6

  Equation 7

**Hydrothermal reaction:**

** Equation 8

Liu et al. [20] synthesised HAp by the hydrothermal method using Ca(OH)2 and CaHPO4.2H2O as follows:

 Equation 9

The initial reaction substances of Ca(OH)2 and CaHPO4 . 2H2O were dissolved in de-ionised water. The study investigated the influences of pH value and treating temperature on the final HAp product morphology [20]. Results indicated that the pH value and hydrothermal temperature were critical for obtaining desirable HAp morphology. Pure HAp phase powders were obtained at temperatures above 120°C and pH 9 without any traces of monetite phases, which were evident at lower temperatures. According to Liu et al. [20], the presence of monetite resulted at a temperature of 140°C if the pH value of the initial solution was not adjusted. Higher pH values (14) produced uniform spherical agglomerates while a lower pH (6) resulting in needle like shaped morphologies.

The most popular hydrothermal technique involves the reaction of calcium monohydrogen phosphate, previously prepared via the precipitation reaction (will be discussed later) between calcium phosphate and orthophosphoric acid, and distilled water in a platinum-lined hydrothermal bomb [16]. HAp produced using hydrothermal synthesis can have high crystallinity because the reaction is performed at high temperature (300°C). However, small amounts of HAp can be obtained from each hydrolysis due to the limited capacity of the platinum-lined hydrothermal bomb. According to Puajindanetr [16], the hydrothermal bomb can crack after several cycles, and iron and chromium ions from the steel bomb may be introduced into the water. Thus, the HAp may contain whitlockite (β-TCP). Control of particle shape and powder size is possible in varying these conditions, such as, temperature and pressure. These may appear as differences in nucleation and particle growth [16].

The hydrothermal technique usually gives HAp with a high degree of crystallinity with a Ca/P ratio close to the stoichiometric value. Their crystal size can be in the range of nanometers to millimetres [20]. Hydrothermal methods using elevated temperature and pressure aqueous solutions allow the synthesis of HAp crystals with a certain shape. Its main disadvantages are the fact that the procedure itself can be quite complicated and the treating temperatures can be comparatively high compared with other alternative synthesis methods [20].

### Hydroylsis Route

HAp is formed from the hydroylysis of various acid calcium phosphate salts (such as, CaHPO4.2H2O, Ca8H2(PO4)6.5H2O or CaCO3) in appropriate environmental conditions. HAp has been synthesised from hydrolysed OCP, with TTCP or Ca(OH)2, according to the following reactions [99]:

  Equation 10

  Equation 11

Young et al. [101] synthesised HAp by refluxing dihydrated calcium hydrogen phosphate in distilled water for one month by the following reaction:

  Equation 12

A similar reaction has been carried out with regards the hydrolysis of calcium monohydrogen phosphate to produce well-crystallised HAp, which was subsequently reacted in a platinum-lined hydrothermal bomb for several days at 300°C [16]. The major disadvantages associated with this technique were (i) the small quantity of HAp production attainable; due to the limitation capacity of the hydrothermal bomb and (ii) impurities of the prepared HAp product due to the contamination of metal from the hydrothermal bomb. Puajindanetr et al. [16] identified two possible hydrolysis techniques using various starting raw material as follows:

1. **Calcium Monohydrogen Phosphate**

The reaction of calcium monohydrogen phosphate (CaHPO4) was prepared using a precipitation reaction between calcium phosphate and orthophosphoric acid, and distilled water in a platinum lined hydrothermal bomb at 300°C to obtain HAp.

1. **Calcium Oxide and Calcium Pyrophosphate**

The first stage of this preparation was a solid-state reaction between calcium oxide (CaO), which was prepared from calcium carbonate (CaCO3), and calcium pyrophosphate (Ca2P2O7), which was subsequently prepared from CaHPO4. The reaction was performed at 1000°C. The product was then ground and loaded into a platinum-lined hydrothermal bomb with distilled water and heated for one month at 500°C and at 103.425 Bar / 10342.20 KPa.

Yamaguchi et al. [102] obtained HAp from brushite, CaHPO4,during their experimentalanalyses. They reported that the product was formed by the structural change of brushite into HAp and subsequent change in its Ca/P ratio. It was also noted that the HAp powder had comparatively low crystallinity, similar to precipitated HAp. Yoon et al. [103] synthesised HAp through the hydrolysis of α-TCP as the precursor.

This method generally produces large single crystals, but is unable to prepare pure HAp phase, producing HAp powders of which are highly non-stoichiometric, with a Ca/P ratio in the range of 1.50 – 1.71. These factors, coupled with its lengthy reaction time, of over a month period, make this an unsuitable approach for use on an industrial scale. The hydrolysis method is ideal in producing small particle sizes, resulting in HAp needles or blades of micron size. The main problem, however, associated with any of these methods as described above is the presence of carbonate ions and/or other impurities of which may be incorporated into the lattice of the crystallised HAp product.

### Sol-Gel Route

Sol-gel processing has attracted much interest in the preparation of special powders and for forming thin coatings and cast or extruded shapes. In sol-gel processing, colloids or molecules in a suspension (that is, a sol) are mixed with a liquid, causing them to join together to form a continuous network (called a gel) [16, 104]. However, polymerisation greatly restricts chemical diffusion and segregation. The gel is dried, calcined and milled to form a powder.

Sol-gel synthesis from ethylene glycol solution of Ca(OAC)2.xH2O and P2O5 butanol solution, produces after sintering at 500ºC, poor and 1000ºC, well crystallised non stoichiometric HAp. Another sol-gel route involves the reaction of P-alkoxide and Ca-alkoxide solutions, which shows ageing as a critical factor in eliminating the formation of CaO at room temperature [104]. HAp powders have also been prepared from a precursor sol formed by mixing a hydrated solution of N-butyl acid phosphate (Ca(NO3)2.4H2O dissolved in 2-methoxyethanol, followed by drying at 175ºC. Figure 4-2 outlines the complete process using the sol-gel reaction route in the preparation of HAp.

Kuriakose, et al. [105] developed a relatively simple method to synthesize pure, stable, stoichiometric nano-crystalline HAp at low temperature by the sol–gel route. Equimolar solutions of Ca(NO3)24H2O and (NH4)2HPO4 dissolved in ethanol were used in the synthesis at 85°C. The product was sintered at various temperatures, such as 400°C, 750°C and 1200°C. The synthesised HAp was found to be stable up to 1200°C without any by-products. In this investigation, ethanol was used as a solvent instead of phosphorous alkoxides and gels, under alkaline conditions to obtain stoichiometric, nanocrystalline HAp. The resultant sol–gel was continuously stirred at a constant pH of 10 (pH was kept constant by adding Ca(OH)2 solution) and at a constant temperature of 85°C for 4 h. After allowing the product to cool, it was kept inside the oven at 40°C overnight. The product was sintered for 2 h at 400°C, again at 750°C and 1200°C.



Figure 4‑2: A flow chart for the sol-gel synthesis of HAp powder

The presence of alcohol seems to provide a thermally stable HAp. The pores in the crystal planes itself helped the material to attain more biocompatibility and enabled the circulation of physiological fluids [105]. Weng et al. [106] developed a synthesis route, using the sol-gel technique for the preparation of HAp powders with its intended application for coatings in the orthopaedic industry. The precursors used to obtain the HAp were an ethylene glycol solution of Ca(OAC)2 . H2O and a butanol solution of P2O5. A stable mixed solution of the two precursors was obtained by adding acetic acid in the HOAC/Ca ratio of 2. The stable mixed solution was poured into a hot stainless steel plate (~200°C) to evaporate the solvent and obtain powders.

The sol–gel route has more advantages than the others, such as high product purity, homogeneous composition and comparatively low synthesis temperature. Reports to date on the sol–gel derived HAp indicate the synthesis of HAp is always accompanied by a secondary phase of CaO [105].

**Analytical comparison of wet chemical methods**

On close inspection of Table 4-4, the precipitation method out weighs, in terms of the advantages it offers in the preparation of HAp powders, in comparison to the hydrothermal or hydrolysis method. The most useful, commercially, appears to be the chemical precipitation method, since it has the capability in producing small particle sizes of a high-purity, resulting in the only by-product of water. Consequently, the prospect of contamination is also eliminated, yielding precipitation as the best option for industrial production with regards cost and production time. In order to achieve HAp precipitates of small particle size and high-purity, the precipitation system and conditions need to be well controlled to obtain reproducibility. The important precipitation process variables include the initial calcium concentration, acid addition rate, stirrer speed, reaction temperature and control of the atmospheric environment. [16, 92]

Table ‑: Analytical overview of the wet chemical methods

|  |  |  |
| --- | --- | --- |
| **METHOD** | **ADVANTAGES** | **DISADVANTAGES** |
| Precipitation: Reaction (1) | 1. Small particle size
2. Good compositional control
3. Ease of production
4. Economic
5. Easily reproducible (through control of the critical precipitation parameters
 | * 1. Precipitated HAp powders are affected by the purity of the calcium nitrate
	2. Requires extensive washing to eliminate the ammonium hydroxide used in its reaction
	3. Low crystallinity (increased with subsequent sintering)

4. Susceptible to agglomeration |
| Precipitation: Reaction (2) | 1. High purity
2. Small particle size
3. Can produce homogenous powders
4. Good compositional control
5. Produces HAp with good sinterability
6. Only by-product is water
7. Ease of production
8. Economic
9. Easily reproducible (through control of the critical precipitation parameters
 | 1. Susceptible to agglomeration
2. Low crystallinity (with increased with subsequent sintering)
 |
| Hydrothermal | 1. Produces highly crystalline HAp
2. Produces HAp powders with a Ca/P ratio close to stoichiometric ratio, 1.667
3. Morphology and particle size of HAp powder can be controlled by adjusting the pH and Temperature of the reaction
 | 1. Risk of contamination is high as the hydrothermal bomb can crack (cyclic fatigue) and release chromium ions into the H2O
2. High heat treating temperatures are required (100 – 1,000ºC)
3. Complicated and lengthy process (1 month)
4. Small amounts of HAp are produced per batch. Unsuitable for production of large quantities of HAp at an industrial scale
 |
| Hydrolysis | 1. Capable of producing small particle sizes of HAp powder.
2. Ideal for producing HAp with needle-like/blade morphology
 | 1. Low crystallinity HAp powders produced compared to precipitation methods (1) and (2)
2. Unable to produce phase-pure HAp. Non-stoichiometric calcium phosphates are produced

3. Complicated and lengthy procedure  |

### Solid-State Reaction Route

Solid state reaction involve solid chemicals, such as monetite (CaHPO4) or Ca3(PO4)2 to react with Ca(OH)2 at a high temperature (>900ºC) to produce HAp, according to the following chemical reactions, which occur [50]:

 Equation 13

 Equation 14

Ceramic tapes have been produced by reacting DCP and calcite at high temperatures (~800ºC) by the following reaction [107]:

 Equation 15

A modification of this reaction has been utilised to produce monoclinic HAp via a carbonate apatite formation, by heating an aqueous slurry of CaCO3 and CaHPO4, under vigorous stirring conditions, with the removal of carbonate by heating, in a vacuum at 1100ºC followed by steaming at 900ºC to ensure complete hydroxylation [107].

Mechanochemical powder synthesis is a solid-state synthesis method that takes advantage of the perturbation of surface-bonded species by pressure to enhance thermodynamic and kinetic reactions between solids. Researchers, like Young et al. [101] mixed calcium carbonate (CaCO3) and calcium monohydrogen phosphate (CaHPO4) and ground the product in an acetone slurry within an agate ball mill. After evaporation of the acetone, the mixture was pressed and placed in platinum crucibles and then slowly heated from room temperature to 1200°C for 10 h in a stream of Potassium Oxide (P2O5) and dried Nitrogen (N2). The product was then reground, pressed and fired again at 1200°C under the stream for at least 8 h. This cycle was repeated three times. In the last cycle, the temperature was kept constant at 900°C for 4 h. Finally, the product was slowly quenched in a water vapour atmosphere to 400°C, cooled in air, ground and homogenized in an agate ball mill. Young et al. [101] found Ca(HO)2 as a contaminant (1 wt. %) in HAp. It was suggested that the reason for this was either a non-stoichiometric ratio of calcium and phosphorous in the first stages of processing, incomplete solid-state reaction or perhaps a combination of both of these factors. Other researchers, however, have varied this process slightly to achieve HAp products of desirable characteristics [91, 94, 108].

Solid-state reactions usually give a stoichiometric and well-crystallised product. The main advantages are simplicity and low cost since conventional milling equipment can be used [108]. Its disadvantages are that the particle size distribution is critically dependent on the milling (grinding) of the calcined HAp product prior to sintering [91].The sinterability of such powders can be low [28, 91, 101]. Also, the contact of the HAp powder with the agate ball mills indicate possible contamination of the end product [16].

Young et al. [101] employed the following method (otherwise known as, the solid-decomposition reaction method) in order to prepare calcium oxide and calcium pyrophosphate as the starting materials to produce HAp via the solid-state reaction. The solid-decomposition reaction involves the thermal decomposition of a complex compound to obtain homogeneous powder and involves the following two initial chemical reactions in order to produce the reactants required:

1. **Calcium oxide preparation:**

  Equation 17

1. **Calcium pyrophospahte preparation:**

  Equation 18

This method has the capability of providing homogeneous, high purity products. However, the process is chemically restricted to certain compositions and is therefore difficult to obtain a precise composition. Table 4-5 outlines the advantages and disadvantages associated with this technique.

Table ‑: Analytical overview of the dry chemical methods

|  |  |  |
| --- | --- | --- |
| **METHOD** | **ADVANTAGES** | **DISADVANTAGES** |
| Solid-state reaction | 1. Successful reports of phase-pure HAp has been reported
 | * 1. Complicated procedure
	2. Reaction temperatures can reach as high as 1390ºC
	3. Long heat treatment times are required.
	4. Sinterability of resultant powders is low
	5. Risk of contamination is high
	6. Milling the product is required prior to sintering.
 |
| Solid-decomposition reaction | * + 1. Homogenous HAp easily attainable
		2. High purity HAp can be produced
 | 1. Restricted to certain compositions and is difficult to obtain a controlled composition |

### Spray Drying Reaction

Dispersion of a solution, containing ions of interest into microscopic volumes and then the removal of the solvent as a vapour forming a salt (that is, the HAp); is an alternative powder production route commonly classified as spray pyrolysis/drying. Atomic scale homogeneity is possible for multi-component systems when the components are of equal solubility or when the salt forms extremely rapidly [109].

Chow et al. [110], prepared nano-sized HAp powders via a spray drying method in which the HAp product was not exposed to any liquid after its formation. The spray drying apparatus consisted of a nozzle that sprayed an acidic calcium phosphate solution in the form of a fine mist into a stream of filtered air flowing through a heated glass column (see Figure 4-3). The water and volatile acid were evaporated by the time the mist reached the end of the column, and the fine particles were collected by an electrostatic precipitator. An important feature of this process is that evaporation of the liquid would lead to in-situ precipitation of HAp that is essentially free of undesired components or impurities. This process required that the solution being sprayed contained only calcium and phosphate ions and an acid component required to solubilize the CaP compound. Fortunately, precipitation of HAp, resulting from evaporation of water in the spray drying process, caused a decrease in solution pH.



Figure ‑: Schematic of spray dry apparatus for direct HAp powder synthesis

This method can produce HAp particles in the range of 10 - 20μm in diameter. Segregation can be reduced as a result of the fast drying that occurs in a matter of milliseconds. Porous calcined agglomerates have been reported to be easily comminuted into the component particles.

### Freeze Drying Reaction

The freeze-drying technique is a process in which droplets formed by spraying a solution with the desired amounts of components are immersed into liquid nitrogen. The frozen materials are collected from the liquid nitrogen and the water ice is sublimed from the frozen materials under reduced pressures. Finally, the freeze-dried materials are heated to form the desired chemical compound [16, 111]. The resulting powder has the advantages of molecular scale homogeneity, due to the flash freezing in the cryogen, and minimal agglomeration, due to the sublimation of the water ice in the freeze-drying process [111]. A theoretical development concerning the freeze-drying technique has been reported recently by McGrath and Laine [112]. They predicted that aqueous droplets may be frozen homogeneously even at ~200 K (273°C) when the droplet diameters are reduced to ~1 mm. On the basis of this theory, the present authors assembled a novel ultrasonic spray freeze drying (USFD) apparatus in which droplets with ~2 mm diameters could be freeze-dried without immersing them in a cryogen [112]. The schematic diagram of the reaction modified reaction chamber used in work carried out by Itatani et al. [111] to prepare various calcium phosphates is shown in Figure 4-4. Calcination of the homogeneous salt yields porous aggregates, which are milled to a micron-size powder with good sinterability.

The four steps in the freeze-drying process are as follows [16]:

1. A mixture of soluble salts containing the desired ratio of metal ions is dissolved in distilled water.
2. The solution is formed into droplets usually 0.1 to 0.5mm in diameter and rapidly frozen so that no compositional segregation can occur and so that the ice crystals that nucleate are very small
3. The water is removed in vacuum by a sublimation process with care to avoid any liquid phase, thus preventing any chance for segregation
4. The resulting powder is then calcined (heat-treated) at a temperature that decomposes the crystallised salts and converts them to very fine crystallites of the desired oxide or compound.



Figure ‑: Overall view of the freeze-drying chamber

In a laboratory scale, the method can be carried out in a suspension solution frozen at liquid nitrogen temperature (-196°C) whilst rotating in a flask. It is then freeze dried under vacuum, usually for 24 hours, to obtain the powder [16].

In summary, vapour phase reaction techniques produce submicron-size, well-dispersed particles entrained within large volumes of gas. The vapour phase method is capable of controlling its particles size based on the reaction conditions, such as temperature, rate and concentration of reaction. This technique, however, has the added complication that a complex collection powder system is required in order to remove the product, such as those used to carry out a solvent evaporation process [16]. An analytical overview of both the vapour phase reactions is presented in Table 4-6.

Table ‑: Analytical overview of the vapour phase reactions

|  |  |  |
| --- | --- | --- |
| **METHOD** | **ADVANTAGES** | **DISADVANTAGES** |
| Gas – solid reaction; | 1. Inexpensive
 | 1. Low purity HAp powders obtained
 |
| Gas – liquid reaction;Gas – Gas reaction | 1. High purity powder producible
2. Small particle size attainable
 | * 1. Expensive to operate
 |

### Novel Routes for Synthesising HAp

#### 4.3.6.1 Emulsion-Microemulsion Routes

Microemulsion is a novel route which has the capacity to produce high purity HAp, in the nanometer range, with a narrow particle size distribution and degree of particle agglomeration. Research [113] on HAp powders produced by the reaction of CaCl2 and (NH4)2HPO4 in bi-continuous and inverse microemulsion and emulsion, with cycloexane, non-ionic surfactant and aqueous solution, after sintering at 1200ºC for 2h, yielding a particle size range of 5 – 10nm for microemulsion, and 10 – 30nm for emulsion. However, while the same reagents by direct reaction in water produced particles in the range of 15 – 200nm.

#### 4.3.6.2 Sonochemical Route

This novel method, where ultrasound, instead of heating is used in the final production of HAp powder [114], produces very fine (20nm) particles, with a narrow size distribution, via the reaction of Ca(OH)2 and H3PO4 with ultrasonics, at a pH = 7, Ca/P molar ratio = 1.67 and T = 42 - 65ºC. By comparison, heating was found to produce a particle size ~180nm.

#### 4.3.6.3 Microwave Irradiation Route

Research carried out by Yang et al. [115], prepared HAp powders in a two step process; through use of the wet chemical precipitation method followed by a novel vapour phase reaction, using microwave irradiation. H3PO4 glucose and Ca(NO3)2.4H2O were used as the starting materials. After the precipitation reaction was complete, the HAp slurry mixture was transferred to a domestic microwave oven (2450 MHz) and irradiated for periods of 5min, 30min, 1h and 2h. Kumar et al. [116] employed the use of microwave irradiation in order to speed up the rate of their hydrolysis reaction to produce calcium deficient HAp. Microwave synthesis of materials offers the advantages of heating throughout the whole volume with the efficient transformation of energy [116].

#### 4.3.6.4 Colloidal Reaction

An alternative method for producing homogenous HAp powders were reported by Yasuda et al. [117]. This research prepared HAp via a colloidal process; a method for precipitating and in-situ stabilising calcium phosphate powders in the presence of a surface active agent. The research aimed to avoid the spontaneous agglomeration between particles on account of its electrostatic, steric or electrosteric repulsive force. The spontaneous agglomeration in a dry powder consolidation process due to van der Waals force induces an inhomogeneous microstructure of sintered materials which result in poor mechanical properties. The addition of a polymer dispersant (polycarboxylic acid ammonium), however, proved effective in preventing the agglomeration of the ceramic powder by electrosteric stabilization. The anionic polyelectrolyte was found to be an effective dispersant to obtain a homogeneous distribution of HAp powders.

### Use of Novel Materials for HAp Synthesis

A novel method in HAp synthesis, using novel precursor or raw materials was reported by Rivera et al. [118] who produced HAp by extracting one its starter materials, CaO, from raw eggshells. Eggshell represents 11% of the total weight of the egg and is composed of calcium carbonate (94%), calcium phosphate (1%), organic matter (4%) and magnesium carbonate (1%). An eggshell consists primarily of a three-layered structure, namely the cuticle, the spongeous layer and the lamellar layer. The cuticle layer, representing the outermost layer consists of a number of proteins. While, the spongeous and lamellar layers form a matrix constituted by fibrous proteins bonded to calcite (calcium carbonate) crystals in the proportion 1:50. After mechanical cleaning of the eggshell surfaces, a two stage heat treatment process was undertaken. A two-stage solid-decomposition reaction to produce CaO powders was carried out at elevated temperatures. The first stage consisted of heating the eggshells to 450°C for 2h at the heating rate of 5°C/min; at this temperature, any organic residue is expected to be destroyed. The second stage consists of heating the samples to 900°C also for 2h but with the heating rate at 0.5°C/min. At this stage, the eggshells transform into calcium oxide by freeing carbon dioxide (CO2) according to the following equation:

  Equation 19

The CaO obtained from the eggshells was then transformed (using the hydrothermal –wet chemical reaction) into HAp in a phosphate solution containing Ca3(PO4)2 in a container designed to reproduce a moist atmosphere. Subsequently, the container was sealed and heated to 1050°C for 3h at the heating rate of 10°C/min. The expected reaction was as follows:

  Equation 20

Once the reaction was completed, the solution was filtered and the resulting material dried overnight at 80°C in an oven. The researchers concluded that the concentration of the HAp obtained by this method could be improved by optimising the concentration of the phosphate solution, the time and the temperature of annealing. The added advantage was that HAp was the only apatite present in the reaction products, apart from minute fractions of certain other calcium compounds. Similar research was conducted by Lee et al. [119], who also successfully fabricated pure HAp using recycled eggshell and phosphoric acid using a wet chemical reaction.

A method in obtaining a phosphorous from a relatively cheap and plentiful source was highlighted by Puajindanetr [16]. This research suggested that HAp can be obtained from rice bran containing phytin as the phosphorous source, as a starting material. The phytin was extracted from oil-less rice bran using sodium hydroxide and calcium oxide to obtain HAp.

Researchers have investigated the conversion of coral into synthetic HAp since 1974 [120]. Coral is similar to cancellous bone in morphology. Full conversion of the coral into HAp has the advantage of retaining favourable pore sizes for bone graft application and improved bioactivity. Again, the poor mechanical properties of the synthetic HAp linit its use in load bearing applications. Unconverted coral (that is, CaCO3), however, is unsuitable for long term implant purposes due to its very high dissolution rate and poor longevity and stability. The coral (*Gergonacea species*) obtained from sea beach of Kotalam at Kerala was used in work carried out by Chattopadhyay et al. [121]. In this work, the coral was washed with distilled water and de-protienised by refluxing for 18hrs in 5% potassium hydroxide solution. The deproteinised coral was washed with distilled water and neutralized by dilute hydrochloric. Wide interest in the use of coral from the Great Barrier Reef is also evident in such works, as carried out by Nissan et al. [122] and Hu et al. [99] who both investigated the use of the Australian coral (of the genus, *Porites species*) in the production of monophasic HAp using a low temperature hydrothermal technique as the conversion method. The exchange reaction with the marine coral species *Porites* is as follows [120]

:  Equation 21

The main advantage to these novel synthesis methods is the fact that mass production may be possible at a low cost while implementing these simple procedures.

## Precipitation Kinetics in HAp Synthesis

Crystallisation and precipitation are similar phenomena in many respects. Crystallisation is the formation of solid particles within a homogeneous phase or simply the forming of crystals. This may occur as the formation of solid particles in a vapour as solidification from a liquid melt, as in the manufacture of large single crystals, or as crystallisation from liquid solution [16, 123]. Precipitation, like all crystallisation processes primarily consists of the three following steps:

1. The creation of supersaturation

2. The generation of nuclei

3. The subsequent growth

Precipitation is the formation of a solid product from a liquid solution and is usually initiated by a change in solution temperature or pressure so as to exceed the solubility limit of the chemical precipitating agent. Seed nuclei may also be present on which the desired phases can precipitate. Impurities can be removed by either retaining them in solution or precipitating them in a separate process. Precipitation, analogous to crystallisation is a nucleation and growth process with Arrhenius control of the kinetics. High nucleation and slow growth are generally required to keep the powder particles small. Factors such as temperature, concentration, pH and the order and rate of mixing all affect the chemical equilibria and the purity and physical nature of the precipitate. While precipitation routes produce homogeneous powders and can be employed to disperse trace additives uniformly, one drawback is that the powders must invariably be dried and calcined leading to the formation of hard agglomerates which require breaking down by extensive milling operations. [123]

### Creation of Supersaturation

A saturated solution may be defined as a solution that is in thermodynamic equilibrium with the solid phase of its solute at a specified temperature. Solutions may contain more dissolved solute than that given by the equilibrium saturation value and are often referred to as ‘supersaturated’. The degree of supersaturation may be defined by the concentration difference [123]:

  Equation 22

Where; C = the actual solution concentration

 Cs = the equilibrium saturation value

Other expressions derived from equation 22 are the supersaturation ratio (S) and the relative supersaturation (α) (which are both dimensionless)

  Equation 23

 ** Equation 24

The driving force for the formation of HAp is the difference in Gibbs free energy, ΔGHA, between the supersaturated solution and the equilibrium solution of HAp and is given by [123]:

  Equation 25

Where *IP* is the ionic product of the precipitating salt, in this case the product of concentrations of (Ca2+)10,(PO43-)6 and (Ca2+)10 ions; *ksp* is the solubility product;  is the number of ions (10 + 6 + 2 = 18); *R* is the universal gas constant and *T* is the absolute temperature. Equation 16 clearly indicates that Δ*GHA* depends on the *IP* and the *ksp* at a given temperature. Ca(OH)2 is a sparingly soluble salt in water, and its solubility decreases with increasing temperature. It is reported that the *ksp* of Ca(OH)2 at 25ºC is 6.6 x 10-6 and the heat of dissociation, Δ*Hr*, of Ca(OH)2 into Ca2+ and 2OH- ions is -17.88kJ/mol. Thus, using the van ‘t Hoff equation, shown in equation 17, are calculated to be 4.60 x 10-6,2.13 x 10-6 and 1.54 x 10-6 at 40, 80, and 100ºC, respectively [123]:

  Equation 26

Where, , is the solubility product at 25ºC,  is the heat of dissociation, *R* is the universal gas constant and *T* is the absolute temperature.

The solubility product of HAp is very low, 2.34 x 10-59 at 37ºC, and its temperature dependencies lie in the range 5 - 37ºC. The solubility product of HAp is reported to decrease with increasing temperature. This relationship is expressed as follows [16, 123]:

  Equation 27

Here, *ksp* (mol9dm-9), is the solubility product and T is the absolute temperature. Higher temperature will give rise to a smaller solubility product of HAp and hence a higher supersaturation degree of the solution [16]. Therefore, it is possible to achieve ideal HAp homogenous nuclei by controlling the pH value and the temperature of the soaking solution. The temperature dependencies of the solubility products are shown in Table 4-7:

Table 4‑7: Solubility Products, Ksp, for various precipitation temperatures. Adapted from [16]

|  |  |
| --- | --- |
| **Temperature (°C)** | **Ks x 1059** |
| 5 | 2.92  0.3 |
| 15 | 3.23  0.25 |
| 25 | 3.04  0.25 |
| 37 | 2.34  0.27 |

Though it is not known in the temperature range above 37ºC, the solubility product is estimated through use of equation 26 for temperatures greater than this [123]. The solubility products however cannot be directly used to measure the supersaturated level (SSL) at any given temperature. The reasons for this are as follows; (i) the applicability for equation 18 is doubtful, (ii) the lack of sufficient solubility data of HAp in the temperature range >37ºC and the wide scatter of *ksp* in the data of HA in aqueous solutions among researchers and (iii) the dependence of solubility/solubility products of HAp with concentrations of Ca2+ and CO3-2 ions and pH as well as the composition and particle size.

The SSL is therefore calculated using equation 28. With the reaction time, the IP increases (to eight orders of magnitude) as a result of the continuous addition of acid, and this will raise the SSL only by 1 order of magnitude at a given temperature [123]:

  Equation 28

However, computing the SSL and Δ*GHA* becomes complicated at different stages, from the beginning to the end of acid addition.

### Kinetics of Nucleation

The rate of nucleation may be defined as the number of new particles formed per unit volume of magma or solid-free original liquid. This quantity is the first kinetic parameter controlling the crystal size distribution (CSD).

New crystal formation is achieved through homogeneous nucleation, heterogeneous nucleation, secondary nucleation and attrition. Homogeneous nucleation is the desired preference in precipitating HAp powder particles and refers to the formation of new crystals from the liquid phase as a result of the supersaturation alone. Heterogeneous (undesirable crystal formation for HAp) nucleation refers to new particle formation resulting from the presence of foreign insoluble material. Secondary nucleation refers to nucleation induced by the presence of suspended crystals of the solute. Attrition is simply the mechanical degradation of suspended crystals in which pieces become growing crystals.

Puajindanetr [16] proposes the rate of homogeneous nucleation as an Arrhenius type relationship:

  Equation 29

Where, *B* is the nucleation rate, *ΔGHA* is the gibb’s free energy of formation of a nucleus, *k* is the boltzmann’s constant, *C* is a proportionality constant and *T* is the absolute temperature. Using appropriate relationships connecting geometry, surface tension, free energy and supersaturation ratio (S); equation 29 becomes:

  Equation 30

Where, *b* is the geometric factor, *s* is the interfacial tension, *v* is the molecular volume, *D* is the solute diffusion coefficient, d is the molecular diameter and *S* is the supersaturation ratio; 

However, equations have also been proposed for a power law form to account for the observation that nucleation does not occur at very low supersaturations [16]:

   Equation 31

Where, *C* is the solute concentration, *Cm* is the concentration greater than the saturation concentration but below which nucleation does not occur, *k* is an empirical constant and *i* is the order of nucleation. In most inorganic systems, it is suggested that *Cm* appears to be very close to *Cs*. Consequently, *Cm* may be taken to equal *Cs* and therefore equation 31 becomes [16]:

  Equation 32

In equation 31 and 32, it is assumed that *k* can be a function of temperature, but that *i* is not a function of temperature. As indicated previously, secondary nucleation describes the formation of nuclei attributable to the influence of the existing macroscopic crystals within the magma. Two types of secondary nucleation have been well documented; (i) one attributable to fluid shear and (ii) the other to collisions between existing crystals with each other (or with agitator blades during milling operations).

Equation 33 represents that of a simple nucleation model. However, the model must also contain a dependence on the frequency and energy of crystal-crystal contact and agitator-crystal contact. Secondary nucleation rates are commonly correlated by empirical relationships such as [16]:

  Equation 33

Where, *MT*is the slurry concentration or magma density,  *s* is the supersaturation, *k* is an empirical constant and *i, j* is the order of nucleation. All quantities and parameters within the above equation are affected by temperature level and the presence of foreign soluble or insoluble material.

### Kinetics of Crystal Growth

The mechanism of crystal growth from a solution requires that solute be transported to the crystal surface and then orientated into the crystal lattice. Two steps are required to achieve this; (i) a diffusional process followed by (ii) a surface reaction. In assuming that solute molecules diffuse to the crystal face and then join the crystal lattice by means of a surface reaction, the following growth rate equation may apply [123]:

  Equation 34

Where, *m* is the crystal mass, *A* is the crystal surface area, *X* is the film thickness, *D* is the diffusivity of solute, *kr* is the reaction rate constant, s is the degree of supersaturation = (*C – Cs*) and *kg* is the proportionality constant =  Equation 35

As the diffusion coefficient increases, the overall rate coefficient approaches kr and the growth rate is controlled by the surface reaction step. Also, if kr is comparatively high, the controlling resistance is that of the diffusion step. Therefore, either step may be considered rate controlling dependent on operating conditions. Each crystal whilst in suspension has been observed to grow at the same rate, regardless of its size, if subjected to the same conditions (that is, uniform supersaturation field and at the same temperature). The ‘ΔL law of crystal growth applies if the agitation is vigorous and the velocities of the crystals of different sizes relative to the solution are the same. When this law is applied, growth rate (*G*) is not a function of crystal dimension length, the total growth of each crystal in the suspension during the same time interval, *t*, is constant [123]:

  Equation 36

The ability to control size and shape of particles by controlling the nucleation and growth is especially attractive as it allows accurate tailoring of the desired powder particles. Achieving the desired size by growth rather than by reduction also makes the production of very fine powder particles more predictable. Homogeneous nucleation of small particles requires that an interface be created between two phases causing an increase in the systems free energy. However, after particle growth has occurred, the interface energy becomes small compared with the volume energy decrease so that the total change in free energy on forming the new phase becomes negative. There are two opposing factors governing the energy of the system for a spherical particle of radius r [123]:

  Equation 37

Where *γ* is the interface energy and *ΔGv* is the change of free energy resulting from transformation from one phase to the other. For nucleation in solids an additional term is included in the equation to account for strain energy resulting from differences in volume between the new phase and the matrix so that it becomes [123]:

  Equation 38

Where the strain energy per unit volume is given by  with ε the strain and b a constant whose value depends on the nucleus shape. The strain energy term  has a profound effect on the process of precipitation of a solid phase from a supersaturated solid solution of different composition and typically causes formation of parallel platelets. This precipitate morphology allows growth to occur with minimum increase in strain energy whereas the formation of thick or spherical particles causes large strains. The energy for nucleation of a precipitate phase depends on the interface structure and orientation. Several types of precipitate may be defined. A coherent precipitate is one in which the planes of atoms are continuous across the interface whereas in a semicoherent precipitate, some or all of the planes of atoms are discontinuous across the interface. For this scenario, the boundary is accommodated by misfit dislocations or other defects. When there is no match between the atomic configuration in the two adjacent phases the precipitate is incoherent and much larger than that of coherent. [16, 123]

In a less surpersaturated solution, there is general agreement that the growth rates are too low to be transport-controlled. Many studies have shown the values of the effective reaction order, *n*, to range from 2.1 to 4.0 in the following equation [123]:

 *R* = *k*σ*n* Equation 39

where *R, k*, and *σ* are growth rate, effective rate constant, and thermodynamic driving force, respectively [49, 123]. This indicates the involvement of a polynucleation mechanism. Values of interfacial tension between HAp surfaces and the solution phases ranged from 10 to 120 mJm.2 [49, 123]. Values of interfacial tension obtained from growth data are greater than those obtained from dissolution kinetics. Growth rate measurement for HAp at very low supersaturation demonstrated that the linear growth rate as a function of driving force in the pH range of 5.0 to 6.5 was different from that in the pH range of 7.0 to 8.5 [16, 123]. As a result, the growth rate of HAp is approximately 2.5 times larger in the pH range of 5.0 to 6.5 than in the pH range of 7.0 to 8.5 even under the same supersaturation. Between these pH ranges, there is the point of zero charge of HA at about pH 7.0, which were verified experimentally. An understanding of the apparent change in the kinetics of HAp growth at pH above and below this value may require elucidation of the role of surface charge on crystallization [16, 123].

## Powder Processing of HAp

Post chemical reaction in the synthesis of HAp, whether in slurry or powder form requires additional processing in order to achieve the desired final HAp powder properties, necessary for its specific medical application. The synthesis route applied to obtain the HAp will ultimately determine the processing steps to follow in achieving the final HAp properties required. These processing steps can include drying, sintering, grinding and crushing, sieving, and sintering treatment for optimum HAp powders to result in a successful coating application. An overview of these steps is discussed in this section.

### Thermal Behaviour of HAp

#### 4.8.1.1 Drying

Drying may be defined as the removal of liquid from a porous material by means of its transport and evaporation into a surrounding unsaturated gas. The evaporation process is relatively energy intensive, and consequently, drying efficiency is an important consideration. Drying must be carefully controlled, since stresses produced by differential shrinkage or gas pressure can result in defects of the final product. Drying involves the transport of energy into the product. This is achieved as liquid is transported through pores to the meniscus, where evaporation occurs, and by vapour transport through pores. In any drying system, it is required that heat energy be brought to the surface of a product while the vapours must be carried away. As a result, in order to fully understand the mechanisms involved in the drying process, evaporation and mass and thermal transport must be considered. [109]

Drying occurs in three stages dependent on the liquid content, in which the drying rate initially increases, remains constant for a certain period and ultimately decreases respectively [124]. These stages are commonly known as (i) the initial period, (ii) the constant rate period (CRP) and (iii) the falling rate period (FRP). Drying curves are plotted as a function of time, or, more commonly in clay-based ceramics, as a function of the moisture content [125].

**4.8.1.2 Spray Drying**

Spray drying (dissimilar to that described in Section 4.3.6) is a process whereby the HAp slurry is sprayed into a warm drying medium to ideally produce spherical powder agglomerates of relative homogeneity. It is considered a relatively efficient process since the material (that is, the slurry) is well dispersed in the drying medium, the diffusion path is shorter, and the high specific surface area contributes to a higher rate of evaporation per unit mass of product [109]. The large surface area-to-volume ratio of the droplets allows rapid water evaporation and the resulting dry powder is separated from the air and stored for later use [126].

The morphology of the HAp particles can vary with spray drying process conditions, such as droplet size, initial solid concentration, viscosity of liquid and drying temperature. Luo et al. [126] have developed a model to predict the crust formation inside slurry droplets at a constant rate period during spray drying as a function of operating parameters and slurry properties. For low-solid-content slurry, they reported on findings that the particles were drawn to the droplet surface along with the capillary induced moisture flow; hence postulating the more favourable use of a high solids content slurry.

There are four stages to the spray drying process [127, 128]:

**Stage One:** Atomisation.

**Stage Two:** Dispersion of the particles in air. The extent, to which the air and spray mix, influences the drying of the particles.

**Stage Three:** Drying of the particles.

**Stage Four:** The final stage is the collection of the powder.

The typical drying system starts with a heated gas source that is introduced into a cylindrical chamber. At the same time, a liquid feed material is also introduced into this drying chamber.

#### Atomisation

Atomisation is undertaken by centrifugal (rotating disc) atomisation or by single-fluid or two-fluid nozzles, similar to melt atomisation [129]. It employs slurry that has either been sieved or milled in order to remove any coarse agglomerates [109, 130]. The largest agglomerate size obtained using a single-fluid nozzle yielded are 600μm and 300μm with rotating discs, indicating that centrifugal atomisation yields the narrowest size distribution [129]. The feed is atomised into a fine spray or mist so that the hot process gas can evaporate the liquid, leaving the solid portion of the feed in a dry particulate form. The liquid stream from the nozzle is broken up by the atomiser into an enormous number of tiny droplets. These droplets are usually known as particles. The particle size is a very important aspect of the process, with the final product usually required to be within a certain range. Spray drying can produce particle size range from ten to a few hundred microns. There are usually coarse and fine dried particles. [127]

The purity of the atomised (especially oxygen content) powders depend upon three principal parameters [131]:

1. Atmosphere inside the melting chamber (highest purity powders give vacuum, then inert gas and lowest purity air atmosphere).
2. Atomisation medium (highest purity powder requires gas as the medium, lowest purity water).
3. Cooling medium in the atomisation tower (highest purity – inert gas cooled, lowest purity – water cooled).

By varying the several parameters (such as, design and configurations of the jets, pressure and volume of the atomising fluid, thickness of the stream of calcium phosphate slurry), it is possible to control the particle size distribution over a wide range. The particle shape is determined largely by the rate of solidification and varies from spherical, if a low heat capacity gas is employed, to highly irregular if water is used. [131]

**Agglomeration**

Agglomeration (otherwise, referred to as granulation) of HAp fine powder particles to ultimately produce HAp particles of appropriate sizes for plasma spraying application onto orthopaedic coatings may also be necessary.

Agglomeration allows enlargement of very fine powders to avert uncontrolled agglomeration and to make the powders free flowing and safe. The bonding mechanisms for agglomeration (controlled as well as uncontrolled) can be:

1. Solid bridges formed by crystallised salts or sintering contacts

2. Immobile liquid bridges formed by viscous binders, adhesives and adsorption layers

3. Mobile liquid bridges (capillary forces)

4. Mechanical Interlocking of powders

The adhesion forces decrease in the sequence given above and increase with decreasing size of the primary particles. The largest agglomerated sizes achieved, through spray drying are 600μm from a single-fluid nozzle, and 300μm with rotating discs, which also yield the narrowest size distribution. In the specific droplet drying process, spray dried agglomerates are often inhomoneously packed and sometimes even take the form of hollow spheres [132].

#### 4.8.1.3 Sintering

Sintering of the HAp powder post spray drying, occurs within a controlled atmospheric environment to a temperature below the melting point but sufficient to allow solid-state diffusion and to permit bonding of the particles. The first stage involves the combustion of air and volatisation of any thermal labile compounds that may interfere with good bonding. The second stage (or high temperature stage) is where the desired solid-state diffusion and bonding between the powder particles occurs. Finally, a cooling period is required to lower the temperature of the HAp product. The cooling period maintains the product in a controlled environment, serving to prevent oxidation that would occur upon direct discharge into air and possible thermal shock from rapid cooling. [132]

The control of sintering behaviour for HAp powders is very important [133]. Highly dense HAp is desirable to prevent the preparation of human fluid into the interfacial area between HAp coating and Ti-alloys in hip joint replacements, while porous HA would be needed for the broken bone replacements in which the ingrowth of natural and artificial bones occurs and strong bond between them can be readily attained [133]. HA powders sintered by Sung et al. [133] at 1000ºC for 1h showed highly dense and almost pore-free morphology. The sintered density value for the sintered HAp was determined as 99.9% of the theoretical value for HAp, owing to the fine particle size that came from the HAp powders synthesised. HAp powders, consisting of very fine particles have a much higher surface area, which is the driving force for solid-state sintering and the amount of surface energy decrease is proportional to the free energy decrease for the sintering reaction. Therefore, sintering behaviour of HAp powders can be controlled by variations of powder particle size. Kim et al. [134] sintered HAp powders with a specific surface area of 68m2/g at 1200ºC for 2h to improve the crystallinity of the final HAp product. It was also noted that prior calcinations at 900ºC for 4h in air with subsequent ball milling were effective in eliminating agglomerates in the HAp powder. Results from Juang et al. [135] showed that sintering treatments increased the average particle size and distribution. The sintering behaviours were investigated by dilatometry and density measurement. Fluidity of the HAp powders and the driving force for sintering were found to dominate the properties. Sintering at 1250ºC resulted in a higher bending strength (about 55MPa) with a finer grain size. Work carried out by Muralithran [136] support this evidence, as scanning electron microscope (SEM) analyses of the sintered microstructure revealed an exponential increase in grain size with increasing sintering temperature.

Muralithran [136] states that the HAp phase was stable when sintered below 1400ºC for 2h. However, sintering at temperatures ≥1400ºC resulted in the decomposition of HAp to form TCP, TTCP and CaO. It is believed that the high humidity content present in the sintering atmosphere slows down decomposition rates by preventing dehydration of the OH- groups from the HAp matrix.

Kim et al. [134] observed that substitutions into the HAp lattice (such as, carbonate, magnesium, silicon) inhibited apatite crystallisation in solution and destabilised the structure of HAp and favoured its thermal conversion into α- or β-TCP. An adverse affect on the sinterability of the HAp was identified.

HAp decomposes to form other calcium phosphates at elevated temperatures. Two mechanisms have been proposed for the decomposition as follows [137]:

 Equation 40

 Equation 41

Of these, the former is the more accepted mechanism. Further heating results in the transformation of β-tricalcium phosphate (β-Ca3(PO4)2), forming α-tricalcium phosphate. This usually requires exposure to temperatures in excess of 1350°C for this phase transformation to take place. Regardless of which mechanism takes place, both result in the formation of soluble or resorbable calcium phosphates, which dissolve when exposed to physiological environments.

### Grinding

The next stage in HAp powder processing is the size reduction of the HAp powder particle agglomerates that resulted upon sintering. Grinding and milling are extensively applied in the field of ceramics for size reduction [114]. The general phenomena during size reduction in the solid state are based on fracture mechanics: the nucleation of cracks, followed by crack propagation and fracture, by which new surfaces are formed. A further decrease of mean particle size can take place only when these processes occur. The kinetic energy within the milling aggregate is partially transformed into mechanical stresses in the material to be disintegrated. The forces acting in these processes cause mainly compression and shear stresses, applied as impact. In ceramic materials, plastic deformation does not take place in coarse particles, but becomes significant for mean particle sizes in the micron range. The main process phenomenon for all grinding and milling processes, however, is crack propagation and fracture, which is accompanied by local, not well defined, temperature rises. These are often defined as thermal ‘spikes’ and can result in anomalous structures. The limit of the minimum obtainable particle size depends on the conditions of the mechanical process as well as on the material itself.

The efficiency of mechanicals size-reduction processes is generally very low. Only about 0.1% of the spent energy in the conventional ball milling process is found in the generated new surfaces of the fine particles. The energy E necessary for the physical process of size reduction is given by [114]:

  Equation 42

Where γ is the specific surface energy and ΔS is the increase of specific surface.

### Sieving

Post sintering, the HAp powders are sieved through the desired mesh size in order to ensure that the correct particle size distribution (that is, that >90% of the HAp powder particles are <100μm) is achieved prior to plasma spraying the HAp powder particles as a HAp coating onto orthopaedic implants [138].

## Calcium Phosphate Coating Application onto HAp

Coating of HAp onto orthopaedic coatings have been applied via numerous techniques; including dip coating into a powder suspension, electron beam evaporation combined with ion beam mixing, electrophoretic deposition, sol-gel, laser ablation, plasma spray, and radio-frequency (rf) sputtering methods.

The main problem  associated  to HAp coatings achieved through  rf  sputtering  is the phosphorous  deficiency and  the amorphous  nature  of  the final HAp coating. The  latter  causes  fast  resorption  of  the HAp  by  the surrounding biological environment. The  ion  beam  sputtering  method,  as  well as  the hot  iso-static  pressing  method  have  a disadvantage associated to its  ability  to  coat  irregular shaped HAp.

The electrophoretic method presents poor adhesion and formation of additional undesirable phases. Plasma  spray  has  been  the  commercial  method  for  preparation  of  calcium  HAp  coatings. However due to the extremely high temperature required, the final coatings may contain traces of TCP, TTCP, OHAp and ACP,  characterised  by its  lack  of  crystallinity  and  poor  adhesion  to  the  substrate. Pure HAp coatings are required for orthopaedic implants, as the presence of other phases and constituents can lead to the acceleration in the degradation of HAp coatings *in vivo*.

Some techniques that have been used in the application of HAp coatings are summarised in the following table [138].

Table ‑: Summary of HAp coating techniques for application onto orthopaedic implants

|  |  |  |  |
| --- | --- | --- | --- |
| **Coating Technique** | **Coating Thickness** | **Advantages** | **Disadvantages** |
| Dip Coating | 0.05-0.5mm | InexpensiveCoatings applied quicklyCan coat complex substrates | Requires high sintering temperaturesThermal expansion mismatch |
| Sputter Coating | 0.02-1µm | Uniform coating thickness on flat substrates | Line of sight techniqueExpensiveTime consumingCannot coat complex substratesProduces amorphous coatings |
| Pulsed Laser Deposition | 0.05- 5µm | As for sputter coating | As for sputter coating |
| Hot Pressing and Hot Isostatic Pressing | 0.2-2.0mm | Produces dense coatings | HP cannot coat complex substratesHigh temperature requiredThermal expansion mismatchElastic property differencesExpensiveRemoval/Interaction of encapsulation material |
| Electrophoretic Deposition | 0.1-2.0mm | Uniform coating thicknessRapid deposition ratesCan coat complex substrates | Difficult to produce crack-free coatingsRequires high sintering temperatures |
| Thermal Spraying | 30-200µm | High deposition rates  | Line of sight techniqueHigh temperatures induce decompositionRapid cooling produces amorphous coatings |
| Sol-Gel | <1µm | Can coat complex shapesLow processing temperaturesRelatively cheap as coatings are very thin | Some processes require controlled atmosphere processingExpensive raw materials |

Of the techniques outlined, thermal spraying, in particular, plasma spraying is the only commercially accepted method for producing FDA approved HAp coatings. While plasma spraying is a well understood process, the control of its process variables is quite complicated. Small changes to one of its processing variables can vastly affect the properties in the final HAp coating. Variations, such as, the powder feedstock may produce soluble coatings. The reason for this is the high processing temperatures encountered by the HAp, which induce decomposition of the HAp powdered material to soluble calcium phosphate compounds.

## Biocompatibilty and Toxicity

HAp continues to show a highly attractive biologic profile. This profile includes a lack of local or systemic toxicity, a lack of inflammatory or foreign-body response to the material in solid or particulate form, a lack of pyrogenic response, an absence of intervening fibrous tissue between implant and bone, and the ability to become bonded to bone directly. This profile probably is generic to all calcium phosphate implant materials [28]. Almost all long-term investigations of calcium phosphate materials have shown positive results *in vivo*. HAp blocks and granules have been studied extensively in orthopaedic and dental animal models, including species, such as, primates, dogs, rabbits, and rats. Results have been uniformly successful with no reported adverse local or systemic response. The underlying basis for the lack of local or systemic toxicity with calcium phosphates is their chemical nature, as they contain only calcium and phosphate ions [28].

## *In vivo* Interfacial Reactions with Bone

The interface remodelling associated with implantation of dense and non resorbable calcium phosphate ceramics in bony tissue can be characterized as presenting normal bone healing processes [28]. Along dense as well as porous materials normally proliferating bony tissue is usually found, deposited directly on the surfaces without the presence of an intervening fibrous tissue layer. The highly biocompatible nature of calcium phosphate materials allow calcium and phosphate ions to be derived from the implant and/or bone. The composition of any solids deposited on the surface of the calcium phosphate implants are believed to be determined by the surrounding physiologic medium, and ultimately, in bone, this medium would generate calcium phosphate solids in the form of biologic apatite. The final result is that calcium phosphate materials become coated with a microscopic layer of bone mineral shortly after implantation [28]*.*

# Summary

*Calcium Phosphates for Medical Applications* provides and in-depth introduction to the various types of calcium phosphates used in medical applications, providing a detailed look at their physicochemical properties and crystallographic structures. The book is divided into four chapters. The first chapter provides a basic introduction to bioceramics; their classification in accordance to their biologic interaction and the compositional classification of bioceramics, in which the family of calcium phosphates belongs. The second chapter discusses the physicochemical properties of biological apatite (that is, natural bone), natural bone cells and bone remodelling by osteoblasts and osteoclasts. The third chapter discusses low and high temperature calcium phosphates, with an illustration of each of their crystallographic structures and a summary of the possible substitutions for each sub-lattice of the calcium apatite. Finally, chapter four examines the physico-chemical properties of HAp and gives an overview of the many techniques, which have been implemented for its production for various medical applications. The most popular method, due to its simplicity, low-cost and lack of additional by-products is discussed further in relation to the kinetics of crystallisation and precipitation. Post-precipitation processing of the HAp is also examined, in respect to its thermal behaviour during drying (that is, clacination) and sintering; grinding and sieving. The chapter concludes with an overview of the types of HAp coating applications presently used in research and industry, with a brief discussion on HAp biocompatibility, toxicity and its *in vivo* interfacial reactions with human bone.

An intensive literature review and optimisation of the wet chemical precipitation technique for the synthesis of monophasic and highly crystalline HAp may be found in Kehoe et al. [139]. This work examines the critical parameters surrounding the chemical precipitation process and studies the significance of each parameter on the final HAp powder properties obtained, such as, phase composition, crystallinity, crystallite size, lattice parameters, mean particle size, particle size distribution and powder morphology.

The book brings the reader to a level of knowledge, where they can understand the techniques implemented to synthesise the various calcium phosphates, their critical parameters and the benefits that each of these have to offer for various medical applications. The book will be an appropriate text for specialist undergraduate courses in materials science and processing, chemistry and any other science or engineering background, who demand information relating to calcium phosphates. It is also intended for postgraduate M.Sc. and Ph.D. students in the biomaterials sciences. Industrial scientists, working in the field of HAp synthesis, and indeed, other calcium phosphates (such as β-TCP) will find the book an invaluable introduction to bring rapid familiarity with the many techniques available for calcium phosphate synthesis of varied composition with the capabilities of each technique for tailoring the desired calcium phosphates to the desired requirements for the medical application to which it is intended.

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