Calcium Orthophosphate Bioceramics

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Abstract

The present review is intended to point the readers' attention to the important subject of calcium orthophosphate bioceramics. Calcium orthophosphates by one-selves appear to be of a special significance for the human beings because they represent the inorganic part of calcified tissues of mammals. Therefore, many types of calcium orthophosphate-based bioceramics possess remarkable biocompatibility and bioactivity. Materials scientists extensively use this property in attempts to construct artificial bone grafts those are either entirely made of or only surface-coated by calcium orthophosphate bioceramics. Namely, self-setting calcium orthophosphate cements are very helpful in filling voids in damaged bones, while metallic implants covered by a surface layer of calcium orthophosphate bioceramics are widely used for hip joint endoprostheses and tooth substitutes. Porous bioceramic scaffolds made of calcium orthophosphates are very promising tools for tissue engineering applications. In this paper, an overview on the current knowledge on calcium orthophosphate bioceramics has been provided.

Introduction

Calcium orthophosphates are the chemical compounds of a special interest in many interdisciplinary fields of science, including geology, chemistry, biology and medicine. According to definition, they consist of three major chemical elements: calcium (oxidation state +2), phosphorus (oxidation state +5) and oxygen (oxidation state –2), as a part of orthophosphate anions. These three chemical elements are present in abundance on the surface of our planet: oxygen is the most widespread chemical element of the earth's surface (47 mass %), calcium occupies the fifth place (3.3 – 3.4 mass %) and phosphorus (0.08 – 0.12 mass %) is among the first twenty of the chemical elements most widespread on our planet [1]. In addition, the chemical composition of many calcium orthophosphates includes hydrogen, either as an acidic orthophosphate anion (for example, HPO_4^{2-} or $H_2PO_4^-$), and/or as incorporated water (for example, $CaHPO_4 \cdot 2H_2O$).

The atomic arrangement of calcium orthophosphates is built up around a network of orthophosphate (PO_4) groups, which give stability color. The vast majority of them are sparingly soluble in water; however, all of them are easily soluble in acids but insoluble in alkaline solutions. The list of known calcium orthophosphates is summarized in Table 1 [2].

Calcium orthophosphate bioceramics

A number of definitions have been developed for the term "biomaterials". The consensus developed by experts in this field is the following: biomaterials are defined as synthetic or natural materials to be used to replace parts of a living system or to function in intimate contact with living tissue [3]. In general, they are intended to interface with biological systems to evaluate, treat, augment or replace any tissue, organ or function of the body and are now used in a number of different applications throughout the body [4, 5]. Bioceramics might be defined as biomaterials of the ceramic origin. In general, it can have structural functions as joint or tissue replacements, be used as coatings to improve the biocompatibility of metal implants and function as resorbable lattices, which provide temporary structures and a framework that

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Table 1.

Properties of the biologically relevant calcium orthophosphates. The solubility is given as the logarithm of the ion product of the given formulae (excluding hydrate water) with concentrations in mol/l [2].

Ca/P molar ratio	Compound	Formula	Solubility at 25 °C, –log(K_s)	Solubility at 37 °C, –log(K_s)	pH stability range in aqueous solutions at 25°C
0.5	Monocalcium phosphate monohydrate (MCPM)	$Ca(H_2PO_4)_2 \cdot H_2O$	1.14	data not found	0.0–2.0
0.5	Monocalcium phosphate anhydrous (MCPA)	$Ca(H_2PO_4)_2$	1.14	data not found	[c]
1.0	Dicalcium phosphate dihydrate (DCPD), mineral brushite	$CaHPO_4 \cdot 2H_2O$	6.59	6.63	2.0–6.0
1.0	Dicalcium phosphate anhydrous (DCPA), mineral monetite	$CaHPO_4$	6.90	7.02	[c]
1.33	Octacalcium phosphate (OCP)	$Ca_8(HPO_4)_2(PO_4)_4 \cdot 5H_2O$	96.6	95.9	5.5–7.0
1.5	α-Tricalcium phosphate (α-TCP)	α-$Ca_3(PO_4)_2$	25.5	25.5	[a]
1.5	β-Tricalcium phosphate (β-TCP)	β-$Ca_3(PO_4)_2$	28.9	29.5	[a]
1.2–2.2	Amorphous calcium phosphate (ACP)	$Ca_xH_y(PO_4)_z \cdot nH_2O$, n = 3–4.5; 15–20% H_2O	[b]	[b]	~5–12 [d]
1.5–1.67	Calcium-deficient hydroxyapatite (CDHA)	$Ca_{10-x}(HPO_4)_x(PO_4)_{6-x}(OH)_{2-x}$ ($0<x<1$)	~85.1	~85.1	6.5–9.5
1.67	Hydroxyapatite (HA or OHAp)	$Ca_{10}(PO_4)_6(OH)_2$	116.8	117.2	9.5–12
1.67	Fluorapatite (FA or FAp)	$Ca_{10}(PO_4)_6F_2$	120.0	119.2	7–12
2.0	Tetracalcium phosphate (TTCP or TetCP), mineral hilgenstockite	$Ca_4(PO_4)_2O$	38–44	37–42	[a]

[a] These compounds cannot be precipitated from aqueous solutions.
[b] Cannot be measured precisely. The comparative extent of dissolution in acidic buffer is: ACP >> α-TCP >> β-TCP > CDHA >> HA > FA [65].
[c] Stable at temperatures above 100°C.
[d] Always metastable.

is dissolved and/or replaced as the body rebuilds tissue. Some types of bioceramics even feature drug-delivery capability [6].

The use of calcium orthophosphates as bioceramics is based upon their chemical similarity with the mineral phase of bones and teeth [7-9]. Therefore, they can be prepared from various sources [10, 11]. According to available literature, the first attempt to use calcium orthophosphate bioceramics (it was TCP) as an artificial material to repair surgically created bone defects in rabbits was performed in 1920 [12]. Unfortunately, up to now, all attempts to synthesize bone grafts for clinical applications featuring the physiological tolerance, biocompatibility and long term stability have had only a relative success, which clearly demonstrates the superiority and complexity of natural calcified tissues [13].

Generally, living organisms might treat artificial implants as bioinert, biotolerant, bioactive or bioresorbable materials [3, 6, 14-16]. Bioinert (*e.g.*, zirconia, alumina, carbon and titanium) and biotolerant (*e.g.*, polymethyl methacrylate, titanium and Co–Cr alloy) materials will evoke a physiological response to form a fibrous capsule, thus, isolating the material from the body. Calcium orthophosphate bioceramics fall into the categories of bioactive and bioresorbable materials [3, 6, 14-16]. A bioactive material will dissolve slightly but promote the formation of a layer of biological apatite before interfacing directly with the tissue at the atomic level, that result in the formation of a direct chemical bond with bone. Such an implant will provide a good stabilization for materials that are subject to mechanical loading. A bioresorbable material will dissolve and allow a newly formed tissue to grow into any surface irregularities but may not necessarily interface directly with the material [6, 16-19]. Implants made of dense HA would be a good example of a bioactive material [20], while porous scaffolds made of biphasic calcium phosphate (BCP, *i.e.*, β-TCP + HA [21-24] or α-TCP + HA [25-29]) or bone grafts made of CDHA and/or ACP [30] appear to be the examples of bioresorbable materials. Unfortunately, having the ceramic nature, any calcium orthophosphate bioceramics alone are notoriously brittle that do not allow them to be used in load-bearing areas [31]. Due to this reason, the current biomedical application is focused on the production of non-load-bearing implants, such as pieces for middle ear surgery, filling of bone defects in oral or orthopedic surgery, as well as coating of dental implants and metallic prosthesis [13]. The mechanical properties of calcium orthophosphate bioceramics were reviewed elsewhere [32, 33]. In addition, there is a good review on the recent developments in processing and surface modification of HA [34].

In spite of the mechanical limitations, biomaterials and bioceramics of calcium orthophosphates are available in various physical forms: particles, blocks (dense or porous), injectable compositions, self-setting cements, coatings on metal implants, composites with polymers, *etc.* [35]. A porous surface provides mechanical fixation in addition to providing with sites on the surface that allow chemical bonding between the bioceramics and bone. For example, porous HA bioceramics can be colonized by bone tissue [36-38]. Therefore, macroporosity (pore size > 100 μm) in calcium orthophosphate bioceramics is intentionally introduced by addition of porogens, which are either volatile or soluble substances (*e.g.*, naphthalene, sucrose, $NaHCO_3$, gelatin, PMMA microbeads) [17, 24, 39-43]. Sintering particles, preferably spheres of equal size, is another way to generate porous 3D bioceramic scaffolds made of calcium orthophosphates. A wetting solution such as polyvinyl alcohol is usually used to aid compaction, which is achieved by cold isostatic pressing the particles into cylinders at approximately 200 MPa [44]. As hardly any effect of macropore size (150, 260, 510 and 1220 μm) was observed on the *in vivo* response [45], there is no need to create bioceramics with very big pores. Microporosity (pore size < 10 μm) results from the sintering process, while dimensions of the pores depend on temperature and sintering time. Creation of the desired porosity calcium orthophosphate bioceramics is a rather complicated engineering task and the interested readers are referred to other papers on this subject [24, 30, 41, 46-52].

The sintering stage appears to be of great importance to produce bioceramics with the required properties. Several processes occur during sintering of calcium orthophosphates. Firstly, moisture, carbonates and all volatile additives remaining from the synthesis stage, such as ammonia, nitrates and any organic compounds, are removed as gaseous products. Secondly, the removal of these gases facilitates the production of dense materials during sintering. Thirdly, these chemical changes are accompanied by a concurrent increase in crystal size and a decrease in the specific surface area. Fourthly, there is the chemical decomposition of all acidic orthophosphates and

their transformation into other phosphates (*e.g.*, $2HPO_4^{2-} \rightarrow P_2O_7^{4-} + H_2O$). Besides, sintering causes toughening [53]. Further details on the sintering process of calcium orthophosphate bioceramics are available elsewhere [17, 32, 54-56].

Studies showed that increasing the specific surface area and pore volume of calcium orthophosphate bioceramics for tissue repair might greatly accelerate the kinetic process of biological apatite deposition and therefore enhance the bone-forming bioactivity. More importantly, the precise control over porosity, pore size and internal pore architecture of bioceramics on different length scales is essential for the understanding of the structure-bioactivity relationship and the rational design of better bone-forming biomaterials [57, 58].

Calcium orthophosphate bioceramics in a number of forms and compositions are currently in use or under consideration in many areas of dentistry and orthopedics, with even more in development. Bulk material, available in dense and porous forms, is used for alveolar ridge augmentation, immediate tooth replacement and maxillofacial reconstruction [54, 59]. Further applications include increment of the hearing ossicles, spine fusion and repair of bone defects [60, 61]. In order to permit growth of new bone into a bone defect, the defect should be filled with a suitable bioresorbable material. Otherwise, ingrowth of fibrous tissue might prevent bone formation within the defect. Today, a variety of calcium orthophosphate bioceramics is available on the market for the treatment of bone defects. As an example, the readers are referred to a thorough physicochemical characterization of 14 calcium phosphate-based bone substitution materials in comparison to natural bone [62]. The commercial and trade-names of several types of calcium orthophosphate bioceramics might be found in literature [62, 63].

Chemically, calcium orthophosphate bioceramics is based on HA, β-TCP and/or BCP (*i.e.*, a composite of HA and α- or β-TCP) [2, 20-29, 54, 55, 63, 64]. General requirements for the ideal bone grafts are as follows: pores of some 100 μm size, a biodegradation rate comparable to the formation of bone tissue (*i.e.*, between a few months and about two years) and the sufficient mechanical stability. When compared to α- and β-TCP, HA is a more stable phase under the physiological conditions, as it has a lower solubility and a slower resorption kinetics [2, 54]. As implants made of calcined HA are present in bone defects many years after implantation, bioceramics made of β-TCP, α-TCP or BCP [24-29, 63-65] is more preferable for medical purposes. According to both observed and measured bone formation parameters, calcium orthophosphates were ranked in order of increasing magnitude as follows: low sintering temperature BCP (rough and smooth) ≈ medium sintering temperature BCP ≈ TCP > calcined low sintering temperature HA > non-calcined low sintering temperature HA > high sintering temperature BCP (rough and smooth) > high sintering temperature HA (calcined and non-calcined) [2]. Fig. 1 shows an example of the commercially available calcium orthophosphate bioceramics suitable for biomedical applications.

Fig. 1. Examples of calcium orthophosphate bioceramics suitable for bone substitution.

Another bone healing concept was introduced with self-hardening bone cements made from calcium orthophosphates [17, 39, 40, 52, 66]. This type of materials might be defined as a low temperature bioceramics. Two major types of the cements are possible. The first one is a dry mixture of two different calcium orthophosphates (a basic one and an acidic one) and the setting reaction occurs according to an acid-base reaction. The second type of the cements is when the initial and final calcium orthophosphates have the same Ca/P molar ratio. Typical examples are ACP with Ca/P molar ratio within 1.50-1.67 and α-TCP: they form CDHA upon contact with an aqueous solution [66]. Upon mixing with water, initial calcium orthophosphate(s) are dissolved and precipitated into less soluble calcium orthophosphates, which causes the cement setting. During the precipitation reaction, new crystals grow and become entangled,

thus providing a mechanical rigidity to the cement. Setting of these cements occurs mostly within the first six hours, yielding an 80% conversion to the final products and a compressive strength of 40-60 MPa. The rate of hardening is influenced by a powder to liquid ratio and addition of other chemicals. Despite a large number of formulations, all calcium orthophosphate cements can only have two different end products: CDHA and DCPD [66].

Calcium orthophosphate cements are biocompartible, bioactive and bioresorbable. The first animal study on calcium orthophosphate cements was performed in 1991: a TTCP + DCPA cement was investigated histologically by implanting disks made of this cement within the heads of nine cats [67, 68]. The structure and composition of the hardened cements is close to that of bone mineral; therefore, they can easily be used by bone remodeling cells for reconstruction of damaged parts of bones [66]. The biomechanical evaluation of calcium orthophosphate cements for use in vertebroplasty might be found elsewhere [69]. Unfortunately, the cements possess a low mechanical strength; this property might be improved by reinforcement with polymers. A good adaptation to the defect geometry is the major advantage of bone cements, when compared to implantation of bulk ceramics and scaffolds [66].

Injectable bone substitutes (IBS), made of calcium orthophosphate bioceramic powders and an aqueous solution of a hydrophilic biodegradable polymer, are also well-known [70-76]. They look like pastes of high viscosity but possessing enough fluidity to be injected into bone defects by a standard syringe with a needle. Creation of the required level of viscosity to prevent IBS from segregation and phase separation during the shelf life is the major task of the polymer in IBS, while calcium orthophosphates is the building material for bone healing. In terms of application, IBS more or less similar to the aforementioned calcium orthophosphate cements but, unlike the cements, IBS do not possess the self-setting abilities since no chemical reactions occur between the components [77]. Besides, there are paste-like formulations consisting of a suspension of pure HA in water prepared by a wet chemical reaction [78-80]. Recently, injectable and macroporous calcium orthophosphate cement scaffold, combining the advantages of IBS and bone cements, has been developed [81]. The future development of both IBS and calcium orthophosphate bone cements is seen in introduction of living cells into their compositions [74, 82].

Coatings of calcium orthophosphate bioceramics on metals are often applied in medicine [83]. Metallic implants are encountered in endoprostheses (total hip joint replacements) and artificial teeth sockets. The requirement for a sufficient mechanical stability necessitates the use of a metallic body for such devices. As metals do not form a mechanically stable link between the implant and bone tissue, ways have been sought to improve the mechanical contact at the interface [84, 85]. The major way is to coat the metal with a surface layer of calcium orthophosphate bioceramics that exhibits bone-bonding ability between the metal and bone [86]. The list of various coating techniques is comprised in Table 2, while the main advantages and drawbacks of each coating technique, as well as the important properties of the deposed calcium orthophosphate bioceramics, are discussed in details elsewhere [17, 83, 87-90]. Clinical results for HA-coated metallic implants revealed that they had much longer life times after implantation than uncoated devices. Namely, HA bioceramic coating as a fixation system of hip implants was found to work well in a short to medium terms (8 years [91], 15 years [92], 17 years [93] and 19 years [94]). The long-term results are awaited with a great interest. The biomedical aspects of osteoconductive coatings for total joint arthroplasty have also been reviewed [95].

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. Researchers therefore have sought ways of combining calcium orthophosphate bioceramics with other materials to tailor properties such as strength and elasticity to meet system requirements. This has led to a large variety of bone substituting composites and hybrid biomaterials made of calcium orthophosphate bioceramics and (bio)organic compounds (usually, polymers, preferably, biodegradable ones). This approach appeared due to the poor mechanical properties (namely: low elasticity, high brittleness, low tensile strength, low fracture toughness and poor impact resistance) of bone substitutes made of calcium orthophosphate bioceramics only [14, 15, 32]. In addition, it is worth mentioning that all biologically formed calcified tissues (bones, teeth, antlers, shells, *etc.*) appear to be very complicated composites of organic and inorganic phases [7-9,

54, 65]. In such composites, the mineral component provides the strength whereas the organic component contributes to the ductility. This combination of strength and ductility leads to an energy absorption prior to failure [96]. A list of the suitable calcium orthophosphate bioceramics (except of MCPM and MCPA – both are too acidic and, therefore, are not biocompatible) is mentioned in Table 1, while there is an even greater choice of biocompatible polymers [97]. Various ways have been already realized to bring these two major components together into composites, like simple mechanical mixing or co-precipitation. Usually, powder forms of calcium orthophosphate bioceramics are used to produce composites. It is also possible to introduce porosity into such composites that is advantageous for most applications as bone substitution material. Such composites might possess the unique properties; for example, there is a report on shape memory properties of poly(D,L-lactide)/HA composites [98].

The topic of the composite materials made of calcium orthophosphate bioceramics and organic/biological compounds was first introduced in 1981 by Prof. William Bonfield, who realized the application potential of calcium orthophosphates as fillers in polymer-bioceramics composites and the move was envisaged towards an improved mechanical performance of HA bioceramics [99]. Composites of polymers and calcium orthophosphate bioceramics can confer favorable mechanical properties, including strength due to the ceramic phase, toughness and plasticity due to the polymer phase, and graded mechanical stiffness. Another advantage of such biomaterials is that they are sufficiently soft and ductile to be shaped by a surgeon in the operating theatre. Nowadays, the synthesis of various types of calcium orthophosphate-based composites and hybrid biomaterials is a strong and very promising research area and the readers are referred to other reviews [17,100-108].

To conclude the subject, the bioactivity mechanism of calcium orthophosphate bioceramics should be described. Strange enough but careful seeking in the literature resulted in only one publication [87], where this mechanism has been briefly described. Therefore, the researchers should rely on the bioactivity mechanism of other inorganic biomaterials, namely of bioactive glasses – the concept introduced by Prof. Larry Hench [14, 15]. The mechanism of bonding of bioactive glasses to living tissue involves a sequence of 11 successive reaction steps. The initial 5 steps occurred on the surface of biomaterials are "chemistry" only, while the remaining 6 steps belong to "biology" because the latter include colonization by osteoblasts, followed by proliferation and differentiation of the cells to form a new bone that had a mechanically strong bond to the implant surface (Fig. 2). Therefore, there is an opinion that in the case of bioactive glasses the border between "dead" and "alive" is located between stages 5 and 6.

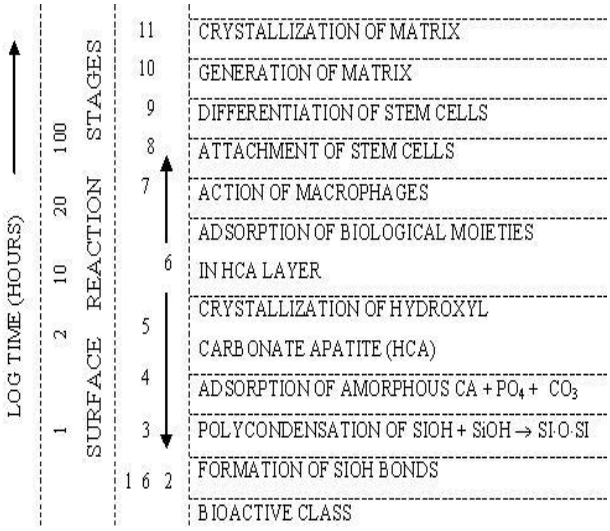

Fig. 2. The sequence of interfacial reactions involved in forming a bond between tissue and bioactive glasses. The border between "dead" and "alive" occurs approximately at stage 6. Reprinted from Ref. [15] with permission.

According to Hench, all bioactive materials "form a bone-like apatite layer on their surfaces in the living body, and bond to bone through this apatite layer. The formation of bone-like apatite on artificial material is induced by functional groups, such as Si–OH (in the case of biological glasses), Ti–OH, Zr–OH, Nb–OH, Ta–OH, –COOH, and – H_2PO_4 (in the case of other materials). These groups have specific structures revealing negatively charge, and induce apatite formation via formations of an amorphous calcium compound, *e.g.*, calcium silicate, calcium titanate and ACP" [14, 15]. For want of anything better, the bioactivity mechanism of calcium orthophosphate bioceramics should also be described by Fig. 2 with omitting of several initial stages, as it was actually made for HA in Ref. [87], where 3 initial chemical stages of the Hench's mechanism were replaced by partial dissolution of HA.

Conclusions and perspectives

At the end of the XX-th century, it became clear that calcium orthophosphate bioceramics by themselves could not give a complete response to the clinical needs of biomaterials for implants. Bioceramics with more demanding properties were required. Namely, in 1998, Prof. Larry Hench published a forecast for the future of biomaterials development [109], where he noted that calcium orthophosphate bioceramics, bioactive glasses and glass ceramics had already improved prostheses lifetime but, unfortunately, any type of prosthesis had mechanical limitations. As the solution, he proposed that biomaterial researchers would need to focus on tissue regeneration instead of tissue replacement. A working hypothesis was announced: "Long-term survivability of prosthesis will be increased by the use of biomaterials that enhance the regeneration of natural tissues" [109]. One path to follow is the regeneration of bone using calcium orthophosphate scaffolds that mimic the structure of biological apatite, bond to bone and in some cases activate the genes within bone cells to stimulate new bone growth [51, 110, 111].

However, what can be said about the future of calcium orthophosphate bioceramics? The major questions on preparation, sintering and scaffold production from the stoichiometric calcium orthophosphates have been answered in the XX-th century. Similar topics for DCPD and CDHA have been investigated in the field of calcium orthophosphate cements [66]. Conversely, calcium orthophosphates of biological origin, including the control of their morphology and interaction of calcium orthophosphate bioceramics with various bio- and organic compounds are not well investigated yet. Small amounts of bone-like apatite powder might be easily prepared by crystallization from SBF and rSBF but what can be said about larger quantities? A standard way of the concentration increasing causes chemical changes in the precipitates. After a necessary technology has been developed, one will have to think on scaffold preparation from this material, keeping in mind that any thermal treatment would destroy bone-like apatite.

Nowadays, the synthesis of various types of hybrid bioceramics, perhaps, is the strongest subject of research. For example, even bioceramic composites of HA with carbon nanotubes already exist [112,113]! In addition, a great attention is paid to manufacturing of calcium orthophosphate cements, multiphase mixtures mimicking as closely as possible the mineral component of biological apatite and the production of calcium orthophosphate bioceramic scaffolds for cells and biochemical factors to be used in tissue engineering. The study of nanostructured and nanocrystalline calcium orthophosphate-based bioceramics, similar to the complex hierarchical structures of hard tissues (bone and teeth), is also a very attractive field [13]. A work along the ecological ways of synthesis of calcium orthophosphate bioceramis might be of a great importance as well [114].

In a close future, the foreseeable application of calcium orthophosphate bioceramics will be as a component of the third generation biomaterials [109,111]. In these biomaterials, the bioceramic component will support cells and/or other biologically active substances (peptides, growth factors, hormones, drugs, *etc*.) to guide regeneration of hard tissues [6, 115-119].

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