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# Calcium phosphates in biomedical applications: materials for the future?

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**Our populations are aging. Some experts predict that 30% of hospital beds will soon be occupied by osteoporosis patients. Statistics show that 20% of patients suffering from an osteoporotic hip fracture do not survive the first year after surgery, all this showing that there is a tremendous need for better therapies for diseased and damaged bone. Human bone consists for about 70% of calcium phosphate (CaP) mineral, therefore CaPs are the materials of choice to repair damaged bone. To do this successfully, the process of CaP biomineralization and the interaction of CaPs and biological environment in the body need to be fully understood. First commercial CaP bone graft substitutes were launched 40 years ago, and they are currently often regarded as 'old biomaterials' or even as an 'obsolete' research topic. Some even talk about 'stones'. The aim of this manuscript is to highlight the tremendous improvements achieved in CaP materials research in the past 15 years, in particular in the field of biomineralization, as carrier for gene or ion delivery, as biologically active agent, and as bone graft substitute. Besides an outstanding biological performance, CaPs are easily and inexpensively produced, are safe, and can be relatively easily certified for clinical use. As such, CaP materials have won their spurs, but they also offer a great promise for the future.**

## Introduction

Calcium phosphates (CaPs; Table 1) are the main constituents of bone and teeth and play as such an essential role in our daily lives. Following the logic that damaged tissue can best be repaired by something with close resemblance, biomaterials based on CaPs were already proposed for fracture treatment in 1920 [1]. CaP biomedical research soared in the 1970s and CaPs were proposed for a broad range of orthopedic and dental applications [2–6] (Table 2). These materials varied from thin coatings on metallic implants to aid implant fixation into bone [7] to sintered CaP to be used as synthetic bone graft substitutes [8]. Truly impressive clinical successes have been achieved with such materials, for example to increase the clinical survival rate of the femoral component of total hip implants [9], to reduce the risk of pin loosening for external fixators [10], or to allow earlier weight bearing after tibia plateau fractures [11]. In some cases, CaPs are even superior to autografts [12]. Nevertheless, all these achievements have become somewhat overshadowed by the advances in the field of polymers for biomedical applications that seem endlessly diverse when it comes to control of composition and related properties (e.g. co-polymers, supramolecular self-assemblies), applicable processing techniques (e.g. additive manufacturing) and functionalization possibilities (e.g. surface micro-and nanostructuring, chemical functionalization).

In the perspective of these recent developments in the field of biomaterials, which have been underlined in a large number of recent review articles (Table 3), the question arises whether CaPs are old biomaterials, functional, but not particularly elegant? Or do they stand the chance to become the materials of the future?

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TABLE 1

**Main calcium orthophosphate compounds (taken from [143]). The first 6 compounds precipitate at room temperature in aqueous systems. The last 6 compounds are obtained by thermal decomposition or thermal synthesis. The 6 columns contain the name, the chemical formula, the Ca to P molar ratio, the mineral name, and the typical acronym, respectively. When $x > 0$ in the chemical composition of 'precipitated hydroxyapatite', one talks also about 'calcium-deficient hydroxyapatite' (CDHA). Generally, $x = 1$ so that CDHA has in most cases the composition $Ca_9(HPO_4)(PO_4)_5OH$.**

| Name | Formula | Ca/P | Mineral | Symbol |
|---|---|---|---|---|
| Monocalcium phosphate monohydrate | $Ca(H_2PO_4)_2{\cdot}H_2O$ | 0.50 | – | MCPM |
| Dicalcium phosphate | $CaHPO_4$ | 1.00 | Monetite | DCPA |
| Dicalcium phosphate dihydrate | $CaHPO_4{\cdot}2H_2O$ | 1.00 | Brushite | DCPD |
| Octocalcium phosphate | $Ca_8H_2(PO_4)_6{\cdot}5H_2O$ | 1.33 | – | OCP |
| Precipitated hydroxyapatite[a] | $Ca_{10-x}(HPO_4)_x(PO_4)_{6-x}(OH)_{2-x}$ | 1.33–1.67 | – | PHA |
| Precipitated amorphous calcium phosphate | $M_u(Ca_3)(HPO_4)_{3v}(PO_4)_{3y}{\cdot}zH_2O$[b,c] | 0.67–1.50 | – | ACP |
| Monocalcium phosphate | $Ca(H_2PO_4)_2$ | 0.50 | – | MCP |
| α-Tricalcium phosphate | $\alpha$-$Ca_3(PO_4)_2$ | 1.50 | – | α-TCP |
| β-Tricalcium phosphate | $\beta$-$Ca_3(PO_4)_2$ | 1.50 | – | β-TCP |
| Sintered hydroxyapatite | $Ca_{10}(PO_4)_6(OH)_2$ | 1.67 | Hydroxyapatite | SHA |
| Oxyapatite | $Ca_{10}(PO_4)_6O$ | 1.67 | – | OXA |
| Tetracalcium phosphate | $Ca_4(PO_4)_2O$ | 2.00 | Hilgenstockite | TetCP |

[a] $x$ may vary between 0 and 2.
[b] $u$ may vary between 0 and 3, $v$ may vary between 0 and 1.5, $y$ may vary between 0 and 0.667, and $z$ is unclear at this point. M is typically a monovalent cation ($Na^+$, $K^+$, $NH_4^+$) which is only present if there is an overall negative charge on the calcium phosphate.
[c] ACP produced in basic conditions has generally $u = 0$, $v = 0$, $y = 0.667$, leading to the following composition: $Ca_3(PO4)_2{\cdot}zH_2O$ where $z = 3$–4.5. In acidic conditions, $u = 3$, $v = 1.5$, $y = 0$, leading to the following composition: $M_3(Ca_3(HPO_4)_{4.5}{\cdot}zH_2O)$ where $z$ is unknown.

Unlike the large majority of both natural and synthetic polymers used in biomedical applications, CaPs are present in the human body and are thus relatively easy to certify. This advantage should not be underestimated at a time when the need for successful and yet affordable strategies for the treatment of diseases and the regeneration of malfunctioning organs and tissues is increasing at a high rate, as a consequence of an aging population in the Western world. CaPs meet these requirements; they can be produced in large quantities, against relatively low cost, they are stable and therefore available off-the-shelf. Nevertheless, their use is also associated with drawbacks, with poor mechanical properties being probably the most relevant one for application in orthopedics and dentistry. This, taken together, shows that additional efforts need to be placed to further advance biomedical strategies based on CaPs, but also that these materials deserve such efforts.

In the current review, we aim to highlight important recent developments in CaP research, divided into the topics biomineralization, nanoparticles for targeted delivery, and bone graft substitution. We also aim to provide an outlook toward the future of CaPs in biomedical applications.

## Biomineralization

Biomineralization can be described as a phenomenon in which a mineral is integrated as a functional and often structural part of living organisms, often in direct and close contact to a matrix forming protein or carbohydrate structure. The superb properties and intriguing complexity of most mineralized structures are indeed a result of the interactions between organic molecules/matrices and the mineral itself [13]. Examples of biominerals found in nature are numerous as described in detail by Lowenstam and Weiner [14]. Most common are the calcium carbonate-based biominerals like aragonite (nacre) and calcite (mussels, exoskeletons of crayfish, *etc.*), CaPs (in vertebrate bone and teeth) and silicates (plants, sea sponges) but also much rarer natural minerals exist. A great number of studies have investigated mineral synthesis under biologically relevant conditions, with the aim to explain the mechanisms behind biological mineral. Crude simplifications of the physicochemical conditions are a necessity in these studies as the complexity of the real biological environment hampers execution of mechanistic studies. In the next chapters, we will focus on developments in the field of CaP biomineralization in both biological and synthetic systems. Important discoveries in the last decade have provided us a deeper understanding of the mechanisms of biological and abiotic CaP mineralization, especially regarding the role of amorphous precursors and charged organic molecules.

### *Bone mineral*

The most prominent representative of CaP biomaterial is vertebrate bone, an intricate composite of collagen, non-collagenous proteins and mineral ordered in a distinct hierarchical fashion [13,15,16]. Bone mineral, which is often referred to as biological apatite or dahlite, is distinctly different from the geological apatite mineral. First of all, bone mineral consists of nanometer-sized platelets or needles [16], incorporated within collagen fibrils, and oriented with the *c*-axis in the direction of the fibril [17]. Additionally, it does not have the hexagonal crystal morphology of geological apatite and is also described as monoclinic apatite [18,19]. Furthermore, bone mineral contains a number of ionic substitutions such as $CO_3^{2-}$ in $OH^-$ (A-substitution) and $PO_4^{3-}$ sites (B-substitution), or $Na^+$, $Sr^{2+}$ and $Mg^{2+}$ in $Ca^{2+}$ sites. In fact, apatite is known for its ability to undergo ionic exchange with metal ions in aqueous solutions [20,21], hence explaining the high variability in bone mineral composition. Also, hydroxide, one of the primary constituents of *hydroxy*apatite, has been reported to be absent in bone mineral [22]. Finally, bone mineral is often described as poorly crystalline, which probably relates to the small size of the crystals as well as residual stresses in the crystal lattice. While amorphous calcium phosphate (ACP), a likely precursor for