Biomaterials for bone regeneration: an overview

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Abstract

This article presents a state of art on synthetic biomaterials currently applied for bone regeneration. A main function unites them is the stimulation of bone regrowth. These biomaterials have several advantages compared to those of natural origin (autograft, allograft and xenograft). A description of their physicochemical and biological properties reflects its interest for orthopedic surgery.

Introduction

In recent years, the use of synthetic bone substitutes has increased rapidly [1]. The orthopaedists and industrialists in this sector have turned their attention to synthetic materials, capable to promoting bone healing and rapidly invaded or replaced by new bone. Currently, there are few surgical therapies that do not use these biomaterials. Thanks to its numerous advantages, this therapeutic process avoids several problems compared to allogeneic or xenogeneic bone substitutes. Certainly, they avoid important blood loss inherent in graft intake and decrease the volume of transfusions.

On the one hand, the use of biomaterials from synthetic origin does not pose any risk of pathogens transmission [2]. They are thus better accepted by patients. On the other hand, they ensure a sufficient quantity to fill or replace the defective bone and avoid a second surgical approach for which there is certain morbidity: pain, risks of infection and embrittlement of the site taken. In economic terms, the use of synthetic substitutes often makes it possible to shorten hospital stays, which are often lengthened by the fact of graft intake. Thanks to their biocompatibility, they are used in oral surgery, periodontology and implantology. Before being put into service, these biomaterials are tested in vitro and in vivo for validation.

In the following, we will give a general description of the most synthetic biomaterials used actually as substitutes for bone regeneration. Given their diverse chemical components and characteristics, there is a wide variety of biomaterials. Currently, the most popular biomaterials in orthopaedics-traumatology sector are the calcium carbonates, phosphocalcic ceramics, composite materials, biopolymers, bioverres and calcium sulphates.

The phosphocalcic ceramics

Thanks to their bioactivity, phosphocalcic ceramic materials are attracting a growing interest in the academic and industrial world. They have very varied forms and often have interesting characteristics.

First, theirs chemicals compositions are close to that of the mineral phase of human bone; they associate covalent and other ionic bonds, they crystallize in different forms and act by their interface on the biological medium in contact, releasing the calcium and phosphate ions. Chemically, these ions will interact with others present on the site, and will build a precipitate of carbonate apatite; indispensable for their cohesion with the bone. Since they have some low mechanical properties, these biomaterials can't be placed in areas subjected to high loads.

Synthetic calcium phosphates are among the most widely used biomaterials in bone or dental surgery, when filling materials are required. They have already been the subject of several fundamental studies which have led to biological and clinical applications [3]. In table 1 we have presented the calcium ortophosphates, their chemical formulas, and their most commonly used Ca / P (calcium /phosphorus) atomic ratios.

Clinically, the most commonly ceramics based on phosphate and calcium, used in bone repair are hydroxyapatite, tricalcium phosphates, biphasic ceramics (BCP) and phosphocalcic cements.

Hydroxyapatite (HA)

Synthetic hydroxyapatite is inorganic compound, of the formula $Ca_{10}(PO_4)_6(OH)_2$. It is osteoconductive, low resorbable and biocompatible. Their stoichiometry has 1.67 Ca/P ratio and they are generally porous. A high porosity of this material improves the osteoconduction, but it is to the detriment of the mechanical qualities.

The synthesis of hydroxyapatite is generally carried out by precipitation under basic conditions and then, sintering at a temperature exceeding 1000ºC [5].

Tricalcium phosphates (β -TCP)

The tricalcium phosphate (β -TCP) is an anhydrous tricalcium phosphate of the formula $Ca_3(PO_4)_2$, is much more soluble than

Table 1. The different families of calcium phosphate [4].

Calcium orthophosphates	Chemical formula	Abbreviation	Ca/P ratio
Monocalcium phosphate anhydrous monohydrate	$Ca(H_2PO_4)_2$ $Ca(H_2PO_4)_2, H_2O$	MCPA MPCM	0,50 0,50
Dicalcium Phosphate Anhydrous (monetite) Dihydrate (brushite)	$Ca_2(HPO_4)_2$ $CaHPO_4, 2H_2O$	DCPA DCPD	1,00 1,00
Octocalcium Phosphate triclinique apatite amorphous	$Ca_8(PO_4)_4(HPO_4)_2, 5H_2O$ $Ca_8(HPO_4)_{2,5}(PO_4)_{3,5}(OH)_{0,5}$ $Ca_8(PO_4)_4(HPO_4)_2, nH_2O$	OCPt OCPa OCPam	1,33 1,33 1,33
Tricalcium phosphate α or β apatite amorphous	$Ca_3(PO_4)_2$ $Ca_9(PO4)_5(HPO_4)(OH)$ $Ca_9(PO_4)_6, nH_2O$	TCP(α,β) TCPa ACP	1,50 1,50 1,50
Hydroxyapatite phosphocalcic	$Ca_{10}(PO_4)_6(OH)_2$	HAP	1,67
Tetracalcium phosphate	$Ca_4(PO_4)_2O$	TTCP	2,00

HA [6]. It is produced by calcination of a mixture of apatite powder deficient in calcium (with a Ca / P ratio equal to 1.5) and naphthalene, which, after sublimation, leaves a porous structure at the origin of the osteoconductive properties of material. Heating at 900ºC of the apatitic powder leads to a β-TCP, which is stable up to 1125ºC [8, 9]. Heating above this temperature leads to a TCP α-form.

The rapid dissolution of β -TCP contributes to the local increase in calcium and phosphate ion concentrations. The high level of these ions induces a precipitation of calcium and phosphate in the form of biological apatite, which promotes mineralization of the extracellular matrix.

Experimentality was in 1920 that Albee and Morisson implanted tricalcium phosphate for the first time in rabbits to test its curative efficacy as a bone substitute.

In literature, several research works have been done to improve the biological properties of β-TCP, either by combining it with another biomaterial. Whether, it is phosphocalcic ceramic, polymer, bioverres, or by substitution of calcium ions, with antimicrobial activity such as Cu^{2+} or Zn^{2+} [10,11], or, by ions, with an inhibitory effect on the resorption of osteoclastic cells such as Sr^{2+}.

Two-Phases Ceramics (BCP)

This ceramic is the combination of hydroxyapatite (HA) and tricalcium phosphate (β-TCP) in different proportions. The HA / β -TCP ratio makes it possible to adapt the phenomenon of resorption of the implant to the kinetics of local remodeling [12]. Porosity is an essential physical characteristic of this biomaterial. It plays a preponderant role in its mechanical and biological properties.

Dicalcium phosphate dihydrate (DCPD) or brushite

Dicalcium phosphate dihydrate (DCPD or brushite) was first introduced into the world of phosphocalcic ceramics in 1987 by Mirtchi and Lemaître [13]. It is a compound of chemical formula $CaHPO_4$, $2H_2O$ of atomic ratio Ca/P=1. This material generally crystallizes in the form of platelets, but can also take on the appearance of needles.

Several authors have identified brushite as a precursor of bone mineralization, including biological apatite [15]. In the field of bone and dental reconstruction materials, brushite cements have been developed [13].

Phosphocalcic Cement

Historically, the first to propose the biomaterial phosphocalcic cement are Legeros et al. (1982) [14]. Since then, many formulations have been developed, studied and marketed [17].

The cements form a family of biomaterials characterized by their galenic formulation. They are an alternative to obtaining high-temperature ceramics. Thus, it can be considered that a cement is composed of a powdery solid phase and a liquid phase which, mixed in the right proportions, set and harden.

Different phosphocalcic cements are currently used in orthopedic or dental surgery, mineral hydraulic cements can be differentiated from polymer cements (or composite cements when they contain mineral filler). Mineral cements have two main classes: the first are apatitic such as TTCP or $CaCO_3$ and the latter are brushite, referring to DCPD or H_3PO_4.

Several phosphocalcic cements are marketed, some examples of cements and their respective compositions are given in table 2 the use of these bioceramics is limited by its low mechanical properties.

Biopolymers

The development of biopolymers has undergone a rapid acceleration in recent years. These biomaterials are formed from natural polymers or from agricultural or microbial origin.

There are generally three main families of biopolymers: natural polymers, polymers of bacterial origin and synthetic polymers.

Natural polymers

Those materials are synthesized by living beings: plants, animals and microorganisms. There are two important families in this category: polysaccharides and proteins [18].

The main sources of polysaccharides are corn, wheat and potato, producing starch. The other family consists of proteins. They come from a wide variety of sources, such as oilseed crops (rapeseed, sunflower, soybean ...), animal tissues (collagen, gelatin ...) or animal products.

Polymers of bacterial origin

These polymers are produced by bacterial fermentation. It is the "in situ" manufacture of polymers that accumulate in the cytoplasm

Table 2. Examples of commercially available phosphocalcic cements [16].

Name of cement	Manufacturer	Composition of the solid phase	Composition of the liquid phase	S/L
Bonesource	Stryker – Leibinger Corporation	TTCP + DCPA	Na_2HPO_4 + NaH_2PO_4	0,25 (g/cm³)
α – BSM (Biobon, Embarc)	ETEX	ACP + DCPD	Physiological serum	0,67 (g/cm³)
Calcibon	Biomet	αTCP + DCPA+ CaCO3 + PHA	Na_2SO_4	
Cerapaste (Primafix)	NGK Spark Plug	TTCP + DCPA	Sodium	
ChronOS Synthes		βTCP + MCPM +MgHPO_4 + MgSO_4	$Na_2H_2P_2O_7$ Hyaluronate de Sodium	
VitalOs	CalciOS	1: βTCP +Na_2H_2P_2O_7 2: MCPM +CaSO_4, 2H_2O	1 : water 2: water+ H_3PO_4	
Eurobone	Kasios	βTCP +Na_4P_2O_7	water+H_2SO_4	

of certain bacteria placed under fermentation conditions. Under conditions of phosphorus, nitrogen, or oxygen deficiencies, excess carbon sources will be biosynthesized by bacteria and accumulated as granules. Polyhydroxyalkanoates (PHAs) are polyesters of bacterial origin, susceptible to degradation by microbes. Depending on the culture and variety conditions of the microorganism used, homo-or copolyesters are formed. The PHA level can reach 80% of the weight of the microorganism [19]. According to their composition, these thermoplastic polymers can be ductile and more or less elastic.

A modern technique is an alternative to the use of organic solvents, based on the application of sodium hypochlorite for 30 to 60 minutes to destroy the organization of the bacterial membrane and release the polyester granules [20]. In an aerated environment, biodegradation of PHAs produces methane and carbon dioxide. The best known are PHB (Poly 3-Hydroxy Butyrate), PHV (Poly Hydroxy Valerate) and PHBV (Poly [3-Hydroxy Butyrate 3-Hydroxy Valerate]). This is a biocompatible material. It also has the advantage of being radio-translucent, thus allowing medical follow-up by imaging. In recent decades, this biomaterial has received a great deal of attention from orthopaedists as a bone implant, new bone can be formed in its contact without a chronic inflammatory response [21].

The Poly (3-hydroxybutyrate) (PHB) is the first poly (3-hydroxyalkanoates) (PHAs) discovered by Maurice Lemoigne at the Institute Pasteur in 1926. It is produced in nature by a wide variety of bacteria that store it as an energy source in a way similar to the storage of fat in humans. Since that discovery more than 150 types of monomers have been identified.

In recent years, clinical demand for biodegradable plastics from renewable resources has grown very rapidly.

PHBV copolymers are good examples of this type of material, belonging to the PHAs which constitute a very promising family of polymers for applications in bone repair. In the following, we will provide an overview of the manufacturing process of the PHBV and some of its properties.

This biomaterial is consisted of hydroxybutyrate (HB) units with between 0 and 24% of the hydroxyvalerate (HV) units occurring randomly throughout the polymer chain. It is naturally produced by bacteria in the form of intracytoplasmic granules, developed in complex substrates such as sugar cane molasses, vegetable oils ... [22]. It can also be produced by chemical synthesis from the polymerization of β-butyrolactone [23,24]. The PHBV copolymer is still in the forefront of commercialization.

Its properties depend on the ratio of the two monomers; it contains 3-hydroxybutanoic acid provides stiffness while 3-hydroxypentanoic acid promotes flexibility.

The biocompatibility of PHBV with bone tissue has been confirmed by a large number of research groups [25]. Baran, E.T. O. et al (2002) found that he has true biological properties in favor of bone regrowth [26]. When degraded in vivo, PHBV gives rise to D-3-hydroxybutyrate, which is already a normal constituent of human blood [27]. Other cell culture experiments in the PHBV with a porous structure [28] have shown that this biopolymer induces the proliferation of fibroblasts within it, with a growth rate similar to that observed in spongy bone.

Synthetic polymers

The best known is PLA (Poly Lactic Acid) from the polymerization of lactic acid molecules. They are called synthetic or chemical-synthetic polymers because of their mode of manufacture. In order to improve their physical and mechanical properties, the biopolymers may be composed of copolymers. This is the case of RESOMER, which is a commercial product in the form of a copolymer of the lactide acid and glycolide dimers.

Currently, these polymers are marketed in the form of powders, granules, viscous pastes, films and blocks. They possess varying mechanical qualities and degradation rates.

The medical applications of biodegradable polymers are very varied and are related to their biological, mechanical, chemical and thermal properties.

The table 3 shows some properties of the most widely used biodegradable polymers in bone substitutes.

Bioverres

The most bioverres consist of oxides or silicates, which are treated to varying degrees by a pressure effect associated with a thermal process (sintering).

The main advantage of bioverres lies in their bioactivity. Extensive experiments on primates have shown that this type of material is highly osteoconductive [29].

The bioverres have a much higher mechanical strength than those of calcium hydroxide or calcium phosphate.

Calcium Sulphate

It is a non-porous biomaterial, very resorbable (1 to 2 months), inorganic and presenting the possibility of including antibiotics. Its chemical formula is $CaSO4$; it corresponds to "plaster of Paris". It is the oldest bone substitutes. It has no osteoconductive activity and has low mechanical strength.

Composite biomaterials

The combination of biomaterials allows obtaining a range of composites with a wide variety of characteristics: mechanical strength, tenacity, bioactivity, biodegradability and biocompatibility.

The composite resorbable phosphocalcic ceramic-biopolymer materials appear as an interesting alternative to materials currently used as bone implants [30]. They have the advantage of combining the rheological and mechanical properties of the polymer matrix and the biocompatibility, bioactivity and osteoconduction of the bioceramic, thus promoting bone regrowth. Several criteria can be used in the classification of these biocomposites: the techniques used for the elaboration, the type of constituent materials, the morphology and the biological response.

Table 3. Properties of some biodegradable polymers [7]**.**

Polymer	Tg : glass transition temperature (°C)	Tf : melting temperature (°C)	E : Young's modulus (GPa)	Tensile strength (MPa)	Degradation time (mondhs)
PCL	-60	58-62	-	100-800 (fibre)	>24
PLLA	60-65	173-178	1.2-3.0 (film) 10-16 (fibre)	28-50 (film) 870-2300 (fibre)	>24
PGA	35-40	225-230	7-14 (fibre)	340-920 (fibre)	6-12
PHB	-2 – 4	120-177	-	20-43	-

Conclusion

This work highlighted the synthetic biomaterials applied as bone substitutes. This field is open to several disciplines (chemistry, biology, and mechanics).

View their property as a biocompatible biomaterial, bioactive and osteoconductive, the phosphocalcic ceramic category occupies the largest share in orthopedic surgery. Biopolymer, also, have considerable value given their mechanical property.

References

1. M Scacchi (2000) "The development of the ITI dental implant system". Part 2: 1998-2000: Steps into the next millennium. *Clin Oral Implants Res*; 11 Suppl 1: 22-32. [Crossref]

2. MJ Joyce, RM Wilkins, SA Brubaker (2009) " Managing musculoskeletal tissues in the hospital/surgical Center". *CTBS, AAOS Now*. [Crossref]

3. K DE Groot (1983) "Ceramic of calcium phosphates: preparation and properties", Bioceramics of Calcium Phosphate, De Groot K. ed, CRC Press, Boca Raton, p.100-114. [Crossref]

4. JC Elliot (1994) "Structure and chemistry of the apatite and other calcium orthophosphates Series", Amsterdam, Elsevier. Editor. [Crossref]

5. RZ Legeros (2002) "Properties of osteoconductive biomaterials: calcium phosphates". *Clin Orthop Relat Res*. (395): 81-98. [Crossref]

6. Daculsi G, Legeros RZ, Nery E, K. Lynch et B. Kerebel (1989) "Transformation of biphasic calcium phosphate ceramics in vivo: ultrastructural and physicochemical characterization". *J. Biomed.Mater. Res.,* 23: 883-94. [Crossref]

7. PS Eggli, W Muller et R.K. Schenk (1988) "Porous hydroxyapatiteand tricalcium phosphate cylinders with two different pore size ranges implanted in the cancellous bone of rabbits. A comparative histomorphometric and histologic study of bony ingrowth and implant substitution". *Clin. Orthop*., 232, 127-38. [Crossref]

8. JH Welch, W Gutt (1961) "High-temperature studies of the system calcium oxyde-phosporus pentoxide"; *J. Chem. Soc.*, pp. 4442-4444. [Crossref]

9. C Rey, C Combes, C Drouet and S Somrani (2008) "Tricalcium phosphate-based ceramics', in 'Bioceramics and their clinical applications', (ed. T. Kokubo), England, Woodhead Publishing.: 326-366. [Crossref]

10. N Matsumoto, K Sato, K Yoshida, K Hashimoto, Y Toda (2009) "Preparation and characterization of beta-tricalcium phosphate co-doped with monovalent and divalent antibacterial metal ions". Acta Biomater; 5: 3157-64. [Crossref]

11. K Yoshida (2006) "Substitution Model of Monovalent (Li, Na, and K), Divalent (Mg), and Trivalent (Al) Metal Ions for β-Tricalcium Phosphate". *J.Am.Ceram.soc*.89: P .688-690. [Crossref]

12. G Daculsi, O Laboux, O Malard, P Weiss (2003) "Current state of the art of biphasic calcium phosphate bioceramics". *J Mater Sci Mater* Med; 14: 195–200. [7]. [Crossref]

13. AA Mirtchi, J Lemaître, N Terao (1989) "Calcium phosphate cements: study of the β-tricalcium phosphate– monocalcium phosphate system", *Biomaterials* 10: 475-480. [Crossref]

14. Legeros RZ (1991) Calcium Phosphates in Oral Biology and Medicine. Vol. 15.

15. MJ Glimcher, LC Bonar, MD Grynpas, WJ Landis, AH Roufosse (1981) "Recent studies of bonemineral: is the amorphous calcium phosphate theory? "*J. Crystal Growth. 53*: 100-119. [Crossref]

16. H Monma, T Takahashi, H Ushio, S Soeda, T Kiyosawa (1988) "Preparation and properties of porous apatite by the hydration and hardening method", *Gypsum and Lime* 212: 25-28. [Crossref]

17. AA Mirtchi, Lemaître J, Terao N (1989) Calcium phosphate cements: study of the β-tricalcium phosphate monocalcium phosphate system, Biomaterials 10: 475-480. [Crossref]

18. ES Stevens (2007) Green Plastics: An Introduction to the New Science of Biodegradable Plastics »[Crossref]

19. Liu, Y. et Wang, M (2007) Thermophysical and mechanical properties of β-tricalcium phosphate reinforced polyhydroxybutyrate and polyhydroxybutyrate-co-hydroxyvalerate composites. Engineering Materials, v. 334-335 II, p. 1217-1220. [Crossref]

20. Von Y. Doi (1990) "Microbial polyesters", *Wiley- Verlagsgesellschaft, Weinheim*. IX, 156 S., DM 74.00, ISBN 3-527-27860-5.

21. GQ Chen, QT Wu (2005) "The application of polyhydroxyalcanoates as tissue engineering materials". *Biomaterials*, vol. 26, pp. 6565-6578. [Crossref]

22. RW Lenz, RH Marchessault (2005) "Bacterial Polyesters: Biosynthesis, Biodegradable Plastics and Biotechnology". *Biomacromolecules*, 6: 1–8. [Crossref]

23. DS Rosa, NT Lotto, DR Lopes, and CGF Guedes (2004) "The use of roughness for evaluating the biodegradation of poly- β-(hydroxybutyrate) and poly-β-(hydroxybutyrate-co-β-valerate*). " Polymer Testing* 23: 3-8. [Crossref]

24. Y Doi, Y Kanesawa, Y Kawaguchi and M Kunioka (1989) "Hydrolytic degradation of microbial poly (hydroyyalkanoates). Makromolecular. *Chemical Rapid Communication*, 10: 227-230.

25. G Torun Kosea, H Kenara, N Hasırcıb, V Hasırcıa (2003) Macroporous poly (3-hydroxybutyrate-co-3-hydroxyvalerate) matrices for bone tissue engineering, *Biomaterials.* 24: 1950 p. [Crossref]

26. Baran ETO, N Zer and V Hasrc (2002) Poly (3-hydroxybutyrate-co-3-hydroxyvalerate) nanocapsule as enzyme carriers for cancer therapy: an in vivo study. *J. Microencapsul* 19: 363-76.

27. RN Reusch (1995) Low molecular weight complexed poly (3-hydroxybutyrate): a dynamic and versatile moleculein vivo. *Can J. Microbiol* 41: 50-4. [Crossref]

28. Rivard CH, C Chaput, S Rhalmi and A Selmani (1996) Bio-absorbable synthetic polyesters and tissue regeneration. A study of three-dimensional proliferation of ovine chondrocytes and osteoblasts,Ann. Chir 50: 651-8 [Crossref]

29. H Ohgushi, Y Dohi, T Yoshikawa, S Tamai, S Tabata, K Okunaga, T Shibuya (1996) Osteogenic differenciation of cultured marrow stromal stem cells on the surface of bioactive glass ceramics. *J. Biomed. Mater. Res* 32: 341-348. [Crossref]

30. KS Jack, S Velayudhan, P Luckman, M Trau, L Grøndahl, J Cooper-White (2009) The fabrication and characterization of biodegradable HA/PHBV nanoparticle-polymer composite scaffolds. *Acta Biomater* 5: 2657-67. [Crossref]