Properties of commonly used calcium phosphate cements in trauma and orthopaedic surgery

Technical note

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Abstract

Since the introduction of calcium phosphate cements (CPCs) in 1985, a number of CPCs became predominantly commercially available for use in trauma and orthopaedic surgery. The aim of this technical note was to provide the relevant knowledge about CPCs that may improve the selection of CPCs for bone defects encountered in trauma and orthopaedic surgery. This includes a classification based on the chemical composition, and details about structural, mechanical and biological properties. Furthermore, the biological performance of each CPC was assessed in an animal study. And finally, a systematic literature search was conducted to provide a comprehensive overview of currently available clinical literature of these CPCs in trauma and orthopaedic surgery.

Keywords

biocompatibility, bone defects, bone graft, calcium phosphate cement, osteoconduction

Introduction

Halve of the population sustains at least one fracture during their lifetime[1], and the majority of these fractures heal successfully. Successful fracture healing requires the following five elements; (i) osteogenic cells (*e.g.*, osteoblasts), (ii) osteoinductive stimuli (*e.g.*, bone morphogenetic proteins); (iii) an osteoconductive matrix; (iv) adequate blood and nutrient supply, and (v) sufficient mechanical support[2]. One or more elements can be compromised due to the existence of a bone defect. Bone defects are treated with bone grafts in order to avoid insufficient fracture healing. Insufficient fracture healing is encountered in 5 to 10% of the fractures, resulting in delayed union, malunion, or non-union[3].

Most commonly used bone graft material is autologous bone. Autologous bone is usually harvested from the iliac crest[4]. However, harvesting autologous bone lengthens the surgical procedure and is associated with complications in 8-39% of patients (*e.g.*, infection, nerve and urethral injury, and postoperative or chronic pain)[5]. An alternative option is to use allogeneic bone. However, allogeneic bone grafting is considered suboptimal since it has biocompatibility disadvantages[6]. The disadvantages of autologous and allogeneic bone grafts have resulted in the development of alternative (synthetic) bone substitute materials. The most interesting group of bone substitute materials is probably calcium phosphate (CaP), because CaPs mimic the mineral part of natural bone[7].

CaP-based bone substitutes have been studied for several decades[8] and they can be categorized into CaP ceramics and CaP cements (CPC)[7]. CaP ceramics are obtained by thermal treatment (sintering), generally resulting in solid grafting materials (*e.g.*, blocks or granules). CPCs, on the other hand, consist of CaP powder that forms a paste upon mixing with a liquid. Mixing is usually done during surgery and the resulting paste becomes solid within several minutes through an isothermic chemical setting reaction. During this setting

reaction, the CPCs can be moulded by the surgeon into any desired shape in order to completely fill the defect.

Since the introduction of CPC in 1985 by Brown and Chow, a number of CPCs have become commercially available for trauma and orthopaedic surgery[9]. Trauma or orthopaedic surgeons are therefore given the opportunity to select a CPC that meets the specific demands of each bone defect that requires bone grafting. The aim of this technical note was to provide a comprehensive overview of knowledge of commonly used CPCs that may improve this selection for bone defects encountered during fracture treatment. The chemical composition and structural, mechanical and biological properties of the four predominantly used CPC products (Table 1) are discussed. In addition, the biological properties were evaluated in a large animal bone defect model. Finally, a systematic literature search was conducted in order to provide insight into the available clinical evidence supporting to use of these CPC products to graft bone defects encountered during fracture treatment.

Calcium phosphate cements

CPCs can be classified based upon the setting product of the CPC into apatite-forming and brushite-forming CPCs. This classification refers to the degree of crystallinity of the CaP formed and has a direct impact on their resorption rate. The resorption of an apatite-forming CPCs is very limited and takes many years, whereas the resorption of brushite-forming CPCs takes place within several months[7]. Commercially available apatite-forming CPCs are BoneSource®, Calcibon®, and Norian SRS®, a brushite-forming CPC is ChronOS™ Inject (Table 2). CPC products are available in several quantities, ranging from 1.5 cc to 30 cc (Table 1), and depending on the CPC product the setting reaction will take five (Calcibon®) or fifteen minutes (Norian SRS®) to be completed once the components are mixed (Table 2).

It is necessary to distinguish CPCs from ceramic CaPs. The ceramic CaPs are acquired from heating (sintering) of calcium phosphate powder up to 800-1300 ºC. This heating results in formation of solid porous blocks or granules that can consist of hydroxyapatite (HA), tricalcium phosphate (TCP) or a combination HA and TCP (composites). Examples of commercially available CaP ceramics are BoneSave®, Camceram®, Cerabone®, ChronOS™, Endobon®, Ostim®, Pro Osteon 500®, and Vitoss® [10].

Structural properties

CPCs are generally described as being osteoconductive[11]. Osteoconduction is defined as the ability of a graft material to function as a scaffold that allows ingrowth of bone and vascular tissues[12]. A scaffold that offers an open porous structure with pore dimensions of 150-500 µm is considered optimal for bone ingrowth[12] . CPCs generally form dense structures with limited porosity (Table 3). *In vitro* micro-CT measurements indicate that the porosity of ChronOS™ Inject is only 5±1 %, and that the porosity of apatite-CPCs does not exceed 5 % (BoneSource® 0.4±0.3 %, Calcibon® 0.9±0.5 %, Norian SRS® 0.3±0.2 %)[13]. Furthermore the majority of these pores is smaller than 150 µm. Average pore size of ChronOS™ Inject is 100 µm, and 18 % of these pores exceed the size of 150 µm. Average pore sizes of other apatite-CPCs is ~50 µm and the fraction that exceeds the size of 150 µm is less than 4 %.

Mechanical properties

The mechanical properties of a bone substitute material should ideally be comparable to the bone being replaced. However, the mechanical properties of bone differ according to their structure (cortical or trabecular bone) and function (weight-bearing or non-weight-bearing). Compression strength of human cortical bone is in the range of 130 to 190 MPa, whereas trabecular bone has a compression strength of 8 to 38 MPa. Apatite-CPCs were found to have

a compression strength in the range of trabecular bone (Table 3); Calcibon® has the highest compression strength (34±7 MPa), followed by Norian SRS® (26±7 MPa) and BoneSource® (14±3 MPa). The brushite-CPC ChronOS™ Inject has a compression strength that is much lower and does not exceed values of 1 MPa[13].

Biological properties

Biological properties of four CPCs were studied in a drill-hole tibia bone defect model in goats[14]. This model allows for a direct comparison of different CPCs. Three months after grafting the drill-hole defects with different CPCs, resorption and bone formation were determined using micro-CT scanning, histology, and fluorochrome labeling (Supplementary Materials and Methods). The study was approved by the institution's Animal Ethics Committee (EUR1540).

The micro-CT scans acquired at three months after grafting showed that drill-hole defects remained adequately filled by apatite-based CPCs (BoneSource®, Calcibon®, and Norian SRS®). This indicates limited resorption of apatite-CPCs whereas using ChronOS™ Inject resulted in a partially remaining bone defect (Figure 1); the latter is most likely the result of fast resorption of this brushite-CPC. Detailed views of defects with CPCs showed that it is difficult to distinguish CPC from native bone as the radiographic density is very close (BoneSource®, Norian SRS®) or even similar (Calcibon®, ChronOS™ Inject) to native bone. These similar radiographic densities obstruct a quantitative analysis from these micro-CT images of newly formed bone and remaining CPC volumes.

Histology confirmed that resorption rate of apatite-CPCs is very limited and that, as a result, these CPCs remained largely intact (Figure 2). Calcibon® still covered almost the complete defect and hardly any bone ingrowth had appeared within three months. Furthermore little activity of bone formation at the CPC-interface, indicated by the

fluorochrome labels, was found in defects grafted with Calcibon®. Although BoneSource® also covered the majority of the defect, some resorption did take place within the three months follow-up. The fact that BoneSource® resorbed slowly was also shown by Welch *et al*[15]. In their study, subchondral femoral bone defects in goats were grafted with BoneSource®, and 38 % of the CPC was still present after two years. A similar resorption pattern was also seen for Norian SRS®. Apelt *et al.* indicated that approximately 5 % of Norian SRS® was resorbed at six months after grafting a comparable subchondral bone defect in sheep[16]. The minimal appearance of fluorochrome labels at CPC-interface of tibia defects grafted with BoneSource® and Norian SRS® also indicate little bone forming activity (Figure 2).

Histology of the only brushite-based CPC, ChronOS™ Inject, indicated indeed that most of CPC was resorbed within three months time. This was also shown by Apelt *et al.*, who found that only 20 % of ChronOS™ Inject was still present six months after grafting subchondral bone defects in sheep[16]. In the periphery of the defect, ChronOS™ Inject had resorbed and was replaced by new bone. After three months, only some bits of CPC were found within the center of the defect (Figure 2). Contrary to the limited bone forming activity found for apatite-CPCs, this brushite-CPC seemed to elect a bone formation activity almost throughout the complete CPC-interface (Figure 2).

Clinical evidence in fracture treatment

A systematic literature search was conducted using the product names of all products (Table 1) as search terms in PubMed database. PubMed database was searched from the earliest date available until October 26, 2012. The following filters were used: species (human), languages (Dutch, English, or German), search fields (Text word). Retrieved manuscripts were only included when they contained original research on CPC use in trauma and orthopaedic related

indications. Manuscripts were excluded when they only contained data describing *in vitro* (*e.g.* cellular response or cadaver studies) or animal experiments, or when the CPC was used in other than trauma and orthopaedic related indications (*e.g.* dental or craniofacial surgery). All references of included manuscripts were reviewed in order to ensure that no relevant papers had been missed.

The systematic literature search resulted in seventy-eight eligible manuscripts. Thirty-three of these manuscripts were selected for final inclusion, including one additional reference (Table 4). The included manuscripts consisted of ten clinical trials, two case-control studies, twenty case-series and one case-report. The four CPC have been used to graft bone defects encountered in treatment of humerus, radius, femur, tibia, calcaneus, vertebra, and odontoid factures (Table 5), as well as bone defect encountered during knee or hip revision surgery and treatment of endochondrale bone cysts.

BoneSource® was subjected to one multicenter prospective randomized trial[17]. Metaphyseal bone defects of thirty-eight fractures, including femur, tibia, calcaneus, humerus, and distal radius fractures were grafted with either BoneSource® or autologous bone. Adequate reduction was maintained in 83 % of the defects treated with BoneSource® versus 67 % of the defect treated with autologous bone. Furthermore, reduction and injection of BoneSource® was compared with reduction and fixation with K-wires as a treatment for displaced distal radius fractures[18]. In their study, all clinical and radiographic parameters were worse after twenty-six weeks in the BoneSource® groups, and they concluded that BoneSource® alone does not provide sufficient fixation after reduction of displaced radial fractures.

Calcibon® is mostly used for grafting defects resulting of (osteoporotic) vertebral fractures. One prospective trial[19], several prospective[20-22] and retrospective[23] case-series all

indicate that grafting with Calcibon® gives comparable results to using polymethylmethacrylaat (PMMA).

Norian SRS® is described in more than twenty manuscripts and has been used as a bone graft for filling bone voids resulting of fractures of the proximal humerus, distal radius, proximal femur, proximal tibia, or calcaneus, as well as in odontoid fractures. Keating *et al.* treated 49 lateral tibia plateau fractures with internal fixation and Norian SRS®. After one year, 95 % had good or excellent Rasmussen knee scores but also 20 % showed radiological evidence of post-traumatic osteoarthritis[24]. In a randomized controlled trial including 323 patients, percutaneous injection of Norian SRS® after closed reduction of displaced distal radius fractures resulted in accelerated rehabilitation and improved clinical outcomes after two years compared with reduction only[25]. Norian SRS® was used for grafting defects resulting from calcaneal fractures and it allowed full weight bearing as early as three weeks after open reduction and internal fixation[26]. Furthermore, the use of Norian SRS® has used in acetabular cup revision surgery (case-report[27]), knee replacement surgery (case-series[28]). One study that was designed to treat enchondromas with Norian SRS® was stopped because three of the four included patients had severe pain after curettage of enchondroma and subsequent grafting with Norian SRS® [29].

ChronOS™ Inject is indicated for use in femur, tibia, calcaneus, humerus, and radius fractures by the manufacturer, however there were no clinical studies found that describe the use of ChronOS™ Inject in this indication (Table 5). ChonOS™ Inject has been used to treat benign bone cysts in children. After treating twenty-four pediatric patients, ChonOS™ Inject was found to be safe and therefore could provide an alternative treatment for benign bone cysts in children[30].

Discussion

Commercially available CPCs have different mechanical, structural and biological properties. Mechanical, structural and biological properties are mainly explained by their chemical composition. Based on chemical composition, CPCs can be classified into apatite-forming and brushite-forming CPCs. This classification may prove helpful in selection of CPCs for bone defects at specific fracture sites. In general, apatite-forming CPCs offer more mechanical strength and have a low resorption rate. Brushite-forming CPCs, on the other hand, offer only limited mechanical strength and have a high resorption rate.

Bone graft materials should offer mechanical support to surrounding bone and soft tissues in order to facilitate fracture healing. Apatite-forming CPCs have compression strengths in the range of trabecular bone (Table 3), and may therefore be most suitable to graft defects of metaphyseal bone defect. Brushite-forming CPCs only offer minimal mechanical strength, and they should therefore only be used in situations in which sufficient mechanical stability can already be acquired using fixation hardware. Overall, CPCs do not possess sufficient mechanical strength to be used in cortical or weight-bearing bone defect.

Micro-CT data indicates that CPCs form a rather dense structure with only a limited amount of pores that may allow bone and vessel ingrowth. The working mechanism in which CPCs can support bone formation is therefore more likely based on gradual resorption of the CPC and subsequent bone formation. Resorption of CPCs is an active process mediated by osteoclasts[7]. The resorption rate is an interplay of chemical composition, structure and volume of the CPC and the availability and activity of osteoclasts. Resorption of CPC should be followed by bone formation until complete regeneration of the bone defects has occurred. The regenerative potential of the surrounding bone should therefore be taken into account, and

when rapid bone formation can be expected, fast resorbing brushite-forming CPCs, such as ChronOS™ Inject, may be preferred over apatite-forming CPCs.

CPCs are mostly studied as a bone substitute material to graft metaphyseal bone defects (Table 5). Grafted metaphyseal bone defects include fractures of the femur, tibia, calcaneus, humerus, radius, and vertebra. The majority of the studies included in the systematic literature search consist of case-series. Case-series can provide relevant knowledge about the specific indication in which a CPC can be used and the relative safety, however they are not the preferred evidence to draw well-founded conclusions on the clinical benefits of using CPCs. Furthermore, the average number of clinical studies performed with each CPC product is surprisingly low, especially for BoneSource®, ChronOS™ Inject and Calcibon® (Table 4). More clinical studies, preferably randomized controlled trials, are therefore desired and can contribute to better understanding of the potential indications and benefits of different CPC products.

Limitations

There are limitations to this work. Firstly, the selection of CPC products was based upon their availability for use in trauma and orthopaedic surgery in the Netherlands. The included products are widely available and used worldwide, which supports a wide relevance of this paper. On the other side, several other CPC products such as Norian Drillable®, HydroSet®, alpha-BSM® and Callos® were not included. Furthermore, biological properties were studied in a tibia drill-hole defect model. This model has the advantage over other models that it allows for evaluation of multiple CPCs within one animal. This makes direct comparison of the studied CPCs more relevant. On the other hand, it model might be a clinically less relevant, since CPC products are predominantly used to graft metaphyseal bone defects. Therefore, whether metaphyseal bone defects can successfully be grafted with the studied

CPC cannot be directly translated from their performance in this model. The biological properties as described here provide important clues that can help to determine which product is most suitable for each type of bone defect encountered during clinical practice. Finally, the systematic literature search may not have been able to retrieve all relevant publications of the included CPC products. This risk is minimized through performing the search with the product name as search term in two different databases (Embase and PubMed) and screening reference lists of the included articles for additional publications (one publication found). Missing publications that did not specify the product name is not considered a limitation, since this systematic search was conducted to provide insight into the available clinical evidence of the commercially available CPC products, helping trauma and orthopaedic surgeons to select the most suitable product. Mentioning the product name was therefore essential for this study.

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References

[1] Brinker MR, O'Connor DP. The incidence of fractures and dislocations referred for orthopaedic services in a capitated population. *J Bone Joint Surg Am* 2004;**86-A**: 290-7.

[2] Giannoudis PV, Einhorn TA, Marsh D. Fracture healing: the diamond concept. *Injury* 2007;**38 Suppl 4**: S3-6.

[3] Einhorn TA. Enhancement of fracture-healing. *J Bone Joint Surg Am* 1995;**77**: 940-56.

[4] Cypher TJ, Grossman JP. Biological principles of bone graft healing. *J Foot Ankle Surg* 1996;**35**: 413-7.

[5] Dimitriou R, Mataliotakis GI, Angoules AG, Kanakaris NK, Giannoudis PV. Complications following autologous bone graft harvesting from the iliac crest and using the RIA: a systematic review. *Injury*;**42 Suppl 2**: S3-15.

[6] Stevenson S, Horowitz M. The response to bone allografts. *J Bone Joint Surg Am* 1992;**74**: 939-50.

[7] Bohner M. Physical and chemical aspects of calcium phosphates used in spinal surgery. *Eur Spine J* 2001;**10 Suppl 2**: S114-21.

[8] Patka P. *Bone replacement by calcium phosphate ceramics: an experimental study*. Amsterdam: Free University Press; 1984.

[9] Larsson S, Hannink G. Injectable bone-graft substitutes: current products, their characteristics and indications, and new developments. *Injury*;**42 Suppl 2**: S30-4.

[10] Van der Stok J, Van Lieshout EMM, El-Massoudi Y, Van Kralingen GH, Patka P. Bone substitutes in the Netherlands - A systematic literature review. *Acta Biomater* 2011;**7**: 739-50.

[11] Hannink G, Arts JJ. Bioresorbability, porosity and mechanical strength of bone substitutes: what is optimal for bone regeneration? *Injury*;**42 Suppl 2**: S22-5.

[12] Blokhuis TJ, Termaat MF, den Boer FC, Patka P, Bakker FC, et al. Properties of calcium phosphate ceramics in relation to their in vivo behavior. *J Trauma* 2000;**48**: 179-86.

[13] Van Lieshout EMM, Van Kralingen GH, El-Massoudi Y, Weinans H, Patka P. Microstructure and biomechanical characteristics of bone substitutes for trauma and orthopaedic surgery. *BMC Musculoskelet Disord* 2011;**12**: 34.

[14] Ooms EM, Wolke JG, van de Heuvel MT, Jeschke B, Jansen JA. Histological evaluation of the bone response to calcium phosphate cement implanted in cortical bone. *Biomaterials* 2003;**24**: 989-1000.

[15] Welch RD, Berry BH, Crawford K, Zhang H, Zobitz M, et al. Subchondral defects in caprine femora augmented with in situ setting hydroxyapatite cement, polymethylmethacrylate, or autogenous bone graft: biomechanical and histomorphological analysis after two-years. *J Orthop Res* 2002;**20**: 464-72.

[16] Apelt D, Theiss F, El-Warrak AO, Zlinszky K, Bettschart-Wolfisberger R, et al. In vivo behavior of three different injectable hydraulic calcium phosphate cements. *Biomaterials* 2004;**25**: 1439-51.

[17] Dickson KF, Friedman J, Buchholz JG, Flandry FD. The use of BoneSource hydroxyapatite cement for traumatic metaphyseal bone void filling. *J Trauma* 2002;**53**: 1103-8.

[18] Jeyam M, Andrew JG, Muir LT, McGovern A. Controlled trial of distal radial fractures treated with a resorbable bone mineral substitute. *J Hand Surg Br* 2002;**27**: 146-9.

[19] Grafe IA, Baier M, Noldge G, Weiss C, Da Fonseca K, et al. Calcium-phosphate and polymethylmethacrylate cement in long-term outcome after kyphoplasty of painful osteoporotic vertebral fractures. *Spine (Phila Pa 1976)* 2008;**33**: 1284-90.

[20] Hillmeier J, Meeder PJ, Noldge G, Kock HJ, Da Fonseca K, et al. [Balloon kyphoplasty of vertebral compression fractures with a new calcium phosphate cement]. *Orthopade* 2004;**33**: 31-9.

[21] Libicher M, Hillmeier J, Liegibel U, Sommer U, Pyerin W, et al. Osseous integration of calcium phosphate in osteoporotic vertebral fractures after kyphoplasty: initial results from a clinical and experimental pilot study. *Osteoporos Int* 2006;**17**: 1208-15.

[22] Maestretti G, Cremer C, Otten P, Jakob RP. Prospective study of standalone balloon kyphoplasty with calcium phosphate cement augmentation in traumatic fractures. *Eur Spine J* 2007;**16**: 601-10.

[23] de Falco R, Scarano E, Di Celmo D, Grasso U, Guarnieri L. Balloon kyphoplasty in traumatic fractures of the thoracolumbar junction. Preliminary experience in 12 cases. *J Neurosurg Sci* 2005;**49**: 147-53.

[24] Keating JF, Hajducka CL, Harper J. Minimal internal fixation and calcium-phosphate cement in the treatment of fractures of the tibial plateau. A pilot study. *J Bone Joint Surg Br* 2003;**85**: 68-73.

[25] Cassidy C, Jupiter JB, Cohen M, Delli-Santi M, Fennell C, et al. Norian SRS cement compared with conventional fixation in distal radial fractures. A randomized study. *J Bone Joint Surg Am* 2003;**85-A**: 2127-37.

[26] Elsner A, Jubel A, Prokop A, Koebke J, Rehm KE, et al. Augmentation of intraarticular calcaneal fractures with injectable calcium phosphate cement: densitometry, histology, and functional outcome of 18 patients. *J Foot Ankle Surg* 2005;**44**: 390-5.

[27] Muller M, Stangl R. [Norian SRS augmentation in revision of acetabular cup of total hip arthroplasty. A follow up of six patients]. *Unfallchirurg* 2006;**109**: 335-8.

[28] Manzotti A, Confalonieri N, Pullen C. Grafting of tibial bone defects in knee replacement using Norian skeletal repair system. *Arch Orthop Trauma Surg* 2006;**126**: 594-8.

[29] Welkerling H, Raith J, Kastner N, Marschall C, Windhager R. Painful soft-tissue reaction to injectable Norian SRS calcium phosphate cement after curettage of enchondromas. *J Bone Joint Surg Br* 2003;**85**: 238-9.

[30] Joeris A, Ondrus S, Planka L, Gal P, Slongo T. ChronOS inject in children with benign bone lesions--does it increase the healing rate? *Eur J Pediatr Surg*;**20**: 24-8.

[31] Robinson CM, Page RS. Severely impacted valgus proximal humeral fractures. Results of operative treatment. *J Bone Joint Surg Am* 2003;**85-A**: 1647-55.

[32] Robinson CM, Page RS. Severely impacted valgus proximal humeral fractures. *J Bone Joint Surg Am* 2004;**86-A Suppl 1**: 143-55.

[33] Abramo A, Tagil M, Geijer M, Kopylov P. Osteotomy of dorsally displaced malunited fractures of the distal radius: no loss of radiographic correction during healing with a minimally invasive fixation technique and an injectable bone substitute. *Acta Orthop* 2008;**79**: 262-8.

[34] Lozano-Calderon S, Moore M, Liebman M, Jupiter JB. Distal radius osteotomy in the elderly patient using angular stable implants and Norian bone cement. *J Hand Surg Am* 2007;**32**: 976-83.

[35] Tyllianakis ME, Panagopoulos A, Giannikas D, Megas P, Lambiris E. Graft-supplemented, augmented external fixation in the treatment of intra-articular distal radial fractures. *Orthopedics* 2006;**29**: 139-44.

[36] Zimmermann R, Gabl M, Lutz M, Angermann P, Gschwentner M, et al. Injectable calcium phosphate bone cement Norian SRS for the treatment of intra-articular

compression fractures of the distal radius in osteoporotic women. *Arch Orthop Trauma Surg* 2003;**123**: 22-7.

[37] Kopylov P, Adalberth K, Jonsson K, Aspenberg P. Norian SRS versus functional treatment in redisplaced distal radial fractures: a randomized study in 20 patients. *J Hand Surg Br* 2002;**27**: 538-41.

[38] Kopylov P, Aspenberg P, Yuan X, Ryd L. Radiostereometric analysis of distal radial fracture displacement during treatment: a randomized study comparing Norian SRS and external fixation in 23 patients. *Acta Orthop Scand* 2001;**72**: 57-61.

[39] Kopylov P, Jonsson K, Thorngren KG, Aspenberg P. Injectable calcium phosphate in the treatment of distal radial fractures. *J Hand Surg Br* 1996;**21**: 768-71.

[40] Kopylov P, Runnqvist K, Jonsson K, Aspenberg P. Norian SRS versus external fixation in redisplaced distal radial fractures. A randomized study in 40 patients. *Acta Orthop Scand* 1999;**70**: 1-5.

[41] Tyllianakis M, Giannikas D, Panagopoulos A, Panagiotopoulos E, Lambiris E. Use of injectable calcium phosphate in the treatment of intra-articular distal radius fractures. *Orthopedics* 2002;**25**: 311-5.

[42] Sanchez-Sotelo J, Munuera L, Madero R. Treatment of fractures of the distal radius with a remodellable bone cement: a prospective, randomised study using Norian SRS. *J Bone Joint Surg Br* 2000;**82**: 856-63.

[43] Jupiter JB, Winters S, Sigman S, Lowe C, Pappas C, et al. Repair of five distal radius fractures with an investigational cancellous bone cement: a preliminary report. *J Orthop Trauma* 1997;**11**: 110-6.

[44] Mattsson P, Alberts A, Dahlberg G, Sohlman M, Hyldahl HC, et al. Resorbable cement for the augmentation of internally-fixed unstable trochanteric fractures. A prospective, randomised multicentre study. *J Bone Joint Surg Br* 2005;**87**: 1203-9.

[45] Mattsson P, Larsson S. Stability of internally fixed femoral neck fractures augmented with resorbable cement. A prospective randomized study using radiostereometry. *Scand J Surg* 2003;**92**: 215-9.

[46] Mattsson P, Larsson S. Unstable trochanteric fractures augmented with calcium phosphate cement. A prospective randomized study using radiostereometry to measure fracture stability. *Scand J Surg* 2004;**93**: 223-8.

[47] Goodman SB, Bauer TW, Carter D, Casteleyn PP, Goldstein SA, et al. Norian SRS cement augmentation in hip fracture treatment. Laboratory and initial clinical results. *Clin Orthop Relat Res* 1998: 42-50.

[48] Jubel A, Andermahr J, Mairhofer J, Prokop A, Hahn U, et al. [Use of the injectable bone cement Norian SRS for tibial plateau fractures. Results of a prospective 30-month follow-up study]. *Orthopade* 2004;**33**: 919-27.

[49] Simpson D, Keating JF. Outcome of tibial plateau fractures managed with calcium phosphate cement. *Injury* 2004;**35**: 913-8.

[50] Engel T, Lill H, Korner J, Verheyden P, Josten C. [Tibial plateau fracture--biodegradable bonecement-augmentation]. *Unfallchirurg* 2003;**106**: 97-101.

[51] Lobenhoffer P, Gerich T, Witte F, Tscherne H. Use of an injectable calcium phosphate bone cement in the treatment of tibial plateau fractures: a prospective study of twenty-six cases with twenty-month mean follow-up. *J Orthop Trauma* 2002;**16**: 143-9.

[52] Wee AT, Wong YS. Percutaneous reduction and injection of Norian bone cement for the treatment of displaced intra-articular calcaneal fractures. *Foot Ankle Spec* 2009;**2**: 98-106.

[53] Schildhauer TA, Bauer TW, Josten C, Muhr G. Open reduction and augmentation of internal fixation with an injectable skeletal cement for the treatment of complex calcaneal fractures. *J Orthop Trauma* 2000;**14**: 309-17.

[54] Harrop JS, Przybylski GJ. Use of an osteoconductive agent (Norian) in anterior surgical management of odontoid fractures. Technical note. *Neurosurg Focus* 2000;**8**: e8.

[55] Mahid SS, Hornung CA, Minor KS, Turina M, Galandiuk S. Systematic reviews and meta-analysis for the surgeon scientist. *Br J Surg* 2006;**93**: 1315-24.

Figure captions

Figure 1: Filling capability of CPCs in a drill-hole defect

Mirco-CT and corresponding Goldner's trichrome histological images. S, substitute material; B, bone. Bar indicates 1 mm.

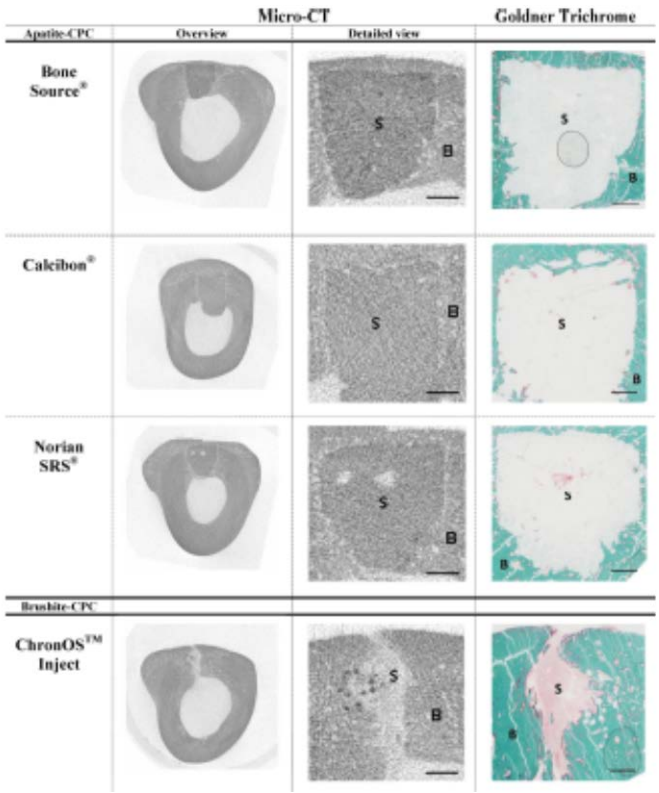
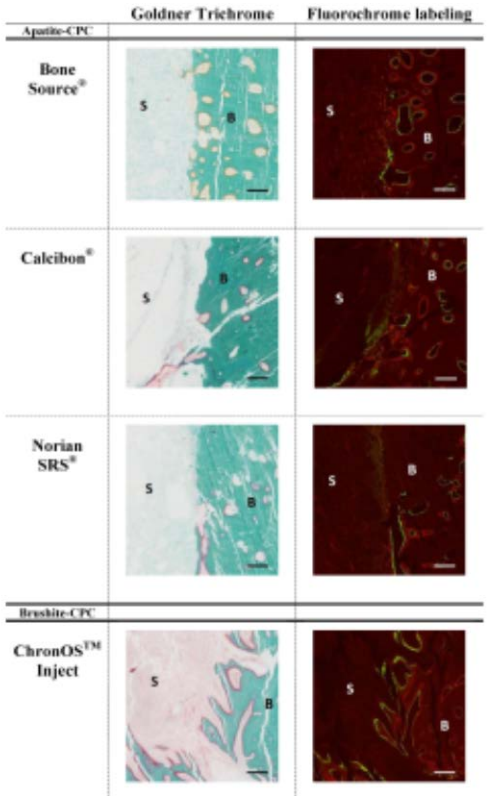


Figure 2: CPC integration and bone forming activity at the CPC-interface

Goldner's trichrome histological images of the CPC-interface and corresponding fluorochrome images. S, substitute material; B, bone. Red label is tetracycline and green label is calcein green. Bar indicates 1 mm.



Supplementary Materials and Methods

Animal experiment

In five skeletally mature female milk goats, each weighing 50-60kg, a drill-hole bone defect model was used in which three holes were created at the diaphysis of the right tibia. Two of the holes were grafted with a CPC and one was left empty, serving as a control. All products were tested in duplicate and randomized over the proximal, distal and middle holes. After three months, the goats were killed and the grafted bone defects were analyzed using micro-CT, histology, and fluorochrome labeling.

Surgical procedure

The operation was performed under general anesthesia induced by an intravenous injection of 0.3ml medetomidine (1mg/ml) followed by 20ml propofol (10mg/ml) and maintained by isoflurane 1.5% through a constant volume ventilator, administered through an endotracheal tube. The goats received prophylactic antibiotics according to the following scheme: 12.5ml amoxicillin (48mg/ml) at the start of the operation and 7.5ml ampicillin (100mg/ml) during the operation; and two and four days after the operation.

Before surgery, the animal was immobilized on its right side and the right limb was shaved, washed, and disinfected with povidone-iodine. A longitudinal incision was made on the medial surface of the tibia, and the bone was exposed by blunt dissection. Three unicortical 2.0mm diameter holes were drilled at low rotational drill speeds and continuous cooling with cold physiologic saline, with an interspace of 2.5cm. The 2.0mm diameter holes were enlarged to 5.0mm diameter and irrigated and packed with sterile cotton gauze, and the calcium phosphate cements were then prepared. Subsequently, the cements were injected into the bone defects and allowed to solidify. Any extruding material was removed. The soft

tissues and skin were closed in separate layers with Vicryl sutures. Post operative analgesia consisted of 0.8ml buprenorphine (300µg/ml) and 1.0ml flunixin (50mg/ml) on day zero to three post surgery.

Fluorochrome labeling

The fluorochrome labels were administered intravenously at 14 days (tetracycline hydrochloride, 30 mg/kg (Sigma)) and four days (calcein, 10 mg/kg intravenously (Sigma)) before they were killed using an overdose pentobarbital.

Micro-CT evaluation

The right tibias were taken out and trimmed to a suitable size for micro-CT scanning. A 9 µm-resolution protocol (75 kV energy, 133 µA current, 1.0 mm Al filter) was used with a SkyScan 1076 micro-CT scanner (Bruker micro-CT N.V., Kontich, Belgium). The CT images were reconstructed using NRecon software version 1.5 (Bruker micro-CT N.V., Kontich, Belgium).

Histological evaluation

After micro-CT scanning, each tibia was trimmed to a size suitable for histological processing. Subsequently, all specimens were fixed in paraformaldehyde (4%) overnight, dehydrated in a graded series of ethanol, and embedded in methylmethacrylate (MMA). After polymerization, undecalcified thin (6µm) sections were made with a heavy duty microtome in a transverse direction through the middle of the defect area. Sections were stained using Goldner's trichrome and evaluated with a light microscope (Olympus BX50). Fluorochrome labels were evaluated in unstained sections using an epifluorescence microscope (Axiovert 200MOT/Carl Zeiss) equipped with a double filter block.

Table 1. Product specifications

Product	Manufacturer	Available volumes
BoneSource®	Stryker	1.5 cc, 3 cc, 6 cc, 15cc, 30cc
Calcibon®	Biomet	4 cc, 8 cc, 16 cc
ChronOS™ Inject	DePuy Synthes	2.5 cc, 5 cc, 10 cc
Norian SRS®	DePuy Synthes	3 cc, 5 cc, 10 cc

Table 2. Chemical composition and mixing properties

Product	Chemical composition	Mixing time	Final setting time	Final setting product [7]
BoneSource®	80% TCP / 20% HA	2-4 m	5-10 m	apatite
Calcibon®	62.5% α-TCP / 26.8% DCPA / 8.9% $CaCO_3$ / 1.8% HA	1 m	5 m	apatite
ChronOS™ Inject	73% β-TCP / 21% MCP.H_2O / 5% MHPT	1 m	11 m	brushite
Norian SRS®	α-TCP / $CaCO_3$ / MCP.H_2O	1 m	15 m	apatite

$CaCO_3$, calcium carbonate; DCPA, dicalcium phosphate anhydrous; HA, hydroxyapatite; MCP.H_2O, monocalcium phosphate monohydrate; MHPT, magnesium hydrogen phosphate trihydrate; TCP, tricalcium phosphate.

Table 3. Structural and mechanical properties

Product	Porosity (%)[13]	Compression strength (MPa)[13]
BoneSource®	0.4	14
Calcibon®	0.9	34
ChronOS™ Inject	4.5	1
Norian SRS®	0.3	26

Table 4: Number of publications retrieved during the systematic literature search

Products	Inclusion	Exclusion					Final
	PubMed Search	In vitro experiments	Animal experiments	Different indication	Other	Additional references	
Apatite-CPC							
BoneSource®	23	9	1	11	1	1	1
Calcibon®	5	1	0	0	0	0	4
Norian SRS®	48	9	0	6	6	0	27
Brushite-CPC							
ChronOS™ Inject	2	1	0	0	0	0	1

Table 5: Clinical evidence for CPC use in fracture surgery

Products	Fracture						
	Proximal Humerus	Distal Radius	Proximal Femur	Proximal Tibia	Calcaneal	Vertebral	Odontoid
Apatite-CPC							
BoneSource®	IV[17]	IV [17, 18]	IV [17]	IV [17]	IV [17]	N.D.	N.D.
Calcibon®	N.D.	x	N.D.	x	x	II[19, 20, 22, 23]	N.D.
Norian SRS®	V[31, 32]	II[25, 33-43]	II[44-47]	V[48-51]	V[26, 52, 53]	N.D.	VI [54]
Brushite-CPC							
ChronOS™ Inject	x	x	x	x	x	N.D.	N.D.

I–VI, the highest clinical level of evidence according to Mahid *et al.* [55] supporting the use for specific indication; x, indicated by manufacturer only; N.D., no data available.