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Comparison of Apatite-Wollastonite Glass-Ceramic and β -tricalcium Phosphate used as Bone Graft Substitutes after Curettage of Bone Cysts

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1. Introduction

The need to treat the bone defects arises throughout the whole spectrum of orthopaedic surgery. It is possible to produce materials with specific mechanical properties, architecture and biodegradability. The essential attributes for regeneration of new osseous tissue include biocompatibility, osteointegration, osteoconductivity, osteoinductivity and osteogenicity. Biocompatibility means that the immunogenic response and foreign body reactions to the implanted material are minimised or absent. Osteointegration is defined as a process leading to close bonding of the newly formed mineralised tissue with the implant material. Beyond these bone regeneration essentials, other matrix properties can influence the suitability of a material for its intended clinical applications, such as surface roughness, the mechanical integrity of the matrix and the matrix's porosity [1]. Several osteoconductive bone graft substitutes are available for clinical application, including coralline hydroxyapatite, collagen-based matrices, calcium phosphates, calcium sulphate and deproteinised bovine bone. These materials vary substantially in terms of their chemical composition, mechanical properties and biodegradability. Orthopaedic surgeons should understand the differences between the various bone graft substitutes to ensure they select a material that provides the desired properties for the intended clinical application.

An ideal synthetic bone graft substitute should be a porous matrix with interconnecting porosity that promotes rapid bone ingrowth, and at the same time, it should possess a sufficient strength to prevent its crushing under physiological loads during osteointegration and healing. Hydroxyapatite ($\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$), β -tricalcium phosphate ($\text{Ca}_3(\text{PO}_4)_2$), their derivatives and combinations are the most commonly used ceramic materials in orthopaedics. While hydroxyapatite ceramic materials provide an osteoconductive matrix for bone ingrowth and ongrowth, slow in-vivo resorption profiles can potentially limit their clinical applications [2]. Although β -tricalcium phosphate ceramic has been studied substantially in animal models and its biocompatibility, osteoconductivity and resorbability have been reported, there have been only limited data regarding long-term outcome of its clinical use in surgery for bone tumours [3-12]. However, the informations about biological responses such as bone bonding and resorption of ceramics are very important in clinical applications [13-15].

The goal of this study was to analyze and compare the clinical and radiological outcome in patients after implantation of the nonresorbable oxyhydroxyapatite glass-ceramic and Beta-Tricalcium phosphate used to fill the defects of long bones after curettage of bone cysts.

2. Materials and methods

2.1 Patients

This retrospective study was approved by the local research ethics committee, and all participants gave their written, informed consent. We evaluated 39 consecutive patients who fulfilled the following inclusion criteria: (i) histologically confirmed unicameral bone cyst, (ii) treatment by curettage of the lesion and implantation of glass-ceramic or beta-tricalcium phosphate, (iii) follow-up after at least 24 months to confirm a static radiographic outcome without recurrence of benign bone lesion. There were 28 male and 11 female patients. The age of the 21 patients with the implanted nonresorbable glass-ceramic at the time of surgery was 4–44 years with an average age of 15 years. The age of the 18 patients with implanted beta-tricalcium phosphate at the time of surgery was 7–30 years with an average age of 14 years. The unicameral bone cysts were located in the humerus (16 patients), femur (13), fibula (4), tibia (2), calcaneus (2), ulna (1), and iliac bone (1). All patients had curettage of the cyst and filling of the resulting bone defect with glass-ceramic (15 male and 6 female) or tricalcium phosphate (13 male and 5 female). Cortical fenestration was carried out using an osteotome and the soft tissue membrane was removed by a curette. After the filling of the bone defect with glass-ceramic or tricalcium phosphate, the cortical window was replaced. Internal fixation was employed in four patients because a pathological fracture occurred in 3 cases and an impending fracture was in 1 patient. Splints or bandages were used postoperatively for patients judged to be at risk of pathological fracture. Full weight bearing was allowed after 8–12 weeks. The patients were scheduled for follow-up evaluations which included clinical and radiographic examinations at 4–6, 10–14 weeks, 6 and 12 months intervals. Thereafter they were seen yearly.

2.2 Filling

A bioactive glass-ceramic material with wollastonite, oxyhydroxyapatite, residual glass phase, and whitlockite at a ratio of 45–30–20–5% (BAS-0, Lasak Ltd., Prague, Czech Republic) was used. The apatite-wollastonite glass-ceramic was dense with a porosity of less than 0.5%. The sintering temperature was 1200°C, the bending strength 170 MPa and the compression strength 400MPa. Granules 2–4mm in diameter were implanted during operations. The amount of the glass-ceramic used ranged from 5 to 45 g.

β -tricalcium phosphate (Poresorb®, Lasak Ltd., Prague, Czech Republic) in granule form with particle size of 0,6 to 2 mm was implanted. The porosity of the interconnected β -tricalcium phosphate scaffold was 35 ± 5 %, the average macropore size was 100 μ m in diameter, the size of micropores was 1–5 μ m, and the sintering temperature was 1180°C. The quantity of implanted β -tricalcium phosphate ranged from 3 to 66 g.

2.3 Clinical and radiographic follow-up

Clinical and radiographic examinations of all patients were carried out at 3–12 years (7 years on average) after implantation of the bioactive glass-ceramic and at 2–8 years (4 years on average) after implantation of tricalcium phosphate. The clinical follow-up included

subjective complaints, objective findings focused on soft tissue status in the area of synthetic bone graft substitute filling, range of movement in adjacent joints and weight-bearing ability of the treated extremity. The radiographs were taken in standard projections and evaluated independently by three investigators. The results were then compared for inter-observer agreement and, in case of differences, the patient's radiograph was reviewed by all three observers together. The radiographic integration of bioactive glass-ceramic was evaluated according to the criteria of Uchida et al. [16] to determine the presence of the markedly seen bridging trabeculae at the recipient bone-implant interface and the amount of incorporation of glass-ceramic. The radiographic integration of tricalcium phosphate was classified according to the criteria of Nigro and Grace [17]. Complete integration was associated with radiographical homogeneity of host bone and bone graft substitute, and trabecular remodelling was seen. Integration was judged to be partial when the formation of a partial gap between synthetic bone graft substitute and host bone was observed. Resorption was defined as gradually decreased radiopacity of the bone graft substitute until complete resorption.

3. Results

3.1 AW glass-ceramic

The clinical evaluation showed that 10 patients had no subjective complaints, 4 reported transient pain, 3 had pain at activity, 2 reported pain during weather changes, 1 woman observed pain during pregnancy, and 1 patient had pain at rest. No restriction in weightbearing of the limb treated was reported by any of the patients. Radiographs obtained immediately after surgery showed radiolucent zones between the implanted glass-ceramic and the surrounding bone. Periodic assessments revealed that, during the period of 3–4.5 months, the radiolucent zones faded and new bone developed. Continuing radiographic observations up to a maximum of 12 years after implantation have shown no evidence of biodegradation of the apatite-wollastonite glass-ceramic material. The distinct bridging bone trabeculae bound to the surface of glass-ceramic granules were observed in all patients (Fig. 1). In the metaphyseal region of long bones, the incorporated glass-ceramic granules still remained distinct and seemed to be connected to each other. However, in the diaphyseal region of long bones, there was little distinction between the glass-ceramic granules and adjacent bone. Increased bone density around the incorporated glass-ceramic material with remodeling of the cortex in the diaphyseal region of long bones was found in six of the nine patients. No degenerative changes were encountered in adjacent joints. No postoperative infections or fractures were observed in our patients. Recurrence of bone cysts was identified in one case. The patient had further curettage of recurrence located in the area surrounding an incorporated glass-ceramic material and the cavity was filled with allogeneic cancellous bone grafts; this patient had no pain. One male with a small residual defect required no further surgery.

3.2 Tricalcium phosphate

Neither postoperative infection nor adverse reaction due to the material were encountered. No patient complained of local pain at final examination and all patients were satisfied with their limb function. Radiographs obtained immediately after surgery demonstrated radiolucent zones between the implanted tricalcium phosphate and the surrounding bone. Over time, radiolucent zones faded and new bone developed in all 18 patients. The mean

a)

b)

c)

d)

e)

Fig. 1. Anteroposterior radiograph of the left proximal femur showing a large unicameral bone cyst in a 13-year-old boy preoperatively (a); complete filling of the curetted cavity with autologous cancellous bone grafts 2 months after surgery (b); recurrence of the cyst 4 years later (c); complete filling of defect with apatite-wollastonite glass-ceramic granules 3 months after curettage of the recurrence (d); bone trabeculae bound to the glass-ceramic granules which still remained distinct 10 years after surgery (e)

period necessary for disappearance of these radiolucent zones was 9 weeks (range 5-13 weeks). Periodic radiographic assessments revealed decreased radiographic density of β -tricalcium phosphate and replacement of β -tricalcium phosphate granules by newly formed bone trabeculae. These processes appear to have started on the periphery of the synthetic filling and progressed centrally. Signs of the implanted β -tricalcium phosphate still remained radiographically at the final follow-up in all 18 cases, but the material was incorporated in the surrounding bone and gradually resorbed.

Postoperative fractures were recorded in two patients with a unicameral bone cyst in the humerus. One male fell 3 weeks after surgery; the fracture was treated conservatively. The other male patient has had a car accident 20 months after surgery; the displaced diaphyseal fracture was treated with open reduction and plate osteosynthesis. In two young patients, growth arrest or deformity were seen before curettage of the cyst and implantation of β -tricalcium phosphate. Premature closure of the physal plate with resulting shortening of the upper extremity was found as a complications after pathological fracture of proximal humerus in one male with unicameral bone cyst. Recurrences of the unicameral bone cyst

were seen only in 2 cases. Both patients had further curettage of recurrence in the area surrounding an incorporated tricalcium phosphate. In the skeletal mature male patient, the cavity was filled with autologous cancellous bone grafts; in other patient, β -tricalcium phosphate was added to the cavity at the time of repeated curettage (Fig. 2).

Fig. 2. Anteroposterior radiograph of the right proximal femur showing a large unicameral bone cyst in a 11-year-old boy preoperatively (a); complete filling of the curetted cavity with β -tricalcium phosphate after surgery (b); decreased radiographic density of β -tricalcium phosphate apparent on the periphery of filling with recurrence of the cyst localized between the implanted material and growth plate 7 months later (c); further progression of the recurrence 13 months after surgery (d); large multicameral recurrence of unicameral bone cyst in 15 years of age (e); β -tricalcium phosphate added to the cavity at the time of repeated curettage (f)

4. Discussion

Conventional bone grafting with an autologous bone harvested from the iliac crest is the current gold standard because the autografts contain marrow cells including nondifferentiated mesenchymal stem cells, osteogenic bone cells, an osteoconductive matrix and osteoinductive proteins. However, the availability of autologous bone grafts is limited, their harvesting requires a further operation with prolonged operative time and is often associated with donor-site morbidity. The processed allograft bone is an attractive alternative to the autologous bone [18]. These allogeneic bone grafts are primarily osteoconductive, but they retain a variable number of osteoinductive proteins. The host immune response to freeze-dried allografts is less vigorous than the response to fresh or fresh-frozen allografts [19]. The transmission of infectious agents from donor to recipient with allogeneic bone grafts is their major risk and a loss of mechanical properties is their important disadvantage. The infectious risk is increased when fresh allografts are used and, therefore, serological testing has to be performed to reduce that risk.

In an effort to overcome limitations of autografts and allografts, the synthetic bone graft substitutes were developed from a variety of materials, including calcium phosphates,

calcium sulphates, bioactive glasses and glass-ceramics which appear to be the ideal substances for use as matrices because the inorganic component of bone is composed of hydroxyapatite. Bioceramics can be divided into three categories: bioinert ceramics (alumina), surface-bioactive ceramics (sintered hydroxyapatite, bioactive glasses and apatite-wollastonite glass-ceramics), and resorbable bioactive ceramics (low-crystalline hydroxyapatite and tricalcium phosphate) [2].

The used dense bioactive glass-ceramic with main crystal phases, oxyhydroxyapatite, and wollastonite, when compared with beta-tricalcium phosphate ceramic, has no microporosity and nanoporosity. After the filling of the bone cavity with 2–4-mm granules of nonresorbable material, new bone tissue ingrowth was seen in the interspaces between granules [20,22]. The nonresorbable bone graft substitutes can offer an excellent permanent function in certain conditions. In contrast, these synthetic materials can also affect tissue repair and function. After osteointegration of the rigid nonresorbable material, the surrounding bone tissue is often mechanically influenced and protected (the stress-shielding phenomenon), thereby changing the local mechanical signals. This adversely affects the surrounding bone tissue. The concentration of load stress at the interface of the rigid implant-bone tissue can generally cause pain or mechanical failure [21, 22]. Equalizing to the mechanical properties of the surrounding bone is essential, because the mechanical signals are important mediators for the differentiation of connective tissue progenitor cells.

The ideal biodegradable bone graft substitutes should fulfill some requirements such as biocompatibility, adequate initial strength and stiffness, retention of mechanical properties throughout sufficient time to assure its biofunctionality and non-toxicity of the degradation by-products [23-25]. The continuous degradation of a resorbable material causes a gradual load transfer to the healing tissue, preventing stress-shielding phenomenon and stimulates the healing and remodeling of bones [22, 26]. The surgeon should be concerned with the mechanical and biological properties of the bone graft substitute as well as the handling and ability to assess healing of the grafted site.

Aside from chemical composition, the microstructure (the volume, density and size of pores and interconnections, the specific surface) plays a role in the bone ingrowth of porous biomaterials. An increase of porosity would make bone ingrowth inside synthetic materials easier but would decrease their biomechanical properties. The interconnections in a porous material act only as pathways for nutritional elements, vascularization and cells between the pores that are the sites for bone tissue growth proceeding from the outside to the inside [2]. Therefore, the size of pores should be larger than the size of interconnections [27]. Macroporosity (pore size $>50\text{ }\mu\text{m}$), microporosity (pore size $<10\text{ }\mu\text{m}$) and pore wall roughness play an essential role in new bone formation [12, 28-33]. The larger surface area can support a higher bone inducing protein adsorption, ion exchange and bone-like apatite formation by dissolution and reprecipitation while the surface roughness enhances attachment, proliferation and differentiation of bone forming cells [2, 32]. Thus, these data imply that more extensive dissolution and reprecipitation of low-crystalline calcium phosphates can cause more osteoconductive and cell-mediated degradation characteristics [34].

Resorption is an important characteristic of bone graft substitutes and can be divided in two mechanisms: solution-mediated dissolution processes and cell-mediated (phagocytic) processes. The degradation attributes of calcium phosphates are dependent on the chemical composition, crystal structure, crystal and grain size, microporosity, neck geometry, and neck dissolution rates of the materials [34]. The density of pores and interconnections which expresses the quantity of connections between pores of porous materials plays a more

important role than their size that is modified during degradation of resorbable bioceramics, whereas the sizes and the densities are equally important in unresorbable biomaterials [2, 27]. Although tricalcium phosphate was incorporated in the surrounding bone and gradually resorbed, signs of the implanted β -tricalcium phosphate still remained radiographically in all cases. In our opinion, the implanted quantity of tricalcium phosphate was higher in our patients compared to other authors [5-7, 15]. More experimental and clinical studies will be required in order to resolve the healing problems of large bone defects combining scaffolds with osteoinductive factors and cell cultures. According to this study, interconnected β -tricalcium phosphate produced better clinical results than apatite-wollastonite glass-ceramic. Interconnected β -tricalcium phosphate is a successful and safe bone graft substitute for the treatment of benign bone tumours and tumour-like lesions because of its biocompatibility and bioresorbability.

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