Microscopic Evaluation of the Natural Coral (Porites spp.) Post-implantation in Sheep Femur

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Introduction

Bone defects resulting from congenital defects, tumour or trauma are conventionally repaired using bone graft[2]. Allogenic and xenogenic bone grafts are used as alternatives but several problems are generally associated with them such as virus transfer, considerable care, high cost and regular immuno-defensive reaction[1]. For all these reasons, bone substitutes are generating growing interest and are frequently used in orthopaedic surgery.

The exoskeleton of some Madreporic corals has remarkable similarities to bone[5]. Thus, it might be appropriate as bone substitute. Previous studies showed that natural coral could be used as biomaterial for onlay grafts in orthopedics and traumatology[8], cranio facial[9] and dental[10]. This study was conducted to evaluate the resorption pattern and the replacement of natural coral by new bone post-implantation microscopically in a large bone defect.

Materials and Methods

Twelve adult, male sheep (15-20kg) were used in this study. The animals were divided into four groups (n=3). Coral blocks (2.5x0.5x0.5cm) were prepared and processed according to the protocol stated by tissue bank of Malaysian Institute of Nuclear Technologies (MINT), Bangi. The freeze-dried coral blocks were then sterilized by gamma irradiation (Model JS8900) at 25 kGy. The large bone defect (2.5x0.5x0.5cm) was created surgically at the left proximal femur on lateral aspect and filled with the coral block. The animals were euthanased at 2, 4, 8 and 12 weeks post-implantation. Samples of the implant were taken and by using the diamond saw (Exakt®, Germany) the bones were cut into small specimens for histological evaluation. The specimens were fixed in 10% neutral buffered formaldehyde. The sections were stained with Toluidine blue and Masson-Goldner Trichome and examined under light microscope.

Results

At two weeks post-implantation, all the implants showed invasion of granulation tissue accompanied by ingrowth of blood vessels into pore region. Abundant osteoblastic activity was seen directly apposed on the surface of the pores implant. Bone began to appear at 4 weeks post-implantation on the surface of the pore regions of the implant. The vacant sites were progressively infiltrated by regenerated bone and new

bone was formed at the place of the resorbed implant as soon as resorption occurred. New bone formations started at the periphery of the implant coral and grow to the center depending on the presence of macroporosity. At 8 weeks, the amount of bone in the pores increased and newly formed bone became matured. Multinucleated giant cells were present on the edges of the coral implant. At 12 weeks, coral implant was completely resorbed and the defect was almost completely filled with bone. The center of implant site still filled with fibrous. Most of the bone tissues were matured and contained abundant osteocytes.

Discussion

Results of this study showed that natural coral gives an excellent bonding with bone. Previous study has shown that fibrovascular tissue begins to invade the porosity immediately after implantation of natural coral[6]. This granulation tissue arises from bone marrow and is accompanied by blood vessels[7]. The osteoconductive capacity of porous coral allows cells attachment and growth through the scaffold of material, characteristic of a good support for cells[6]. The initial invasion of coral by blood and bone marrow cells with subsequent vascularization is a determinant factor for bone regeneration[5]. The phase of calcium carbonate in the coral is considered to be a biologic asset in rapid initiation of bone formation[14]. Following the initial dissolution phase, the appearance of a calcium phosphate layer occurs on coral [11,13]. Guillemin et al (1987) studied the capacity of coral implants to heal canine femoral defects. Both cortical and spongy bone defect were at least partially filled with new bone after 8 weeks, while the implants underwent continuous resorption. In our experience, complete resorption and replacement by newly formed bone was observed at the end of study period. Previous studies have shown that porous coral completely degrades after 3 month when placed in rabbit tibia[10]. Our findings showed that its degradation is due to a phagocytic process mediated by multinucleated giant cells such as osteoclasts like cells. Osteoclasts contain abundant carbonic anhydrase, an enzyme that locally lowers the pH at the osteoclasts-implants, dissolving the calcium carbonate matrix4. Braye et al. (1996) reported that the resorption is most active in the bone-implant contact areas and proceeds centripetally. Similar finding was found in this study.

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