USE OF BIPHASIC CALCIUM PHOSPHATE IN REPAIR OF LONG BONE FRACTURE WITH BONE LOSS IN DOGS

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ABSTRACT

The present study was conducted on 8 clinical cases of dogs presented to the Department of Veterinary Surgery and Radiology, Bombay Veterinary College, Parel, Mumbai and an affiliated Bai Sakarabai Dinshaw Petit Hospital with history of an accident or limping. Radiographs were taken and 8 dogs showing complete fracture with probability of bone loss were included in the study. The long bone fracture segments were reduced by open method. Stable internal fixation of the fractured segments was achieved using locking compression plate (LCP). Guided tissue regeneration (GTR) was achieved using a Vicryl mesh (Polyglactin 910) at periphery of fracture enclosing the gap due to bone loss. The gap between the fractured segments resulting into bone loss was filled with biphasic calcium phosphate. On 90th post-operative case 2, 5 and 6 showed complete radiographic union. Ceramics are promising materials for tissue engineering because they offer three-dimensional support and serve as a scaffold for cell proliferation, cell differentiation and ultimately for bone formation. Biphasic calcium phosphate (BCP) ceramics are composed of hydroxyapatite (HA) and β-tricalcium phosphate (β-TCP). HA has excellent biocompatibility and bioactivity and can be directly bonded to the host bone, while β-TCP has suitable degradation rate that matches the growth rate of newly formed bone.

Key words: Biphasic calcium phosphate, fracture, bone loss and callus formation

Introduction

Segmental fracture is a fracture of long bone at two levels, with intermediate fragment having an intact tubular or split structure. It can occur due to falling from a height or trapping of the limb. This results into a larger "butterfly" fragment (splinter) or comminution of the concave surface which is associated with severe soft tissue injuries, bone loss or bone defect which can perceive a tremendous challenge (Motsitsi 2006 and Hulse and Hyman, 2003). Autogenous bone grafting is considered the "gold standard" for filling bone defects. But, due to the limited availability and donor site morbidity of bone autografts and the risk of possible immune responses, disease transmission and the cost of allografts the use of synthetic bioactive materials opens new possibilities for clinical application, (Nandi *et al*., 2008). Therefore, bone regeneration by means of tissue engineering has attracted increasing interest. Ceramics are promising materials for tissue engineering because they offer three-dimensional support and serve as a scaffold for cell proliferation, cell differentiation and ultimately for bone formation. Biphasic calcium phosphate (BCP) ceramics are composed of hydroxyapatite (HA) and β-tricalcium phosphate (β-TCP). HA has excellent biocompatibility and bioactivity and can be directly bonded to the host bone, while β-TCP has suitable degradation rate that matches the growth rate of newly formed bone (Nie *et al*., 2012). The present study was undertaken to evaluate the role and effectiveness of biphasic calcium phosphate in repair of long bone fracture with bone loss in dogs. On 60th post-operative case 1, 3, 4 and 7 showed complete radiographic union at this time (Fig. 10). The biphasic calcium phosphate granules were completely incorporated in the callus.

Materials and Methods

The present study was conducted on 8 clinical cases of dogs presented to the Department of Veterinary Surgery and Radiology, Bombay Veterinary College, Parel, Mumbai and an affiliated Bai Sakarabai Dinshaw Petit Hospital with history of an accident or limping. Radiographs were taken and 8 dogs showing complete fracture with probability of bone loss were included in the study. Prior to general anaesthesia, in all the dogs, Atropine sulphate was administered @ 0.02 mg/kg s/c, followed by a pre-anaesthetic i.e. Inj. Diazepam hydrochloride @ 1 mg/kg i/v. Induction of anaesthesia was done with Inj. Propofol @ 4 mg/kg i/v followed by its maintenance with Isoflurane at 2% to 2.5%. The long bone fracture segments were reduced by open method. Stable internal fixation of the fractured segments was achieved using locking compression plate (LCP).

Case 1: The dog showed history of limping with left hind limb. Radiographic examination revealed multiple fracture of left femur (Fig.1). An eight holed locking compression plate was used to immobilize the fracture segments.

Case 2 : The dog was presented with a history of automobile accident where he was unable to bear weight on right hind limb. Radiographic examination revealed multiple fracture of right tibia-fibula. A ten holed locking compression plate was used to immobilize the fracture segments.

Case 3 : The dog was presented with a history where the dog's limb was stuck in the door. He was limping with left hind limb. Radiographic examination revealed complete oblique diaphyseal fracture of left femur (Fig. 3). A seven holed locking compression plate was used to immobilize the fracture segments.

Case 4 : The dog showed history of jumping from a height which resulted into limping with left hind limb. Radiographic examination revealed multiple fracture of left tibia-fibula. An eight holed locking compression plate was used to immobilize the fracture segments.

Case 5: The dog was presented with a history of hit by a truck and was unable to bear weight on left fore limb. Radiographic examination revealed complete diaphyseal transverse fracture of left radius-ulna. An eight holed locking compression plate was used to immobilize the fracture segments.

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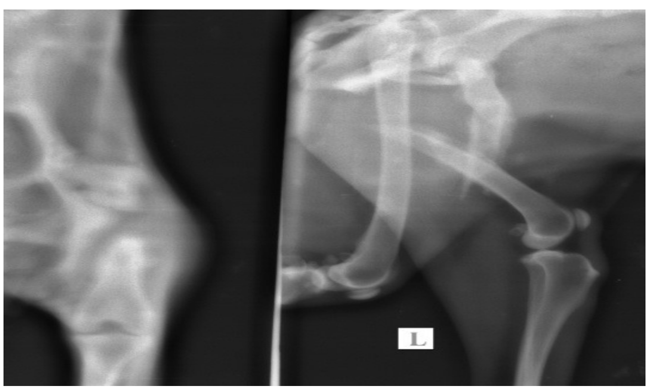
Case 6: The dog was presented with a history of jumping from a height. Radiographic examination revealed complete oblique distal 3^{rd} fracture of right femur. A seven holed locking compression plate was used to immobilize the fracture segments.

Case 7: The dog was presented with a history of hit by a four wheeler and was unable to bear weight on right fore limb. Radiographic examination revealed complete diaphyseal oblique fracture of right radius-ulna. A seven holed locking compression plate was used to immobilize the fracture segments.

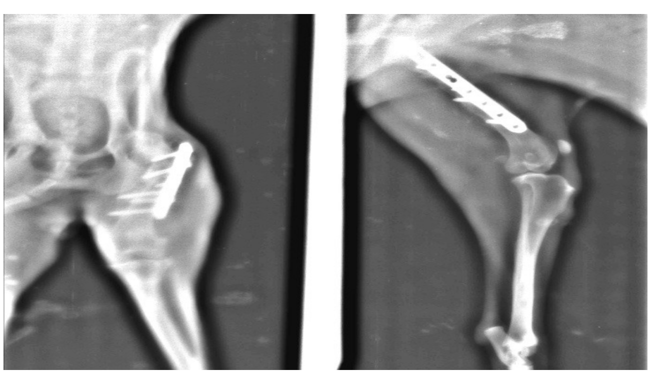
Case 8: The dog showed history of jumping from a height which resulted into limping with right hind limb. Radiographic examination revealed multiple fracture of right femur. An eight holed locking compression plate was used to immobilize the fracture segments.

Guided tissue regeneration (GTR) was achieved using a Vicryl mesh (Polyglactin 910) at periphery of fracture enclosing the gap due to bone loss (Fig. 5). The gap between the fractured segments resulting into bone loss was filled with biphasic calcium phosphate (Sybograf™-Plus) which is 70% hydroxyapetite and 30% β-tricalcium phosphate having granule size of 600 µ to 700µ (Fig. 6).

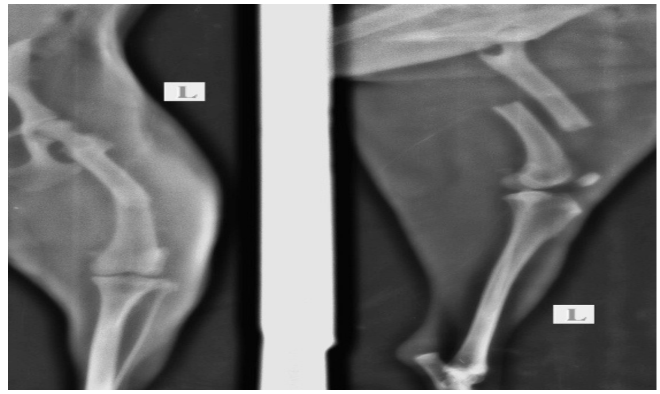
Post-operatively, Elizabeth collar was applied around the neck of the dogs to prevent self-mutilation and injury to surgical site. Cotton bandages were applied to the site to avoid injuries to the limb. All the dogs were administered, Cefotaxime at the dose rate of 20 mg/kg body weight for 10 post-operative days. Analgesics i.e. Meloxicam at the dose rate of 0.2 mg/kg body weight were administered intramuscularly, for 5 days post-operatively. Movement of the limb was restricted by giving complete rest for 15 days.



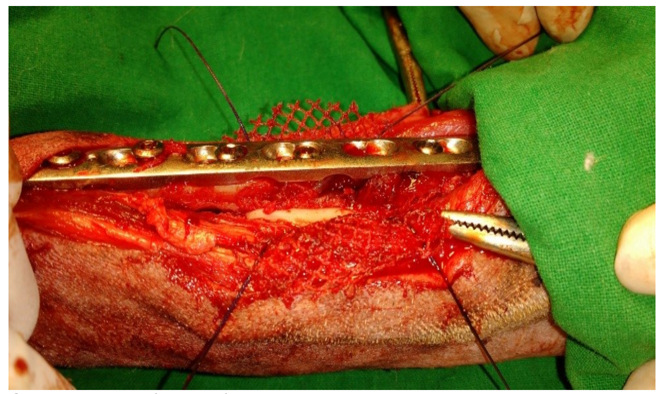
Case 1: Fig. 1: Pre-operative radiographic examination revealed multiple fracture of left femur



Case1: Fig. 2: (Day 60): Complete radiographic union at this time



Case 3: Fig. 3: Pre-operative radiographic examination revealed complete oblique diaphyseal fracture of left femur



Case 3: Fig. 4: (Day 90): complete radiographic union

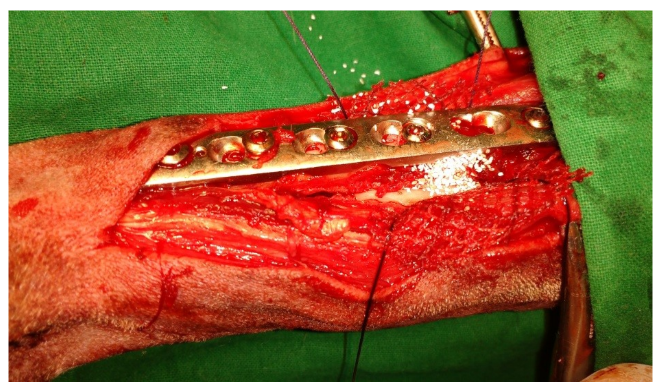


Fig.5: Guided tissue regeneration (GTR) was achieved using a Vicryl mesh (Polyglactin 910) at periphery of fracture enclosing the gap due to bone loss

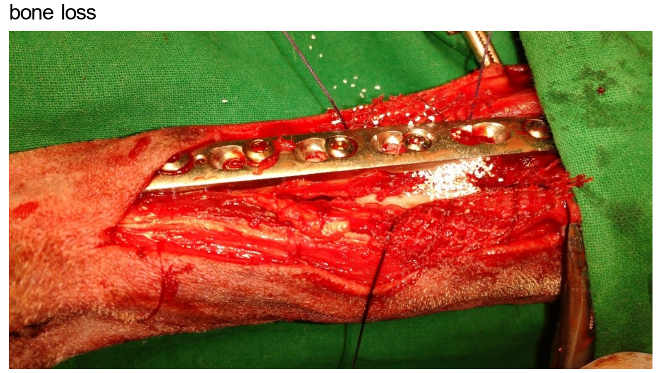


Fig.6: The gap between the fractured segments resulting into bone loss was filled with biphasic calcium phosphate (Sybograf™– Plus)

Radiograph was taken a day prior to surgery of the affected bone in appropriate position to evaluate the position of the fractured fragments along with bone loss and the type of the fracture. Post-operatively, radiographs were taken immediately after surgery and subsequently on day 15th, 30th, 60th and 90th. Based on periodic radiographs, implant position, union of fragments and callus formation, fracture healing was assessed and recorded. Wound healing was evaluated up to 10 days following surgery. Weight bearing of affected limb was recorded pre-operatively and then on 7th, 14th, 30th, 60th and 90th day post-operatively, to monitor the progress of fracture healing. The weight bearing was assessed and categorized using classification devised by Fox *et al.* (1995) as: excellent weight bearing without lameness, good-slight lameness principally after exercise, fair-slight to moderate lameness, but consistently weight bearing and poor-intermittent or consistent weight bearing lameness.

Results and Discussion

In case 1, 3, 4 and 7 slight lameness was seen on 7th post-operative day while partial weight bearing was seen by 15th post-operative day and excellent limb usage without lameness was seen on 30th post-operative day. The gap between the fractured segments was less in all these cases. A possible explanation for better weight bearing in these cases could be that as the gap was small, biphasic calcium phosphate allowed for earlier bridging of the fracture line, and as a result weight bearing forces were possibly transmitted from the proximal fragment to the distal fragment through the bridged callus without straining the implant. This is supported by the findings of Hwang *et al.* (2012), who reported that biphasic calcium phosphate showed highest amount of new bone formation within bone defects as compared to other ceramic bone graft substitutes.

In case 2, 5 and 6 and 8, consistent weight bearing lameness was seen on 15th post-operative day. Complete weight bearing was observed on 60th post-operative day. The inferior weight bearing grade in these cases could be due to the increased gap between the fractured segments leading to more bone loss, thereby allowing movement of fracture fragments leading to inflammation and pain as well prevention of callus formation. Sirin *et al.* (2013) also made similar observations with respect to trauma of the forelimb.

Radiographs taken immediately after surgery revealed anatomical reduction of fracture in all the cases but there was no callus formation. On 15th post-operative day, radiographs revealed the implants to be well in place and stable in all the cases. In case 1, 3, 4 and 7 mild periosteal reaction around the fracture site was aggressive. Ragsdale *et al.* (1981) reported that the activated periosteum has a deceptive anatomic constancy .The change involves the production of matrix and in the process, proliferation and sacrifice of cells. When the demand for a reaction is excessive, nearby extraperiosteal soft tissue serves as a ready source for additional modulating cells, just as it does for fracture callus. The configuration of a periosteal reaction is an index of the nature and intensity of the inciting process. On 60th post-operative case 1, 3, 4 and 7 showed complete radiographic union at this time (Fig. 2). The biphasic calcium phosphate granules were completely incorporated in the callus. Manjubala *et al.* (2005) also reported similar findings in dogs wherein process of ossification was noticed to start at 30 days with the use of biphasic calcium phosphate.

In case 2, 5, 6 and 8 where the gap was slightly big, mild periosteal reaction was seen around 30th post-operative day. Singh *et al.* (2011) reported early radiographic union by 6 weeks post operatively in similar cases. On 90th post-operative case 2, 5 and 6 showed complete radiographic union (Fig. 4). Agrawal (2007) reported bridging callus forms at a stage when the periosteal and endosteal calluses from either end of the fracture segments approach each other and come in contact forming the bridging callus. Thus, bridging callus formation starts peripherally and progressively moves towards the center of the fracture and between the fracture gap. Manjubala *et al.* (2005) studied bone in-growth induced by biphasic calcium phosphate ceramic in experimentally created 4 mm circular defects on lateral aspect of femur of dogs. Radiography revealed that the process of ossification started after 4 weeks and defect was completely filled with the new woven bone after 12 weeks. Histologicaly, osteoblast formation was evident while collagenous fibrous matrix and complete haversian system were observed after 12 weeks, therefore suggesting the osteoconductive and osteoinductive potential of biphasic calcium phosphate. Biphasic calcium phosphate allowed earlier weight bearing by day 30 a and callus formation also occured faster with radiographic union achieved by day 60. Further, there were no complications to BCP and hence it is suitable for hastening fracture healing.

References

Agrawal S (2007) Interlocking nailing for fracture fixation with a modified aiming device in dogs. M.V.Sc Thesis submitted to Jawaharlal Nehru Krishi Vishwavidyalaya, Jabalpur.

Fox SM, Bray JC, Guerin SR and Burbridge HM (1995) Antebrachial deformitiesin the dog: treatment with external fixation. *J. Small Anim. Pract.* **36**: 315-320.

Hulse D and Hyman B (2003) Fracture biology and biomechanics. In: Textbook of small animal surgery, 3rd ed.,W.B. Saunders Co., Philadelphia, pp: 1790.

Hwang J, Park J, Lee J, Jung U, Kim C, Cho K, Lee Y and Choi S (2012) Comparative evaluation of three calcium phosphate synthetic block bone graft materials for bone regeneration in rabbit calvaria. *J. Biomed. Mater. Res. Appl. Biomater.* **100**(8): 2044-2052.

Manjubala I, Sastry TP and Kumar RVS (2005) Bone in-growth induced by biphasic calcium phosphate ceramic in femoral defect of dogs. *J. Biomater. Appl.* **19**(4): 341-360.

Motsisi NS (2006) Management of segmental tibial fractures. *East Central African J. Surg.* **12**(2): 72-76.

Nandi SK, Kundu B, Ghosh SK and Basu D (2008) Efficacy of nano-hydroxyapatite prepared by an aqueous solution combustion technique in healing bone defects of goat. *J. Vet. Sci.* **9**(2): 183-191.

Nie L, Suo J, Zou P and Feng S (2012) Preparation and properties of biphasic calcium phosphate scaffolds multiply coated with HA/ PLLA nanocomposites for bone tissue engineering applications. *J. Nanomater.* **11**: 32-43.

Ragsdale BD, Madewell JE and Sweet DE (1981) Radiologic and pathologic analysis of solitary bone lesions Part II: periosteal reactions. *Radiol. Clin. North Am.* ***19***: 749-783.

Singh K, Thakur N, Mohindroo J, Gopinathan A and Mahajan AK (2011) Successful surgical repair of bilateral humerus fracture with modified T-plate and end threaded intramedullary pin in a dog. *Indian J. Vet. Surg.* **32**(1): 67.

Sirin OS, Kaya U and Olcay B (2013) Clinical and Radiological Outcomes of Locking Compression Plate System in Dogs with Diaphyseal Fractures: 32 Cases. *J. Faculty Vet. Med. Kafkas Univ.* **19**: 13-18.