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Use of osteogenic protein-1 in the management of a nonunion radial fracture in a Squirrel Monkey (*Saimiri boliviensis*)*

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Keywords

Osteogenic protein-1, bone morphogenetic proteins, nonunion fracture

Osteogenic protein-1 (OP-1 or bone morphogenetic protein 7) was used in the management of a nonunion fracture of the radius in a Squirrel Monkey (*Saimiri boliviensis*). Osteogenic protein-1 was used in preference to autogenous cancellous bone as the small size of the monkey would have limited the quantity of cancellous bone and predisposed to morbidity at the harvest site. This is the first report of the clinical use of OP-1 in veterinary surgery.

Summary

A Squirrel Monkey (*Saimiri boliviensis*) presented with comminuted, proximal diaphyseal fractures of the left radius and ulna which were repaired with a type II acrylic external fixateur and intramedullary pin, respectively. The external fixateur was removed six weeks postoperatively as the antebrachium was stable despite radiographic evidence that the radial fracture had failed to completely heal. Eight months postoperatively, radiographic examination revealed a nonunion fracture of the proximal radial diaphysis. The nonunion fracture was stabilised with a miniplate and screws, and osteogenic protein-1 (OP-1) (bone morphogenetic protein-7) was inserted to augment bony union. Radiographic evidence of fracture healing was present four weeks postoperatively. Bone morphogenetic proteins, in particular OP-1, can be used as a substitute for autogenous cancellous bone if there is an insufficient quantity of cancellous bone or harvesting will result in unacceptable morbidity.

Introduction

Bone morphogenetic proteins (BMPs) are members of the transforming growth factor- β superfamily (9, 17). They are regulators of organ, cartilage and bone differentiation during embryogenesis, and essential for the regeneration of cartilage and bone in postnatal life (17). Fifteen BMPs have been identified however only BMP-2, BMP-4 and BMP-7 have a significant osteoinductive effect (3, 9, 17, 18, 20).

Osteogenic protein-1 (OP-1), or BMP-7, has an important role in natural fracture healing and has been identified in both endochondral and intramembranous fracture healing (17). Fracture healing involves a complex interaction of many local and systemic regulatory factors (3). Only a small number of primitive cells express BMPs during the early phases of fracture healing. However, as endochondral ossification progresses, the levels of BMPs increase markedly, especially as osteoblasts lay down woven bone on the chondroid matrix (3, 17, 20). The expression of

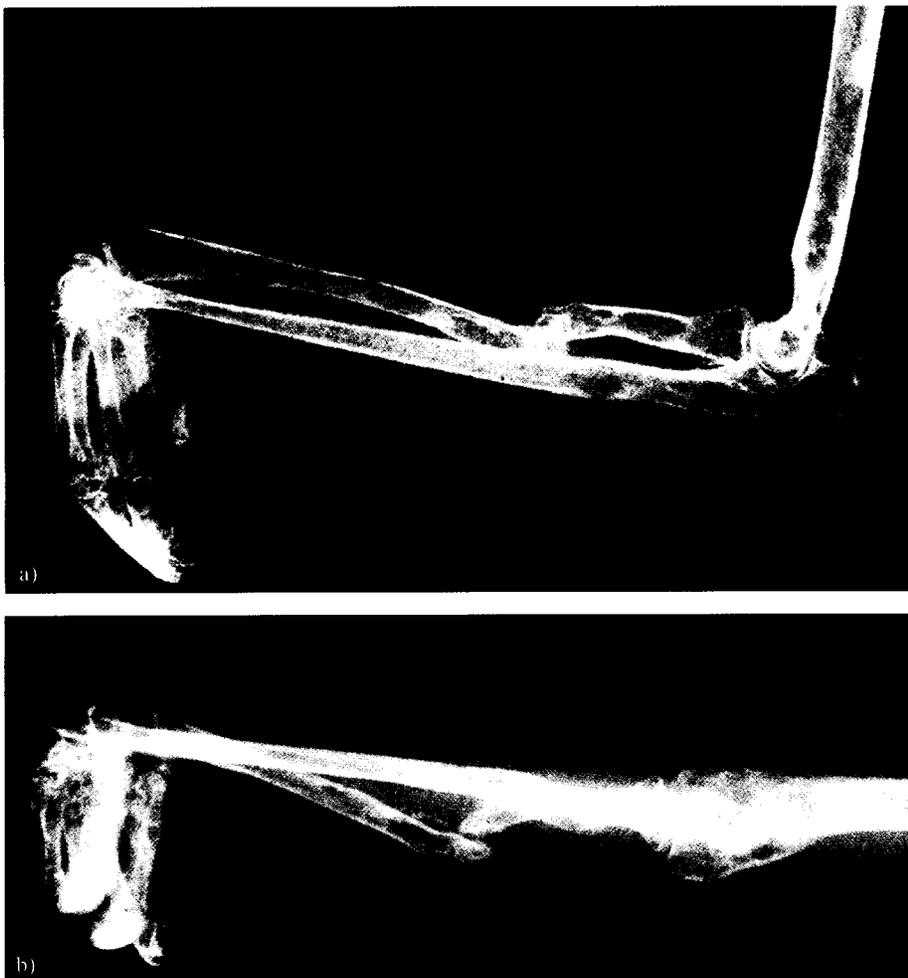


Fig. 1 Carpal contracture and a poorly vascular nonunion fracture of the proximal radius is evident on a) lateral and b) craniocaudal radiographs eight months postoperatively.

BMPs is reduced as the cartilaginous component of the callus matures, the number of immature cells decreases and lamellar bone replaces primitive bone (3, 17, 20). Bone morphogenetic proteins are integrally involved in chemotaxis, mitosis and differentiation of cells leading to cartilage and bone formation (3, 18).

Osteogenic protein-1, when combined with a bioresorbable carrier matrix, initiates the recruitment, attachment, proliferation and differentiation of mesenchymal cells, and results in the formation of new bone containing fully functional bone marrow elements (9). In vivo studies in rabbits, dogs and non-human primates, with critical and non-critical sized defects in the axial and appendicular skeleton, show that OP-1 accelerates the bone healing process, improves the performance of autografts and allografts, promotes

spinal fusion and enhances the osseous integration of metal implants (11).

Bone morphogenetic proteins are not species specific, due to homology between mammalian species, and hence recombinant human OP-1, produced in large quantities through DNA hybridisation techniques, can be used in a variety of animals including dogs and non-human primates (9). Osteogenic protein-1 does not provide structural support and hence concurrent rigid immobilization of the fracture is necessary (9). Furthermore, OP-1 must be carefully applied to the fracture site as it may induce heterotopic bone formation (9).

Case Report

A six-year-old, 600 g, female Squirrel Monkey (*Saimiri boliviensis*)

presented with an open fracture of the left antebrachium. General anaesthesia was induced with isoflurane and oxygen through a mask, and maintained with 2.5 percent isoflurane, oxygen and nitrous oxide via an uncuffed endotracheal tube and Bain nonrebreathing system. Intravenous fluids (lactated Ringer's solution) were administered at 6 ml/h for the duration of general anaesthesia. Cephazolin (15 mg IV) was administered for antimicrobial prophylaxis and analgesia was achieved with carprofen (2.5 mg SC) and a continuous rate infusion of fentanyl (0.03 ml/min IV).

Lateral and craniocaudal radiographs of the left antebrachium revealed minimally displaced, comminuted fractures of the proximal radius and ulna. The opposite limb was radiographed to assess normal conformation of the antebrachium.

The ulna was approached through a lateral incision. The fracture was reduced and stabilised with a retrograde 1.0 mm intramedullary pin. The radial fracture was approached through a medial incision. The small diameter of the bone in comparison to the 1.5 mm veterinary cuttable and dynamic compression plates precluded the use of a bone plate for stabilisation of the fracture. A sample of tissue from the fracture site was biopsied and submitted for microbiology. Bacteria were not observed on either a Gram stain or culture. The radial fracture was stabilised with a type II external skeletal fixateur constructed from 1.0 mm smooth fixation pins and acrylic connecting bars. Subcutaneous tissue was closed using 4-0 poliglecaprone 25 (Monocryl, Ethicon, Johnson & Johnson) in a continuous pattern.

Postoperative radiographs revealed adequate alignment of the radial and ulnar fractures. The monkey tolerated the external fixateur but chewed off her left thumb and index finger. Radiographically, the radial fracture had not healed six weeks postoperatively. However, the external fixateur was removed as the left antebrachium was stable and arm use was satisfactory despite the elbow and carpus being maintained in a semi-flexed position.

The monkey was housed as a display animal at Taronga Zoo and continued

Fig. 2 The radial fracture has been immobilised with a 1.2 mm 6-hole cuttable titanium miniplate and 1.2 mm cortical screws. Osteogenic protein-1 paste is being carefully injected into the fracture site.



Fig. 3 Postoperative lateral radiograph of the left antebrachium showing good alignment of the fracture ends and the fracture gap which was filled with osteogenic protein-1.

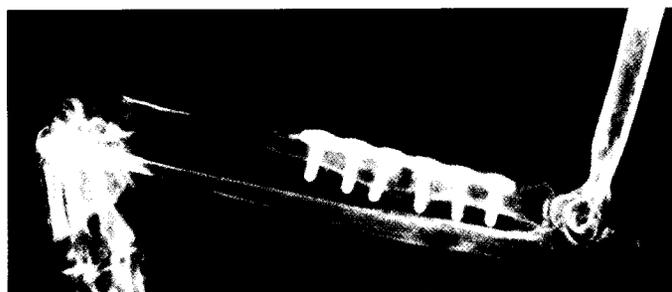
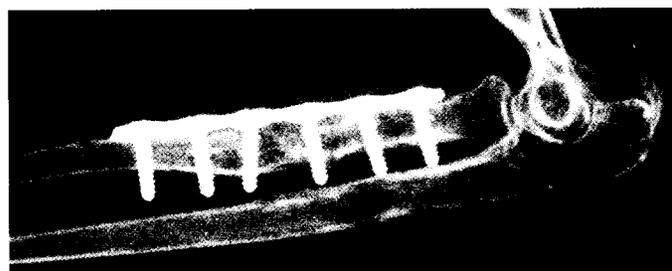


Fig. 4 Four weeks postoperatively, the fracture has healed.



to use the injured arm for feeding purposes. Radiographic examination six months after surgery revealed a delayed union of the radial fracture. A delayed union, rather than a nonunion, was diagnosed based on the strict definition of bony union in human orthopaedic surgery (AJ Shimmin). However, the importation of OP-1 was organised as further surgery was considered likely. The monkey was re-examined eight months postoperatively.

A similar anaesthetic and analgesic protocol was used although subcutaneous instead of intravenous fluids

were administered as a peripheral vein could not be catheterised. Marked and unreducible carpal contracture was evident. The range of motion in both left and right elbow joints was similar. Neurological examination could not be performed as the monkey was not tractable. Lateral and craniocaudal radiographs of the left antebrachium revealed a nonunion fracture of the proximal radial diaphysis (Fig. 1a and 1b). The ulnar fracture had healed.

The radial fracture was approached through a craniomedial incision. Fibrous tissue between the fracture ends was removed with rongeurs and the

medullary canals opened with a 1.1 mm drill bit. The fracture was reduced and stabilised with a 6-hole 1.2 mm titanium cuttable miniplate (Liebinger, Stryker, Australia) and 1.2 mm titanium cortical screws in a neutral fashion (Fig. 2). The fracture gap was filled with 1.5 ml of OP-1 paste (Stryker Biotech, Australia). The OP-1 paste was carefully contoured to the fracture site to minimise the risk of ectopic bone formation in adjacent muscle and subcutaneous tissue. The muscle and subcutaneous tissue were closed with a continuous pattern of 5-0 polyglactin 910 (Vicryl, Ethicon, Johnson & Johnson).

Good alignment of the radial fracture was demonstrated on postoperative radiographs (Fig. 3). Recovery from surgery was uneventful although carpal contracture had not improved and arm use remains unchanged six months after surgery. Fracture union was evident four weeks postoperatively (Fig. 4).

Discussion

To the best of the authors' knowledge, this is the first report of the clinical use of OP-1 in veterinary medicine. However, OP-1 has been used successfully in humans with nonunion fractures of the tibia (11) and BMP-2 has been used in the management of a nonunion fracture of the femur in a dog (16).

Differentiating between a delayed union and nonunion fracture can be difficult. In the present case, nonunion was diagnosed by the absence of radiographic bridging of the fracture eight months after initial reduction and stabilisation of the fracture. The nonunion fracture of the radius was probably caused by premature removal of the external skeletal fixateur due to stability of the antebrachium and successful union of the ulnar fracture. The open and comminuted fracture may have also contributed to the development of the nonunion fracture (22). Movement between the fracture ends disrupts the ingrowth of capillary and fibroblastic buds and delays revascularisation (22). Constant interfragmentary instability results in failure of mineralisation of the bridging fibrocartilage as mesenchymal

cells differentiate into fibroblasts and chondroblasts rather than osteoblasts (22). Interfragmentary fibrocartilage also acts as an avascular barrier preventing penetration of vascular resorptive channels and endochondral ossification (22).

Surgical intervention is usually indicated for the treatment of nonunion fractures (22). Surgical management includes removal of the interfragmentary fibrocartilage, decortication of the fracture ends, rigid immobilisation of the fracture and the use of an autogenous cancellous bone graft to promote revascularisation and osteoinduction. However, in the present case, OP-1 was used in preference to an autogenous cancellous bone graft as the small size of the patient may have limited the amount of cancellous bone available and increased the risk of donor site morbidity. Iatrogenic fracture of the humerus has been reported in a dog following harvest of a cancellous bone graft (13) and, in humans, up to 20 percent show symptoms of pain, hypersensitivity and anaesthesia at the cancellous bone harvest site (9).

Bone morphogenetic proteins provide a viable alternative to bone grafts in the management of nonunion fractures. Mesenchymal progenitor cells, capable of differentiating into chondroblasts and osteoblasts, have been identified in the fibrocartilage of a chronic nonunion fracture model in dogs (4). Moreover, in segmental defect and spinal fusion models in animals, the rate and quality of bone deposition and biomechanical performance were superior in OP-1 treated animals compared to those treated with autogenous cancellous bone grafts (5-8, 10, 12, 14, 19, 21).

The aetiology of carpal contracture in the present case remains unknown. Traumatic or iatrogenic injury to the posterior interosseus branch of the radial nerve, which courses between the radial head and supinator muscle, may have been a contributory factor (15). The posterior interosseus nerve may have been damaged by the proximal radial fracture or placement of the fixation pins. Neurological examination and electromyography were not performed but posterior interosseus nerve palsy can cause hyperaesthesia or paraesthesia of the thumb and index finger,

radial deviation of the hand and carpal contracture (15). Self-amputation of the thumb and index finger in the present case is suggestive of hyperaesthesia caused by posterior interosseus nerve damage. Fracture disease, with fibrosis and contracture of muscles and tendons (2), and Volkmann's ischaemic contracture, as a result of compartment syndrome in the antebrachial musculature (1), may have also been involved in the development of carpal contracture in the present case.

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Editorial Comment

The referees of this paper and the Editor-in-Chief had some difficulty in accepting the delay in treatment of this animal and expressed their concern to the authors. The correspondence published in a letter to the Editor [page IX] was received from the senior author and gives an understanding of the difficulties experienced. The explanation is deemed to be satisfactory.

Editor-in-Chief