# Radiographic evaluation of the absorption of bovine bone screws coated with bovine bone morphogenetic protein (BMP) inserted in the humerus of dogs

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# Abstract

Allografts are produced from natural sources and have been used in orthopedic repair procedures for a long time in humans and animals. Since 1952, hundreds of orthopedic surgeries have been performed with success using donated tissues and materials from bone banks. The purpose of the present study was to verify the absorption time of xenografts screws made of bovine cortical bone, inserted in the greater tubercule. The humerus of seven dogs in comparison with stainless steel screws. The results showed that screws made from bovine cortical bone used in the humerus of dogs were incorporated within a period of 30 to 60 days with no severe periosteum reaction.

#### **Key-words:**

Dog. Transplantation, heterologous. Bone and bones. Prostheses and implants.

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## Introduction

Allografts are originated from natural sources and have been used in skeletal repair for a long time. Their successful utilization was first reported by Macewen in 1887, but until 1940 bone grafts were viewed with caution by surgeons for reasons such as difficulties in bone-graft remodeling and in finding donors, in addition to lack of means for preserving the materials <sup>1</sup>.

Since 1952, hundreds of surgeries have been performed with success using donated tissues and materials from bone banks.<sup>2</sup> This type of surgery progressed between 1985 and 1995, with more than 500,000 surgeries using allografts being performed in the U.S alone.

Notwithstanding the advances, due to the high costs and difficulties in obtaining viable materials in large quantities, treatment of bone lesions with lyophilized human matrix is being substituted in Europe and in Brazil by products of bovine origin.<sup>3</sup>

Lewandrowski et al.4, in an

experimental study on tibial fractures in sheep, reported good results with the utilization of xenografts as substitutes for bone segments.

Choi-Inhyuk et al.<sup>5</sup> evaluated the use of xenografts of goat origin in the repair of injuries produced experimentally in the fibula of dogs and noted that the xenograft underwent osteoconduction in the callus formation in only 4 animals.

Pascalev<sup>6</sup> compared radiographically in dogs the behavior of three different types of grafts for the repair of fractures in the fibula: autografts of medular bone, allografts submitted to freeze-drying and xenografts preserved under freeze-drying conditions. It was reported that the best osteoinduction occurred in the first group and that, after two weeks, the autografts were reabsorbed but showed discrete callus formation.

In a comparative study on bovine cortical grafts containing or not Bone Morphogenetic Protein in fractures of the dog ulna, Tuominen et al.<sup>2</sup> reported that both grafts can be used as physical substitutes for bone,

however they do not induce osteogenesis.

The use of xenografts in the form of pins, screws and plates for the fixation of fractures is still an area which has been little explored in worldwide literature.

Wander et al.<sup>7</sup> presented one of the few papers on the use of xenografts in the form of pins for the correction of fractures of the humerus in pigeons, obtaining good results in comparison to stainless steel pins.

Studies using implants made of absorbable materials are particularly important in surgeries of the joints where exposure of part of the material in the joint surface is required. Since these implants are considered more biological than metal implants, there is no need for a new surgical procedure for their removal.

Due to lack of literature on implants manufactured from bovine cortical bone and as there are no studies on the degradation time of these implants, we tried to research pins manufactured from bovine cortical bone implanted in dog humerus. For this reason, the purpose of the present study was to verify the absorption time, through radiographic exams, of xenografts screws made of bovine cortical bone, inserted in the greater tubercule of humerus of dogs in comparison with stainless steel screws.

#### **Materials and Methods**

Seven healthy male dogs, weighing between 10 and 20 kg and up to seven years of age, from the São Paulo Zoonosis Center were used.

Absorption time of the bovine cortical bone screws coated with Bone Morphogenetic Protein (BMP) (Figure 1) was evaluated through radiographic exams performed 30, 60 and 90 days postsurgery.

Animals were fasted 12 hours for food and received water until 6 hours before surgery. The animals were submitted to the most adequate anesthetic protocol depending on their clinical condition.

The screws were inserted in the greater tubercule humerus. The animals were prepared and draped in a sterile pattern and two skin incisions were made over the lateral face of the humerus on both sides. Divulsion of adjacent tissues allowed exposure of the bone surfaces that were 2 cm distal from the scapular-humeral joint. After elevation of the muscles and periosteum adjacent to the bones, holes were made using a 4.5 mm diameter drill. Subsequently, screw threads for the holes were made using a 5.5mm screw tap and the 6mm screws were inserted.

According to convention, cortical bone

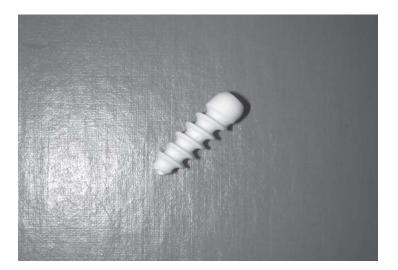


Figure 1 - Photography of the cortical bone screw

screws were inserted in the right limb and stainless steel screws in the left limb. For the closure of the surgical incisions prethreaded needles with 3-0 nylon thread were used.

For pain relief, Carpofren (Rimadyl – Pfizer S.A.) was administered to all the animals at the dose of 4.4 mg/kg every 24 hours, during 5 days.

Immediately after surgery and 10, 30, 60 and 90 days postsurgery, the animals underwent radiographic exams to evaluate bone reaction around the screws.

#### Results

In all dogs x-rays performed immediately after surgery showed that the screws were placed correctly in the greater tubercule humerus (Figure 2). In dog number one, at 30 days after surgery the bone screw was still visible on the X-ray and it was possible to differentiate the tunnel made by the drill for the insertion of the screw as well as the screw itself.

X-rays taken on the 60th day after



Figure 2- Radiograph immediately after the surgery, in the animal number two, showing the cortical bone screw inserted in the greater tubercule humeri



Figure 3 - Radiograph 30 days after surgery, in the animal number two, showing the absence of the cortical bone screw, and the hole made to insert the screw in tubercule humeri

surgery showed no evidence of the screw or the orifice where it was inserted. In the place where the screw had originally been lodged in the humerus no radiographic sign of bone lesion was observed. The same was noted at 90 days.

Similar results were found in animals number 4 and 5 which represented 42.8% of the cases under study.

At 90 days in animals number 3 and 4 corresponding to 28.5% of the animals evaluated, it was observed that the head of the screw was broken and found loose near the muscles. The screw head was found, in the soft tissue up to the third period of observation (90 days).

At 30 days, the bone screw in the tuberculum humerus of animal number 2 was no longer visible, nor the orifice made for the insertion of the screw. Similar results were obtained in animals number 3, 6 and 7, totalizing 57.2% of the cases evaluated (Figure 3).

Radiograph exams showed that the stainless steel screws were present in all animals under evaluation at all observation time points, with no periosteum reaction in the region of the implants.

## **Discussion**

The use of serial radiograph exams as a mean of evaluating the incorporation of the xenograft screws was adequate, as shown by Choi-Inhyuk et al.<sup>5</sup>, Pascalev<sup>6</sup>and Wander et al..<sup>7</sup>

The results obtained from the present study were similar to those obtained by Wander et al. who used xenografts as orthopedic implant. The xenografts, due to its characteristics, remained firmly attached to the receptor bone and, aided by the absence of mobility as well as by the extensive contact between the bone and the xenograft itself, allowed the incorporation of the screw to the receptor bone in a short period of time.

The different results obtained by Tuominen et al.<sup>2</sup>, Lewandrowski et al.<sup>4</sup> and

Choi-Inhyuk et al.<sup>5</sup> using xenografts, can probably be explained by the way the grafts were used. In all these studies, the authors tried to use the graft as a physical substitute for bone segmental defects and were not worried about the stability between the bone of the receptor and the graft; this may have caused the different results vis-a-vis the present study.

The same occurred with Pascalev<sup>6</sup> who used the same experimental model to compare xenografts and autografts.

The absorption time of the xenografts, in the present study, was longer than that reported by Pascalev<sup>6</sup> probably due to the larger size of the grafts used in our study.

Nevertheless, the phenomena of bone repair that occurred in this experiment will be evaluated in future studies because, although Tuominen et al.<sup>2</sup> demonstrated that xenografts produced reduced osteogenesis, only through histological analysis of the area of the screw insertion it will be possible to affirm if screw integration with the bone really occurred or if there was a simple absorption and primary callus osseus formation. Nevertheless, the authors agree that due to the short time of callus formation and lack of periosteum reaction in the region of the screw insertion and absorption, the most accepted hypothesis is that of osteointegration of the organic and mineral material of the screw with the receptor bone.

In spite of radiographic evidence of complete incorporation of the xenograft screw to the receptor bone, and lack of extensive periosteum reaction, this research requires a second study, i.e. a histological analysis of the insertion area of the screws to evaluate cellular phenomena which may have occurred.

#### **Conclusions**

It was possible to conclude through this study that screws made of bovine cortical bone coated with BMP used in the humerus of dogs were incorporated within a period of 30 to 60 days with no severe periosteum reaction and which was observed through conventional radiographic exam.

Avaliação radiográfica da absorção de parafusos de osso cortical bovino com bone morphogenetic protein (BMP) inseridos em úmero de cão

# Resumo

Os aloenxertos, são retirados de fontes naturais e são utilizados em cirurgias ortopédicas em humanos e em animais há muitos anos. Desde 1952 centenas de cirurgias ortopédicas foram realizadas com sucesso usando-se tecidos e materiais doados de banco de ossos. O propósito do presente estudo é verificar o tempo de absorção, através de exames radiográficos, de parafusos confeccionados com cortical óssea bovina, inserida no tubérculo maior do úmero de sete cães, em comparação com parafusos de aço. Os resultados demonstraram que o parafuso de cortical bovino foi incorporado entre um período de 30 a 60 dias sem reação periostal exuberante.

# References

- 1 BUSH, L. F. The use of homogenous bone grafts: a preliminary report on the bone bank. **Journal of Bone and Joint Surgery**, v. 29, n. 3, p. 620-628, 1947.
- 2 TUOMINEN, T. et al. Bovine bone implant with bovine bone morphogenetic protein in healing a canine ulnar defect. **International Orthopaedics**, v. 25, n. 1, p. 5-8, 2001
- 3 TAGA, R. et al. Reparo de defeito ósseo perene em crânio de cobaia pela aplicação de osseobond. **Revista Brasileira de Implantes**, jan. fev., p. 13-20, 1997.
- 4 LEWANDROWSKI, K. U. et al. Incorporation of perforated and demineralized cortical bone allografts. Part II: A mechanical and histologic evaluation. **Biomedical Materials and Engineering,** v. 11, n. 3, p. 209-219, 2001.
- 5 CHOI- InHYUK et al. Effectiveness of transplantation by freeze-dried bone of goat to dogs. **Korean Journal of Veterinary Clinical Medicine**, v. 15, n. 2, p. 442-449, 1998.
- 6 PASCALEV, M. Radiological studies on three types bone grafts in dogs. **Bulgarian Journal of Veterinary Medicine**, v. 1, n. 1, p. 41-48, 1998.
- 7 WANDER, K. W. et al. Fracture healing after stabilization with intramedullary xenograft cortical bone pins: a study in pigeons, **Veterinary Surgery**, v. 29, n. 3, p. 237-244, 2000.

#### Palavras-chave:

Cães. Transplante heterólogo. Osso e ossos. Próteses e implantes.