The effect of immobilization on reconstruction of mandibular defect using free iliac crest bone graft in dogs

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Abstract: Aim: Evaluation of the effect of two fixation devices (plates and fixation wires) for immobilizing of a free iliac crest bone graft in an induced defect in dog's mandible. **Materials and Methods**: Sample of this study was fourteen apparently healthy adult mongrel dogs. All cases showed normal appearance and function except two cases. **Results**:In these cases, the results were unsatisfactory. Radiographic evaluation revealed that the defective bone gaps appeared to be bridged by newly formed osseous tissue in 12 cases at one and three months postoperatively. Histologically, the bone defect was filled with newly formed bone trabeculae at three months postoperatively in comparison with the assessment at one month. **Conclusion**: no significant difference was found between both fixation devices (plates and fixation wires). Iliac crest bone graft could be used with great success to reconstruct the mandibular defects in dogs. [Nature and Science 2010;8(11):52-58]. (ISSN: 1545-0740).

Key words: fixation devices; iliac crest bone graft; radiographic evaluation

1-Introduction:

Surgical reconstruction for mandibular defects still remains extremely challenging. Mandibular defects are a common problem encountered by oral and maxillofacial surgeons. Such defects result from congenital malformations, cancer, trauma, and infections. Various osteogenic methods, including autogenous bone grafting (free bone grafting and vascularised grafting of compound tissue flaps), alloplastic and allogeneic materials, have met with limited success, (Fennis et al., 2002; Fennis et al., 2005; Huh et al., 2005 and Zhoua et al., 2008).

Autograft is the most commonly used type of bone graft. It can come from a variety of areas, including the iliac crest, distal femur, proximal tibia, fibula, distal radius, and olecranon. The iliac crest is the most common source of autograft bone due to its richness with progenitor cells and growth factors, limited donor site morbidity, fair bone quantity, and relative ease of harvest. Cancellous, corticocancellous, or vascularized bone may be harvested, but cancellous graft is most commonly used (Panagiotis, 2005 and Sen and Miclau 2007). The advantage of using bone autograft is that there is no risk of disease transmission, and that it is easily available (Jones, 2005).

The aim of the present study was to evaluate the effect of two fixation devices (plates and fixation wires) used for immobilizing a free iliac crest bone graft in an induced defect in dog's mandible depending upon clinical findings,radiographic and histopathological examinations.

2-Materials and methods

2.1.Materials

2.1.1.Sample:

Fourteen apparently healthy adult mongrel dogs 2- 4 years old were used in this experiment at the Department of Surgery, Anesthesiology and Radiology, Faculty of Veterinary Medicine, Suez Canal University. The animal's weight ranged between 12-18 kgs. A model of 2 cm defect was made in the mandible by osteoctomizing the 3rd and 4th mandibular premolar teeth of the dogs. Subsequently, reconstruction was carried out using an autogenous iliac crest graft. Dogs were assigned into two groups each of 7 dogs. In group I, the bone grafts were immobilized with plates and screws, while the bone grafts in group II were immobilized using fixation wires.

2.1.2.Drugs:

Intramuscular injection of Gentamycine Sulphate (Gentamycine 10% injection, El Nasr Pharmaceutical Chemicals Co., Abu Zaabal, Egypt) 0.5 ml/kg body weight was performed twice daily for 7 successive days postoperatively to prevent postoperative infection. Voltaren (Declofenac sodium, 75 mg ampoules, Novartis Pharma S.A.E. Cairo Egypt) 75 mg IM was injected every 12 hours for successive days to alleviate postoperative pain.

2.3.Methods:

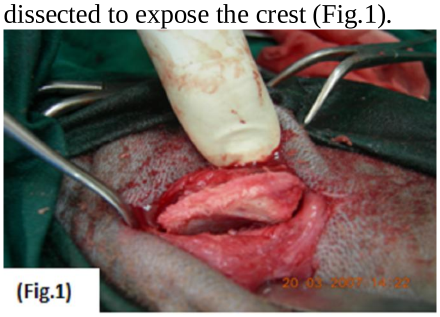
2.3.1.Before the operation

Food was with-held twelve hours before the operation. About 10-15 minutes prior to the induction of general anesthesia, each dog was premedicated with intramuscular injection of chloropromazine hydrochloride in a dose rate of 1 mg/kg. (Hall et al., 2001).

2.3.2.The surgical operation

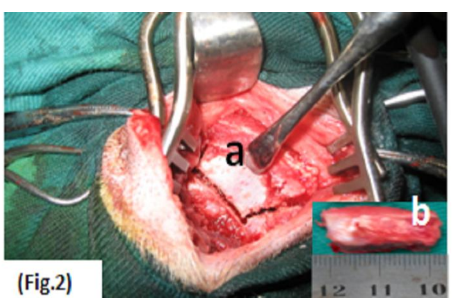
The surgical site was then clipped, shaved and disinfected with povidone iodine solution (Betadine: Povidone- Iodine U.S.P. 10%, El Nile-Co.). General anesthesia was then conducted by intravenous injection of thiopental sodium 2.5% until the main reflexes were abolished. The whole animal, except the sites of operation, was draped with sterile towel. Balanced electrolyte (Sodium chloride 0.9%) solution in a dose 10 ml/kg/hr was administered during surgery.

The dog was placed on its lateral recumbency. An incision of 5 cm was made just lateral to the palpable right dorsal crest of the ilium. The subcutaneous tissue, fat, and fascia were dissected to expose the crest (Fig.1).



(Fig.1): Exposure of the iliac crest after skin incision.

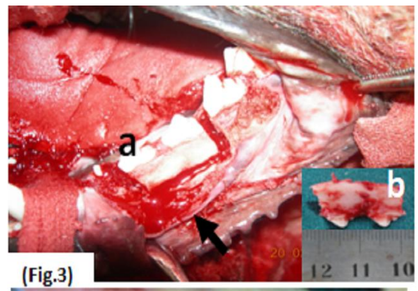
A longitudinal incision was made in the dorsolateral rim of the crest and the musculature was elevated on both sides, including the middle gluteal muscle origin. The dorsal rim was osteomized flatly from the cranial aspect to the caudal direction (Fig. 2).



(Fig. 2): Osteotomy of iliac crest (a) and the osteoctomized bone graft (b).

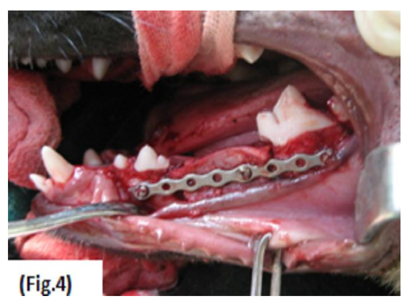
The graft was separated and kept between gauze moistened with saline solution. Fascia, fat and subcutaneous tissue were opposed again using polyglactin 910 No. 0 in a simple continuous pattern. Skin edges were sutured using silk No. 0 in a simple interrupted pattern.

In the dog at the same position, the recipient area was prepared for operation. The mandible was exposed using an intraoral approach. The buccal mucosa was carefully stripped and dissected to expose the bony mandible. The right 3^{rd} and 4^{th} premolar teeth of each dog had been unilaterally osteoctomized to create bone defects measuring 2 cm (Fig.3).

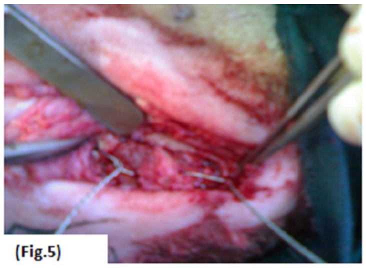


(Fig.3): Osteotomy of 3^{rd} and 4^{th} mandibular premolar teeth after stripping and dissection of buccal mucosa (arrow) (a). The osteoctomized the 3^{rd} and 4^{th} mandibular premolar teeth (b).

Intraoperative irregation with saline solution is used to prevent thermal injury to bone. After osteoctomizing the segment, the iliac crest graft was implanted and the mandible was stabilized by plate with screws for group I (Fig. 4) and stainless wires for group II (Fig. 5).



(Fig.4): Fixation of the iliac crest graft at mandibular defect using plate and screws



(Fig.5): Fixation of the iliac crest graft at mandibular defect using stainless wire

The wound was abundantly irrigated with saline solution and closed using silk suture material in simple interrupted pattern (Fig. 6).

(Fig.6): Suturing the buccal mucosa using 0 silk in simple interrupted suture pattern

After recovery, dogs were allowed for water free choice and continued to receive intravenous fluids for 48 hours postoperatively. Dogs were fed on a soft diet for seven days then gradually returned to full feed over the next week. Each dog was observed till the end of the experiment.

2.3.3. Clinical examination:

Dogs were observed for appetite and mastication. Donor and recipient sites were inspected for haematoma, wound dehiscence or sepsis.

2.3.4.Radiographic evaluation:

Radiograph films were taken for each dog immediately as well as, at one, two and three months postoperatively to verify and document the proper position of fixation devices and bone formation at the recipient site. Moreover, x-ray films were made at the donor site at three months postoperatively to follow the process of healing.

2.3.5. Histological evaluation:

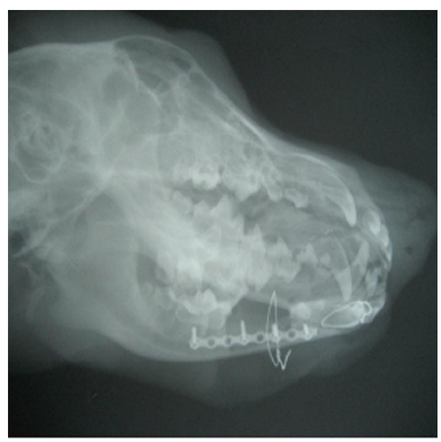
The animals were divided into two categories. The first category was sacrificed after one month while the second was sacrificed after 3 months postoperatively. Mandibles were dissected and resected after euthanasia of the animals using an over dose of thiopental sodium. The specimens were sectioned and fixed in 10% neutral buffered formalin for 1 week and decalcified in 20% formic acid in distilled water for 3 weeks. They were processed by routine histological procedure to obtain tissue paraffin blocks. They were sectioned along the sagittal axis at five µm thickness and Massion trichorme staining technique was used (Bancroft and Gamble, 2004) and the slides were observed under the light microscope

3-Results

All dogs appeared clinically normal depending on good appetite and normal mastication within 5–7 days postoperatively. All cases showed stability of their mandibles except two cases one from each group. Both cases suffered from loss of the graft with the development of sepsis and fracture of the mandibles at 18 days in the case of group I and 21 days in the case of group II postoperatively. At the donor site, only three cases suffered from wound dehiscence and infection.

3.1.Radiographic evaluation

Radiographic evaluation revealed that the seat of the graft appeared as radiolucent areas at mandibular defect one month postoperatively **(Fig.7)**.



(Fig.7): Oblique radiographic view in a dog of group I, showing radiolucent area at mandibular defect one month ostoperatively

Thereafter, bone healing, as manifested by the obliteration of the bone gaps, was particularly observed. This was well illustrated by the presence of radio-opaque areas at the mandibular defects at 2 and 3 months (**Figs.8 &9)**.

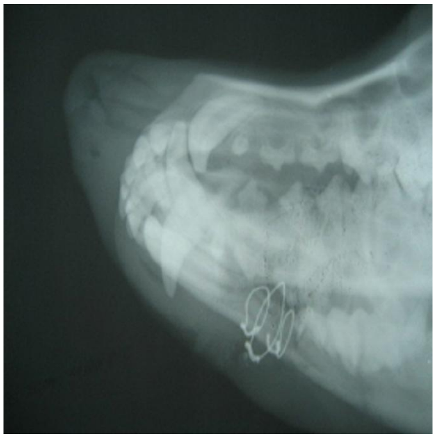


Fig.8): Oblique radiographic view in a dog of group II showing radio-opaque areas at the mandibular defect gap 2 months postoperatively.

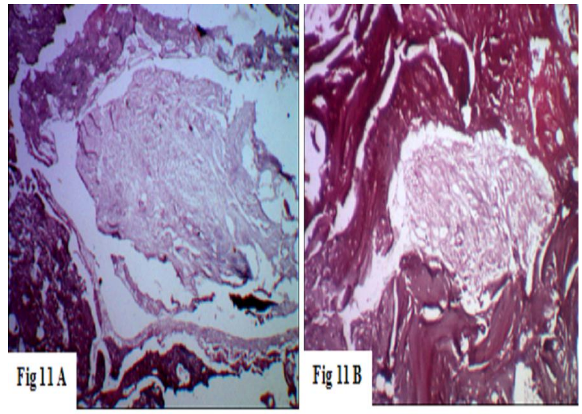
(Fig.9): Oblique radiographic view in a dog of group I, showing well illustrated radio-opaque areas at the mandibular defects gap 3 months postoperatively

Complete obliteration of the defective donor site was observed radio graphically three months postoperatively **(Fig.10)**.

Fig.10):Ventrodorsal radiograph view in a dog of group II showing bone healing at the donor site 3months postoperatively (arrow).

3.2.Histopathologic assessment

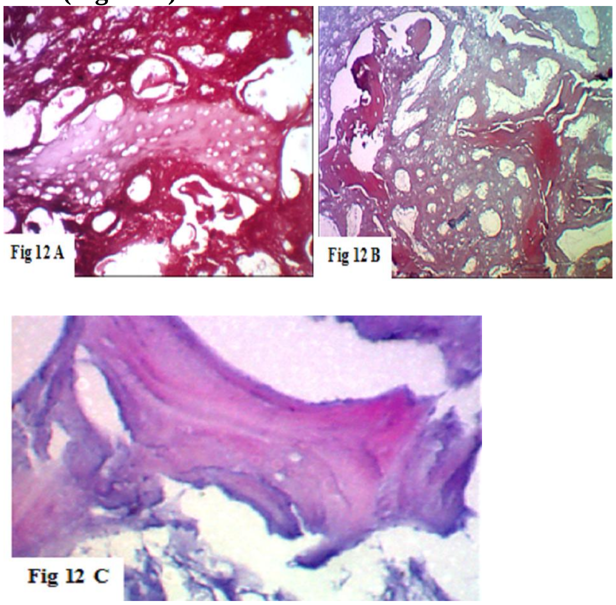
Histopathologic assessment revealed in group I and II, one month postoperatively, the bone defects were filled with fine and thin trabeculae of new bone that were highly vascularized. In the central part of the bone defect, the granulation tissues were observed as they were surrounded by newly formed bone trabeculae **(Fig. 11A)**.



(Fig 11): A photomicrograph of group I after one month showing A. thin bone trabeculation surrounding large mass of granulation tissue filling the central of bony defect. B. the number of formed bone is higher in group I than in group II. There is no sign of presence of inflammatory cells in both in group I &II. (Masson trichrome, x 100)

In group I, The number of newly formed trabeculae was greater than that in group II **(Fig. 11B)**. There were no inflammatory cells observed in the bone defect **(Fig. 11B)**. Linear concentrations of osteoblasts were present alongside. The newly formed trabeculae were located at the entrance of the defect.

In group I and II, three months postoperatively, the bone defect was filled with thicker newly formed bone trabeculae accompanied with a smaller number of blood vessels than those at one month **(Figs. 12A&B)**. The entrance of the osseous defect was covered by thin layer of compact bone tissue. Newly formed bone trabeculae were observed not only at the edge, but also in the central part of the bone defects. Coarse fibered woven bone was also observed which are characterized by large sized, high concentration and irregular arrangement of osteocyte **(Fig. 12A)**. Direct structure union was visible between old and new bone, with no inflammatory cells, and very little fibrous connective tissue present locally in group I **(Fig. 12B)**. Many reversal lines were observed at the periphery of bone trabeculae with its concave side toward the bone surface. A thin layer of new bone was laid down over the reversal line indicating formation of new bone **(Fig. 12C)**.



(Fig. 12): A photomicrograph of group II showing A. woven bone (arrows) with its large sized, high concentration osteocyte surrounded by well organized and formed bone trabeculas. B. in group I area of thick old mature bone arrows in direct union with thin newly formed bone trabeculas. C. many reversal lines indicating bone remodeling and in periphery new bone are formed. (Masson trichrome, X 200).

4-Discussion

Autogeneic bone grafts represent the "gold standard" of bone transplantation. The most common source of autograft is cancellous bone from the iliac crest. It is rapidly incorporated into the host site (Jones, 2005). This incorporation is due to its osteogenic properties, the abundance of growth factors, and the large surface area that provides for bone formation to occur. The cells present in the donor graft are capable of responding to local stimuli and releasing growth factors of their own, which accelerate angiogenesis and bone formation (Lemperle et al., 1998 and Khan et al., 2005).

The adult mongrel dog offers a near ideal animal model for the study of mandibular discontinuity defects. In the present study, it was possible to create defects of 2cm length in the mandibles. This animal has the advantage of allowing the creation of such large bone defects. This result was supported by Huh et al. (2005) who mentioned that critical size defect presents a suitable model for evaluating the usefulness of treatments because bone regeneration in a critical size defect only occurs in the presence of osteogenic materials.

The goal of the fixation is to achieve bony union across a fracture or osteotomy. Therefore, an ideal fixation technique will result in the maintenance of the proper alignment of bones during the healing process, without preventing function. With careful attention to principles and with proper case selection, miniplating, compression plating, lag screw fixation, and reconstruction plating will all provide this outcome (Bilgili and Kurum, 2003 and Sverzut et al., 2008).

In the present study, 12 out 14 cases showed good immobilization without significant difference between groups depending on clinical examination, radiographic evaluation and histological examination. Clinical findings and radiological evaluation showed no plate fractures and screw loosening. This is well illustrated by the presence of radio-opaque areas at the mandibular defects at 2 and 3 months postoperatively. This suggested that plates and wires successfully held the graft. This result was in agreement with Huh et al.,(2005).

On the other hand, two cases suffered from loss of the graft with the development of sepsis and fracture of the mandibles at 18 days in the case of group I and 21 days in the case of group II postoperatively due to fixation failure. Although the exact reason is not known, it might be caused by chewing movement on hard material which had eroded the mucosa and had become exposed to the oral environment. These results were supported by Bilgili and Kurum (2003) and Sverzut et al. (2008) who explained that any fixation hold the fragments together and facilitated bone healing if the mandible was to remain unloaded at all times. Unfortunately, this is not the case. Even the act of swallowing loads the mandible. For this reason, proper fixation will overcome these forces and keep fragments immobilized even when loaded in function.

Results of the present study demonstrated that methods (plates and screw or wire fixation) used to stabilize the bone graft at mandibular defect allowed the ‘gap healing’ pattern of bone repair to occur, although unremarkable histological differences were noticed between both groups. As after one month, the granulation tissue healing surrounded by parts of woven bone which was seen in both groups and also the absence of inflammatory cells were indicating bone healing. However, after three months direct bone communication is seen between old and new bone, also bone trabeculae were observed. Moreover, the presence of many reversal lines also indicated bone formation and healing. This is in accordance with Spiessl et al. (1989), who demonstrated that using of 2.0 mm miniplates comparing with compression plates in the dog mandible, both types of plates induced not only direct bone repair but also indirect repair by callus formation. In addition, Mckibbin (1989) reported that bone tissue has two remarkable properties; it can alter its mechanical characteristics in response to changes in functional demand and it also has the capacity to heal itself through a healing process resulting not in a scar, but in an actual reconstitution of the injured tissue. Moreover, Rasubala et al. (2004) stated that the healing process is affected by many variables, including the extent of damage to soft and hard tissue, the vascular supply caused by both the fracture and the fracture treatment.

In the present study, at 3 months, woven bone formation appeared as regions of improved organization, remodeling into more mature, parallel-fibered bone was found in the bone defect promoting the union of the parent bone edges. This finding was observed by Reitzik and Schoorl (1983) in monkeys at 6 weeks and Rasubala et al. (2004) in rats as early as 3 weeks. Meanwhile, Quereshy et al. (2000) observed in dogs at 12 weeks islands of haphazard woven bone lined by osteoblasts-forming osteoid matrix. However, Sverzut et al. (2008) found that woven bone formation occured after 6 weeks in adult mongrel dogs.

In our study, group I showed slight superior tissue healing than group II in the form of presence of thicker and denser mature bone trabeculae than group II and this was in consistent with Reitzik & Schoorl (1983) and Hochuli-Vieira et al. (2005) who demonstrated that rigid internal fixation of fractures and osteotomies in the craniomaxillofacial region was mandatory and better for optimal bone repair than external fixation.

To qualify as a bone graft, material must provide at least one of three basic functions: osteogenesis, osteoconduction, and osteoinduction. In the present study, iliac crest bone graft could fulfill these properties. This is in accordance with Sen and Miclau (2007) who reported that iliac crest bone graft remains the only clinically available graft source that is osteogenic, osteoinductive, and osteoconductive.

It could be concluded that although there was no significant difference between both fixation devices (plates and fixation wires), the plates fixation was superior to wire fixation that was represented by presence of thicker and denser mature bone trabeculae. Iliac crest bone graft is a good clinically available graft source so it could be used with great success to reconstruct the mandibular defects in dogs.

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