

Radiological and histopathological study of the effect of omental pedicle flap on the transverse and oblique rib fracture in dogs

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Abstract

The aim of this study was to estimate the possibility of the effects of omental pedicle flap on improving the transverse and oblique rib fracture healing. Twenty two old dogs were used in this study. The animals were divided into two equal groups. In first group transverse rib fracture was induced, while in second group obliquely fracture induced. Each group were divided into two equal subgroups depending on covering by omental pedicle flap (treated) or without covering (control). Atropine sulphate 0.04 mg/kg B.W as a premedication followed 15 minutes later by a mixture of ketamine hydrochloride and xylazine hydrochloride 15 mg, 5 mg/kg B.W. intramuscular respectively as a general anesthesia. The histopathological study at a period of 1, 2, 3 and 4 weeks post-operation were revealed that in first group, the simple compact bone formation at 4 week in treated cases, when compared with the very simple formation of compact bone in control cases. While in second group simple compact bone formation appear at 3 week in treated cases, while compact bone was seen at 4 week in control cases. The radiological findings of the two main groups were coincide with the histopathological results, which appeared that the degree of healing in second group better than in first group specially in treated cases.

Keywords: Radiography, Omental, Flap, Rib, Fracture.

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دراسة شعاعية نسجية مرضية لتأثير طية من الثرب على الكسور المستعرضة والمائلة للاضلاع في الكلاب

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الخلاصة

هدفت الدراسة الحالية لتقييم استخدام قطعة من الثرب لتحسين التئام الكسور الميترضة والمائلة للاضلاع في الكلاب. استخدم ٢٢ كلباً في هذه الدراسة. قسمت الحيوانات الى مجموعتين متساويتين؛ في المجموعة الاولى تم عمل كسر مستعرض في الضلع في حين تم عمل كسر مائل في الضلع لحيوانات المجموعة الثانية. قسمت بعدها حيوانات المجموعة الواحدة الى مجموعتين متساويتين اعتماداً على استخدام قطعة الثرب (مجموعة المعالجة) او عدم استخدام الثرب (السيطرة). اعطي عقار الاتروبين سلفيت بجرعة ٠,٠٤ ملغم/كغم من وزن الجسم وبعد ١٥ دقيقة اعطي مزيج الكيتامين مع الزايلازين وجرعة ١٥ ملغم و ٥ ملغم/كغم من الوزن الجسم على التوالي في العضل كمخدر عام. اظهرت نتائج الدراسة النسجية المرضية للفترات ١، ٢، ٣ و ٤ اسابيع بعد العمليات الجراحية ان في المجموعة الاولى قد تكون العظم في الاسبوع الرابع في مجموعة المعاملة في حين لم يظهر سوى تعظم بسيط في مجموعة السيطرة. بينما في المجموعة الثانية فقد تكون العظم في الاسبوع الثالث في مجموعة المعاملة في حين ظهر التعظم في مجموعة السيطرة في الاسبوع الرابع. كانت نتائج الدراسة الشعاعية مطابقة لنتائج الدراسة النسجية المرضية حيث ظهر ان درجة الالتئام في المجموعة الثانية كانت افضل من المجموعة الاولى خصوصاً في حيوانات المعالجة.

Introduction

Multiple rib fracture carry with them higher morbidity and mortality rates and in survivors long term morbidity is significant, operative rib stabilization has a role in treatment and studies suggest improved short and long term outcomes. The surgical stabilization by kirshner wires, stainless steel wire on both of a flail chest was indicated in patient who required a thoracotomy because of associated thoracic injuries and in patient with extensive antero-lateral failed chest and progressive dislocation of the fracture ribs in order to prevent lateral chest wall deformity and a consequent restructure disorder (1-4). Rib fractures can also be marker of sever trauma and aortic trisection is well described a cause of death (5). Rib fracture may be accompanied by one or more of the following sign: cough, cyanosis, chest distortion or subcutaneous emphysema (6). Rib fracture repair has been performed at selected center around the world for more than 50 year, the potential indications for rib fracture repair include flail chest, painful, chest wall deformity, movable rib fractures refractory to conventional pain management (7). Also rib fracture fixed by using absorbable plates and screws, thoracoscopic assistance also was used in rib fracture repair (8). Some methods use in ribs stabilization was reconstruction plate(s), self –tapping cortical screw and cerclage wire, Steinman pins and cerclage wire (3). Early surgical stabilization may result in shorter intensive care unit stay with lower morbidity and prevention of pulmonary restrictive complications resulting in working in capacity (2).

Omentum, it is a highly vascular organ with a rich source of angiogenic factors that promote the growth blood vessels into whatever tissue it placed close to omentum can be used as a plastic and disinfecting material in combination with laser radiation in the management of chronic osteomyelitis of the ribs and sternum (9,10). The objective of our investigation was to estimate possibilities for application of pedicle flap of greater omentum in improving the transverse and obliquely rib fractures healing.

Materials and methods

All operation were performed on 1-4 years old dogs from both sexes, weighting 14-22 kg were maintained under standardized condition with free access to water and diet. Twenty two experimental animals were divided into two equal groups, ribs fracture was induced on the last four ribs of both side and repeated on unbroken rib in same animal at interval of long period. In first group transverse rib fracture was induced, while in second group obliquely fracture induced. Each group were divided into two equal subgroups depending on covering by omentum pedical flap

(treated) or without covering (control). Antibiotic therapy penicillin- streptomycin was started before one hour of the operative. Anesthesia was induced by intramuscular injection of 0.04 mg /kg B.W. atropine sulphate as a premedication followed 15 minutes later by a mixture of ketamine hydrochloride 15 mg/kg B.W and xylazine hydrochloride 5mg/kg B.W intramuscularly.

The rib was explored by direct incision over its, incision of subcutaneous tissue and rib fracture was made at a suitable distance from costo-vertebral articulation. The fracture lines were made transversely with long axis of rib in first group and obliquely with long axis of the rib in second group. Drill and one hole was made from each side of fracture ends at about 1cm from edge. Stainless steel orthopedic wire was used to fix the fracture line by introducing through a hole on each side and twisting into each other over the fracture site.

In treated subgroups of transverse and obliquely fracture, the omental pedicle flaps was carried out after laparotomy and extend through a subcutaneous canal, then fixing by multiple stitches with muscles over the fracture site by absorbable suture material (catgut 2.0). The muscle and skin were closed as a routine mannar. Penicillin streptomycin was given at a dose of 10.000 I.U 20mg/kg B.W intramuscular respectively for four days post operation.

Radiological and histopathological studies were performed weekly to evaluate the degree of healing between the main groups. The experimental animals were anaesthetized at the 1, 2, 3 and 4 weeks after operation and partials resection of the ribs which included the fracture site were resected for histopathological and radiological study, some radiography were done on alive animals while others on resected ribs. The sites of resection were closed. The resected ribs decalcified, embedded in paraffin and cut into 4-5 micrometer, Hematoxylin and Eosin staining were used

Results

In all experimental animals no signs of sever complications as well as the animals were tolerated the surgical operation, in some cases, rib fracture was accompanied with mild cough and slight cellulites, especially association with opening of pleural cavity during rib fixation. At biopsy on a period mentioned above show that sever adhesion in between fracture site with muscles and omentum in treated cases while this adhesion in less degree between fracture site and muscles in control cases.

The histopathological examination of biopsy at the period of 1, 2, 3 and 4th weeks of the two groups were summarized on (table1 and 2).

The radiological findings of the experimental rib fracture which obtained before taken rib biopsy or after partial rib resection were summarized in table 3 and table 4.

Table 1: Show the histopathological findings of first group (transverse control and treated).

Time	Transverse (control)	Transverse (treated)
1 week	The histopathological section showed, fibrinous network at the site of fracture, partially converted into granulation tissue. Simple formation of trabecular bone was seen, which surrounding by fibrous connective tissue (figure 1).	Organization of blood clot with formation of granulation tissue which consist of collagen fiber and fibroplasias. Simple formations of trabecular bone which surrounding by dense connective tissue (figure 2).
2 week	Large area of trabecular bone, mixed with cartilage and fibrous connective tissue (figure 3).	Mixed area of trabecular bone, cartilage tissue which surrounding by fibrous connective tissue (figure 4).
3 weeks	Large area of trabecular bone which surrounding by fibrous tissue. Small area of cartilage tissue adjacent to trabecular bone (figure 5).	Large area of mature trabecular bone surrounding by fibrous connective tissue (figure 6).
4 week	Start to formation of compact bone with the large patches of cartilage tissue, also fibrous connective tissue seen around trabecular bone (figure 7).	Simple formation of compact bone also area of cartilage tissue surrounding by fibrous connective tissue were seen (figure 8).

Table 2: Show the histopathological finding of second group (Obliquely control and treated).

Time	Obliquely (control)	Obliquely (treated)
1 week	The Histopathological section showed, fibrinous network infiltrated with inflammatory cells. Simple Formation of trabecular bone with thick layer of fibrous connective tissue around its (figure 9).	Organization of blood clot, simple formation of trabecular bone, with active proliferation of periosteum (figure 10).
2 week	Large area of trabecular bone, surrounding by cartilage and fibrous tissue (figure 11).	Large area of trabecular bone, surrounding by fibrous and cartilage tissue (figure 12).
3 weeks	Large area of trabecular bone surrounded by cartilage and fibrous connective tissue.	Simple formation of compact bone, cartilage and fibrous tissue also seen (figure 14).
4 week	Large areas of trabecular bone, and formation of compact bone which surrounding by fibrous connective tissue (figure 15).	Formation of compact bone as well as large area of trabecular bone surrounding by fibrous connective tissue.

Table 3: Show the radiological finding of first group (transverse control and treated).

Time	Transverse (control)	Transverse (treated)
1 week	Show clear fracture line (figure 13).	Show clear fracture line (figure,14)
2 week	Simple callus formation in between the fracture ends, but fracture line visible (figure 15).	Show callus formation in between the fractured ends, but fracture line still visible (figure 16).
3 weeks	Show callus formation, but fracture line still visible (figure 17).	Callus formation in between the fractured ends but fracture line still visible (figure 18).
4 week	Show clear callus formation in between fractured ends. fracture line still visible (figure 19).	Show more dense callus formation in between fractured ends fracture line very slight visible (figure 20).

Table 4: Show the radiological finding of second group (Obliquely control and treated).

Time	Obliquely (control)	Obliquely (treated)
1 week	Radiological finding showed clear fracture line (figure 21).	Radiological finding showed clear fracture line (figure 22).
2 week	Show callus formation in between the fractured ends, but fracture line still visible (figure 23).	Show callus formation in between the fractured ends, but fracture line still visible (figure 24).
3 weeks	Show callus formation in between the fractured ends, but fracture line still visible (figure 25).	Show callus formation in between the fractured ends, but fracture line slight visible.
4 week	Show dense callus formation in between fractured ends. the fracture line slight visible.	Show thick callus formation in between fractured ends, fracture line semi-unvisible (figure 26).

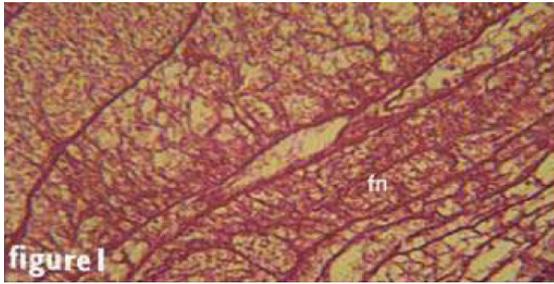


Figure 1: Histopathological section show, fibrin network (fn) oneweek post operation on first group (transverse control). H&E X 40.

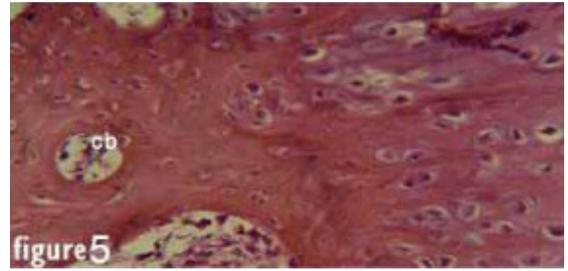


Figure 5: Histopathological section show, compact bone (cb) and cartilage tissue four week on first group (transverse control). H&E X 40.

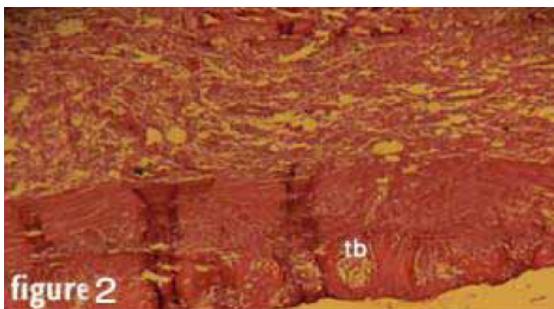


Figure 2: Histopathological section show, start formation of trabecular bone (tb) one week on first group (transverse treated). H&E X 40.

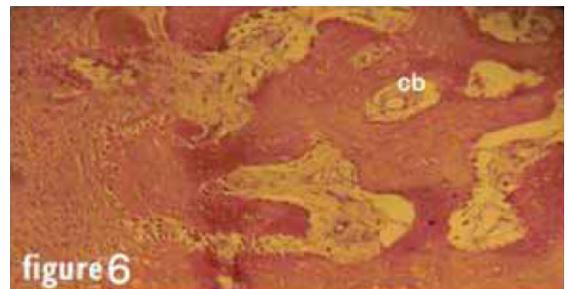


Figure 6: Histopathological section show, fibrous tissue, trabecular bone and compact bone (cb) four week on first group (transvers treated) H&E 10.

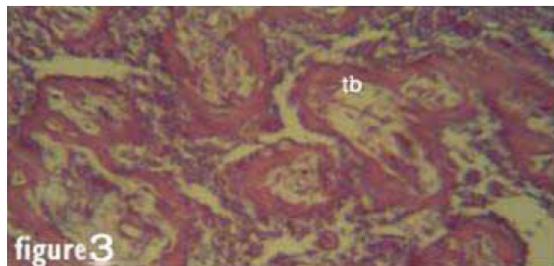


Figure 3: Histopathological section show, trabecular bone (tb) two week on first group (transverse control). H&E X40.

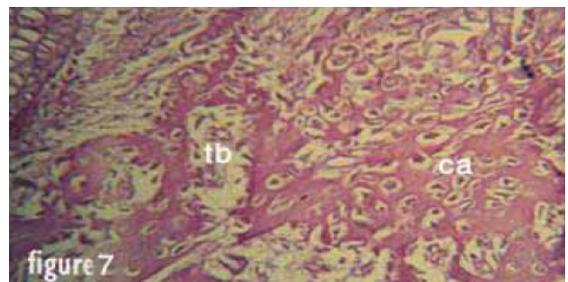


Figure 7: Histopathological section show, cartilage (ca) and trabecular bone (tb) one week on second group (oblique control). H&E X40.

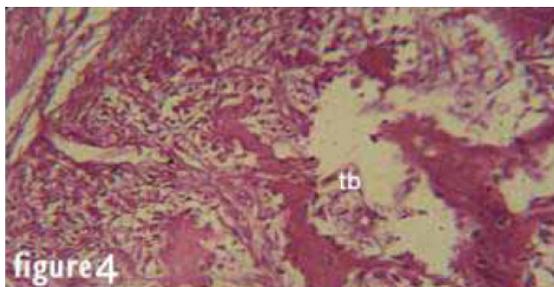


Figure 4: Histopathological section show, fibrous tissue and trabecular bone (tb) two week on first group (transverse treated). H&E X 40.

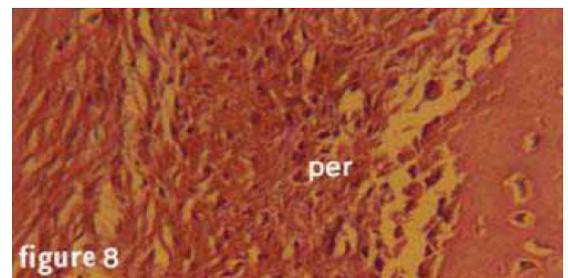


Figure 8: Histopathological section show, proliferation of periosteum one week on second group (oblique treated). H&EX40.

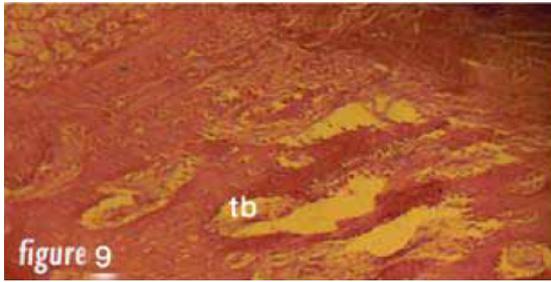


Figure 9: Histopathological section show, trabecular bone (tb), cartilage and fibrous tissue two week on second group (oblique control). H&E X 10.

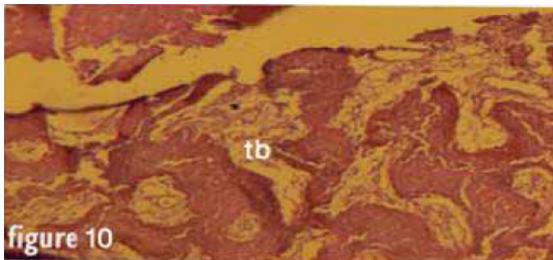


Figure 10: Histopathological section show, trabecular bone (tb) two week on second group (oblique treated). H&E X 10.

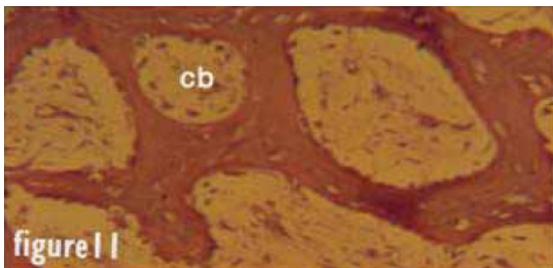


Figure 11: Histopathological section show, trabecular bone and compact bone (cb) four week on second group (oblique control). H&EX 40.

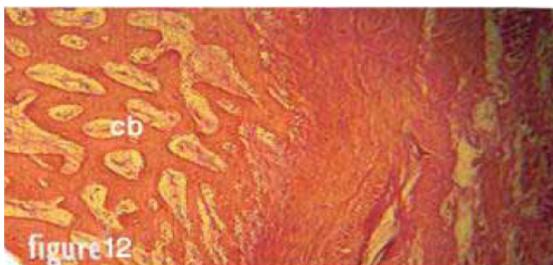


Figure 12: Histopathological section show, fibrous tissue, trabecular bone and compact bone (cb) four week on second group (oblique treated).H&E X 10



Figure 13: Radiograph of the rib show clear fracture line one week post operation on first group (transverse control).



Figure 14: Radiograph of the rib show clear fracture line one week post operation on first group (transverse treated).



Figure 15: Radiograph of the rib show callus formation in between the fracture ends but fracture line visible two week post operation on first group (transverse control).



Figure 16: Radiograph of the rib show callus formation in between the fractured ends but fracture line still visible two week post operation on first group (transverse treated).



Figure 17: Radiograph of the rib show callus formation in between fractured ends but fracture line still visible three week post operation on first group (transverse control).



Figure 21: Radiograph of the rib show clear fracture line one week post operation on second group (oblique control).



Figure 18: Radiograph of the rib show Callus formation in between the fractured ends but fracture line still visible three week post operation on first group (transverse treated).



Figure 22: Radiograph of the rib show clear fracture line one week post operation on second group (oblique treated).



Figure 19: Radiograph of the rib show clear callus formation in between fractured ends. Fracture line still visible four week post operation on first group (transverse control).



Figure 23: Radiograph of the rib show callus formation in between the fractured ends but fracture line still visible two week post operation on second group (oblique control).



Figure 20: Radiograph of the rib show more dense callus formation in between fractured ends. fracture line very slight visible four week post operation on first group(transverse treated).



Figure 24: Radiograph of the rib show callus formation in between the fractured ends but fracture line still visible two week post operation on oblique treated group.



Figure 25: Radiograph of the rib show callus formation in between the fractured ends but fracture line still visible three week post operation on second group (oblique control).



Figure 26: Radiograph of the rib show thick callus formation in between fractured ends, fracture line semi-unvisible four week post operation on second group (oblique treated).

Discussion

In this research we suggested that early surgical fixation of rib fractures lead to restores normal mechanical ventilation and decrease post operation complications such as penetration lung lobe rupture of large blood vessels or affected of heart and mediastinal region (11). This facts documented in our study were found that postoperation complications of rib fracture rare may be due to immediately fixation of rib fracture, this agree with other workers (6). The adhesion of muscle and omentum around the fracture site in first and second treated sub groups were given more fixation to the fracture site, where compared with control sub groups. This coincide with other authors (9,12) whom said that the omentum, it's a highly vascular organ with a rich source of angiogenic factors that promote the growth of blood vessels into what ever tissue its placed closed to. The omental graft was converted into fibrous connective tissue which help of fracture site fixation and also regard as a scaffold of osteoblasts proliferation (13). The histopathological findings of the first group revealed that there is no bigger different between the control and treated cases in the first and second week that include organization of blood clot which rapidly in treated than control subgroup with formation of cartilage tissue and

trabecular bone, while at the 3rd week large area of mature trabecular bone which formed in treated cases while still present immature trabecular bone with cartilage tissue at this period in control cases. At the 4th week simple compact bone which formed in control cases but more mature in treated cases at the same period this results refers the affect of omentum on enhance fracture healing because capability for the formation of osteogenic tissue and revascularization with the tissue it contact, this results coincide with (14) and also we agree with (15) whom said that the greater omentum has well developed osteogenic potential.

The histopathological examination of the treated cases of the second group at the first week show organization of blood clot and active proliferation of periosteum with simple formation of trabecular bone and in the second week complete organization of blood clot and trabecular bone formation with small area of cartilage tissue also seen, while in the control cases show inflammatory cells with thick layer of fibrous connective tissue and until the second week the organization of blood clot which not complete, and large area of trabecular bone, cartilage tissue also seen. At the third week in the treated cases show that started to formation of compact bone that increase in width and maturity in the 4th week, while at the 3rd week in control cases show cartilage tissue still present with large area of trabecular bone also present while the compact bone start formation at the 4th week. This result refers to the affect of omentum on the fracture site by stimulation of un differentiation mesenchymal cells at the site of fracture by local growth factor metaplastically altered in the osteoblast cells that increase the osteogenic cells number (15). Omentum contain stem cell, that can differentiated into a variety of cell type and good source of angiogenic factor like vascular growth factor (9) that provide oxygen at site of fracture and stimulate mesenchymal cell to differentiate into osteoblast cell to form trabecular bone with small area of cartilage tissue when compare with control cases.

In comparative between two group we found that at the first week in 2nd group treated cases highly periosteal proliferation, start of trabecular bone formation and complete organization of blood clot at 2nd week while still present of organization in first group at the similar time, at 3rd week show start formation of compact bone in 2nd group treated cases while not formed in first group at the same period in treated cases and at the 4th week in second group of treated cases present of large area of trabecular bone and well developed compact bone while in first group at the similar period small area of simple compact bone.

The results of histopathological findings may indicate that there is relationship in between the fracture shape and speed healing. Which may be the obliquely fracture line, provide large area, which suitable for proliferation of blood vessels from surrounding tissue and migration into the fracture line. On other hands the histopathological result

revealed that the role of omental pedicle flap for enhance fracture healing on treated cases of the first and second group when compared with first and second control sub group.

The radiological findings of the two main groups were coincided with the results of the hisopathological study, show that, the fracture line visible at the 1st, 2nd and 3rd weeks of both first and second main groups. Except that at 3rd weeks of the treated second group, which appeared more dense callus formation indicated mature trabecular bone and started to formation of compact bone while other sub groups at the period mention above, the trabecular bone and cartilage tissue and fibrous connective tissue not well good visible in X-ray. While in 4th weeks in two main groups show more dense callus formation in between the ends of fracture ribs, but more clearly in second group (treated subgroup). This indicated that the compact bone more mature that the other subgroups. In conclusion of this study revealed that the omental pedicle flap was enhanced rib fracture healing, and the healing of rib fracture obliquely line was appeared more speed than transverse fracture

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