# COMPARATIVE HISTOMORPHOLOGICAL STUDY OF INTESTINAL ANASTOMOSIS BY LAPAROSCOPIC VS CONVENTIONAL METHODS IN DOGS.

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# ABSTRACT

This study was conducted to evaluate the histomorphological changes occurred during intestinal anastomosis performed by laparoscopic Vs conventional techniques in eighteen dogs, which were divided randomly into three groups. In group 1, intestinal anastomosis was performed extracorporeally by laparoscopic-assisted surgery, during which a loop of the small intestine was exteriorized through a mini-laparotomy opening and was surgically resected and anastomsed by simple interrupted sutures; in group 2, intestinal anastomosis was performed by intracorporeal laparoscopic surgery; and in group 3, intestinal anastomosis was performed by conventional laparotomy. Intestinal biopsies were obtained from the site of anastomosis in all dogs at days 15 and 30 following the operation.

Normal regeneration of the structural constituents of the intestine was observed 15 days following anastomosis in animals of the first and second groups compared to villous atrophy associated with delayed and partial regeneration of the intestinal mucosa that were manifested in animals of the third group. In addition, histopathological changes indicative of serosal adhesion were observed in two animals of the latter group. These results showed that intestinal anastomosis achieved by laparoscopic surgery revealed better and faster healing rates compared to intestinal anastomosis achieved by conventional laparoptomy.

# **INTRODUCTION**

Improvement in the outcome of intestinal anastomosis can be achieved through a better understanding of the basic mechanisms and patterns of intestinal healing following intestinal anastomosis. This better understanding can be realized through experimental studies in animal models using different suture materials, mechanical aids and techniques. It is on the basis of these experimental studies that improvements in human intestinal anastomoses have been made (1). Laparoscopic surgery for colon and rectal diseases has increased in importance, currently both malignant and benign intestinal diseases are being treated with minimally invasive surgery because of the touted advantages regarding cosmesis, reduced post operative stay, faster return to habitual activities, as well as similar safety and feasibility when compared to open colon and rectal surgery (2, 3).

Today, intestinal anastomosis can be constructed intracorporeally or extracorporeally in a laparoscopic-assisted manner (4). Laparoscopy offers an advantage in that there is less exposure of peritoneal organs to the atmosphere, thus reducing the consequent potential for intra abdominal infection; it prevents drying of the peritoneal surfaces which may be responsible for tissue ischemia (5).

The aim of this study was to evaluate the histomorphological outcome following intestinal anastomosis by laparoscopic Vs conventional techniques.

# **MATERIALS AND METHODS**

## 1. Experiment design

Eighteen local breed male and female dogs, of 15 - 25 kg weight and 1.5 - 3 years age, were used in this experiment. The dogs were divided randomly and equally into three groups. All dogs were healthy and free of congenital or acquired diseases as indicated by their physical and clinical examination. The animals were treated for internal and external parasites and they were housed during the experiment period in the College of Veterinary Medicine, University of Mosul.

#### 2. Anesthesia

The dogs were fasted 24 hours prior to surgery. During this period, the ventral abdominal wall, extending longitudinally from the xiphisternum to the pubis and bilaterally as far as the flanks, was aseptically prepared for the operation. Preanesthetic medication with atropine sulphate at a dose of 0.04 mg/kg body weight was given intramuscularly. Ten minutes later, a mixture of Xylazine 2% and Ketamine 5%, was given by intramuscular injection at a dose of 5mg/kg body weight and 15 mg/kg body weight, respectively.

### 3. Intestinal anastomosis

- a. In animals of the first group, the intestinal anastomosis was performed extracorporeally using laparoscopic-assisted surgery by exteriorization of a 5 cm-long jejunal loop together with its mesenteric fold through a mini-laparotomy incision (of 1.5 2 cm diameter) made in the umbilical region. This jejunal loop was partially resected and then anastomsed by simple interrupted sutures using polygalactin size 3/0. At last, the jejunal loop was placed back into the abdominal cavity through the mini-laparotomy incision.
- b. In animals of the second group, the intestinal anastomosis was carried out using intracorporeal laparoscopic surgery by suspending a jejunal loop of a sufficient length together with its mesenteric fold into the ventral abdominal wall by a laparoscopicassisted insertion of a series of 4-6 silk sutures through the abdominal wall into the anti-mesenteric surface of the selected jejunal loop. The suspended jejunal loop was grasped by two Babcock graspers (inserted together with a plastic entrapment sac through an already performed mini-laparotomy incision). The mesenteric blood vessels on either sides of the grasped jejunal loop were ligated and the jejunal loop together with its mesenteric fold was partially resected. The resected part was dragged into a plastic entrapment sac which was closed and placed temporarily within the abdominal cavity. Prior to anastomosis, the suspending sutures were loosened and the two ends of the partially resected jejunal loop, which was still grasped by the Babcock graspers, were sutured together by simple continuous suture using 3-0 polygalactin starting from the mesenteric border. The mesenteric fold was finally sutured by simple continuous mattress and the entrapment plastic sac was exteriorized through the minilaparotomy incision.
- c. In animals of the control group (third group), the intestinal anastomosis was performed by the conventional laparotomy technique, during which an incision sufficient to allow full exploration of the abdomen was made through the skin and lina alba. Following that a 5 cm-long jejunal loop was exteriorized, partially resected and then anastomosed by simple interrupted mattress using 3-0 polygalactin sutures. Finally the laparotomy incision was closed by routine manner.

#### 4. Biopsy collection and histological preparations

Biopsies of 0.5 cm length and width were obtained from all dogs at days 15 and 30 after anastomosis by conventional laparotomy. The biopsies were obtained from the site of anastomasis and incubated in10% neutral buffered formaline for 48 hours. Following that, they were exposed to a series of histological preparations prior to sectioning and staining with hematoxylin and eosin stains (6).

## RESULTS

Histopathological examinations for the intestinal biopsies revealed that the rate of healing at the anastomotic sites was faster and of better-quality in animals of the first group in which anastomosis was performed extracorporeally by laparoscopic assisted surgery, and in animals of the second group in which intestinal anastomosis was experienced by intracorporeal laparoscopic surgery in comparison with those of the third group which were exposed to conventional intestinal anastomosis.

Normal regeneration of the intestinal mucosa was observed 15 days following anastomosis in animals of the first group associated with insignificant pathological alterations represented by slight fibrous connective tissue proliferation and mild inflammatory cell infiltration in the submucosal layer (Figure 1). Similar results, i.e., normal mucosal regeneration associated with insignificant fibrous connective tissue proliferation and slight inflammatory cell infiltration in the submucosal layer, were observed 15 days following anastomosis in animals of the second group (Figure 2) with the exception of one animal which showed marked thickening of the serosal layer due to fibrous connective tissue proliferation, precipitation of edematous fluid and infiltration of mononuclear inflammatory cells (Figure 3).

On the other hand, villous atrophy and/or delayed and partial regeneration of the intestinal mucosa at the site of anastomosis associated with intensive fibrous connective tissue proliferation and marked inflammatory cell infiltration in the submucosal layer were manifested in animals of the third group (Figures 4, 5 and 6). In addition, the histopathological examination of the intestinal biopsies obtained from two animals in this group showed focal destruction of the smooth muscle fibers in the muscularis externa layer associated with blood vessels congestion, fat necrosis and marked infiltration of inflammatory cells within the serosal layer which also showed partial loss of its mesothelial integrity, an indication of serosal adhesion at the anastomosis site (Figures 7 and 8).

### DISCUSSION

The results of the present study indicate a rapid and better-quality rate of healing in animals of the first and second groups in which the intestinal resection and anastomosis were performed by laparoscopic-assisted extracorporeal surgery and intracorporeal laparoscopic surgery respectively, as indicated by the normal regeneration of the intestinal mucosa which was evident on day 15 following anastomosis and the insignificant pathological alterations that were observed at the site of the anastomosis. Such a satisfied results can be attributed to the circumstances of the laparoscopic surgery which comprise several advantages over the conventional laparoptomy such as; the small surgical incisions required to undertake the operation, the avoidance of cutting the abdominal muscles as the incisions are made in between through the muscle fibers, the minimal risk of incisional hernias, and the less opportunity of hospital-acquired infections (7, 8). However, an exception to this satisfied result was observed in one animal in the second group which showed a marked thickening of the serosal layer due to fibrous connective tissue proliferation, precipitation of edematous fluid and infiltration of mononuclear inflammatory cells. This displeased exception can be ascribed to the possibility of loosening and opening of the stitches at the anastomosis site resulting in leakage of the intestinal contents and peritoneal contamination. This possibility is predictable in all groups but is particularly expected in animals of the second group where the intestinal anastomosis was carried out by intracorporeal laparoscopic surgery in which the suturing technique and the numbers of stitches may not be secure and adequate as they are in the hand-sewn extracorporeal surgery (9). Intracorporeal laparoscopic surgery is safe in the hands of laparoscopically experienced surgeons (10, 11, 12).

In animals of the third group (in which intestinal anastomosis was performed by conventional laparotomy), histopathological changes indicative of a slower and lesser-quality rate of healing was observed in comparison with that observed in animals of the first and second groups. These histopathological changes were represented by villous atrophy and/or delayed and partial regeneration of the intestinal mucosa at the site of anastomosis associated with intensive fibrous connective tissue proliferation and marked inflammatory cell infiltration in the submucosal layer. This finding could be attributed to the extensive trauma that associates the large surgical incision through the abdomen and the higher possibility of contamination and intra-abdominal infection due to exposure of abdominal organs to the atmosphere (5). It is in agreement with findings of other authors (7, 8, 13) who mentioned that intestinal anastomosis performed by laporoscopic surgery revealed shorter hospital stay

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and improved cosmetic results compared to intestinal anastomosis achieved by conventional laparotomy. In addition, two animals in the third group showed focal destruction of the smooth muscle fibers in the muscularis externa layer and marked infiltration of inflammatory cells within the serosal layer which also showed partial loss of its mesothelial integrity. These pathological changes pointed out serosal adhesion at the site of anastomosis due to the extensive trauma that associate the conventional laparoptomy as indicated by the fat necrosis which was evident at the site of anastomosis in these two animals.

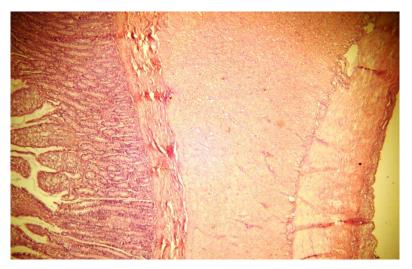


Figure 1: A hisotomicrograph of a biopsy obtained from the anastomosis site 15 days following anastomosis in the jejunum of a dog in group 1. It shows normal epithelization and restoring of the intestinal villi associated with slight fibrous connective tissue proliferation and mild inflammatory cell infiltration in the submucosal layer. H & E stain, X 100.

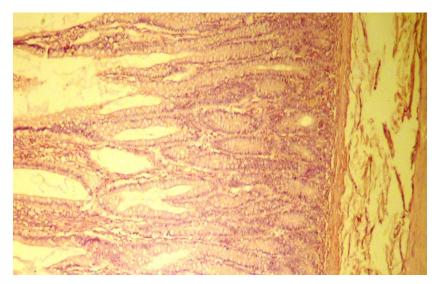


Figure 2: A hisotomicrograph of a biopsy obtained from the anastomosis site in the jejunum of a dog in group 2 showing normal epithelization and restoring of the intestinal villi 15 days following anastomosis. H and E stain, X 200.

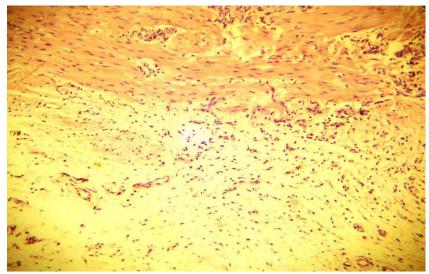


Figure 3: A hisotomicrograph of a biopsy obtained on day 30 from the anastomosis site in the jejunum of a dog in group 3. It shows marked thickening of the serosal layer due to fibrous connective tissue proliferation, precipitation of edematous fluid and infiltration of mononuclear inflammatory cells. Angiogenesis in the muscularis externa and serosal layers is also evident. H & E, X 200.

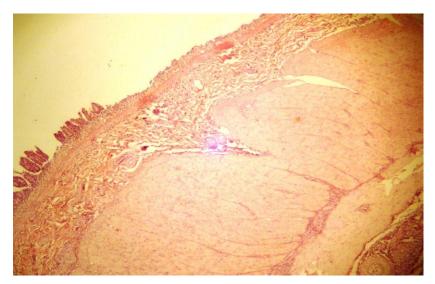


Figure 4: A Histomicrograph of a biopsy obtained on day 15 from the anastomosis site in the jejunum of a dog in group 3. It shows marked atrophy and loss of the intestinal villi over the anastomosis site associated with intensive connective tissue proliferation in the submucosal layer. H & E stain, X 100.

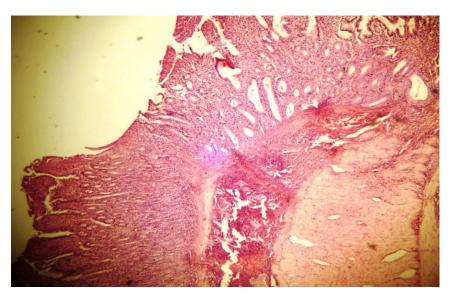


Figure 5: A histomicrograph of a biopsy obtained from the anastomosis site in the jejunum of a dog in group 3. It shows partial renewal of the intestinal villi 30 days following anastomosis associated with blood vessel congestion, connective tissue proliferation and marked infiltration of inflammatory cells in the submucosal layer. H & E stain, X 100.

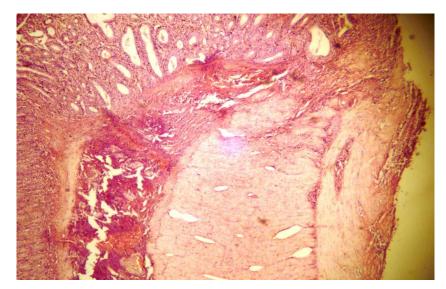


Figure 6: Higher magnification view of the histomicrograph illustrated in the previous figure. There are B.V. congestion, connective tissue proliferation and marked infiltration of inflammatory cells in the submucosal layer and angiogenesis and focal areas of inflammatory infiltrates in the muscularis externa layer. H & E stain, X 200.

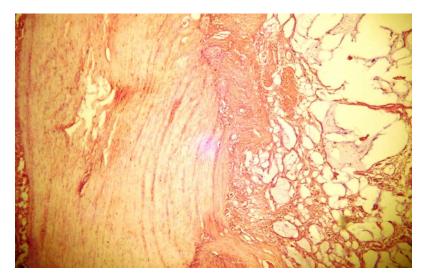


Figure 7: A histomicrograph of a biopsy obtained on day 30 from the anastomosis site in the jejunum of a dog in group 3. There are focal destruction of the smooth muscle fibers in the muscularis externa layer and blood vessels congestion and fat necrosis in the serosal layer. H & E stain, X 100.

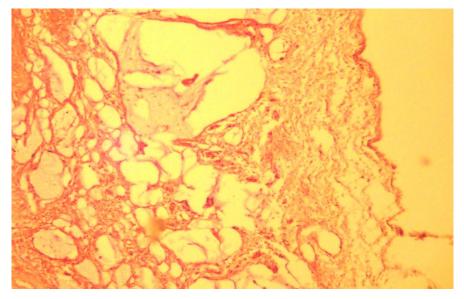


Figure 8: Higher magnification view of deeper layers of the tissue section illustrated in the previous figure. It shows fat necrosis, as indicated by the pinkish coloration of the cytoplasm of some fat cells, associated with marked infiltration of inflammatory cells within the serosal layer which suffers from partial loss of its mesothelial integrity. X 200.

دراسة مظهرية نسيجية مقارنة لجراحة التفمم المعوي المنظارية والتقليدية في الكلاب بهجت طيفور عباس \*؛ علي حسين حسن \*\*؛ عثمان جلال علي \* فرع الجراحة والأمراض التناسلية؛ \*\*فرع التشريح والأمراض؛ كلية الطب البيطري، جامعة السليمانية السليمانية، إقليم كردستان – جمهورية العراق

#### الخلاصة

صممت هذه الدراسة لأجل تقييم التغيرات المظهرية النسيجية بعد إجراء جراحة التغمم المعوي المنظارية والتقليدية. قسم 18 حيوان من فصيلة الكلاب بشكل عشوائي الى ثلاث مجاميع: أجريت عملية التغمم المعوي في حيوانات المجموعة الأولى خارج الجسم بواسطة تقنية الجراحة المنظارية المساعدة، حيث تم خلالها إخراج جزء من الأمعاء الدقيقة الى خارج الجسم من فتحة جراحية صغيرة في ميوانات المجموعة الأولى من فتحة جراحية صغيرة في حيوانات المجموعة الأولى من فتحة جراحية صغيرة الجراح جزء من الأمعاء الدقيقة الى خارج الجسم من فتحة جراحية صغيرة في حيوانات المجموعة الأولى من فتحة جراحية صغيرة في حيوانات المجموعة الأولى من فتحة جراحية صغيرة في حيدار البطن وتم أجراء قطع جراحي لهذا الجزء من الأمعاء الدقيقة ومن ثم فُمَّم بواسطة غرزات جراحية متقطعة بسيطة. في حيوانات المجموعة الثانية أنجزت عملية التغمم المعوي بواسطة تقنية الجراحة المنظارية المناعية أنجزت عملية التفمم المعوي بواسطة تقنية الجراحة المنظارية المحموعة الثانية أنجزت عملية التفمم المعوي بواسطة تقنية الجراحة. لمنظارية المحموعة الثانية أنجزت عملية التفمم المعوي بواسطة تقنية الجراحة المنظارية المنظارية المعاع وراحي لهذا الجزء من الأمعاء الدقيقة ومن ثم فُمًم بواسطة غرزات جرزات جراحية من المعام المعوي بولية القيمة المعوي بواسطة تقنية المنظارية غرزات جراحية من عروات المجموعة الثانية أنجزت عملية التفمم المعوي بواسطة تقنية الجراحة المنظارية داخل الجسم. اما في حيوانات المجموعة الثالثة فقد أجريت عملية التقمم المعوي بطريقة فتح البطن التقليدية. تم أخذ خزعات نسيجية من موقع جراحة التفمم المعوي من جميع حيوانات التجرية في الأيام 15 و 30 بعد الجراحة.

لوحظ حدوث إلنتام طبيعي للمكونات التركيبية لجدار الأمعاء خلال 15 يوم بعد إجراء جراحة النغمم المعوي في حيوانات المجموعتين الأولى والثانية بالمقارنة مع حيوانات المجموعة الثالثة التي لوحظ فيها ضمورالزغابات المعوية مصحوباً بالتئام جزئي ومتأخر في الطبقة المخاطية للأمعاء. وفضلاً عن ذلك لوحظت تغيرات مرضية نسيجية تدل على وجود التصاقات في الطبقة المصلية في إثنين من حيوانات المجموعة الأخيرة. دلَت نتائج هذه الدراسة على أنَ جراحة التقمم المعوي المنظارية قد اظهرت معدلات إلتئام افضل وأسرع بالمقارنة مع جراحة التقمم المعوي التقليدية.

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