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Comparison of Four Gastrotomy Closure Techniques in Dogs

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Abstract

This study aimed to evaluate and compare four double-layer closure techniques for canine gastrotomy. Twenty adult clinically healthy mongrel dogs of both sexes were involved in the present study. Dogs were randomly allocated into four groups; A, B, C, and D (n=5 for each group) based on the used technique for gastrotomy closure. In group A, the first layer of the stomach wall (mucosa and submucosa) and the second layer (tunica muscularis and tunica serosa) were closed in simple continuous suture pattern. In group B, both layers were closed using inverting suture pattern. In group C, the first layer was closed by inverting suture pattern, while the second layer was closed by simple continuous suture pattern. In the group D, the first layer was closed by simple continuous suture pattern, while the second layer was closed by stabling using the skin staplers. Physiological parameters (rectal temperature, heart rate, and respiratory rate) and the body weight were taken preoperatively, 7 and 14 days post-operatively. Dogs were euthanatized 14 days, postoperatively. The four techniques were evaluated for the procedure time, closure efficiency, postoperative complications, and histopathology. Results concluded that using double layer closure technique for gastrotomy wound was beneficial because the first suture line (first layer) provides homeostasis of the mucosa and submucosa, and the second suture line (outer layer) prevents leakage of gastric contents. The different closure techniques of gastrotomy wound in the present study gave satisfactory results, however, closing the first layer by simple continuous suture and the second layer skin staplers is considered to be the superior.

KEYWORDS
Canine, Gastrotomy, Stomach, Closure, Suture, Stapling

INTRODUCTION

The stomach acts as a reservoir that controls the size and rate of passage of ingesta into the small intestine, initiates the digestion of protein and fat, and the absorption of vitamins and minerals (Fossum and Hedlund, 2003). The most common indications for canine gastrotomy include gastric foreign bodies, neoplasia, gastric dilatation-volvulus, ulcerations, and surgical biopsies (Hayes, 2009; Williard, 2012; Hobday *et al*., 2014). Many surgical techniques have been described for closure of gastrointestinal incisions including different suture materials and patterns (Hardy, 1990; Fossum and Hedlund, 2003).

The large luminal diameter, abundant blood supply and rapid healing of the stomach provide many different options for gastric closure (Ellison, 2015). Engaging the submucosa and mucosa with continuous inverting pattern will generally ensure adequate healing of the stomach. The second layer should engage the serosa, muscularis and submucosa (Tobias and Johnston, 2012). The continuous Cushing or Lembert pattern has been recommended for closure of the second layer. However, the simple continuous pattern has been preferred in certain circumstances since less tissue is inverted (Ellison, 2015).

Although, the stomach is capable of rapid healing, due to its rich blood supply and paucity of intraluminal bacteria, wound breakdown and leakage are the most serious and catastrophic complications of surgery on the gastrointestinal tract (Allen *et al*., 1992; Shales *et al*., 2005). Dehiscence of a wound of the gastrointestinal tract often leads to generalized bacterial peritonitis and potentially death (Ellison, 2015). Consequently, technical failures and factors that negatively affect gastrointestinal healing are of great clinical significance to the surgeon (Ellison, 2011).

To the best of our knowledge, the available literature lacks a detailed study to compare different techniques of gastrotomy wound closure in dogs. Consequently, the present study aimed to evaluate and compare four closure techniques of canine gastrotomy. These techniques were evaluated for the procedure time, clinically, closure efficiency, postoperative complications, and histopathology.

MATERIALS AND METHODS

Ethical Approval

The present study was approved by the National Ethical Committee of The Faculty of Veterinary Medicine, Assiut University, Assiut, Egypt, according to the OIE standards for use of an-

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imals in research.

Experimental Animals

Twenty adult clinically healthy mongrel dogs of both sexes (13 males and 7 non-pregnant, non-lactating females) were involved in the present study. Their ages ranged from 1.5 to 3 years and their average body weight was 14.45 kg (range 12-20 kg). Dogs were housed in individual standard cages with food and water supplied ad libitum.

Dogs were randomly allocated into four groups; A, B, C, and D (n.= 5 dogs for each group) based on the used technique for stomach closure. In group A, the first layer of the stomach wall (mucosa and submucosa) and the second layer (tunica muscularis and tunica serosa) were closed in simple continuous suture pattern. In group B, both layers were closed using inverting suture pattern. In group C, the first layer was closed by inverting suture pattern, while the second layer was closed by simple continuous suture pattern. In group D, the first layer was closed by simple continuous suture pattern, while the second layer was closed by stabling using the skin staplers.

Body weight (kg) and physiological parameters included rectal temperature (RT, ºC), heart rate (HR, beat/min), and respiratory rate (RR, breath/min) were recorded pre-operatively and on the 7th and 14th days post-operatively. Dogs were euthanized 14 days, postoperatively. The four techniques were evaluated for the procedure time (min), clinically, closure efficiency, postoperative complications, and histopathology.

Surgical Techniques

On the day of surgery, dogs were fasted for 12 hours. Gastrotomy procedure was operated under the effect of intramuscular 2 mg/kg Xylazine HCl 2 % (Xyla-Ject, ADWIA Co., SAE, Egypt) and intravenous 10 mg/kg Ketamine HCl 5 % (Ketamine, Sigma-Tec Pharmaceutical Industries, SAE, Egypt). The animal was positioned on dorsal recumbency, prepared for aseptic surgery, and draped except for the surgical site.

The stomach was approached via a pre-umbilical midline 8-cm abdominal incision. The stomach was exteriorized and isolated by sterile wet surgical sponge. Two stay stitches were applied 1 cm away from both ends of the proposed incision site to prevent accidental gastric content overflow from the incision. A 5-cm incision was made midway between the greater and lesser curvatures of the stomach. In all dogs, the stomach was closed in two layers; the first layer incorporated the mucosa/submucosa, while the second layer included the serosa/musculosa using the same suture material (Polyglycolic Acid # 2-0, M-NATUR®, International Sutures Manufacturing Co., Egypt).

In group (A), the first and second layers were closed using simple continuous suture pattern. In group (B), both layers were closed using inverting suture pattern (Cushing sutures). In group (C), the first layer was closed by inverting suture (Cushing sutures), while the second was closed by simple continuous suture. In group (D), first layer was closed by simple continuous sutures, while the second layer was closed by disposable skin stapler F-35R (AdvaMed, Ningbo Advan Electrical Co., Ltd., Industrial Development Zone, Fuhai Town, CiXi City, Zhejiang, China).

The exteriorized stomach was flushed by sterile warm normal saline and reduced into the abdomen. Abdominal wall was closed in 3 layers. The parietal peritoneum with the linea alba as a layer and the subcutis as a second layer were sutured by simple continuous sutures using polyglycolic acid # 2-0. The skin was closed by simple interrupted sutures using silk #2-0.

Post-operative Evaluation

Dogs were daily examined for 14 days postoperatively for presence of any swellings, exudates from the operation sites, sepsis, patency of skin stitches, or any other complications (pyrexia, anorexia, and vomiting).

Postmortem Evaluation

Dogs were euthanized on day 14 post-operatively by intravenous administration of sodium pentobarbital at a dose of 100 mg/kg (Fatal-Plus® Vortech Pharmaceuticals, Dearborn, Michigan, USA). The operated stomachs were examined grossly for

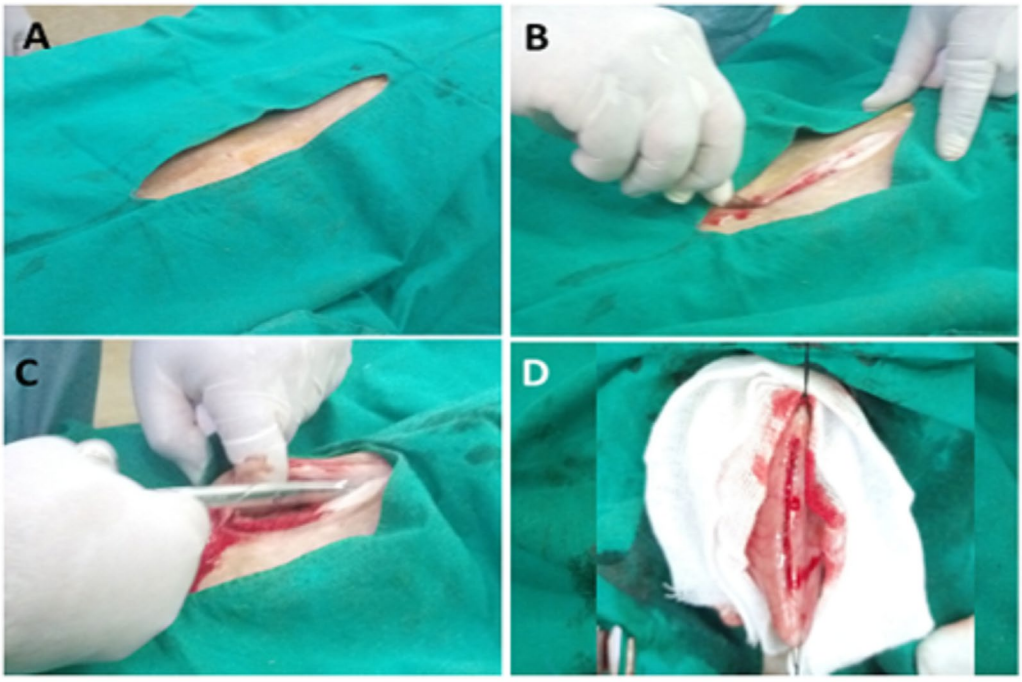

Fig. 1. Gastrotomy procedures: (A) draping of the surgical site, (B) initial pre-umbilical skin incision, (C) widening of celiotomy incision using a pair of scissors, and (D) exteriorization of the stomach and its isolation by sterile sponge and application of two stay stitches before its incision.

wound healing, leakage of gastric contents, wound dehiscence, peritonitis, and presence of adhesions. The gastrotomy site was also inspected from the mucosal aspect for the wound healing and presence of ulcerations. A scoring system (0 to 4) was developed to record the degree of adhesion between the operated stomach and the surrounding viscera. The score (0); no adhesions at all, score (1); mild adhesions between less than half of the line of gastrotomy site and surrounding viscera, score (2); moderate adhesions between nearly half of gastrotomy site and surrounding viscera, score (3); severe adhesions between two thirds to three fourths of the gastrotomy site and the surrounding viscera, and score (4); extensive adhesions between the whole gastrotomy site and the surrounding viscera.

Tissue samples were collected from the gastrotomy sites for histopathological evaluation. Specimens (0.5 × 0.5 cm) were fixed in 10 % neutral buffered formalin. The fixed specimens were dehydrated in graded alcohol series, cleared, embedded in paraffin and sectioned at 3-5 µm thick sections using Richert Leica RM 2125 Microtome, Germany. The obtained sections were stained with Harris Haematoxyline and Eosin stain (H&E). The stained sections were examined using a LeitzDialux 20 microscope. Images were taken using a Canon digital camera (CandisonPowershot A95). A score system was developed to evaluate stages of wound healing in different groups. The inflammatory stage of wound healing was given a score system from 0 to 4. Where, 0 means absence of inflammation; 1 means presence of mild inflammatory process; 2 means presence of moderate inflammatory process; 3 means presence of severe inflammation; and 4 means presence of extensive inflammatory process.

Stage of connective tissue proliferation was given a score system from 0 to 4. Where, 0 means absence of connective tissue proliferation; 1 means presence of mild degree of connective tissue proliferation; 2 means presence of moderate proliferation of connective tissue; 3 means presence of severe degree of connective tissue proliferation; and 4 means presence of extensive connective tissue proliferation. The presence of remodeling stage or gap formation was given score of 1, while absence of these features was given score of 0.

Statistical Analysis

The data were expressed as mean±SE and were analyzed by ANOVA using the GLM procedure of SAS (SAS institute, 2009). When treatment effects were significant, differences between least squares means were tested using Duncan's multiple-range test and the differences were considered significant at the level of $P < 0.05$.

RESULTS

Clinical Findings

There was a non-significant ($P>0.05$) increase in the mean body weight of all dogs during the study period (Table 1). There were non-significant changes in the mean heart and respiratory rates of dogs during the study period. There was a significant ($p=0.0016$) increase in the rectal temperature on the 7th day, postoperatively. The changes in rectal temperature were non-significant at POD14 (Table 1).

There was a significant ($P<0.0001$) decrease in hear rate of dogs in group B when compared to other closure techniques (A, C and D) (Table 2). Dogs in group C had significantly ($p=0.016$) higher respiratory rate than those in group B, while respiratory rate of dogs in other groups (A, C and D) did show significant changes (Table 2). There was a significant ($p<0.0001$) variation in rectal temperature among dogs of the four groups. The least recorded rectal temperature was in dogs of group C followed by those of group B and the highest recorded rectal temperature was in dogs A and D (Table 2).

Surgical Procedure Time

The procedure time lasted the longest time in dogs of group B, while the procedure time was the shortest in dogs group D. The variations in procedure time were significant among dogs of different groups ($P<0.0001$) (Table 3).

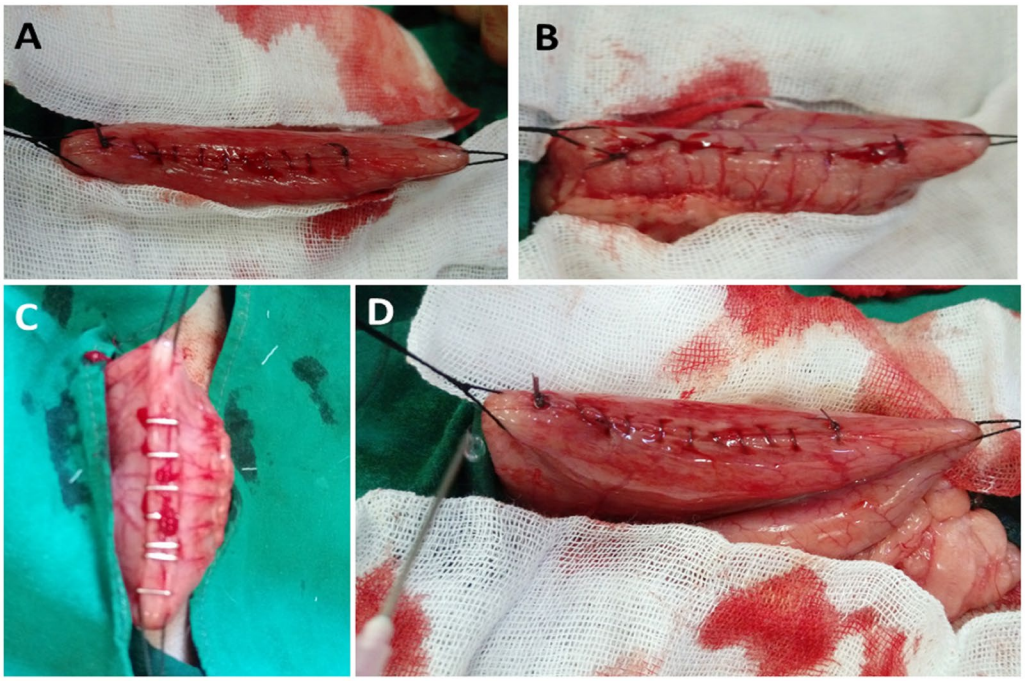

Fig. 2. Gastrotomy procedures: closure of seromuscular layer with (A) simple continuous sutures, (B) Cushing sutures, (C) skin staples, and (D) flushing of the gastrotomy site with sterile normal saline before its reduction into the abdominal cavity.

Post-operative Follow-up

Gastrotomy procedures were tolerated by all dogs. However, they were anorexic, dizzy and depressed following surgery, but they regained their normal feed intake and activity with no observed complications by the first postoperative day (POD1).

Gross Evaluation of Gastrotomy Wound

In all treatment groups, the inspection of gastrotomy site showed normal wound healing with no leakage of gastric contents, wound dehiscence, or peritonitis except that one of the dogs in the treatment group (B) died from peritonitis. In group (D), no staples were missing from the incision site.

The score of adhesions between gastrotomy site and omentum was the highest in dogs of group C, while the lowest score of adhesions was recorded in dogs of groups A and D (Table 3). There were significant (P=0.013) differences in mean scores of adhesions between dogs of different closure techniques (Table 3 and Fig. 3).

Histopathological Evaluation

The mean scores of wound healing stages in the four treatment groups were summarized in Table (3). In group (A), the proliferation of the epithelial layer of the mucosa and submucosa was complete but in a typical process. The inflammatory process was moderate (score 2), while the proliferation of granulation tissue with obviously seen (score 3). At the muscularis layer the inflammatory process was moderate (score 2), while there was

Table 1. Body weight and physiological parameters of dogs before and after gastrotomy in all dogs (n.=20).

Time	BW (kg)		HR (beats/min)		RR (times/min)		RT (˚C)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
PreOper.	13.12	0.3	61.5	3.37	45.83	5.4	38.52^{b}	0.11
POD7	13.7	0.32	58.17	3.75	51	5.71	39.03^{a}	0.16
POD14	14.05	0.37	56.55	3.23	50	5.46	38.72^{b}	0.25

Data are expressed as Mean ±SE.
PreOper.: Preoperatively; POD7: Postoperative day 7; POD14: Postoperative day 14. In the same column, values of similar letters have non-significant changes (p>0.05), while values of different letters has significant changes (p<0.05).

Table 2. Body weight and physiological parameters of dogs after gastrotomy in dogs of the four closure techniques (n.=5 in each group).

Group	BW (kg)		HR (beats/min)		RR (times/min)		RT (˚C)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE
A	15.16	0.17	62.89^{a}	3.47	48.00^{ab}	7.09	39.08^{a}	0.15
B	12.84	0.18	46.25^{b}	4.38	34.50^{b}	2.2	38.66^{b}	0.14
C	12.93	0.22	66.00^{a}	2.29	62.22^{a}	6.64	38.16^{c}	0.21
D	13.43	0.32	58.67^{a}	2.52	49.33^{ab}	4.22	39.11^{a}	0.18

Data are expressed as Mean ±SE.
In the same column, values of similar letters have non-significant changes (p>0.05), while values of different letters have significant changes (p<0.05).

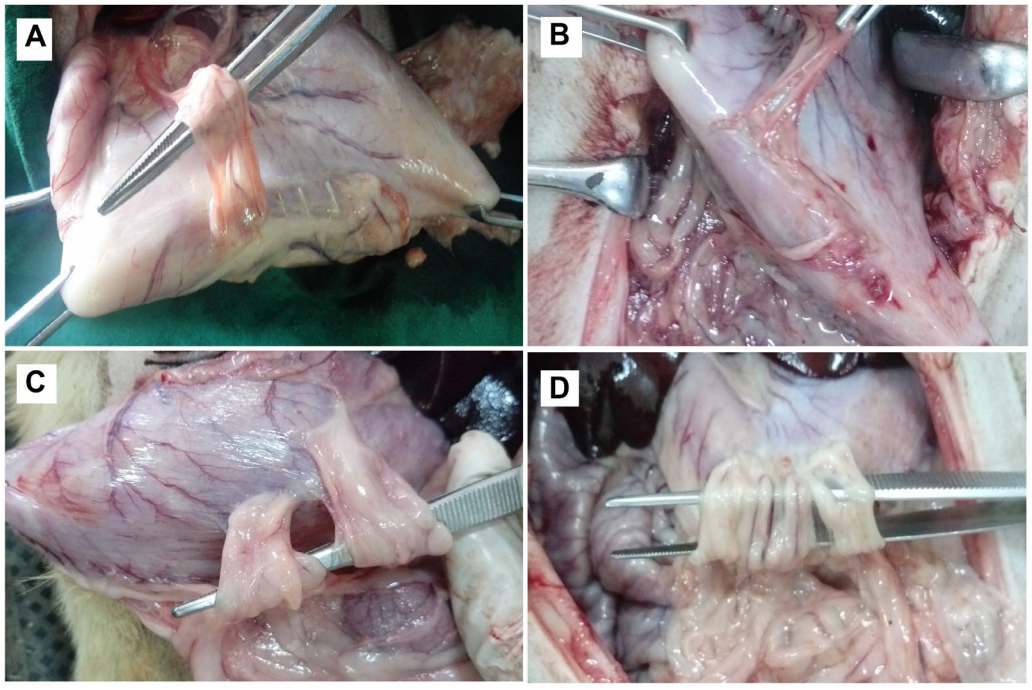

Fig. 3. Postmortem pictures showing different scores of adhesions between gastrotomy site and omentum; (A) score 1 in a dog of group D, (B) score 2 in a dog of group A, (C) score 3 in a dog of group A, and (D) score 4 in a dog of group B.

huge proliferation of connective tissue (score 3) with presence of macrophage cell proliferation. In group B, the mucosal layer showed presence of connective tissue proliferation in some areas (score 2) indicating healing by substitution, while other areas showed atypical regeneration. In the muscularis layer, the inflammatory and proliferation processes were obviously seen (score 3). In group C, the mucosal layer, the gap of incision was obviously seen (score 1) in some places with presence of extensive inflammatory cell reaction (score 4). In the muscularis layer, there was a huge proliferation of connective tissue (score 4) indicating retardation of healing process. In group D, the mucosal layer, it is more or less similar to group A (score 2). In the muscularis layer, the remodeling process was obviously seen (score 1) indicating a complete healing process.

DISCUSSION

A variety of surgical techniques have been described for closure of gastrointestinal incision (Silberstein and Rolandelli, 2012). The present study evaluated four closure techniques for gastrotomy in dogs to choose the one with the shortest surgery time, lowest cost and minimum postoperative complications.

Table 3. Gastrotomy procedure time, score of adhesions and histopathological evaluation scores of dogs in different groups (n.= 5 dogs in each group).

Group	Procedure Time	Adhesions Score	Histopathology			
			Inflammatory reaction	CT Proliferation	Remodeling	Gap
A	16±0.58^{c}	1.66±0.36^{b}	2	3	0	0
B	25±0.47^{a}	2.5±0.5^{ab}	3	2	0	0
C	20±0.65^{b}	3.66±0.29^{a}	4	4	0	1
D	13±0.38^{d}	1.66±0.42^{b}	2	2	1	0

Data are expressed as Mean ±SE.
CT: Connective Tissue. In the same column, values of similar letters have non-significant changes ($p>0.05$), while values of different letters has significant changes ($p<0.05$).

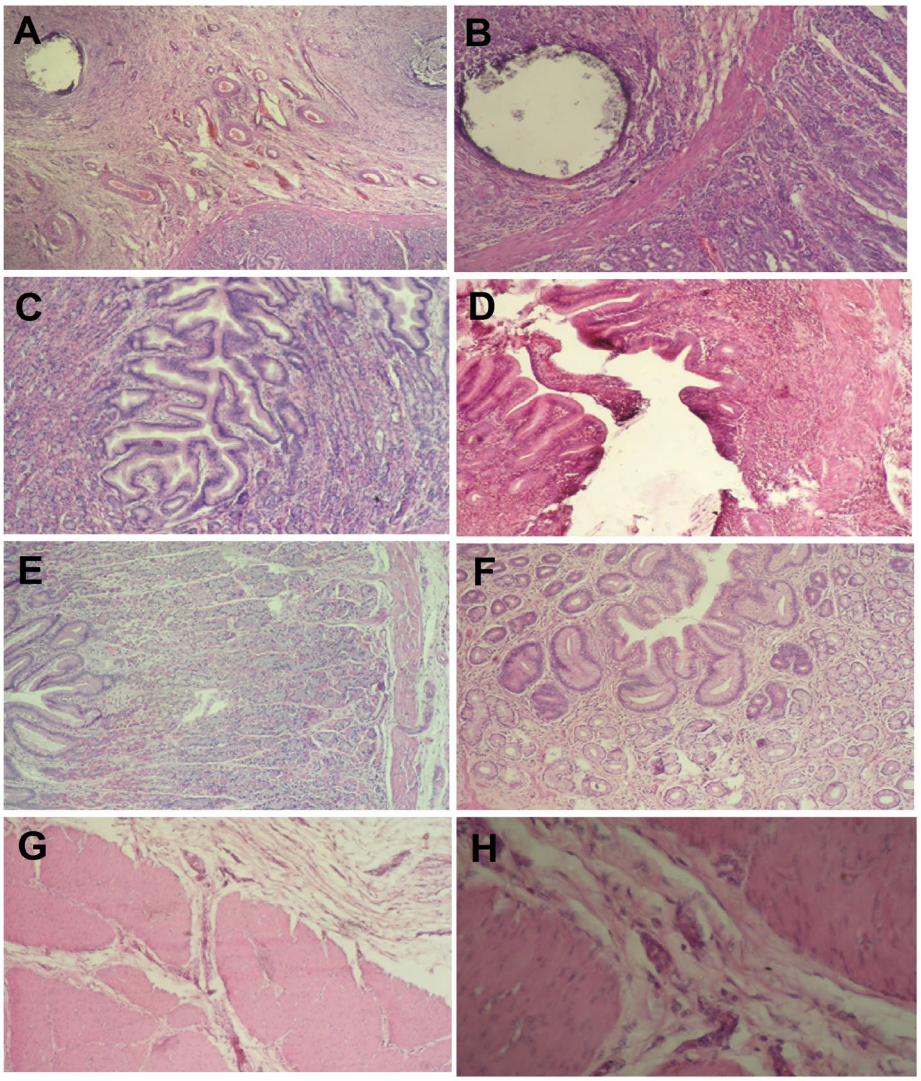

Figure 4. Photomicrographs are taken from the site of suture of stomach in dogs. In dogs of groups A, B and C: The mucosal and submucosal layers shows inflammatory reaction and granulation tissue formation around the suture materials, H&E 10×10 (A). Marked inflammatory reaction formed of neutrophil and macrophage cell infiltration surrounding the suture material, H&E 10 × 10 (B). Typical regeneration of the mucosal layer is clear, H&E 10×40 (C). In a dog of group C: The mucosal layers show intense inflammatory cell reaction with the presence of incision gap, H&E 10×10, (D). In dogs of group D: The mucosal and submucosal layer shows minimal inflammatory cell reaction with absence of granulation tissue formation, H&E 10×5 (E). The mucosal layer shows regeneration of the gastric glands. Some of the gastric glands are regenerated atypically, H&E 10×40 (F). The serosal and muscularis layer at the site of staples shows presence of minimal connective tissue formation, H&E 10×5 (G). The site of staple, note the presence of remodeling connective tissue at the site of incision H&E 10×40 (H).

In the present study, gastrotomy procedures were tolerated by dogs of all groups. The procedure time lasted the longest time in dogs in which the stomach was sutured by double layer of inverting sutures, while the procedure time was the shortest in dogs in which the stomach was closed by a layer of simple continuous sutures followed by a second layer of staples. Gastrotomy site showed complete wound healing with no leakage of gastric contents and no wound dehiscence. There were significant differences in mean scores of adhesions between dogs of different closure techniques. Closing the first layer by simple continuous suture and the second layer skin staplers is considered to be the superior.

Antibiotics were not given, postoperatively, in the present study. Antibiotics are not typically given postoperatively with gastric surgery unless gastric perforation is present or spillage occurs during procedures (Ellison, 2015).

In the present study, clinical findings showed variable differences among dogs of the four groups. There was insignificant increase in the body weight of all dogs, while there were significant changes in heart rate, respiratory rate and rectal temperature among the animals groups. These changes in physiological parameters may be attributed to fear and anxiety of the animals during recording of these parameters.

The procedure time lasted the longest time in dogs of group B, while the procedures time was the shortest in dogs of group D. These variations in procedure time were indeed due to the type of closure technique, which were two layers of inverting patterns in group B, while, in group D, the technique was performed by two layers, in which, the first layer was closed by simple continuous suture, while the second layer was closed by skin staplers.

It has been found that engaging the submucosa and mucosa with continuous inverting pattern in the first layer ensured adequate healing of the stomach (Tobias and Johnston, 2012; Ellison, 2015). In all groups of this study, the submucosa and mucosa were sutured as first layer either by simple continuous (in group A and D) or continuous inverting (in group B and C) technique.

The use of linear stapling devices is well described in the literature, but application can be challenging in smaller veterinary patients due to their organ size, cost of equipment and lack of familiarity with the technique or the availability of the stapling instruments for veterinary surgeons (Ullman *et al*., 1991; Tobias, 2007). In the present study, stapling technique was applied on the group D, where it was available in markets with low price, easy applied with the surgeon with short procedure time and resulted in relatively good wound healing when compared to other closure techniques especially from the histopathological score of wound healing.

The risk of leakage from gastrotomy wound is usually greatest in the first 72–96 hours postoperatively (Ellison, 2015). Inspection of gastrotomy site of all groups showed good healing without wound dehiscence or leakage of the gastric contents. These results of the current study were due to good wound closure with two layers of suture pattern as well as prompt wound healing due to the abundant blood supply of gastric wall.

Peritonitis is a life-threatening complication that can happen in 13 to 20% of the cases due to severe contamination during surgery and/or leakage of the suture line (Monnet, 2015). Peritonitis resulted in death of one dog (in group B) of the present study and constituted 5% of the total numbers of dogs. Good hygienic measures, minimizing the trauma to the tissues and observing the tissue viability were important to reduce the risk of peritonitis.

Other complications include vomiting, anorexia, ulceration at gastrotomy sites, and gastric outlet obstruction (Fossum and Hedlund, 2003) were not recorded in dogs of the present study.

Adhesions between gastrotomy site and omentum were recorded in all groups. The lowest score was recorded in groups A and D. Type of suture technique play a role on the degree of adhesions. Immediately after wounding, platelets aggregate, the coagulation mechanism is activated and fibrin clots are deposited to control hemorrhage (Ellison, 1989). Fibrin has adhesive properties and may be converted to fibrous adhesions.

The process of wound healing is divided into three overlapping phases; inflammation, proliferation, and remodeling (Witte and Barbul, 1997; Singer and Clark, 1999). Histopathological examination of gastrotomy wounds of the current study 14 days following surgery showed presence of inflammation and connective tissue proliferation in dogs of all groups, while remodeling was recorded in dogs of group D only, which considered a sign of complete wound healing. These results indicated that complete and good healing was observed in case of gastrotomy wound closure using stapler technique in the second layer.

CONCLUSION

Using double layer closure technique for gastrotomy wound is beneficial because the first suture line (first layer) provides homeostasis of the mucosa and submucosa, and the second suture line (outer layer) prevents leakage of gastric contents. The different closure techniques of gastrotomy wound in the present study gave satisfactory results, however, closing the first layer by simple continuous suture and the second layer skin staplers is considered to be the superior.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

Allen, D.A., Smeak, D.D., Schertel, E.R., 1992. Prevalence of small intestinal dehiscence and associated clinical factors: a retrospective study of 121 dogs. Journal of American Animal Hospital Association 28, 70-76.

Ellison, G.W., 1989. Wound healing in the gastrointestinal tract. Seminars in Veterinary Medicine and Surgery (Small Animal) 4, 287–98.

Ellison, G.W., 2011. Complications of Gastrointestinal Surgery in Companion Animals. Veterinary Clinics of North America Small Animal Practice 41, 915–934.

Ellison, G.W., 2015. The stomach. In: BSAVA Manual of Abdominal Surgery. Second edition. Editors: Williams JM, Niles JD. British Small Animal Veterinary Association Publications. Gloucester, England. pp. 64-88.

Fossum, T.W., Hedlund, C.S., 2003. Gastric and intestinal surgery. The Veterinary Clinics, Small Animal Practice 33, 1117–1145.

Hardy, K.J., 1990. Non-suture anastomosis: the historical development. Australian and New Zealand Journal of Surgery 60, 625-633.

Hayes, G., 2009. Gastrointestinal foreign bodies in dogs and cats: a retrospective study of 208 cases. Journal of Small Animal Practice 50, 576-583.

Hobday, M.M., Pachtinger, G.E., Drobatz, K.J., Syring, R.S., 2014. Linear versus non-linear gastrointestinal foreign bodies in 499 dogs: clinical presentation, management and short-term outcome. Journal of Small Animal Practice 55, 560-565.

Monnet, E., 2015. The proceedings of the 40thWorld Small Animal Veterinary Association Congress, Bangkok, Thailand I 15-18 May, 2015, pp. 286-189.

Shales, C.J., Warren, J., Anderson, D.M., 2005. Complications following full thickness small intestinal biopsy in 66 dogs: a retrospective study. Journal of Small Animal Practice 46, 317-321.

Silberstein, I., Rolandelli, R., 2012. Suturing, stapling, and tissue adhesive. In: Shackelford's Surgery of the Alimentary Tract. 7th edn. Ed C. J. Yeo. Elsevier Saunders, Philadelphia, PA, USA. pp 920-928.

Singer, A. J., Clark, R.A., 1999. Cutaneous wound healing. New England Journal of Medicine 341, 738-746.

Tobias K.M., Johnston S.A., 2012. Veterinary Surgery: Small Animal Volume 2, Saunders, p. 1490.

Tobias, K.M., 2007. Surgical stapling devices in veterinary medicine: a review. Veterinary Surgery 36, 341-349.

Ullman, S.L., Pavletic, M.M., Clark, G.N., 1991. Open intestinal anastomosis with surgical stapling equipment in 24 dogs and cats. Veterinary Surgery 20, 385-391.

Williard, M.D., 2012. Alimentary neoplasia in geriatric dogs and cats. Veterinary Clinics of North America: Small Animal Practice 42, 693-706.

Witte, M.B., Barbul, A., 1997. General principles of wound healing. Surgical Clinics of North America 77, 509-528.