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A Comparative Analysis of Different Gastrotomy Closure Techniques in Dog Models Using Barbed, Skin Stapler, and Polyglactin 910 Suture

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Abstract

Generation, according to molecular studies that demonstrated stronger angiogenic factor production and fewer proinflammatory cytokines. The polyglactin 910 suture studies that demonstrated stronger angiogenic factor production and fewer proinflammatory cytokines. The polyglactin 910 suture caused inflammation and epithelium regeneration problems. Skin staplers cause more wound inflammation than absorbable barbed but less than polyglactin 910. Absorbable barbed suture caused inflammation than absorbable barbed but less than polyglactin 910. Absorbable barbed sutures cause more wound inflammation than absorbable barbed but less than polyglactin 910. Absorbable barbed sutures cause the fastest and safest way to close the stomach with speeding tissue regeneration.

Keywords: Gastrotomy, Barbed suture, Polyglactin910 Suture, Skin stapler, Dog.

Introduction

Gastrotomy in pet animals may be necessary for Gastrotomy in pet animals may be necessary for various reasons, such as foreign body removal [1-3], correction of gastric dilatation volvulus [4],tumor excision [5], or the management of certain conditions gastrointestinal (GIT) such as gastropathy hypertrophic and gastroduodenal ulceration, gastric retention and, biopsy [6]. While gastrotomy can be a life-saving procedure, it is not possible without potential complications.

Infection is the most important surgical complication. Like any surgical procedure, gastrotomy carries a risk of infection at the incision site or within the abdominal cavity. Leakage of gastric contents from the surgical site is one of the primary concerns following gastrotomy [7-10]. This can lead to peritonitis [11], and inflammation of the

abdominal cavity. Bleeding can occur during or after the gastrotomy procedure. Some animals may experience delayed wound healing or wound dehiscence, where the surgical incision reopens. This can be due to factors such as poor tissue quality, infection, or excessive tension on the wound [12].

GIT disturbances are the most important postoperative complications after gastrotomy, dogs and cats may experience vomiting, diarrhea, or a decreased appetite. Gastric dilation after gastrotomy can cause discomfort, compromised blood flow, and potential complications requiring urgent treatment [13]. Adhesions can cause intestinal obstruction or interfere with normal GIT function. Type of suture technique plays 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 [14].

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In gastrotomy procedures, the primary goals are to achieve secure wound closure and promote optimal healing without compromising the integrity of the stomach [15].

Albert, in 1977, described vicryl suture as the best superior surgical material used in GIT surgery due to its biological character and its absorption rate.

Polyglactin 910 (Vicryl), an absorbable braided multifilament suture, is the most appropriate suture material selection, so the surgeons are using it as the most common, simple, and traditional technique for GIT surgery [16].

The GIT incision is closed in the conventional method by the cushing suture pattern in one layer, which provides an approximate apposition of the gastric wound [17, 18]. The hand-sewn suture is acceptable as it provides a safe and efficient incision close, but it has a dangerous risk factor as it may cause narrowing in the GIT lumen [19]. Additionally, the suturing consumed more time [20].

Barbed sutures are a type of suture material that has small, evenly spaced barbs along its length. These barbs allow for a self-anchoring effect, eliminating the need for traditional knot tying.

The barbe size is related to the size of the suture [21]. Using knotless suture materials has a significant decrease in time consumption during surgery [22]. Without tying knots, the suture's self-anchoring, unidirectional barbs enable stable tissue apposition in continuous patterns, which provide efficient incision closure in minimal time [23], avoiding the slipping of the suture due to barbs which restrict backward movement [24] as About 50% of laparoscopically tied sutures rupture due to knot slippage, even in the hands of experienced surgeons [22, 25]. During suturing, two or three bites needed to be extended after the incision end point, as an already-constructed end has been embedded into these suture devices In 2013, Nicole. demonstrated that the [26]. absorbable barbed suture provides a safe and efficient incision for GIT closure with minimal time. Recently absorbable barbed sutures have been considered game-changers in GIT surgery because they provide perfect apposition of the wound lips [23] with minimal tissue manipulation, thereby decreasing fatal complications following the procedure and improving healing, which speeds up the restoration of normal GIT motility and function. Thus, absorbable barbed suture is considered a breakthrough in spite of its pricey cost [27] that improve wound healing due to decrease of ischemia, and less suture extrusion [28].

While absorbable barbed sutures have gained popularity in certain surgical procedures, their use in gastrotomy procedures is not widespread practice. The digestive tract, including the stomach, is a dynamic organ that undergoes continuous movement

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and peristalsis. The use of absorbable sutures in gastrotomy raises concerns due to the potential for tissue damage or irritation caused by the barbs. The barbs may increase the risk of tissue trauma, inflammation, or even perforation within the GIT tract [29].

Skin stapler in gastrotomy procedures provide safety [30], efficient closure, less operation time [31], and a reduction in cost. in addition to easy performing [32]. So that, surgeons create an interrupted, inverting seromuscular single layer using stainless steel skin staples. Nowadays, Skin staplers are an acceptable method in dogs GIT surgery [33-36].

The aim of this work was to assess and compare the efficiency of knotless self-anchoring absorbable barbed sutures, skin staplers with a conventional hand-sewn suture using polyglactin 910 sutures in gastrotomy wounds in dogs.

Material and Methods

Experimental animals

In this study, fifteen male and fifteen female dogs (mean body weight was 14 kg) were used. A complete physical examination [respiratory rate (RR) (15-30 breaths/minute), heart rate (HR) (60-100 beats/ minute), rectal temperature (RT) (38-38.5 °C)] was performed for only the clinically apparently healthy dogs. Dogs were inhabited one week before surgery. Sufficient food and ad libitum water were provided for every animal housed in an individual box. All animals are kept in separate boxes. Eight hours before surgery, all of the dogs were fasting. The dogs were split into three groups, each consisting of five male and five female dogs. Group (A) had a gastric incision closed using a stainlesssteel skin stapler, Group (B) had a control group with a gastric incision closed using a Vicryl suture, and Group (C) had a gastric incision closed using a absorbable barbed suture. Prior to surgery, the dogs were fasted for approximately eight hours.

Surgery

After administering of an intravenous injection of 1 mg/kg xylazine hydrochloride (Xylaject 2%, ADWIA, Egypt) to sedate the dogs, 10 mg/kg of ketamine hydrochloride (Ketamax-50, Troikaa Pharmaceuticals Ltd., India). The abdomen was routinely aseptically prepared for surgery. Afterward a ventral midline laparotomy incision was made, and in order to reduce the risk of gastric fluid contamination, the stomach was extracted from the abdominal cavity and is isolated using a laparotomy sponge. Next, the stomach body's avascular region between the greater and lesser curvatures was cut open by a scalpel to create a 10-cm gastrotomy incision. Three separate procedures were then used to close the gastrotomy incision in one layer. In group A, the stomach incision was closed with stainless steel pins that had an interspace of roughly 2 mm using a skin stapler. Vicryl sutures (2/0) were used to close the incision in group B using an inverted Cushing suture pattern. Group C underwent absorbable barbed sutures with unidirectional self-anchoring stitches to seal the stomach incision (Fig. 1). The wound was then rinsed with a warm, sterile saline solution (0.9% NaCl). The completion time of each gastrotomy closure was recorded. Ultimately, the abdominal wall was closed routinely. Every animal was closely observed until they had recovered from the anesthetic effects.

Aftercare and follow-up

Prior to being allowed to walk around freely, the dogs were kept under close watch. There was restricted access to freshwater six to twelve hours after surgery. Little servings of solid food were given every six hours for twelve hours after surgery, and after thirty-six hours, the diet was gradually increased to a regular one. Seventy-two hours following surgery, the dog was given Ringer's solution (60 milliliters per kilogram of body weight, IV) daily. To prevent infection and minimize inflammation, administer an intramuscular injection (I.M.) of a prophylactic dose of Ceftriaxone (50mg/24 hours) (EPICO (Egyptian International Pharmaceutical Industries), Egypt) for three days following surgery, as well as Meloxicam (Anticox2, ADWIA, Egypt) at a dose of 0.2 mg/kg BW.

Clinical evaluation

The dogs were clinically assessed every day before and after operation. Fever, vomiting, anorexia, lethargy, diarrhea, and problems with wound healing were noted. Palpating the stomach on the left side of the abdominal wall was part of the general examination. Throughout the study period, RT, HR and RR were recorded at scheduled times to assess any infection or inflammation as well as body weight was examined on the weigh machine.

Gastric emptying time

Positive-contrast radiography was used to measure the stomach emptying time 21 days after surgery. Each dog was orally administered 10ml barium sulfate solution (70%) per kg of body weight. Every thirty minutes, until the stomachs of all the dogs were completely empty, positive contrast radiographic images were collected.

Hematological Evaluations

Test tubes containing anticoagulants were used to collect blood for hematological analysis in days 0, 5, and 21 after surgery. The blood work includes measuring the hemoglobin level, red blood cells, white blood cells (WBCs), and the differential leucocytic count (total leucocytes, neutrophils, lymphocytes, eosinophils, and macrophages) to investigate any infections or blood loss-related anemia.

Methods for histopathology

Specimen collection

On the 21^{st} postoperative day, the abdominal cavity was explored to detect adhesions, leakage, or collections of clotted blood. Stomach biopsy was taken from all groups under general anesthesia. Samples were fixed in neutral buffered formalin (10%). Following the processes of dehydration and clearing, the tissues were fixed in paraffin and sectioned at a thickness of 5 µm. The serial sections were stained using Masson's trichrome stain and hematoxylin and eosin [37]. Other specimens were collected and emersed in liquid nitrogen for RNA Isolation.

Methods used for immunohistochemistry [38]

The serial sections were dewaxed, hydrated and immersed in an antigen retrieval (EDTA solution, PH 8). The slides were then treated with hydrogen peroxide 0.3% and protein block, followed by incubation with anti-NF-KB P65 (Santa cruz, Cat# (F-6): sc-8008, 1:100 dilution) and TGF-β (Invitrogen, USA, Catalog # PA1-9574). The slides were rinsed three times with PBS, incubated with secondary antirabbit IgG antibodies (Envision+system HRP;Dako) for 30 minutes at room temperature, visualized with di-aminobenzidine commercial kits (liquid DAB+ substrate chromogen system ; Dako) and finally counter stained with May's haematoxylin. As a negative control procedure, the primary antibody was replaced by normal mouse serum antibody of NF-KB P65 and TGF-β. The reaction of both markers was expressed as the percentage of positive immunostaining per mm² using ImageJ analysis software (NIH, USA).

Methods for Realtime PCR

The relative expression of the mRNA of the VEGFA, MMP9, IL6, and MCP-1 genes was assessed using real-time PCR. Initially, an RNeasy Mini kit (Qiagen) was used to obtain total RNA from liver tissue. 1% agarose gel electrophoresis and Nanodrop, respectively, were used to evaluate the integrity and purity of the RNA. Subsequently, 4 mg of the extracted RNA underwent reverse transcription with Quantiscript reverse transcriptase to produce cDNA. Third, using the StepOnePlus realtime PCR system (Applied Biosystem, USA) and QuantiTect SYBR Green qPCR Master Mix along with gene-specific primers created using the Primer 3 web tool based on the published rat sequence, the generated cDNA was utilized as a template for a realtime PCR reaction. The target genes' critical threshold (Ct) values were normalized using the internal control's (β actin) Ct values.

VEGFA gene [39]: Forward: 5'-CGTGCCCACTGAGGAGTT-3', Reverse: 5'-GCCTTGATGAGGTTTGATCC-3'

MMP2 gene [39]: Forward: 5'-GAGCGAGGGTACCCCAAG-3', Reverse: 5'-GCTCCAATTAAAGGCAGCAT-3'

IL6 gene [40]: Forward: 5'-CCCACCAGGAACGAAAGAGA-3', Reverse: 5'-CTTGTGGAGAGGGAGTTCATAGC-3'

MCP-1 gene [40]: Forward 5'-GAGTCACCAGCAGCAAGTGT-3', Reverse 5'-TGGGTTTGGCTTTTCTTGTC-3'

β-actin gene [41]: Forward 5'-GCCAACCGTGAGAAGATGACT', Reverse, 5'-CCCAGAGTCCATGACAATACCAG-3'

Statistical analysis

Statistical analysis was done by SPSS v27 (IBM©, Chicago, IL, USA). The Shapiro-Wilks test and histograms were used to evaluate the normality of the distribution of data. Quantitative parametric data were presented as mean and standard deviation (SD) and were analyzed by one way ANOVA (F) test followed by post hoc test (Tukey). Qualitative variables were presented as frequency and percentage (%) and were analyzed utilizing the Chisquare test. A two tailed P value < 0.05 was considered statistically significant.

Sample size calculation

The sample size calculation was performed using G. power 3.1.9.2 (Universitat Kiel, Germany). The sample size was calculated based on the following considerations: 0.05 α error and 80% power of the study to demonstrate 10% increase in gastric closure time with stainless-steel skin stapler than polyglactin 910 (mean 10.75 min and SD 0.8 min) and absorbable barbed sutures (mean 8.4 min and SD 0.6 min) according to a previous study [42]. Two dogs were added to each group to overcome dropout. Therefore, 10 dogs were allocated in each group.

Results

Surgery

Surgical time was significantly different among stapler, polyglactin 910 and barded materials (P<0.001). Surgical time was significantly higher with polyglactin 910 material and absorbable barbed material than stapler material and with polyglactin 910 material than barbed material (P <0.001). Regarding stapler material, the mean value of surgical time was 45.8 ± 12.89 sec. Regarding polyglactin 910 material, the mean value of surgical time was 228 ± 29.17 sec. Regarding absorbable barbed material, the mean value of surgical time was 95.1 ± 6.72 sec. (Table 1)

Clinical findings

Every surgical operation was successfully completed without any postoperative issues. All thirty dogs survived, and there were no reported deaths. Following the surgeries, each animal maintained good overall health. Close monitoring of the animals' physical parameters, including RR, HR, RT, and body weight, revealed that they remained within the normal range. No significant changes were observed in these physical criteria, and there were no notable postoperative complications such as wound dehiscence or leakage. The incision site showed no signs of pus discharge or other indications of infection. The wounds healed perfectly, without any signs of inflammation or skin swelling. Throughout the investigation, the dogs exhibited normal defecation and food intake, without any lethargy, anorexia, vomiting, or fever within 5 days after the surgery. Furthermore, they gradually returned to their normal activity levels .

Radiographic Findings

The stomach emptying time was assessed 21 days after the surgery using contrast radiography with a positive agent (barium sulphate). As shown in Fig. 2 and Fig. 3, there was no significant difference observed between group B that ranged from 2 to 3 hours with a mean value 2.4 ± 0.42 hours and group C that ranged from 2.5 to 3.5 hours with a mean value 3.2 ± 0.45 hours. However, group A (skin stapler) exhibited a significant longer duration for the evacuation of stomach contents. As a result, the longest stomach emptying time was observed in group A that ranged from 4 to 5 hours with a mean value 4.6 ± 0.42 hours.

Gross findings and necropsy

In groups B and C, no adhesions were observed between the stomach and the omentum, body wall, or other organs. However, in Group A, minimal adhesion was visually observed between the abdominal wall at the incision site and the omentum (Fig. 4).

Hematological Evaluation

CBC at day 0, and 5 were insignificantly different between all groups. Regarding CBC at day 21, RBC, HB, HCT, PLT, WBCs, basophils, eosinophils and neutrophil were insignificantly different among three materials. Lymphocytes was significantly higher in stapler material and absorbable barbed material than polyglactin 910 material (P value =0.005 and <0.001) and was insignificantly different between stapler material and absorbable barbed material. Macrophages were significantly lower in stapler material and absorbable barbed material than polyglactin 910 material (P < 0.05) and was insignificantly different between stapler material and absorbable barbed material (Table 2).

On the first day of sample collection, there were no significant variations observed in the hematological parameters between the different groups. This included the RBCs, HB, Hematocrit values, as well as the components of the differential leucocytic count (total leukocytes, neutrophils, lymphocytes, eosinophils, macrophages). On the 5th day post-operation, noticeable neutrophilia and leukocytosis were observed compared to the initial day of the study. Additionally, a decrease in RBC count and HB level was noted. However, there were no significant differences observed in the hematological data on the 5th day between the different groups, except for a noticeable increase in TLC in group B (polyglactin 910). By the 21st day post-operation, both the group A (stapler) and group C (barbed) exhibited a mild but non-significant reduction in RBC count and HB level, along with a normal leukogram. However, the group using polyglactin 910 (vicryl) sutures still showed leukocytosis with neutrophilia and monocytosis, which was statistically significant (P < 0.05).

Histopathological findings

H&E-Staining

Histopathological evaluation of the healing effects of different suture materials, including polyglactin 910 (Vicryl), skin stapler, and absorbable barbed sutures, on gastrotomy closure revealed distinct healing responses across the different layers of the stomach. Polyglactin 910 (Vicryl) sutures demonstrated clear intestinal regeneration with some congestion and exhibited an intense submucosal inflammatory reaction, characterized by an abundance of neutrophils and macrophages, especially in the peri suture area. Gastric gland regeneration was evident, and the submucosal regions surrounding the Vicryl material showed a notable tissue reaction characterized by the presence of macrophages. The myenteric plexus demonstrated normal histological features, with normal cytoplasm and nucleus and a prominent nucleolus. In the case of skin stapler sutures, the mucosal layer exhibited incomplete regeneration, with decreased thickness of regenerative mucosa and a low number of gastric glands, accompanied by wide interstitial fibrosis between the gastric glands. Regenerative changes were observed in the mucosa, indicating proliferative epithelial cells. The submucosal tissue showed insufficient healing capacity, with dilated lymphatics, interstitial fibrosis, marked edema, and mild collagen deposition, especially in the peri suture area.

Barbed sutures, being non-absorbable, demonstrated complete epithelial regeneration in the mucosal layer, even with persistently congested blood capillaries. Submucosal regions displayed a wound cavity at the suture site with low inflammatory cell infiltration surrounded by mature collagen fiber deposition, although with less remodeling compared to other materials. Overall, absorbable barbed sutures exhibited the most favorable healing response, with complete epithelization of the mucosal lining and diminished vascular and inflammatory changes. In contrast, stapler sutures showed the least favorable outcomes, with incomplete mucosal regeneration and significant tissue reactions in the submucosa. Polyglactin 910 (Vicryl) sutures showed an intermediate response, with clear intestinal regeneration but notable inflammation around the suture material (Fig. 5).

Masson's trichrome stain

Masson's trichrome staining was conducted to assess the fibrous connective tissue and collagen deposition within the mucosa and lamina propria surrounding the suture material. In animals sutured with a stapler, the mucosa exhibited peri glandular fibrosis, and prominent observed fibrosis was seen around the staples and extending into the lamina propria. Meanwhile, other surrounding areas displayed separated bundles of connective tissue. Polyglactin910 (Vicryl) material demonstrated a reduction in interstitial fibrosis between gastric glands, primarily restricted to the region beneath the covering epithelium. The vicinity around the polyglactin 910 fibre exhibited marked fibrous connective tissue proliferation. Barbed suture led to a remarkable decrease in mucosal fibrosis, displaying a normal pattern, particularly parallel to the glandular structure. The area surrounding the absorbable barbed suture displayed intense, regular, and uniformed connective tissue fibres (Fig. 6).

Immunohistochemical Findings (NF- κ B-p65 & TGF- β 1)

Nuclear Factor Immune Staining (NF-KB (P65) was prominently observed within the inflammatory cells surrounding the suture materials, particularly evident in macrophages and lymphocytes around the diffusely distributed around staples, the polyglactin910 (Vicryl) fibre, and markedly decreased in the absorbable barbed suture. Quantitative scoring revealed a significant increase in the number of positively stained NF-kB-p65 cells in the group B (Polyglactin910) compared to the group A (skin stapler) (P < 0.01). Group C (barbed suture) exhibited a marked decrease in NF-KB-p65 cells compared to both group A (skin stapler) (P <0.01) and the group B(polyglactin910) (P < 0.001). Transforming growth factor β (TGF- β 1) exhibited immunostaining marked in the group B(polyglactin910), followed by group C (barbed) and then group A (skin stapler). In multiple comparisons, a significant increase in the expression was observed in the group B(polyglactin910) compared to group A (skin stapler) (P < 0.001) and barbed (P < 0.05) groups. Conversely, group C (barbed) showed an increased expression compared to group A (skin stapler) (P < 0.01) (Fig 7).

Realtime PCR

The mRNA expression levels of IL6, MMP2, MCP1, and VEGFA within the sutured gastric mucosa were analyzed for group A (skin stapler), group B (polyglactin 910), and group C (barbed). Regarding IL6 expression, group B (polyglactin 910) showed a significant increase compared to both group A (skin stapler) (P < 0.001) and group C (barbed) (P < 0.01). However, no significant difference was observed between group C (barbed) and group A (skin stapler). In terms of MMP2 expression, group B (polyglactin 910) exhibited a significant increase compared to group C (barbed) (P < 0.05), while group A (skin stapler) showed a significant elevation compared to group C (barbed). There was no significant difference between group A (skin stapler) and group C (barbed). On the other hand, the mRNA expression levels of MCP1 were significantly higher in group B (polyglactin 910) compared to both group A (skin stapler) (P < 0.05) and group C (barbed). Group A (skin stapler) also showed higher expression levels than group C (barbed) (P < 0.01). Furthermore, VEGFA expression levels were significantly higher in group B (polyglactin 910) compared to both group A (skin stapler) (P < 0.01) and group C (barbed) (P < 0.05). Additionally, group A (skin stapler) exhibited significantly higher expression levels than group C (barbed) (P < 0.01) (Fig. 8).

Discussion

A gastrotomy is one of the most common surgeries in pet animals, as it is urgently indicated for the removal of foreign bodies, gastric dilatationvolvulus, and surgical biopsy [43].

Gastrotomy closure techniques are very crucial, which help to avoid post-operative complication e.g., peritonitis. Different techniques may be considered to close gastric incisions including different suture materials, stapling technique and use of adhesive glue [1].

In the current study, skin staplers group showed significantly lower surgical closure time than barbed group and polyglactin 910 group and polyglactin 910 group needed the longer time. In agreement with our results, Abdelkader et al. [42] found that the procedure time was the shortest in dogs with skin staplers. In the same line Ghazy and Gomaa [44] found that gastric closure time with absorbable barbed sutures was significantly faster than with polyglactin 910.

According to our results, in groups barbed and polyglactin 910, no adhesions were observed between the stomach and the omentum, body wall, or other organs. While, in skin stapler group, minimal adhesion was visually observed between the abdominal wall at the incision site and the omentum.

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 [14]. 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 [45].

Supporting our results, Abdelkader et al. [42] showed that the lowest score of adhesions was recorded in dogs of skin staplers group.

This came in line with Ghazy and Gomaa [44] reported that there was no adhesions at all between the stomach and other organs, omentum, or body wall in barbed and polyglactin 910 groups.

Also, Balas et al. [46] demonstrated that polyglactin-910-chitosan gel showed less adhesive than polypropylene control.

There were no significant differences observed in the hematological data on the first day or fifth day between the distinct groups, except for a noticeable increase in TLC at fifth day in polyglactin 910 group. By the 21st day post-operation, both stapler group and barbed group exhibited a mild but nonsignificant reduction in RBC count and HB level, along with a normal leukogram. While polyglactin 910 (Vicryle) group still showed leukocytosis with neutrophilia and monocytosis, which was statistically significant.

In the same context, Collins and Simons [47]showed that complete blood count results indicated a chronic inflammatory process with mild non-regenerative anemia and a shift in the leukocyte percentages with neutrophils comprising 73% of the total white blood cells and lymphocytes only 22%.

Regarding histopathological evaluation, barbed sutures exhibited the most favorable healing response, with complete epithelization of the mucosal lining and diminished vascular and inflammatory changes. In contrast, stapler sutures showed the least favorable outcomes, with incomplete mucosal regeneration and significant tissue reactions in the submucosa. Polyglactin 910 (Vicryle) sutures showed an intermediate response, with clear intestinal regeneration but notable inflammation around the suture material. In the same line demonstrated that Ghazy and Gomaa [44] barbed suture group showed a complete healing process with complete regeneration represented in normal gastric mucosa, normal gastric glands, and normal blood vessels. Polyglactin 910 showed clear inflammatory signs during the healing process represented in dilated blood vessels engorged with blood cells, severe inflammatory cells infiltration, but with normal gastric glands.

However, Abdelkader et al. [42] demonstrated that in the muscularis layer, the remodeling process was obviously seen (score 1) indicating a complete healing process.

In animals sutured with a stapler, the mucosa exhibited peri glandular fibrosis, and prominent observed fibrosis was seen around the staples and extending into the lamina propria. Meanwhile, other surrounding areas displayed separated bundles of connective tissue. Polyglactin910 (Vicryle) material demonstrated a reduction in interstitial fibrosis between gastric glands, primarily restricted to the region beneath the covering epithelium. The vicinity around the polyglactin910 fiber exhibited marked fibrous connective tissue proliferation. Barbed suture led to a remarkable decrease in mucosal fibrosis, displaying a normal pattern, particularly parallel to the glandular structure. The area surrounding the barbed suture displayed intense, regular, and uniformed connective tissue fibers.

Polyglactin 910 is designed to be absorbed by the body through hydrolysis within 56 to 70 days, leaving minimal foreign material behind. This can lead to a reduced inflammatory response and, subsequently, less fibrotic tissue formation [47].

Materials that remain in the body for extended periods can cause a foreign body reaction, leading to fibrosis as part of the healing process. Since Polyglactin 910 is absorbed over a couple of months, it minimizes this reaction and the development of fibrotic tissue [48].

In this study, NF- κ B-p65 was prominently observed within the inflammatory cells surrounding the suture materials, particularly evident in macrophages and lymphocytes around the staples, significantly increase around the polyglactin910 (Vicryle) fiber, and markedly decreased in the barbed suture.

The transcription factor NF- κ B regulates multiple aspects of innate and adaptive immune functions and serves as a pivotal mediator of inflammatory responses. NF- κ B induces the expression of various pro-inflammatory genes, including those encoding cytokines and chemokines, and participates in inflammasome regulation. In addition, NF- κ B plays a critical role in regulating the survival, activation and differentiation of innate immune cells and inflammatory T cells. Consequently, deregulated NF- κ B activation contributes to the pathogenic processes of various inflammatory diseases [49]. NF- κ B signaling during tissue repair is associated with the macrophages that drive the inflammatory phase of healing [50]

In the present study, there was a significant increase in the expression of TGF- β 1 in polyglactin910 group compared to skin stapler group and barbed group and barbed group showed an increased expression compared to skin stapler group.

TGF- β which is a pleiotropic cytokine with potent regulatory and inflammatory activity, represent the higher inflammatory reaction with polyglactin910 [51]. The surface area of Vicryle suture material might be larger than that of skin staples or barbed sutures due to its braided nature. This could lead to more extensive contact with tissues and a different absorption profile, which might contribute to a higher expression of TGF- β 1 during the inflammatory and proliferative phases of healing [52].

Regarding IL6 and MMP2 expressions, polyglactin 910 group showed a significant increase compared to both skin stapler group and barbed group with no significant difference observed between barbed group and skin stapler group. Elevated levels of IL-6 and MMP2 can, however, be a double-edged sword. While necessary for normal wound healing, their overexpression can also lead to chronic inflammation and excessive matrix degradation, causing poor healing outcomes such as ulceration or fibrosis. In chronic wounds, persistent inflammation can lead to sustained high levels of IL-6, and the continuous activation of MMPs can result in excessive tissue breakdown and the inhibition of normal healing processes [53, 54].

The mRNA expression levels of MCP1 and VEGFA expression levels were significantly higher in polyglactin 910 group compared to both skin stapler group and barbed group. Also, the skin stapler group showed higher expression levels than barbed group.

During wound healing progranulin mRNA levels are upregulated in the dermis for at least 10 days following the wound [55].

MCP-1 protein which is encoded by MCP-1 gene is involved in various processes, such as inflammation, wound healing, fibrosis, and formation of vessels [56].

In the current study, the longest stomach emptying time was observed in skin stapler group with no significant difference observed between polyglactin 910 group and barbed group.

Skin staplers cause more tissue trauma and inflammation compared to polyglactin 910 group and barbed group, potentially leading to a more pronounced effect on gastric motility and delayed stomach emptying [57].

This finding is supported by Ghazy and Gomaa [44] who illustrated that time of stomach emptying showed no significant difference between the barbed and polyglactin 910 groups.

Conclusions

In dogs, barbed sutures are the most effective and safe for gastric closure in a brief time, accelerating tissue healing with no postoperative complications.

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Contributions of the authors

The authors' team made a substantial contribution to this study. Every author has thoroughly reviewed

the manuscript and has given their consent to its contents. Every author unanimously approved the final manuscript prior to its submission.

Ethics approval statement

This study follows the ethics guidelines of the Faculty of Veterinary Medicine, Kafr El-Sheikh University, Egypt

Disclosure statement

No potential conflict of interest was reported by the authors.

Data availability statement

The datasets during and/or analysed during the current study available from the corresponding author on reasonable request.

Acknowledgement: Not applicable

TABLE 1. Surgical time (in seconds) of the studied groups

	Group A (Stapler material) (n=10)	Group B (Polyglactin 910) (n=10)	Group C (Barbed material) (n=10)	Р	Post hoc
Surgical time (sec)	45.8 ± 12.89	228 ± 29.17	95.1 ± 6.72	<0.001*	P1<0.001* P2<0.001* P3<0.001*

Data are presented as mean \pm SD. *: Significant as P value ≤ 0.05 . P1:P value between Stapler material and Vicryl material, P2:P value between Stapler material and Barbed material and P3: P value between Vicryl material and Barbed material.

TABLE 2. CBC at day 0, 5 and 21 of the studied groups

	Group A	Group B	Group C (Barbed material)	Р
	(Stapler material)	(polyglactin 910)		
44.1.0	(n=10)	(n=10)	(n=10)	
At day 0	52.1.66	5 2 1 01	5.5.1.95	0.020
RBC (million)	5.2±1.66	5.3±1.81	5.5±1.85	0.920
HB (g/dl)	13.7±4.7	13.8±4.82	14.1±4.83	0.083
HCT (%)	39.7±13.12	40.8±13.59	41.8±14.21	0.944
PLT (mcL)	283.4±98.35	280.3±95.87	279.9±94.9	0.996
WBCs (millions)	7.6±2.55	7.3 ± 2.34	7.3±2.39	0.953
Basophils (%)	0.3 ± 0.09	0.3±0.1	0.4±0.12	0.113
Eosinophils (%)	1.5±0.46	1.2±0.39	1.2±0.38	0.247
Neutrophil (%)	55.8±19.12	56.1±19.27	53.2±18.03	0.933
Lymphocytes (%)	27.6±9.2	27.5±9.35	30.4±9.88	0.734
Macrophages (%)	3.9±1.19	4.1±1.28	4.1±1.38	0.889
At day 5				
RBC (million)	4.5±1.48	4.7 ± 1.6	4.7 ± 1.64	0.934
HB (g/dl)	11.1±3.64	11.8 ± 3.9	12.2 ± 4.14	0.819
HCT (%)	32.3±10.43	33 ± 10.69	36.8 ± 12.58	0.636
PLT (mcL)	200.4±66.9	217.7 ± 74.44	228.1 ± 77.25	0.695
WBCs (millions)	14.8±4.94	17.4 ± 5.87	13.8 ± 4.61	0.294
Basophils (%)	0.5±0.17	0.5 ± 0.16	0.4 ± 0.14	0.297
Eosinophils (%)	1.6±0.54	1.7 ± 0.52	1.6 ± 0.53	0.909
Neutrophil (%)	62.3 ± 21.2*	64.8 ± 22.34*	62.1 ± 21.49*	0.952
Lymphocytes (%)	19.5 ± 6.53	17.2 ± 5.91	21.6 ± 7.36	0.353
Macrophages (%)	5.4 ± 1.73	6.1 ± 1.85	4.6 ± 1.48	0.147
At day 21				
RBC (million)	5±1.7	5.4±1.85	5.4±1.88	0.848
HB (g/dl)	12.5±4.33	13.1±4.43	13.2±4.5	0.920
HCT (%)	37.4±12.86	39.5±13.56	40.1±13.78	0.897
PLT (mcL)	235.7±81.59	246.4±84.61	254.1±86.74	0.887
WBCs (millions)	11.1±3.7	13.1±4.39	9.2±3	0.084
Basophils (%)	0.3±0.09	0.3±0.1	0.4±0.12	0.113
Eosinophils (%)	1.3±0.42	1.4±0.45	1.2±0.41	0.672
Neutrophil (%)	50.4±17.07*	62.3±21.27*	43.7±14.74*	0.080
Lymphocytes	32.7±10.27	17.2±5.58	40.2±12.94	<0.0001*
(%)	<i>P1=</i> 0.005*, <i>P2=</i> 0.231, <i>P3<</i> 0.001*			
Macrophages	6.1±1.93*	9.4±3.07*	5.2±1.57*	0.001*
(%)		=0.008*, P2=0.640, P3<0.00		_

Data are presented as mean \pm SD. RBC: red blood cell, HG: haemoglobin, HCT: haematocrit test, PLT: platelet, WBC: white blood cell.





Fig. 1. Photos showing type of suture used in each of the three groups



Fig. 2. Contrast radiology by barium sulfate in lateral view of the three groups

Group A	Group B	Group C
3) minutes	30 minutes	30 minutes
60 minutes	60 minutes	60 minutes
90 minutes	90 minutes	90 minutes
120 minutes	120 minutes	120 minutes
150 minutes	150 minutes	150 minutes

Fig. 3. Contrast radiology by barium sulfate in dorsal view of the three groups



Fig. 4. Fibrous adhesion (A) following Stapler material). No detected adhesion following either polyglactin 910 (B) or following Barbed material (C)



Fig. 5. The histopathological photomicrographs of the suture, polygalactin 910 and barbed suture groups. Stapler group showing less mucosal regeneration (white arrowhead), submucosal oedema (white arrow). Polyglactin 910 suture group showing marked mucosal regeneration with congestion (white arrowhead) and focal leucocytic infiltration (black arrowhead), intense submucosal inflammation (white arrow) and normal myentric plexus (black arrow). Barbed suture group showed marked mucosal regeneration with mild congestion (white arrowhead), regular collagen around the suture (white arrow) and normal myentric plexus (black arrow), sm means suture material and mp means myentric plexus, H&E stain, Bar= 200 μm



Fig. 6. The photomicrographs of the gastric mucosa of the suture, polyglactin 910 and barbed suture groups from stained sections with Masson's trichrome stain. Stapler group showing intense mucosal fibrosis (white arrowhead) and marked submucosal fibrosis (white arrow). Polyglactin 910 suture group showing marked decrease of mucosal fibrosis (white arrowhead) and marked submucosal fibrosis (white arrow). Barbed suture group showed marked decrease of mucosal fibrosis (white arrowhead), regular collagen around the suture (white arrow), sm means suture material, Masson's trichrome stain, Bar= 200 μm



Fig. 7. The photomicrographs of the immunostained sections of the gastric mucosa of the group A (skin stapler), group B (polyglactin 910) and group C (barbed), (A) Remarkable decrease of NFκB-p65 immunostained was noted in group C (barbed) (white arrowheads indicates the positive cells within the different groups), NFκB-p65 IHC, Bar= 50 µm, (B) Remarkable increase of TGF-β1 immunostained was noted in group B (polyglactin 910) (white arrowheads indicates the positive expression within the different groups), TGF-β1 IHC, Bar= 50 µm



Fig. 8. The mRNA expression levels of IL6, MMP2, MCP1 and VEGFA within the sutured gastric mucosa of the group A (skin stapler), group B(polyglactin 910) and group C (barbed)

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تحليل مقارن لتقنيات مختلفة لإغلاق فتحة المعدة في نماذج الكلاب باستخدام الخيوط الشانكة، ودباسة الجلد، وخيوط بولي جلاكتين 910

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الملخص

يتطلب استئصال المعدة، و هو جراحة بيطرية شائعة، إغلاقًا فعالًا لضمان سرعة الشفاء والحد الأدنى من المشاكل. تُعدَ الدباسات المسننة والجلدية بدائل لخيوط بولي جلاكتين 910، المستخدمة على نطاق واسع. قورنت دباسات الجلد، وخيوط بولي جلاكتين 910 القياسية المخيطة يدويًا، والخيوط المسننة ذاتية التثبيت بدون عقد، المستخدمة في استئصال معدة الكلاب.

خضع ثلاثون كلبًا بالغًا سليمًا (المونغريل) لإجراءات فتح معدة للمقارنة التجريبية. وُزَ عت هذه العمليات إلى مجموعات أ، ب، وج. أغلق فتح المعدة بدباسة جلد من نوع ميريل ميروس، مقاس 35 وات. استُخدم بولي جلاكتين 910 (فيكريل، إيثيكون، 2/0) في المجموعة ب، بينما استُخدمت خيوط إيثيكون المسننة القابلة للامتصاص في المجموعة ج. قللت الخيوط المسننة الالتهاب وعززت تجديد الأنسجة، وفقًا لدر اسات جزيئية أظهرت إنتاجًا أقوى لعوامل تكوين الأوعية الدموية وقلة السيتوكينات المؤيدة للالتهابات. تسببت خيوط بولي جلاكتين 910 في التهاب ومشاكل في تجديد الأفرعية تسبب دباسات الجلد التهابًا في الجروح أكثر من الخيوط المشوكة القابلة للامتصاص، ولكن أقل من بولي جلاكتين 910 سرّعت الخيوط المشوكة القابلة للامتصاص إغلاق المعدة بشكل ملحوظ من 910 في التهاب ومشاكل في تجديد الظهارة. تسبب دباسات الجلد التهابًا في الجروح أكثر من الخيوط المشوكة القابلة للامتصاص، ولكن أقل من بولي جلاكتين 910 سرّعت الخيوط المشوكة القابلة للامتصاص إغلاق المعدة بشكل ملحوظ. خضعت جميع المجموعات الألاش لإغلاق آمن الشق المعدة، لم تُسجل أي مضاعفات أو وفيات كارثية بعد الجراحة. في الختام، تُعد الخيوط المشوكة أسرح عن المراحق أمانًا لإغلاق المعدة، حيث تُسرّع تجديد الأنسجة، وفيًا لعراصة على المعرف منه المراحق في من يولي المراحق أكثر الطرق

الكلمات الدالة: بَصْع المعدة؛ خيوط مشوكة؛ خيوط بولى جلاكتين 910؛ دباسات الجلد؛ كلب.