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# EVALUATION OF HEALING OF EXPERIMENTALLY-INDUCED VERTICAL EYELID WOUNDS AFTER SINGLE OR DOUBLE LAYER CLOSURE IN DOGS

**Short Title:** Healing Of Experimentally-Induced Vertical Eyelid Wounds

RASHA EZZ-ELDEEN ABDELKADER [1]; NASHWA HAMAD [2]; MOHAMED SEMIEKA [3] AND AHMED F. AHMED [4*]

[1] Cardiology Hospital, Faculty of Medicine, Assiut University, Assiut 71526, Egypt. ashoushezz@gmail.com

[2] Lecturer of Pathology and Clinical Pathology, Faculty of Veterinary Medicine, Assiut University, Assiut 71526, Egypt. nashwahamad@aun.edu.eg

[3] Professor Emeritus of Surgery, Anesthesiology and Radiology, Faculty of Veterinary Medicine, Assiut University, Assiut 71526, Egypt. mohamedms@aun.edu.eg

[4] Professor and Department Head of Surgery, Anesthesiology and Radiology, Faculty of Veterinary Medicine, Assiut University, Assiut 71526, Egypt, **Fax:** 002-088-2080501

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## ABSTRACT

This study aimed to evaluate the healing of experimentally-induced vertical eyelid wounds after the application of one or double-row sutures for closure. Twelve adult healthy mongrel dogs were generally anesthetized then a vertical eyelid incision was inflicted. Dogs were then randomly divided into two groups (*n*=6 each). Wounds in group A were sutured in a single layer of simple continuous sutures using Silk #3-0. However, wounds in group B were sutured in a double layer of simple continuous sutures. The first layer using Polyglycolic acid #3-0 was applied in the conjunctiva and tarsal plate. The second layer was applied to the skin and muscle layer using 3/0 silk sutures. Wound healing was evaluated by monitoring ophthalmic signs, gross and histopathological examinations. Results revealed the presence of ophthalmic signs with varying scores that were not significant between groups. Grossly, eyelid wound healing was superior in wounds sutured in a double layer, although, mean values of gross evaluation scores were not significantly different between groups. Histopathology revealed incomplete healing of wounds with the presence of gaps in skin and conjunctival aspects of wounds closed in a single layer. In contrast, in wounds closed in a double layer, the epithelial gap was completely closed with normal epithelium at the skin aspect of the eyelid and abundant well-organized collagen fibers with minimal inflammatory infiltration were present at the wound site. In conclusion, using a double layer of simple continuous sutures is superior in healing of vertical eyelid wounds in dogs. More time for healing is necessary for eyelid wounds closed in a single layer of simple continuous sutures.

***Keywords:*** canine, eyelid, wound, closure, healing

*Corresponding author:* Ahmed F. Ahmed
*E-mail address:* Afahmed70@aun.edu.eg; afahmed1970@gmail.com
*Present address:* Professor and Department Head of Surgery, Anesthesiology and Radiology, Faculty of Veterinary Medicine, Assiut University, Assiut 71526, Egypt.

## INTRODUCTION

Four main structures constitute the eyelids; a fibrous tarsal plate (which gives some structural rigidity to the eyelid), muscles responsible for blinking, skin on the outer surface (which is very thin, mobile, and pliable compared with skin elsewhere), and palpebral conjunctiva on the inner surface (Stades and van der Woerdt, 2021). The eyelids have numerous functions that are absolutely vital for pain-free normal vision (Maggs, 2013). The eyelids have two main layers; an inner layer that includes the conjunctiva and underlying connective tissue and an outer layer that is composed of haired skin and underlying orbicularis muscle. Meibomian glands are lipid-secreting exocrine glands present on the eyelid margins.

Eyelid injuries most commonly occur as a result of trauma such as bite wounds or road traffic accidents. Reconstructive surgery should be performed as soon as the patient is stable because full-thickness lacerations disrupt normal eyelid anatomy and function (Manning, 2014, Esson, 2015). Following thorough saline irrigation, the wounds and conjunctival sac were cleaned with diluted povidone–iodine solution not scrubbed. The eyelid skin has an excellent blood supply, which usually ensures the healing of even contaminated wounds (Manning, 2014).

For meticulous evaluation and treatment, sedation or anesthesia may be required especially in fractious animals. Proper wound management by gentle debridement, removal of foreign material, accumulated blood and secretions, and careful cleaning using normal saline (Esson, 2015).

Debridement of devitalized tissues should be minimized to preserve the normal integrity of the eyelid margin and the subcutaneous blood vessels. Recent wounds can usually be debrided and repaired immediately while older wounds may be delayed in the repair to treat blepharitis and/or infection before debridement and closure. However, prompt surgical repair in two layers is required in full-thickness eyelid lacerations and left to heal by second intention in order to ensure accurate apposition and long-term functionality (Aquino, 2008).

A two-layer closure of the full-thickness eyelid laceration is adopted. The first deep layer of continuous absorbable suture material is used to close the conjunctival and connective tissue layers. A second layer of Interrupted non-absorbable sutures is used to close the outer skin and muscular layers. Accurate apposition of the eyelid margin with obliteration or drainage of the dead space is critical in order to prevent subsequent keratopathy. Lacerations at the medial *canthus* with incorporation of the nasolacrimal duct and/or *canaliculi*, these structures are identified and may be stented. Routine systemic antimicrobial, anti-inflammatory, and analgesic care is required as postoperative care (Aquino, 2008).

According to available literature, there are no detailed study/studies to evaluate the effect of suture patterns either single or double layer on the healing of eyelid wounds in dogs. Consequently, the present study aimed to evaluate the healing of experimentally-induced vertical eyelid wounds after the application of one or double-row suture patterns for closure. The wound healing is evaluated grossly, monitoring of ophthalmic signs and histopathologically.

## MATERIALS AND METHODS

### I. Ethical Approval

This study was approved by the Research Ethics Committee (REC) of the Faculty of Veterinary Medicine-Assiut University No. 06/2023/0112 in accordance with the Egyptian by laws and OIE animal welfare standards for animal care and use in research and education.

The experiment was carried out at the Department of Surgery, Anesthesiology and

Radiology, Veterinary Teaching Hospital, Faculty of Veterinary Medicine, Assiut University, Assiut, Egypt.

**II. Experimental Animals**

Twelve adult healthy mongrel dogs of both sexes (8 males and 4 non-pregnant, non-lactating females) were selected and housed in separate standard cages. The average body weight of the dogs was 13.97 kg (range 11.5-18 kg). The dogs were provided daily with adequate amounts of feed and water *ad libitum*, weighed and kept for two weeks to adapt. The dogs were dewormed by subcutaneous injection of 1 ml/50 kg Ivermectin and randomly allocated into two treatment groups (A and B) each with 6 animals as shown below.

**III. Animal Preparation for Surgery:**

On the day of surgery, each dog was weighed to calculate the dose of anesthetic drugs and examined clinically to record the physiological parameters including rectal temperature (RT), heart rate (HR), and respiratory rate (RR). Dogs were fasted for 12 hours and operated under general anesthesia by intramuscular injection of 2 mg/kg Xylazine Hcl 2% (Xyla-Ject, ADWIA Co., SAE, Egypt) and 10min later by intravenous injection of 10 mg/kg Ketamine Hcl 5% (Ketamine, Sigma-Tec Pharmaceutical Industries, SAE, Egypt). With the animal on lateral recumbency, the upper eyelid area was prepared aseptically using the appropriate aseptic technique by clipping and shaving of hair and application of Povidone-iodine solution 10% (Betadine, El-Nile Co. for Pharmaceutical and Chemical Industries, Cairo, Egypt) and draped except for the surgical site.

**IV. Surgical Techniques (Figs. 1 and 2)**

Dogs were randomly divided into 2 groups according to the suture pattern either single or double layer that was used for wound closure.

**Group A, *n*=6:**

A 1.5-cm vertical wound was inflected by a scalpel blade at the upper eyelid midway between the medial and lateral canthi and included the eyelid margin. The wound was then closed in a single layer of simple continuous sutures using 3/0 silk sutures (Silk # 3-0, M-NATUR®, International Sutures Manufacturing Co., Egypt).

**Group B, *n*=6:**

A 1.5-cm vertical wound was inflected by a scalpel blade at the upper eyelid midway between the medial and lateral *canthi* and included the eyelid margin. The wound was then closed in double layers of simple continuous sutures. The first layer using 3/0 Polyglycolic Acid (Vicryl # 3-0, M-NATUR®, International Sutures Manufacturing Co., Egypt) was applied to the conjunctiva and tarsal plate. The second layer was applied to the skin and muscle layer using 3/0 silk sutures.

**V. Post-operative Care:**

Animals were given non-steroidal anti-inflammatory drugs for 3 days. An initial dose of 0.2 mg/kg, IV, q 24 h (Meloxicam, Amriya for Pharmaceutical Industries, Egypt) was given on the day of surgery followed by an oral dose of 0.1 mg/kg q 24 h (Melocam, Amoun Pharmaceutical, Egypt) for two more days. Topical antibiotic (tobramycin) eye ointment (Tobrin, Epico, Tenth of Ramadan City, Egypt) was applied q12h for three days, postoperatively. Skin stitches were removed 10 days after surgery. Animals were monitored after surgery and during the recovery period.

Eyes were examined daily for 14 days postoperatively for the presence of any swellings, exudates from the operation site, patency of skin stitches, or any other complications.

**VI. Evaluation Criteria Include:**

1. **Ophthalmic symptoms:** Ophthalmic symptoms and their severity were monitored after surgery for the presence of eyelid edema and swelling, epiphora, photophobia, ocular

discharge, keratitis and/or conjunctivitis. A score system (from 1 to 4) was developed to numerically express the severity of these symptoms. Score 1= absence of the symptom. Score 2= mild symptom. Score 3= moderate symptom. Score 4= severe symptom.

**2. Gross evaluation of the wounds**: The eyelid wounds were evaluated at the end of the study by examining the wound alignment, cosmetic appearance of the eyelid and scar tissue formation. A developed score system from 1 to 5 was used to numerically describe the gross outcome of different techniques. Score 1= the eyelid is properly apposed without mal-alignment with minimum scar tissue formation at the wound site and considered cosmetically acceptable. Score 2= slight mal-alignment and distortion of the eyelid at the wound site with slight scar tissue formation but still cosmetically acceptable. Score 3= Noticeable distortion of the eyelid at the wound site with scar tissue formation and the wound is cosmetically unacceptable. Score 4= wound infection with purulent exudation and partial wound dehiscence. Score 5= Complete wound breakdown and dehiscence.

3. **Histopathology and Histochemistry:**
At days 7 and 15 post-wounding, 3 animals from groups A (wounds closed in one layer) and B (wounds closed in two layers) at each time point were randomly selected and euthanized by intravenous administration of sodium pentobarbital at a dose of 100 mg/kg (Fatal-Plus® Vortech Pharmaceuticals, Dearborn, Michigan, USA). For histopathological evaluation, full-thickness eyelid biopsies from wounds, including the surrounding skin of the incision sites, were taken from both groups at each time point. The collected tissue specimens were fixed for 72 hours in a 10% neutral buffered formalin solution at room temperature, dehydrated through an ascending graded sequence of ethanol, cleared in methyl benzoate, and then embedded in paraffin wax (Sigma Aldrich, USA). Tissue slices were cut at 5 µm thickness using a microtome (Richert Leica RM 2125, Germany), mounted on glass slides, and stained with hematoxylin and eosin (H&E) for blinded histopathological evaluation of wounds (Bancroft and Gamble, 2008). Afterward, all stained slides were examined by a light microscope (Olympus CX31, Japan), and images were taken by a digital camera (Olympus Camedia C-5060, Japan). A histopathological scoring system was employed in a blinded manner to assess the extent of re-epithelialization, the presence of inflammatory cells, newly formed blood vessels, and fibroblasts according to a semiquantitative scoring system: 0 (absent), 1 (minimal), 2 (mild), 3 (moderate), and 4 (marked) (Farghali *et al.*, 2017). The scoring was accomplished in three sections of three different animals from each group at each time point. For histochemical staining, another set of formalin-fixed paraffin sections was stained with Gomori's trichrome stain for assessment of total collagen content (Eissa *et al.*, 2023). The staining process was conducted following the protocol described by the manufacturer. Next, the stained slides were inspected using a light microscope to detect collagen staining with a green color. The collagen content in trichrome-stained slides was assessed based on a scale of 0 - 3. A score of 0 indicated the absence of collagen fibers. A score of 3 indicated plentiful total collagen. The organized collagen fibers appeared parallel and wavy with a consistent green color, whereas the less organized fibers had variations in color and infiltration in parallel fibers or breaks (Farghali *et al.*, 2017).

**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

Dogs tolerated the surgical operation and did not exhibit any adverse effects. They returned to normal feed intake gradually after the operation and continued throughout the experiment.

**1. Ophthalmic symptoms:**

Ophthalmic symptoms were recorded in dogs of the two groups (Figure 2). Eyelid swelling, epiphora and photophobia were observed starting from day 2 to 4 of surgery. Ocular discharge and conjunctivitis were recorded starting day 3 to day 7 of surgery. However, dogs did not show signs of keratitis. Although dogs showed variations in the severity of these ophthalmic symptoms, there were no significant differences ($P$>0.05) between the mean score values of the two groups (Table 1).

**2. Gross evaluation of the wounds:**

There was no significance ($P$>0.05)between the mean gross evaluation score of the healed eyelid wounds among different groups. However, the mean gross score value of dogs with vertical eyelid wounds that closed in double layers was superior (Table 1). Wound infection and partial dehiscence (score 4) were recorded in one dog (1/6) with vertical eyelid wounds sutured in one layer and one dog (1/6) with vertical eyelid wounds sutured with double layers (Figure 3). Moreover, complete wound breakdown and dehiscence (score 5) were recorded in two dogs (2/6) with vertical eyelid wounds sutured with double layers (Figure 3).

**3. Histopathology:**

On day 7 after suturing, examination of wounds closed in single and double layers revealed that the healing process in both groups was in the proliferation phase, with slight variations in the degree of healing progression. In wounds closed in a single layer, the epithelium at the wound margins began to proliferate, leaving a clearly visible wound gap at the outer (skin) aspect of the eyelid. Epithelial proliferation was not observed at the inner (palpebral conjunctiva) aspect, which revealed a large gap between the wound edges containing necrotic debris. A moderate amount of granulation tissue was noticed along the incision site from the outer to the inner aspects of the eyelid, which partially occluded the wound area. There was diffuse hemorrhage and extensive infiltration of inflammatory cells, particularly neutrophils and macrophages, within loose connective tissue. Also, few fibroblasts and numerous hyperemic new blood vessels were seen. A marked inflammatory reaction composed of necrotic debris, neutrophils and mononuclear cells was seen around the suture tract. On the other hand, the wounds closed in a double layer displayed prominent epidermal regeneration at the skin aspect of the eyelid. However, at the conjunctiva aspect, a wound base consisted of granulation tissue with marked inflammatory cells and hyperemic blood vessels were covered by slightly proliferated epithelium at the wound edges. In addition, the wound area along the incision site was entirely occluded by a large amount of granulation tissue, composed of copious inflammatory cells, a moderate number of new blood vessels, and fibroblasts. Diffuse hemorrhage at the wound site was also observed. The suture material was surrounded by collagen fibers with a moderate inflammatory reaction (Figure 4).

On day 14 after suturing, in wounds closed in one layer, marked re-epithelialization covered almost the entire surface of the wound area. However, an epithelial gap was still observed at the conjunctival surface. The wound site was occupied by granulation tissue with abundant inflammatory cells and neovascularization. The suture tract was surrounded by collagen fibers with moderate inflammatory infiltrates. In contrast, in wounds closed in a double layer, the epithelial gap was completely closed with normal epithelium at the skin aspect of the eyelid. One out of three animals examined in this group showed incomplete closure of the epithelial gap at the conjunctival aspect. At the wound site, abundant collagen fibers, which were more organized with minimal

inflammatory infiltration, were seen. The suture material was surrounded by collagen fiber with minimal inflammatory reaction and abundant fibroblasts (Figure 5).

Semiquantitative scoring of the histopathological findings revealed that there was a reduction with non-significant differences in the number of new blood vessels in wounds closed in a double layer at days 7 and 14 after suturing compared to wounds closed in a single layer. On day 7, the number of inflammatory cells in wounds closed in a double layer decreased compared to those closed in a single layer. This decrease in the number of inflammatory cells became statistically significant at day 14 ($p < 0.05$). The re-epithelialization process was more obvious at both the skin and conjunctival aspects in wounds closed in a double layer than those closed in a single layer (Figure 6).

**Histochemical evaluation**

Histochemical evaluation of collagen deposition in wounds was accomplished using trichrome stain. The collagen fibers in wounds closed in single and double layers were loose and disorganized on day 7 after suturing. However, its amount in wounds closed in a double layer was significantly higher ($p < 0.05$). At day 14, the amount of tightly packed, more organized, and intensively stained collagen fibers was significantly higher ($p < 0.05$) in double-layered wounds than in single-layered ones (Figure 7).

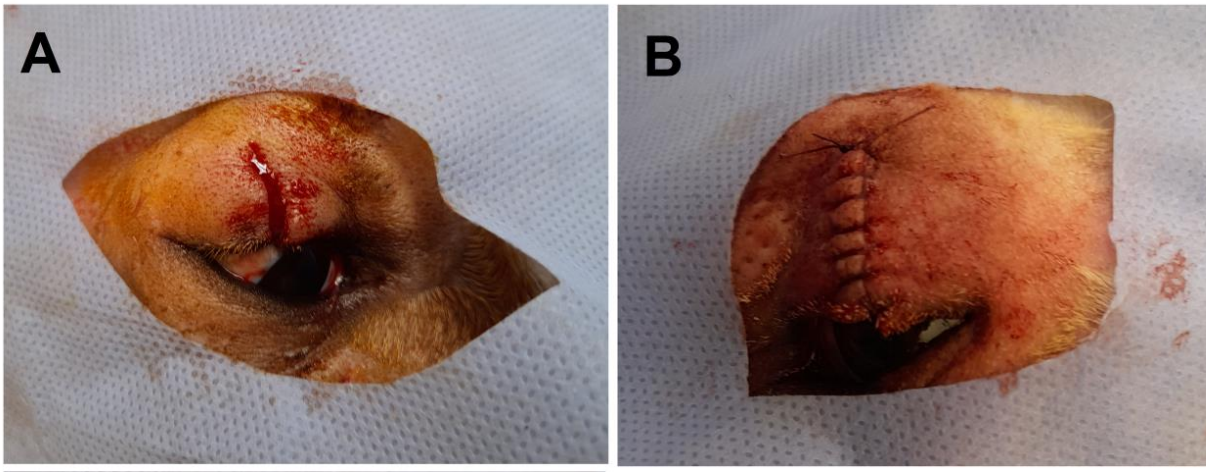


**Figure 1:** Induction of a vertical eyelid incision (A), closing the wound in a double layer of simple continuous sutures (B).

**Figure 2:** Ophthalmic symptoms recorded in a dog in the form of moderate mucopurulent discharge in the eye of a dog with a vertical eyelid wound sutured in one layer.

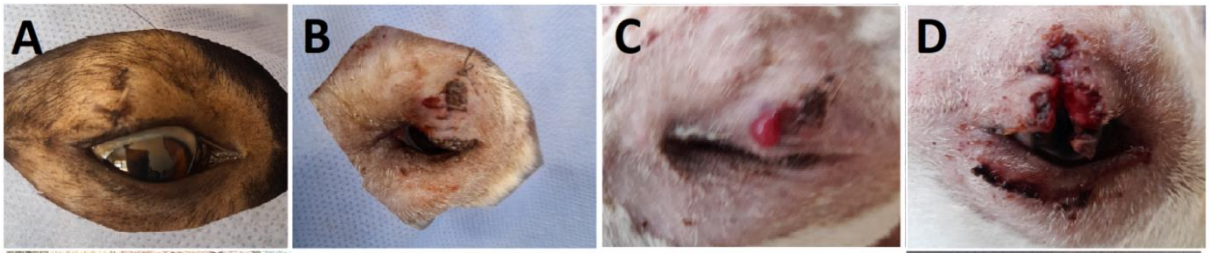


**Figure 3:** Gross evaluation of the healed eyelid wounds in dogs of different groups. Score 1 in a dog with a vertical eyelid wound sutured in one layer (A). Score 2 in a dog with a vertical eyelid wound sutured in two layers (B). Score 4 was recorded in a dog with a vertical eyelid wound sutured in one layer (C). Score 5 was recorded in a dog with a vertical eyelid wound sutured in one layer (D).

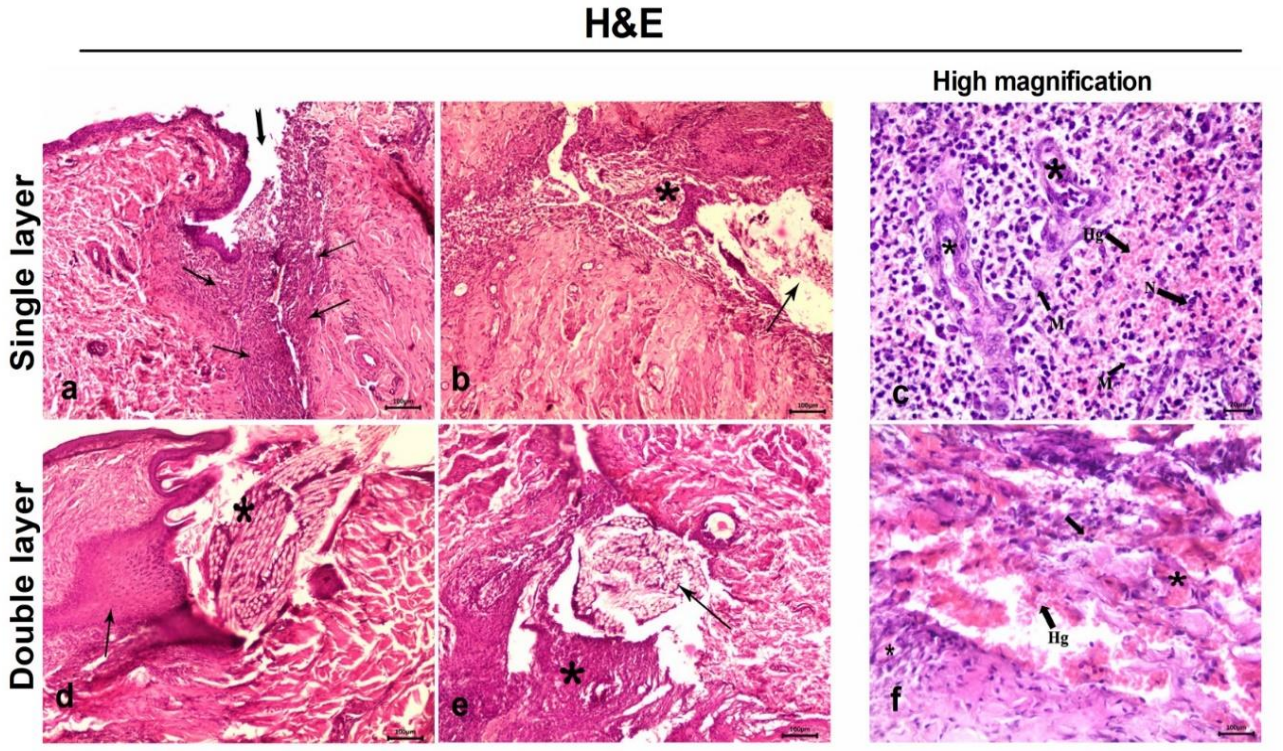


**Figure 4:** Histopathological evaluation of eyelid wounds closed in single and double layers at day 7 after suturing. (a) depicting granulation tissue partially closed the incision site (arrow), and a large epithelial gap (notched arrow). (b) depicting abundant inflammatory cell infiltration (asterisk) around the suture tract (arrow). (c) depicting neovascularization (asterisk), diffuse hemorrhage (Hg), neutrophils (N), and macrophages (M). (d) depicting markedly proliferated epithelium at the wound edge (arrow) and suture tract opens to the skin surface (asterisk). (e) depicting suture material (arrow) surrounded by collagen fibers with moderate inflammatory reaction (asterisk). (f) depicting neovascularization (asterisk), diffuse hemorrhage (Hg), and mononuclear inflammatory cellular infiltration (arrow).

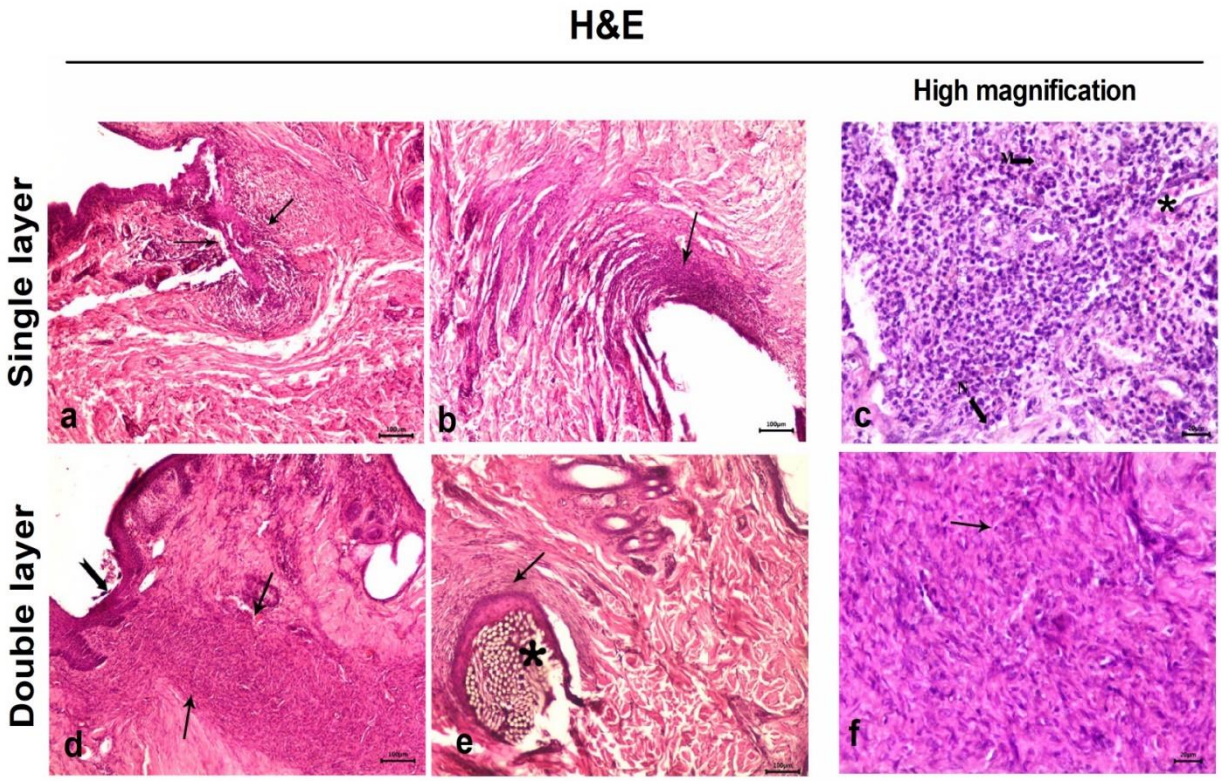


**Figure 5:** Histopathological evaluation of eyelid wounds closed in single and double layers at day 14 after suturing. (a) depicting granulation tissue occupying the wound site (arrow) which is markedly covered by regenerated epithelium. (b) depicting collagen fibers moderately infiltrated with inflammatory cells around the suture tract (arrow). (c) depicting neovascularization (asterisk), neutrophils (N), and macrophages (M). (d) depicting regenerated normal epithelium (notched arrow) completely covering the wound area (arrow). (e) depicting suture material (asterisk) surrounded by abundant collagen fibers with minimal inflammatory reaction (arrow). (f) depicting abundant collagen fibers and minimal inflammatory cell reaction (arrow).

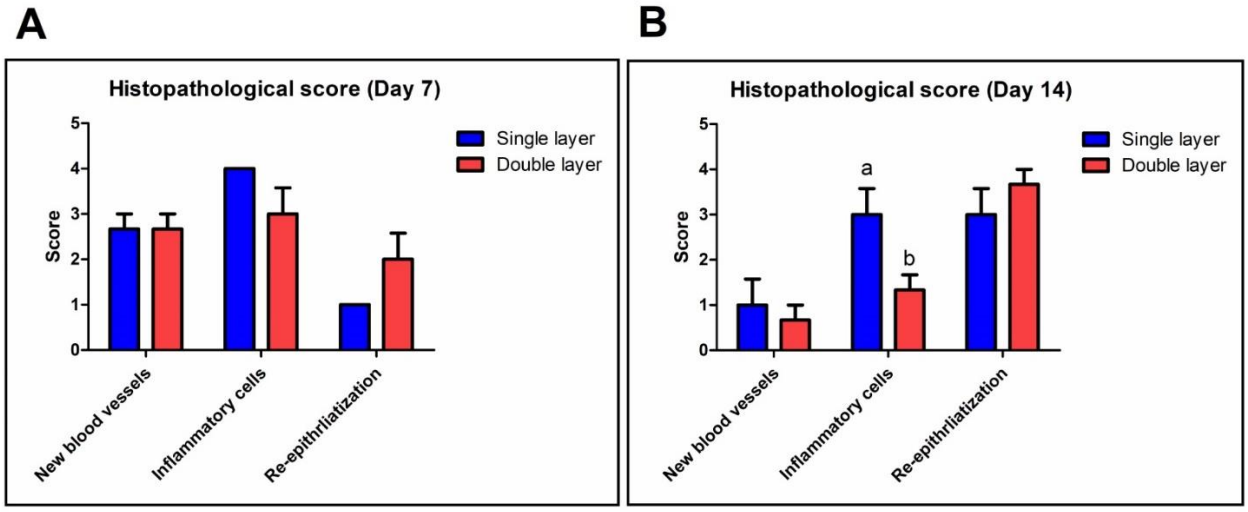


**Figure 6:** Histopathological scoring of new blood vessels, inflammatory cells and re-epithelialization on days 7 (A) and 14 (B) after suturing. The values are expressed as

mean± SEM (n=3). Different superscripts (a, b) above the bars indicate significant differences ($P < 0.05$).

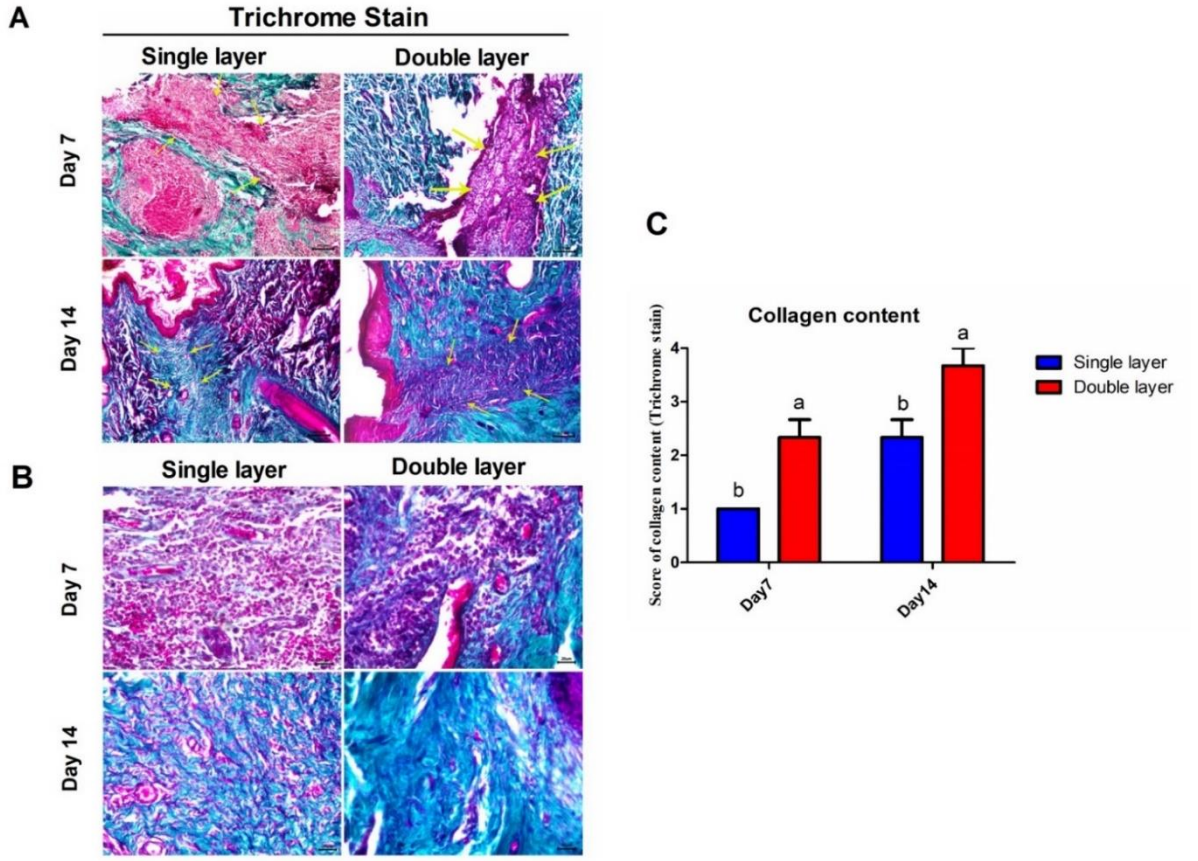


**Figure 7:** Histochemical evaluation of eyelid wounds closed in single and double layers. (A) and (B) Trichrome stained sections on days 7 and 14 after suturing displaying the blue-colored collagen content at the wound area (between yellow arrows). Scale bars in (A) =100µm and (B) = 20µm. (C) Semiquantitative scoring of the collagen content at the wound area. Results are expressed as mean ± SEM (*n*=3). Different superscripts (a, b) above the bars indicate significant differences ($P < 0.05$).

## DISCUSSION

Because eyelid injuries are frequent in dogs, this study was designed to compare the healing outcomes of suturing the eyelid with one or two layers of a simple continuous suture pattern. Ophthalmic signs were recorded in dogs of both groups with nonsignificant variations between the two groups. Although there is no significance between the mean gross healing score of the two groups, healing of the eyelid wound sutured by a double layer was superior. The superiority was confirmed by histopathology.

Although ophthalmic symptoms were currently recorded in dogs, they were insignificantly different between groups. Ophthalmic signs such as blepharedema, blepharospasm, chemosis, and conjunctival hyperemia are usually associated with eyelid trauma and blepheritis (Martins *et al.*, 2016). Moreover, when an eyelid injury is noted, a thorough search must be made for concurrent injuries to the cornea, sclera, and nasolacrimal apparatus in particular, and to the globe as a whole (Maggs, 2013). Currently, keratitis was not observed in the eyes of all dogs. Irritation of the corneal surface as a result of contact with the suture materials was not evident in this study. It may be due to the minimal amount of suture materials at the conjunctival aspect of the wounds as well as the presence of the suture knots at the cutaneous aspect of the eyelids. Keratitis is avoided by placing knots external to the free rim of the lid margin to avoid contact with the cornea (Martin, 2005).

Moreover, in double-layer closure, knots should be avoided or placed beneath the conjunctiva. The skin, together with the *orbicularis oculi* muscle, is closed using nonabsorbable monofilament sutures. Absorbable material may be used in aggressive or anesthesia-risk patients (Gelatt and Gelatt, 2011).

It has been cited that eyelids have an excellent blood supply, and injuries to them heal rapidly when repaired correctly (Maggs, 2013). Gross evaluation revealed that healing of the eyelid wounds of the current study was relatively rapid within the first week of the experiment. However, histopathological examination in the first week revealed that the wound was in the proliferation phase with slight variations in the degree of healing progression and clearly visible wound gap at the outer (skin) aspect and a large gap between the wound edges contained necrotic debris at the conjunctival aspect.

On the other hand, eyelid swelling was currently evident after surgery. This extensive eyelid edema and distortion was attributed to the rich blood supply of the eyelids and usually occurs after even relatively minor injury or rough and excessive tissue handling during surgery (Peña and Garcia, 1999; Lewin, 2003; Maggs, 2013).

The gross outcome of the eyelid wounds including the cosmetic appearance was variable among dogs of both groups. However, the mean score of the gross appearance of the eyelid wound closed in double layer (Group B) was superior. Closing the eyelid wound in a single layer (group A), although improperly closing the wound, has been cited to have less operation time and consuming a small amount of anesthesia and suture material (Martin, 2005; van der Woerdt, 2004). Closing the eyelid wound in a double layer in the present study consumed more time and suture material. However, the outcome, although insignificant, was better than the wounds sutured in a single layer. It has been cited that standard two layers of sutures with soft, braided sutures are used in sterile eyelid wounds (Martin, 2005, Maggs, 2013). Polyglycolic acid (Vicryl) and silk were used to close the eyelid wound of the present study and are considered soft and braided suture materials. Cannulation of the lacrimal puncta was not indicated in the current study because the inflected incision was vertical and positioned away from the course of the puncta. In another study including wounds involving the medial canthal region, the nasolacrimal puncta should be identified and cannulated, and the nasolacrimal apparatus flushed to ensure patency (Stades and van der Woerdt, 2021).

Marked re-epithelialization covered the almost entire surface of the wound with the presence of epithelial gaps at the conjunctival surface and granulation tissue with abundant inflammatory cells and neovascularization was observed histologically in a one-layer closed wound. In contrast, in wounds closed in a double layer, the epithelial gap was completely closed with normal epithelium at the skin aspect of the eyelid and abundant well-organized collagen fibers with minimal inflammatory infiltration were present at the wound site. This histologic finding may indicate that eyelid wounds sutured in one layer necessitated more time for healing than those sutured in a double layer. This finding was also proved by the semiquantitative scoring of the histochemical findings.

Wound infection was recorded in one dog of each group in the present study. Bacterial flora in the conjunctival sac and surrounding area have been reported to readily invade this area, but severe postoperative septic blepharitis is uncommon if a course of systemic antibiotics with a good spectrum of activity against Gram-positive organisms is instituted (Maggs, 2013). Antibiotic eye ointment was currently used in addition to the extensive blood supply of the eyelid had limited secondary infection.

Unexpectedly, wound dehiscence was currently recorded in two dogs with their eyelid wounds closed in double layers.

Severe pruritus and self-trauma have been cited to occur during wound healing, especially if inflammation is present as a result of infection, tissue trauma, or tight sutures (Aquino, 2008). Control of these primary factors, provision of an Elizabethan collar and postoperative wound hygiene as well as the use of warm packs and analgesics are essential to minimize this problem (Stades and van der Woerdt, 2021).

## CONCLUSION:

Using a double layer of simple continuous sutures is superior in the healing of vertical eyelid wounds in dogs. More time for wound healing is necessary for eyelid wounds closed in a single layer of simple continuous sutures.

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# تقييم إلتئام جروح جفن العين الرأسية المستحدثة تجريبيا بعد خياطتها بطبقة أو طبقتين في الكلاب

***رشا عز الدين عبد القادر ، نشوى حماد ، محمد سميكة ، أحمد فتحي***

E-mail: afahmed1970@gmail.com Assiut University website: www.aun.edu.eg

هدفت الدراسة الى تقييم إلتئام الجروح الرأسية في جفن العين والمستحدثة تجريبيا بعد خياطتها بطبقة أو طبقتين في الكلاب، تم استخدام ١٢ كلب بتخديرها تخديرا عاما وإحداث جرح رأسي في الجفن العلوى للعين، وتم تقسيم الكلاب عشوائيا إلى مجموعتين (أ و ب)، تم خياطة الجرح فى كلاب المجموعة (أ) بطبقة واحدة من الغرز المستمرة البسيطة بخيط الحرير مقاس (٣/صفر)، بينما تم خياطة الجرح فى كلاب المجموعة الثانية بطبقتين من الغرز المستمرة البسيطة، الأولى باستخدام خيط الفيكريل مقاس (٣/صفر) لخياطة الملتحمة والصفيحة الترسية، والثانية باستخدام خيط الحرير مقاس (٣/صفر) لخياطة الجلد والعضلات، وتم تقييم التئام الجروح بمراقبة ظهور الأعراض الإلتهابية للعين، التقييم العياني للجروح، التقييم الهستوباثولوجي والهستوكيميائي. أظهرت النتائج تفوق غير معنوى لمعدل التئام الجروح المخيطة بطبقتين عن تلك المخيطة بطبقة واحدة عند تقييمها عيانيا، وأظهر الفحص الهستوباثولوجي التئام غير كامل مع وجود فراغات عند السطح الجلدي والملتحمي للجفن للجروح المخيطة بطبقة واحدة، بينما التأمت الجروح المخيطة بطبقتين إلتأما كاملا مع اختفاء الفراغات مع تكون نسيج طلائي طبيعي وتكون ألياف كولاجين منتظمة مع وجود ارتشاح ضئيل للخلايا الالتهابية. خلصت الدراسة الى تفوق الخياطة بطبقتين على الخياطة بطبقة واحدة لجروح جفن العين الرأسية فى الكلاب، مع احتياج الجروح المخيطة بطبقة واحدة الى وقت أطول لتحقيق إلتئاما كاملا.

**كلمات مفتاحية:** الفصيلة الكلبية ، جفن العين ، جروح ، خياطة ، إلتئام