



## One-Stage Technique for Rumenal Fistulation in Rams

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### Abstract

Rumenal fistula is very essential in nutrition and physiology experimental research studies in sheep. Permanent placement of the cannula without complication is a challenge. This experimental study aimed to provide a detailed description of modified one-stage technique for rumenal fistula in sheep to simplify the surgery, decrease the post-operative complications, and lengthen the functional life of the cannula. As well as reporting the postoperative complications related to the surgery as well as short and long term monitoring. 10 healthy rams were surgically prepared for left side laparotomy in lateral recumbent position. The rumen was grasped, anchored to the subcutaneous tissue, and incised to fit the custom-made cannula. The rumenal wall was sutured directly to the skin after insertion of the cannula, and a metal retainer flange was pushed to secure the cannula in place. No early post-surgical complications. None fistula got a bigger size, and none cannula was rejected, or had to be repositioned. The one-stage technique used in this study to fix a custom-made cannula in its position recommended to experimental research on sheep nutrition and physiology.

### Key words:

Sheep, rumenal fistula, one-stage technique

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## 1. INTRODUCTION

Rumenal fistulation (RF) is a permanent surgically created fistula between the dorsal rumenal sac and the body wall in the left paralumbar fossa with an intra-rumenal cannula fitted in the fistula (Azizi et al., 2007). RF has been performed for experimental purposes (studies on rumenal digestion), and for therapeutic purposes (temporary symptomatic relief of chronic rumenal tympany) (Turner et al., 1989 and Azizi et al., 2007). RF is preferred than stomach tube sampling, especially when repeated sampling is required, as it prevents the contamination of rumenal sample with saliva and gives a typical sample of rumenal fluid (Shen and Song, 2012).

There are three techniques to perform the RF, a one-stage method, a two-stage method, and Schalk and Amadon technique (Hecker, 1969; Dougherty, 1981; Lumley et al., 1990; Muzzi et al., 2009; Stedile et al., 2008; and Azizi et al., 2007). *The first is one-stage technique*, the rumen is exteriorized through a circular (Haskell, 2002; Laflin and Gnad, 2008) or vertical (Ducharme, 1990; Noordsy and Ames, 2006) skin incision in the left paralumbar fossa, incised, and

sutured to the skin. The cannula inserted into the rumenal opening at the same day of surgery. *The second is two-stage technique*, the rumen is exteriorized through a circular (Dougherty, 1981) or vertical (Azizi, et al., 2007) skin incision in the left paralumbar fossa. The rumenal wall (without opening the rumen) can be sutured to the skin along the edge of a laparotomy incision to form an adhesion between the rumen and the skin (Dougherty, 1981; Azizi et al., 2007) or a metal clamp is applied to a fold of the exteriorized rumenal wall through a laparotomy incision and then allowing a fistula to develop by gradual necrosis and sloughing of the fold and adhesion of the rumen to skin (Hecker, 1974). The cannula inserted 7-12 days after surgery to allow adhesion formation between the rumenal wall and the skin. *The third technique* was first described by Schalk (1928) for cattle and modified for sheep by Hecker (1969), a fold of rumen wall is exteriorized and clamped between two metal bars, the fold necrotizes and sloughs, and a fistula between the lumen of the rumen and the outside is created. RF in sheep usually performed in a one-stage method

(Dougherty, 1955; Hecker, 1974; Thyfault et al., 1975; and Dougherty, 1981). The two-stage method is usually recommended for cattle, but other researchers used this method in sheep as well (Quin et al., 1938; Philipson et al., 1939).

The abdominal wall incision and stabilization of the rumenal cannula in the RF are a big challenge to the surgeon, especially in sheep due to the weak abdominal wall musculature. The main postoperative complication is the leakage of rumenal fluid between the surgical wound edges due to scarring (Komarek, 1981), necrosis of the tissue (Harrison et al., 1957), the damage of the low quality material used as rubber cannulas (Dougherty, 1955; Kondos, 1967), or when using a cannula of rigid material as the surgical incision must be bigger in size to let the cannula pass in it (Harmon and Richards, 1997). Moreover, infection and peritonitis are the most important postoperative complications that may possibly tend to severe crisis and death (Azizi et al., 2007).

Little data on safety or efficacy of RF in sheep are available, only one study using the two-stage method reported 1/10 sheep with suture rupture, wound dehiscence and the cannula too loose to stay permanently in place (Azizi et al., 2007).

This study was designed to provide a detailed description of modified one-stage technique for RF in sheep to simplify the surgery, decrease the post-operative complications, and lengthen the functional life of the cannula. As well as reporting the postoperative short and long term monitoring, care and complications related to the surgery.

## **2. Materials and methods**

### **2.1. Animals**

The study includes 10 healthy rams. All included rams weight  $45 \pm 5$  kg, age 15-22 months old, and with no previous history of gastrointestinal dysfunction.

### **2.2. Rumenal cannula**

A custom-made rubber cannula (Fig.1), not commercially available, was designed as follow:

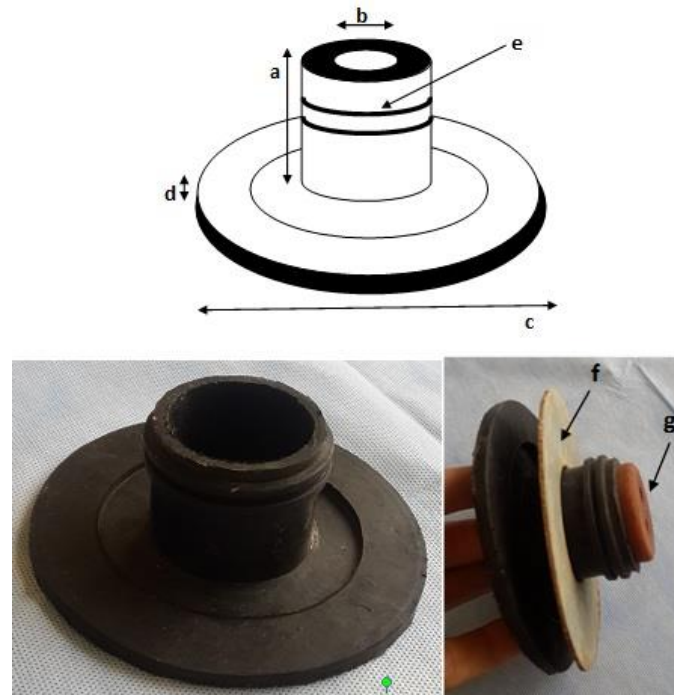
- a. Rubber radiator hose length 4.5 cm
- b. Rubber radiator hose internal diameter 4 cm
- c. Rubber Inner flange diameter 12 cm
- d. Rubber Inner flange thickness 0.5 cm
- e. Rubber ridge to retain the metal outer flange
- f. Metal outer retainer flange diameter 13 cm
- g. Stopper

### **2.3. Surgical protocol (Rumenal Fistulation)**

The experimental protocol was approved by the Animal Care Committee of Maryout Research Station, Desert Research Center. All rams were deemed healthy by physical examination and hematological tests. The animals were fasted for 24 hours, and water was withheld for 12 hours prior to surgery. All rams were sedated by intravenous (I.V) administration of xylazine Hcl (Xyla-Ject®, ADWIA Pharmaceuticals Co. Cairo, Egypt) at a dosage of 0.1 mg/kg B.W., and were restrained on operation table in the right lateral recumbent position.

Following surgical preparation and local desensitization of the left flank region, a circular skin incision (the same diameter as of the radiator hose of the cannula) was made in the mid paralumbar fossa 4 cm ventral to the transverse processes of the lumbar vertebrae and 2 cm caudal to the last rib (Fig.2). The incision was continued through the delicate subcutaneous tissue. The external abdominal oblique muscle, internal abdominal oblique muscle, and transverse abdominal muscle were dissected bluntly along the direction of fibers (grid technique) (Fig.2). The dorsal sac of rumen was exposed after sharp incision of the parietal peritoneum.

The dorso-lateral portion of the sac was then grasped with a pair of Allis forceps and pulled through the abdominal wall incision (Fig.3). This portion was anchored to the subcutaneous tissue using No. 1 Vicryle in continuous circumferential sutures passed seromuscularly in the rumenal wall. The exposed portion of the rumenal wall was incised circularly (gauged strictly enough to allow the rubber radiator hose of the cannula) at the mid-point.



**Fig. 1: Custom-made rumenal cannula.**

a: Rubber radiator hose length.

c: Rubber Inner flange diameter.

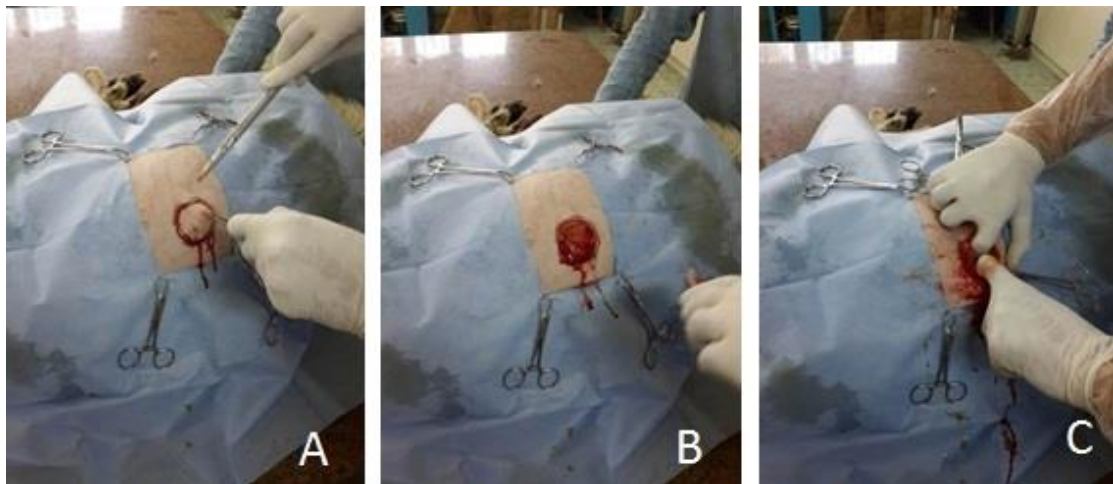
e: Rubber ridge to retain the metal outer flange.

g: stopper

b: Rubber radiator hose internal diameter.

d: Rubber Inner flange thickness.

f: Metal outer retainer flange diameter.

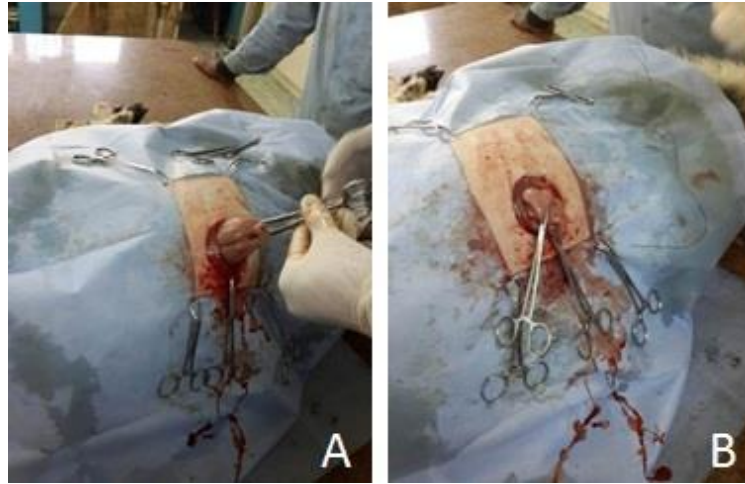


**Fig. 2: Laparotomy.**

A. Skin incision

B. Exposing abdominal muscles

C. Grid technique (opening abdominal muscles bluntly along their fiber direction)



**Fig. 3: anchoring of the rumen (before opening the rumen) to the subcutaneous tissue using Vicryle size 1 in circumferential continuous pattern.**

- A. Exteriorization of the rumen
- B. Anchoring of the rumenal wall to the subcutaneous tissue

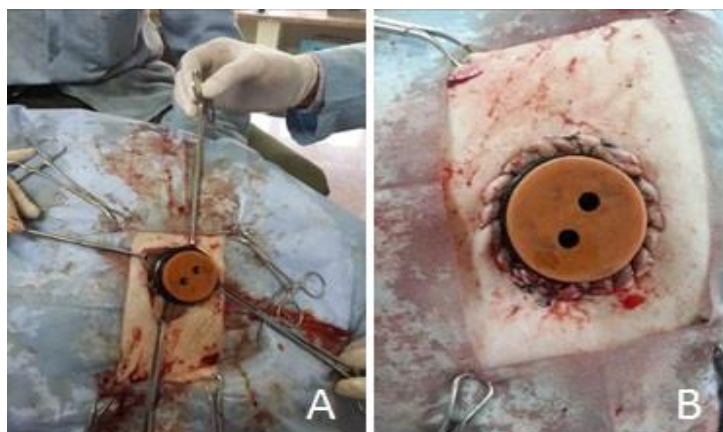


**Fig. 4: Folding the inner flange of the rumenal cannula before insertion into the rumen**

The cannula was thoroughly lubricated with gel to facilitate its advancement into the rumenal stoma smoothly. The rubber inner flange of the cannula was folded and pushed through the radiator hose from inside to outside (Fig.4).

The folded cannula inserted manually into the anchored rumen, it was carefully unfolded by

pushing the folded inner flange from outside to inside the radiator hose to secure the rumenal stoma close to the abdominal wall. The rumenal stoma was then sutured to the skin using No. 1 Vicryle in continuous circumferential pattern (Fig.5) with 2 runs 12-6 and 6-12 o'clock.



**Fig. 5: Cannula insertion and suturing the rumen to the skin**

A: Cannula insertion into the rumen

B: Suturing the rumenal wall to the skin in a circumferential pattern using Vicryle size 1



**Fig. 6: Pushing the Metal retainer flange over the neck of radiator hose in its socket to insure the stability of the cannula**

After that the stainless steel sealing disc (Metal retainer flange) was pushed over the neck of radiator hose in its socket to insure the stability of the cannula (Fig. 6). A rubber stopper used to close the cannula opening.

#### **2.4. Postsurgical procedures**

Postsurgical medical treatment included the use of systemic antibiotics (Pentomycine<sup>®</sup>, AM Trading, 1ml/25kg BW, twice daily), anti-inflammatory (Finadyne<sup>®</sup>, Schering-Plough, 1ml/45 kg BW) for 3-5 successive postoperative days.

##### **2.4.1. Short term monitoring**

The surgical site washed daily with antiseptic solution and inspected for any signs of swelling, bleeding, irritation or infection. Physical examinations (feed intake, body temperature,

respiration rate, and rumen contractions) were performed twice daily for the first 24 hours post-operatively, then daily for 7 days. The cannula washed weekly with antiseptic solution.

##### **2.4.2. Long term monitoring**

Body weight of each animal was obtained weekly for 6 months. The cannula, stopper, and the wound were checked daily to confirm stability of the cannula in its place without leakage.

### **3. RESULTS**

The sedation, anesthesia and restraint used in the current study were fairly adequate to accomplish the procedure.

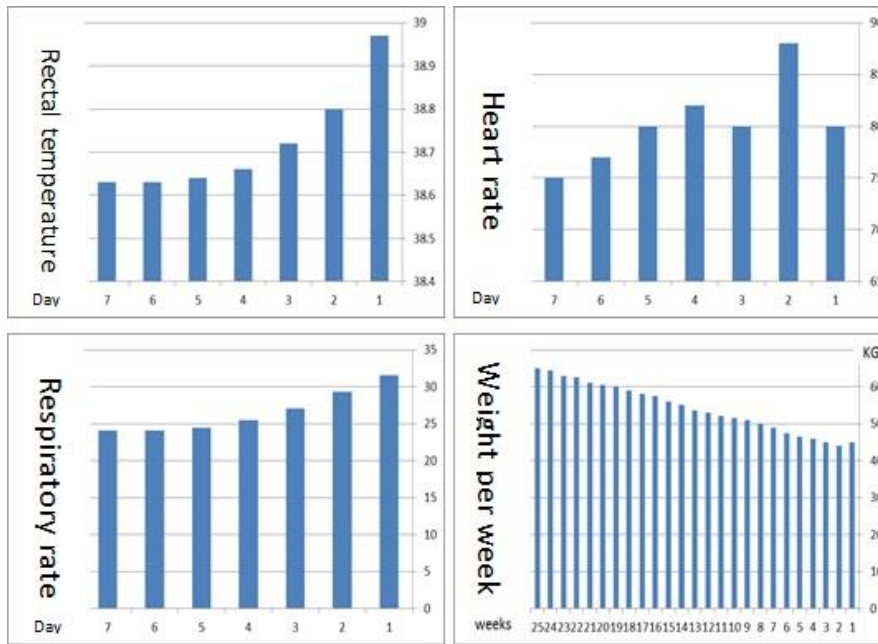
All rams recovered quickly and consumed their food 12 hours after surgery



According to the clinical observations, no early post-surgical complications (hemorrhage, local inflammation, infection, wound dehiscence, or suture abscess in the surgical site) were occurred. No signs of pain were reported. All mean values of physical examinations (rectal temperature, respiratory rate, heart rate, rumenal movements) were within range in

the first 7 post-operative days (Fig. 7). All animals maintained their body weight in the first week post-surgery and then gradually gained weight (Fig. 7).

Six weeks after surgery, a well-defined fistula was completely formed in all animals without any superficial necrosis, discharge or irritation (Fig. 8).



**Fig. 7: Rectal temperature, Respiratory rate, and Heart rate for 7 post-operative days. And weight gaining for 25 post-operative weeks**



**Fig. 8: A well-defined fistula (6 weeks after surgery), showing the healing between rumenal wall and the skin (the arrow) without any complications.**



**Fig. 9: Rumenal fistula 6 months after surgery**

In the long-term observations (6 months after surgery), the cannulas remained fixed in position in all rams. None fistula got bigger in size, and none cannula was rejected, or had to be repositioned (Fig. 9). All animals remained in good health condition and no further complications with the fistula sites up to 6 months after the surgery.

Complete cicatrization and adhesions among the rumen and the abdominal wall were observed. There was no leakage of the rumenal fluid to the abdominal cavity and no cases of peritonitis occurred. No rumenal contents leakage was observed between the cannula and the fistula.

The custom-made cannula was tolerated by all sheep. Only one ram died suddenly after 14 days of surgery. No sign of intra-peritoneal infection was observed on inspection of the carcasses, but there was adhesion between the reticulum and the peritoneum.

#### **4. DISCUSSION**

Nutrition and physiology experimental research studies are dependent on rumen fistulation (Thyfaul 1975). During these long time studies, permanent placement of the cannula and its stability are a challenge.

RF in sheep usually performed in a one-stage method (Dougherty, 1955; Hecker, 1974; Thyfault et al., 1975; and Dougherty, 1981). The two-stage method is usually recommended for cattle, but other researchers used this method in sheep as well (Quin et al., 1938; Philipson et al., 1939).

All two stage RF techniques are time consuming to the veterinary surgeons, in which we had to do a second rumenotomy, after 7-12 days, to implant the

cannula (Azizi et al., 2007). As an advantage, it is important to point that one-stage technique used in current study is very easy and faster. Also the appropriate surgical technique used in this study showed lack of wound complication and systemic disorders throughout the follow up.

Blunt dissection (using grid technique) of the abdominal muscles in their fiber directions allows a firm and stable fit around the cannula. The bluntly separated muscular structure takes a valve-like action (Turner et al., 1989). The function of this reconstructed muscle structure might offer a better fitting and securing of the cannula in its position in comparison with the traditionally-created opening.

The inflammatory response following trauma results in fibrin deposition and formation of fibrinous adhesion between contacting surfaces within 2 to 4 days (Tol van den, 2001). Abrading rumenal serosa with a piece of surgical gauze enhances the adhesion formation between rumen and skin (Martineau et al 2015). In current study, rumenal anchoring to the subcutaneous tissue before rumenal fistulization simulate the abrading effect done by Martineau et al, 2015, and this can explain the smooth and rapid healing of RF within 10 days without complications. Durmic et al. 2015, stated that, rumenal fistula in sheep physiologically remodeled within several weeks after the surgery and the wound form a broad, strong scar.

The Weight and size of inner flange play an important role in rumenal cannula stability (Mc Sweeney, 1989). In current study, using a relatively tough and thick inner washer (0.5 cm in thickness) to strengthening thickness of the interior flange of the cannula, resulted in permanent cannula stability

The pliability of the cannula used in the current study was important for placing the cannula into the rumenal stoma. The use of flexible cannulas provides better anatomical adjustment and is efficient for retrieval of rumenal content with minimal risk of local necrosis and peritonitis (Stedile et al., 2008). Using a cannula of rigid material make the surgical incision must be bigger in size to let the cannula pass in it (Harmon and Richards, 1997), the flexible rubber cannula used in this study could be folded to fit a small size skin and rumenal stoma.

All rumenal cannulas leaks and frequent adjustment should be applied, Dougherty 1981. In our study, the flexibility of the rumenal cannula enables to put it exactly and securely in the small sized rumenal opening without slippage or dropping (the circular incision of the skin and the rumen were gauged strictly enough to allow the rubber radiator hose of the cannula). No fistula got a bigger size, and none cannula was rejected, or had to be repositioned.

Hemorrhage, local inflammation, surgical site infections, subcutaneous emphysema, wound dehiscence, and dropping of cannulas are the most common post-surgical complications of RF in ruminants (Azizi et al., 2007; Muzzi et al., 2009). In spite of that, none of such complications occurred in the current study, this was supposedly due to the appropriate adhesion formed between skin and rumen.

According to the clinical observations, and long term postoperative monitoring, all cannula was efficiently secured without any surgical complication.

## 5. CONCLUSION

The one-stage technique used in this study to fix a custom-made cannula in its position revealed no slippage of the cannula and no critical postoperative complications were seen in all animals.

This technique is recommended to experimental research on sheep nutrition and physiology.

### Disclosure

The author declares no conflict of interest related to this report.

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