Sterilization of Male Dogs by Bilateral Electrocoagulation of Testicular Blood Vessels

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ABSTRACT

Key words: Dog, sterilization, electrocautery, testicular blood vessels, testosterone

Canine overpopulation is a worldwide problem especially in rural area for this reason many researches have been performed to choose the most effective and less invasive technique for sterilization of male dogs with minimal complications. This study aimed to evaluate the possible outcomes and complications following bilateral electrocoagulation of testicular blood supply, as a method of sterilization of male dogs. Six stray male dogs were used in this study. Electrocoagulation of the testicular blood supply was induced through a prescrotal 1-2 cm incision length followed by separation of spermatic cord and exposure of testicular blood vessels. The outcomes were measured depending on clinical signs, Histopathological evaluation, ultrasonographical evaluation and testosterone level estimation. Electrocoagulation of testicles was accompanied by a very short prescrotal incision with less operation time. Some difficulties were relevant during dissection and separation of testicular blood supply with mild to moderate bleeding. Some inflammatory reactions followed by marked testicular atrophy were observed. Severe coagulative necrosis of seminephrous tubules and remnants only of mature spermatzoa were found. Ultrasonographical findings revealed marked decrease in testicular dimensions with uniform hypoechoic appearance of testicular matrix. It could be concluded that bilateral Electrocoagulation was proved safe and effective method for dog sterilization without any complication.

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1. INTRODUCTION

Surgical sterilization of dogs and cats is a method of rendering animals unable to reproduce and completely sterile. Aim of sterilization is to control overpopulation problem and to prevent diseases associated with the reproductive system, such as mammary neoplasia and prostatic hyperplasia (Olson et al., 1986; Jana et al., 2005, Jana et al., 2007). Sterilization could be performed by surgical and non surgical method as classified by Huber et al., (1986). Surgical sterilization (elective gonadectomy) includes traditional castration, early age gonadectomy and vasectomy (Howe, 2006). Non-surgical Sterilization induced by intratesticular injection of chemical substances as Calcium chloride and zinc gluconate that cause destruction of testicular cellsand subsequent permanent infertility (Samanta, 1998; Jana et al., 2002; Levy et al., 2008, Abu-Ahmed et al., 2015). Many complications were recorded after both surgical and chemical sterilization as scrotal bruising, hemorrhage, swelling and sever pain (Phillips and Leeds, 1976; Booth, 2003, Tobias, 2010). Another methods were used for rendering testes nonfunctional in scrotum by prevention of its blood supply and this occurred by either ligation, transection and torsion of spermatic cord or by cautery of blood vessels (Turner and Brown, 1993; Booth, 2003; Baba et al., 2013, Abu-Ahmed et al., 2012). Electro surgery is a term used to describe the passage of high frequency current through the tissue to create a desired tissue effect (Wu et al., 2000). Tissue effects that can be achieved with electro surgery can be divided into three basic groups: cutting, fulguration, and desiccation. Electrosurgical cutting divides tissue with electric sparks that direct intense heat to the tissue over a very limited surface area, producing maximum current density and delivering the greatest amount of heat over a very short time (Massarweh et al, 2006). The most widely function of electrocaogulation is to achieve haemostasis during surgical procedures, this effect is achieved by direct application of heat via active electrode to the vessel causing coagulation of blood...
proteins (Memon, 1994). This study was designed to evaluate transection of testicular blood supply by electrocoagulation instead of spermatic cord ligation as a method of sterilization in dogs.

2. MATERIALS AND METHODS

2.1. Animals:
The present experimental study was carried out on 6 apparently healthy, sexually intact, adult mixed breed male dogs having body weights of 12 - 31 kg and age of 3-10 years. All procedures were performed at the Department of Surgery, Faculty of Veterinary Medicine Alexandria University.

2.2. Animal Preparation and Anesthesia:
Dogs were subjected to physical examination to assess their fitness for the study. They were fasted for 12 hours prior to surgery, while water was left adlibitum. All dogs were premedicated with 2 mg/kg body weight of Xylazine hydrochloride (Xylaject 20 mg/ml, ADWIA Co. S.A.E., Egypt) injected intramuscular and 0.04 mg/kg body weight of atropine sulphate (Atropine 10 mg/ml, ADWIA Co. S.A.E., Egypt) injected intramuscular. After 15 minutes, general anesthesia was induced and maintained with 5 mg/kg body weight of Ketamine hydrochloride (Ketamine 50 mg/ml, Rotexmedica, Trittau, Germany) injected intramuscular in the hind limb especially at thigh muscle after its routine preparation for aseptic injection.

2.3. Surgical Procedures:
Each dog was positioned in dorsal recumbency and the scrotal and prescrotal area aseptically prepared for surgery. Two parallel incisions (1-1.5 cm each) were made through skin at the prescrotal area over the spermatic cord, using blunt dissection each spermatic cord was exteriorized over an artery forceps. After careful incision of tunica virginals the vas deference was identified then bluntly separated, segment of about 2cm of the vascular portion of spermatic cord was clamped by using two artery forceps then apply the cautery device (SURTRONE 400 HP, 80 °c) in between until complete separation, the two ends of blood vessels were checked for bleeding before reposition. Skin incision was closed by simple interrupted suture using silk (fig., 1).

2.4. Clinical evaluation:
The animals kept under observation for 2 months, during the first week the animals checked for signs of pain, swelling, wound dehiscence. Testicular size was observed every week till the end of experiment.

2.5. Testosterone level estimation:
Testosterone levels were measured in each dog just before surgery and at 2 months post operation, after that each dog was injected with (5,000 USP Units) of human chorionic gonadotrophine (HCG) then testosterone levels were measured again 1 and 24 hours post IM injection of HCG (Pregnyl®, Baxter Oncology GmbH, Germany).

Fig. (1): electro cauterization and transection of testicular blood Supply: A- Showing animal prepared for aseptic surgery, B- show skin incision at prescrotal region, C- exposure of spermatic cord by using artery forceps, D- The testicular blood vessels (arrow) separated from vas deference, E- application of diathermy device, F- complete transection of blood vessels.
2.6. Ultrasonographical Imaging:
Changes in testicular size (length and width) were evaluated ultrasonographically at (1, 1.5, 2 months) post cautery. Ultrasound beam was adjusted before imaging to 5 MHz’s. Skin was aseptically prepared and sufficient amount of sonographic gel was then applied immediately before application of the Micro-convex transducer.

2.7. Histopathological Evaluation:
Testes were removed from scrotum by scrotal castration 2months after surgery for Histopathological examination. Tissue specimen obtained and fixed in 10% neutral buffered formalin solution for at least 24 h. The fixed specimens were processed through the conventional paraffin embedding technique. Paraffin sections of 5 μm thickness were prepared and stained with hemotoxylin and eosin (HE) then read under optical microscope.

3- RESULTS
3.1. Clinical evaluation and post-operative complication:
Spermatic cord was easily exteriorized through 1-1.5 cm incision length. The operative time was 12±2.5 min. There was not bleeding post electrocoagulation of blood vessels except one case, which was controlled immediately by application of diathermy device again otherwise only scanty bleeding from skin, and sub cut was observed (table, 1). Moderate scrotal swelling was observed during first three days then subsided at fifth day post operation, reduction of testicular size was observed after that until 2-month post- surgery. Mild signs of pain in form of licking of wound were observed in first 2 days. Wounds in all cases were almost closed, dry and take short time to heal.

3.2. Testosterone analysis:
As shown in table (2) testosterone levels were significantly decreased at 2 months post electrocoagulation. After injection of HCG testosterone level, remain unchanged from the pre-injection value.

3.4. Ultrasoundographical evaluation:
Ultrasonographic findings post operation revealed marked reduction of testicular dimensions which were ø= 9.7 mm, ø= 34.7 mm after 1 month. And after 1.5 month are ø1 = 20.8 mm, ø2 = 5.4mm while after 2 months the testicular dimensions are ø= 10.4 mm, ø=7mm (fig., 2).

<table>
<thead>
<tr>
<th>Item</th>
<th>Blood vessels Cauterization</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incision length</td>
<td>1-1.5 cm</td>
</tr>
<tr>
<td>Operation time</td>
<td>12±2.5 minutes</td>
</tr>
<tr>
<td>Intra-operative bleeding</td>
<td>Mild bleeding</td>
</tr>
<tr>
<td>Easiness and Equipment</td>
<td>Some difficulty</td>
</tr>
</tbody>
</table>

<p>| Table (2): show testosterone level before and post operative along with injection of HCG |
|----------------------------------|-------------------------------|</p>
<table>
<thead>
<tr>
<th>Cauterization of blood supply</th>
<th>Before treatment</th>
<th>2 month after treatment</th>
<th>1hr After GNRH injection</th>
<th>24hr After GNRH injection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cauterization of blood supply</td>
<td>4.50±0.19</td>
<td>0.03±0.00b</td>
<td>0.03±0.00c</td>
<td>0.03±0.00c</td>
</tr>
</tbody>
</table>

Fig (2): Ultrasound imaging of testicle exposed to Blood vessel cauterization after A: 1 month, B: 1.5 month, C: 2months. Note: 1- Marked reduction of testicular dimensions with time, 2- hypo echoic uniform appearance of testicular matrix.
4.5. Histopathological findings:

**Macroscopically** the testes appeared severely atrophied in (4 animals) while in other 2 animals only remnants of testicular tissue were found in scrotum. The atrophied testes have yellowish white coloration.

**Microscopically:** The testes showed massive coagulative necrosis of all seminephrous tubules as well as collecting tubules represented by increased Eosinophilic and loss of basophiles in almost all spermatogonia cells. Some of the tubules showed remnants of mature spermatozoa. The interstitial tissue showed proteniuos edema with Fibrinous Eosinophilic network (fig.4).

4. DISCUSSION

Although surgical methods of sterilization have remained the mainstay in dogs they were accompanied by many disadvantages and post-operative complications such as hemorrhage, infection and dehiscence (Adin, 2011). Several alternative methods have been developed recently depending on obliteration of blood supply to the testes rendering it nonfunctional in situ either by ligation and/or transection of spermatic cord which resulting in coagulative necrosis to the testicular cells (Abu-Ahmed et al., 2012, Baba et al, 2013). Electrocoagulation of testicular blood vessels was used in this study to block blood supply to the testes. Spermatic cord was easily manipulated and exteriorized from 1-1.5 cm incision length. Moderate scrotal swelling was observed during first three days then subsided at fifth day post operation, this was attributed to minimal trauma from incision and manipulation of spermatic cord, on other hand, ligation of spermatic cord resulted in marked swelling which subsided after 9 to 15 days post operation (Ponvijay, 2007 and okwee-Acai et al., 2008), this swelling was due testicular edema. Wounds in all cases were almost closed, dry and take short time for healing. On other hand wounds post pinhole technique in dogs were wet, inflamed and septic in some cases was recorded by Abd-El wahed et al., (2014). Licking of wound was observed during the first day post-surgery which considered as a normal response to tissue injury. Advantages of the blood cauterization include shorter operative time, less tissue injury, less postoperative swelling and pain and a lower complication rate As ligation like pinhole has different complication as severe pain (marked increase of cortisol), swelling and inflammation (Booth, 2003). Blood vessels cauterization in this study resulted in mild pain during the procedure and early follow-up period. Blood vessels cauterization can be utilized if a large number of animals are to be sterilized in a small period of time with less surgical interventions. During cauterization, smooth muscle relaxation is needed to perform surgery. The combination of xylazine at 2mg/kg and ketamine at 5 mg/kg was sufficient to achieve

![Fig (3): Macroscopical findings of atrophied testes. A: Atrophied testis within the scrotum, B: Severe adhesions between the testicle and its tunicae and scrotum, C: removed testes note its small size, D: on opening testes have yellowish white colors.](image1)

![Fig (4): Histopathological picture of the testes after 60 days of blood vessels cauterization.](image2)
anaesthesia. Naccarato and Hunter (1979) and Kumar 

et al. (2011) have also used xylazine and ketamine for
effective anaesthesia in male dogs. All animals
recovered smoothly after anaesthesia and survived the
surgery. All animals started taking food and water
normally. Marked reduction of testicular size was
recorded all over the observational period which was
nearly disappeared in some cases this result was
confirmed by ultrasonographical examination which
revealed that testicular dimensions were decreased
markedly at 2 month post operation. Significant
reduction of testosterone level was recorded in our
study, this was due to degeneration of leydig cells by
ischemic necrosis of testes, and the same result was
recorded by Ponvijay (2007); okwee-Acai et al.,
(2008), Baba et al., (2008). A useful non- invasive
technique for determining the presence of testicular
tissue in cryptorchidism and questionable animals is
the endocrine analysis (Jean et al., 1992, Cox, 1993).
The test based on the increased production of
testosterone in response to the injection of HCG which
stimulates the leydig cells as do LH (Morrow, 1986,
Turkstra et al., 2005). Testosterone remained
unchanged in our research even after injection of HCG
which ensured absence of survived leydig cells. The
moderate disturbance of blood supply of testes could
cause testicular malfunction, in rats 5 hours reduction
of approximately 70% of normal value may be
sufficient to induce death of spermatogonia and early
spermatocytes (Bergh et al., 2001). While in dogs 10
hours of ischemia resulted in the elimination of all
leydig cells and the testicular elements were replaced
by connective tissue (Smith, 1955). In this study the
histological findings revealed massive coagulative
necrosis of all seminiferous tubules as well as
collecting tubules with interstitial edema these
findings confirmed the successful sterilization.

5. CONCLUSION

According to the result of clinical appearance,
testosterone analysis only or with GNRH,
ultrasonography of the testes and pathological picture;
the electrocoagulation of testicular blood vessels
proved to be an alternative technique over other
conventional methods of Sterilization in dogs.

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