

CANINE TESTICULAR HISTOPATHOLOGY ENSUING CHEMICAL CASTRATION

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ABSTRACT

Castration in domestic and companion animals plays an important role in controlling the animal population, aggressive male behavior, and producing meat consistently acceptable to consumers. Surgical castration is widely used method, but it is more expensive and painful if not well managed. This research focused on using 10% iodine and 1% polidocanol foam as intratesticular injections to achieve castration in adult dogs owned by a client in the Lwandeti village in Kakamega County, Kenya. Two dogs received 1mL of 1% polidocanol, while one dog received 1ml of 10% Lugol's iodine in each of their testicles. Surgically harvested testicles after three months for histological examination. The histological examination revealed that the seminiferous tubules of testes that received 10% Lugol's iodine had cellular infiltration and fibrin deposition. Those that received 1% polidocanol showed moderate to severe cellular infiltration but less than the iodine group. Concluded that polidocanol and iodine as a chemical method can be used instead of surgical castration.

Keywords: Dog, Chemical castration, Iodine, Polidocanol.

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1. INTRODUCTION

Contraception plays a major role in prevention of unintended population growth and abortion (Roth and Amory 2016). In dogs, for example, control of dog population through castration and spaying helps in control of rabies-a serious zoonotic disease in developing countries (Schildecker et al. 2017; Tenzin et al. 2015). According to Kwoba et al. (2019), domestic dogs transmit 99% of the estimated 59,000 human rabies deaths occurring globally annually. There are various methods, both surgical and nonsurgical, that are currently used as contraception methods in domestic animals.

Surgical castration has been the most common contraception method used in male dogs, together with other livestock. It has been shown to reduce the incidence of prostate adenomas, neoplasms and prostatic hyperplasia (Ong et al. 2017). However, animal rights activists argue that castration is against animal welfare (Ajadi and Gazal 2016). This has been supported by scientific studies. A study in the United States by Sundburg et al. (2016) on effect of orchiectomy revealed that the procedure increases the risk of autoimmune diseases such as atopic dermatitis, autoimmune hemolytic anemia, hypoadrenocorticism, hypothyroidism, immune-mediated thrombocytopenia and inflammatory bowel disease. Apart from the risks that surgical castration poses, it is time consuming and has a high cost due to the need of surgical skill and equipment (Ibrahim et al. 2016).

As animal activists, supported by various scientific materials/chemical substances, argue against orchiectomy alternative methods to male permanent contraception have been carried out. One of the methods is the use of chemicals that are injected directly into the testis or through the epididymal tissue. The use of chemical sterility is inexpensive and technically easier compared to orchiectomy therefore is suitable for mass sterilization. These chemical agents induce azoospermia resulting into infertility (Fagundes et al. 2014). Chemical agents that have shown efficacy as chemical sterilant include Zinc gluconate, Calcium chloride and Sodium chloride. In a study using cats, Fagundes et al. (2014) demonstrated that zinc gluconate causes atrophy and dilation of seminiferous tubules, a decrease in the number of germ cells, and incomplete spermatogenesis. Sertoli cells had various degrees of cytoplasmic vacuolization. Intertubular tissue revealed active fibroblasts, collagen deposition, and inflammatory cells. Calcium chloride was used in a study by (Jana and Samanta 2006; Jana and Samanta 2007) at levels of 5, 10 and 15 mg per testis per kg body weight and proved efficacious in dogs and albino rats. Canpolat et al. (2016) used 20% hypertonic saline to achieve focal coagulation necrosis in testicular tissue together with diffuse and severe degenerative changes in seminiferous tubules in non-adult dogs. The aim of this study is to test the efficacy of povidone iodine and polidocanol as chemical sterilant.

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2. MATERIALS AND METHODS

2.1. Test Products

A good chemical sterilant is one which effectively arrests spermatogenesis and androgenesis as well as the libido without having toxic effects (Ibrahim et al. 2016). The products to be used are: 1% polidocanol (LGM Pharma, USA) and 10% Lugol's iodine (MEDS, Nairobi-Kenya). Polidocanol has been used as a sclerosing agent widely (Nastasa et al. 2015; Das et al. 2016). At 1-2%, Lugol's iodine has been used for uterine irrigation. At stronger concentration than this it induces an inflammatory response resulting in loss of functionality of an organ (Brinsko 2001). The study used either 1ml of 1% polidocanol or 1ml 10% iodine injection into each testicle to evaluate their effect on the testicular morphology and function in dogs.

2.2. Animals

The study used a total of three male adult dogs owned by a client in Lwandeti village of Kakamega County, Kenya. The dogs were in good health as per the guidelines by the vet surgeon on the project. One dog (approximately 2 years having weight 8 kg) received 1ml of 10% i Lugol's odine into either of its testicles while the remaining two dogs (approximately 2 years having weight 7-8 kg) received intratesticular injection of 1ml of 1% polidocanol into each of their testicles.

2.3. Intratesticular Injection

The dogs were sedated using an intramuscular combination of xylazine (LGM Pharma, USA) at 1.0mg/kg body weight and ketamine at 1.0mg/kg body weight as indicated by Silva et al. (2020). The testicles were cleaved and cleaned using 70% ethyl alcohol before the test products were injected into the testicles. The dogs were followed up for three months after injection of the chemicals.

2.4. Testicular Histopathology

After the three-month study period, the dogs were surgically castrated to harvest the testicles for histopathology. The dogs were anaesthetized using a combination of 1.0mg/kg bwt xylazine and 1.0mg/kg bwt ketamine intramuscularly and skin anesthetized using 2% lidocaine (Silva et al. 2020). The testicles were extracted, cut in half and immersed in 10% formalin (pH 7.0) and transported to the lab for histopathological analysis. Testicles were then be trimmed, dehydrated in a series of graded alcohol solution, and embedded in plastic resin of glycol methacrylate (Historesin, Leica, Wetzlar, Germany). Histologic sections (4-5µm) were stained with Hematoxylin and Eosin and analyzed morphologically.

3. RESULTS AND DISCUSSION

Histological sections of the two testicles that were injected with 10% Lugol's iodine showed massive cellular infiltration in the seminiferous tubules with fibrin deposits. From the microscopic view, more than 90% of the tubules atrophied, as shown in Fig. 1A and 1B. In the four testicles that received polidocanol 1%, there was moderate to severe mononuclear cell infiltration as shown in Fig. 2A, 2B and 2D.



Fig. IA and IB showing mononuclear cell infiltration in seminiferous tubule following injection with 10% Lugol's iodine (H&E stain; 100X and 400X, respectively).

This current study demonstrates that iodine at 10% can be used as a chemical sterilant as shown by Jana and Samanta (2007) and Pereira et al. (2018) in which calcium chloride was the chemical used. The substances initiate an inflammatory reaction that leads to fibrosis, making the seminiferous tubules sealed and therefore unable to carry out their physiological function (Oliveira et al. 2013). In the study by Oliveira et al. (2013) in which they used zinc gluconate in cats there was significant azoospermia and decrease in penile spines, with no significant alteration on testosterone levels. In 2018, a study by Pereira et al. (2018) on histological effects following intratesticular injection

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of calcium chloride in bulls at various concentrations showed coagulation necrosis of seminiferous tubules and interstitial cells was observed, mainly in the medial portion in all groups. Some animals presented total absence of tubular formations in all the studied groups. Inflammatory infiltrate, fibroplasia and neovascularization were found to be predominantly discrete (Pereira et al. 2018). Polidocanol foam at 1% showed moderate to severe inflammatory reaction and atrophy of seminiferous tubules. This could be attributed to a lower concentration of the drug being used (Oliveira et al. 2013).



Fig. 2A, 2B and 2C: showing severe to moderate mononuclear cell infiltration in the seminiferous tubules following intratesticular injection with 1% polidocanol (H&E stain; 100X).

Conclusion: The study makes the following conclusions, i) 10% Lugol's iodine has potential as a chemical sterilant to be used instead of surgical castration and ii) at 1%, polidocanol has moderate potential a chemical sterilant instead of surgical castration.

Author's Contribution: NWW conceived the idea of the study. NWW, JM and RK performed the study. NWW and JM wrote the manuscript. All authors revised and approved the final version of the manuscript.

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