

## EFFECTS OF CASTRATION ON PENILE AND URETHRAL DEVELOPMENT IN AWASSI LAMBS

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### Summary

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The objective of this study was to determine the effect of castration on penile and urethral development in Awassi lambs. Twenty 1–2-weeks old Awassi lambs were divided randomly into 4 groups. Groups 1, 2, and 3 were castrated at 2 weeks, 3 and 5 months of age, respectively. Group 4 was not castrated (control). To assess penile and urethral development, penile length, penile diameter and urethral cross sectional area were measured at 3 sites (proximal sigmoid flexure, distal sigmoid flexure, and glans penis) using digital image analysis. The penis of control animals was significantly longer and larger in diameter compared to that in castrated animals at 2 weeks and 3 months of age. The urethral cross sectional area in control animals was significantly larger at the 3 selected sites compared to groups 1 and 2. The results of this study indicate that castration affects negatively the normal development of the penis and urethra and that Awassi lambs, castrated at an early age have smaller penises and narrower urethras.

**Key words:** Awassi lambs, castration, development, penis, urethra

### INTRODUCTION

In all mammalian species, development, growth and maintenance of urogenital organs are under the influence of androgens, namely testosterone (Louca *et al.*, 1977; Wilson *et al.*, 1995). Previous studies on the effects of castration and therefore the lack of sex hormones on the growth and development of male ruminants have shown numerous contradictory findings. Castration of male ruminants resulted in increased distal limb bone length, and shorter vertebral column resulting in taller animals and shorter body length than intact counterparts (Brannang, 1971) In small ruminants, castration by surgical or non-surgical means has been

used to improve the quality and palatability of meat (Jeremiah, 2000). In addition, the effect of castration on growth and feed conversion rates, and meat quality in small ruminants has been extensively studied (Turton, 1969; Field 1971; Louca *et al.*, 1977). Because of the proposed possible reduction in penis size and urethral diameter, the practice of early age castration in ruminants has been thought to predispose to obstructive urolithiasis (Oheme & Tillman, 1965). Clinically, obstructive urolithiasis is a serious, potentially life-threatening disease that results from the obstruction of the urethra by calculi. In the United States, estimates of

feedyard lamb losses due to obstructive urolithiasis are over 4% of all lamb deaths (Kimberling & Arnold, 1983; Floyd, 1989; Smith & Sherman, 1994). The condition is diagnosed primarily in castrated males, especially those castrated early in life (Oheme & Tillman, 1965). Apparently, there is no sex predisposition for calculi formation. However, clinical urolithiasis occurs in the male because of anatomical and hormonal differences (Kimberling & Arnold, 1983; Floyd, 1989; Smith & Sherman, 1994). Although, many other factors including management practices, nutrition and environmental factors are involved in the etiology of the condition in small ruminants, the role of early age castration as a predisposing factor remains to be elucidated (Oheme & Tillman, 1965). There is limited information available in the current literature regarding the effect of castration on penile and urethral development in lambs and hence the potential role of castration in the incidence of this economically important disease. Therefore, the purpose of this study was to evaluate the effect of early age castration on penile and urethral development in Awassi lambs.

## MATERIALS AND METHODS

### *Animals*

Twenty healthy male Awassi lambs (1–2 weeks old;  $7 \pm 0.57$  kg BW) were selected and randomly assigned to 4 groups of 5 lambs each. Lambs in groups 1, 2 and 3 were castrated at 2 weeks, 3 months and 5 months of age, respectively. Surgical castration was performed as described by Tibary and Van Metre (2004). Lambs in group 4 were not castrated and served as control. Each lamb was udder-fed freely from its own mother until weaning (2

months of age). After weaning, lambs were housed separately and offered a barley-based diet containing 16% crude protein.

### *Slaughter*

Animals were humanely slaughtered for meat consumption at 7 months of age. The entire urogenital tract including the urinary bladder, urethra and the whole penis up to the glans was carefully removed.

### *Penile length and diameter*

The penile length was measured using a roller. The penile diameter (PD) was measured at 3 different regions – proximal sigmoid flexure (PrF), distal sigmoid flexure (DsF), and glans penis (GsP) using a digital micrometer caliper (Japan).

### *Tissue sample collection*

Approximately 1 cm<sup>3</sup> cross sectional tissue samples were taken from each penis from 3 regions (PrF, DsF, and GsP). Tissue samples were placed in 10% buffered formalin for further processing. All trimmed samples were processed for histopathologic examination.

### *Urethral cross sectional area (CSA) measurement*

Urethral CSA measurement was performed as previously described with some modifications (Chen & Farese, 2002). Briefly, the urethral histological sections were viewed at 4 x magnifications, and images were obtained with a Nikon eclipse 200 microscope with digital camera (Nikon, Japan). The images were imported into Adobe PhotoShop CS2 (Adobe Systems, San Jose, CA, USA). Following selection of the luminal area of the urethra using the magnetic lasso tool, the pixel numbers were obtained using the histogram command (expanded view).

CSAs were expressed by the computer as pixels per mm<sup>2</sup>. Results were directly imported into a spreadsheet program (Excel, Microsoft Inc., Redmond, WA, USA) for analysis. To determine the conversion factor, a digital image of 1 mm micrometer slide was captured at same magnification power and light density. Each mm<sup>2</sup> of the digital image equaled ~1180 pixels. The calculated areas were multiplied by a conversion factor of 8.47×e<sup>-4</sup> to determine the CSA of the urethra in μm<sup>2</sup>. Values less than 100 μm<sup>2</sup> were assumed to represent artifacts from the image conversion process and were excluded from analysis. To further confirm our results, we used image analysis software (ImageJ, NIH Image software, USA).

*Statistical analysis*

Data were expressed in mean ± SD. Mean values of the penile length, penile diameter, and urethral CSA were compared by one way ANOVA for multiple groups.

When significant difference was identified, Bonferroni's post test was used. Statistical analyses were performed using Graphpad Prism for Windows (Graphpad, San Diego, CA, USA). The differences were considered significant at P< 0.05.

RESULTS

*Penile length*

The penile length in control group was significantly higher than the penile length in groups 1, 2 (P<0.001) and 3 (P<0.05). The penile length in group 3 was significantly (P<0.01) higher than that in group 1.

*Penile diameter*

At PrF region, the penile diameter in control group was significantly larger than the respective penile diameters in group 1 (P<0.001) and group 2 (P<0.05) (Table 1). The value in group 2 was significantly

**Table 1.** Means ± SD of urethral cross sectional area (UCSA) and penile diameter (PeD) at the proximal sigmoid flexure (PrF), distal sigmoid flexure (DsF), and glans penis (GsP)

Group	PrF		DsF		GsP	
	UCSA (mm)	PeD (cm)	UCSA (mm)	PeD (cm)	UCSA (mm)	PeD (cm)
1	0.02± 0.006 <sup>a,b</sup>	4.08± 1.56 <sup>a,b,c</sup>	0.02± 0.01 <sup>b,c</sup>	4.83± 2.07 <sup>b,c</sup>	0.02± 0.02 <sup>b,c</sup>	4.55± 1.13 <sup>a,b,c</sup>
2	0.04± 0.02 <sup>c</sup>	7.5± 0.99 <sup>c</sup>	0.05± 0.008 <sup>c</sup>	8.16± 2.07	0.07± 0.01 <sup>c</sup>	8.15± 1.22 <sup>c</sup>
3	0.07± 0.02 <sup>f</sup>	8.48± 0.65	0.07± 0.01 <sup>f</sup>	8.93± 0.64	0.08± 0.02 <sup>f</sup>	9.15± 0.85 <sup>f</sup>
Control	0.12± 0.05	10.85± 1.12	0.13± 0.04	11.53± 0.97	0.17± 0.02	12± 0.86

Significant differences: <sup>a</sup> between groups 1 and 2; <sup>b</sup> between groups 1 and 3; <sup>c</sup> between group 1 and the control group; <sup>d</sup> between groups 2 and 3; <sup>e</sup> between group 2 and the control group; <sup>f</sup> between group 3 and control group.

( $P < 0.05$ ) higher than that in group 1 whereas the value in group 1 was significantly ( $P < 0.01$ ) smaller compared to that in group 3.

At DsF region, the penile diameter in control group was significantly ( $P < 0.01$ ) larger than that in group 1. The value in group 1 was significantly ( $P < 0.05$ ) smaller than that in group 3.

At GsP region, the penile diameter in control group was significantly larger than those in group 1 ( $P < 0.001$ ), group 2 ( $P < 0.01$ ), and group 3 ( $P < 0.05$ ). The value in group 1 was significantly lower compared to groups 2 ( $P < 0.01$ ) and 3 ( $P < 0.05$ ).

#### *Urethral CSA*

At PrF region, the urethral CSA of controls was significantly larger ( $P < 0.01$ ) than those in groups 1, 2 and 3. Significant difference ( $P < 0.05$ ) was also noted at the same region between groups 1 and 3 (Table 1).

At DsF region, the urethral CSA in the control group was significantly ( $P < 0.001$ ) larger compared to group 1. The respective values in groups 2 and 3 were significantly ( $P < 0.01$ ) smaller than that in controls (Table 1).

At GsP region, the control urethral CSA was significantly higher related to groups 1 ( $P < 0.001$ ), 2 and 3 ( $P < 0.01$ ). A significant difference ( $P < 0.05$ ) was also noted between groups 1 and 3.

#### DISCUSSION

In this study, the effects of castration on penile and urethral development in Awassi lambs were investigated. Surgical castration was used in this study because it is one of the most common techniques used in the field that gives immediate results by removing the source of endoge-

nous androgens (Tibary & Van Metre, 2004).

Few studies have compared penile and urethral luminal size in castrated and uncastrated ruminants (March & Safford, 1957; Baily, 1975; Kumar *et al.*, 1982). In addition, the beneficial effects of delaying castration to an older age on the penile and urethral size have been documented in calves (March & Safford, 1957). In agreement with these studies, we found significant increases in penile and urethral luminal size in uncastrated lambs compared to counterparts castrated at an early age. Even when lambs were castrated at 5 months of age, a significant increase in penile and urethral luminal sizes were noted indicating a beneficial effect of delaying castration allowing more time for growth and development of urogenital organs under the influence of testosterone.

Previous studies on Barbari goats and steers found that castrated animals have smaller penises and narrower urethral lumens (March & Safford, 1957; Baily, 1975; Kumar *et al.*, 1982). Obviously, the lack of testosterone effect by castration resulted in poor development in these androgen-dependent organs.

Measurement of urethral CSA is clinically important to elucidate a possible etiological link between the common practice of early age castration and obstructive urolithiasis in feedlot lambs. The PrF, DsF and GsP are the commonest sites for calculi obstruction (Oheme & Tillman, 1965).

In this study, urethral CSA was determined at these regions. We found that the urethral CSA at PrF, DsF and GsP sites in lambs castrated at an early age were significantly lower than those in lambs castrated at 5 months of age and in uncastrated lambs. This effect makes these sites

predisposed to potential obstruction by urinary calculi.

In conclusion, the effect of castration on penile and urethral development in Awassi lambs has been documented in this study. The exact mechanism by which testosterone and androgen receptors regulate the development of these organs needs further investigation. In addition, the link between clinical obstructive urolithiasis and the common practice of early age castration in ruminants warrants more controlled studies to compare the incidence of urethral obstruction by calculi between castrated and uncastrated animals.

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