www.czasopisma.pan.pl PAN www.journals.pan.pl
POLSKA AKADEMIA NAUK

Polish Journal of Veterinary Sciences Vol. 22, No. 1 (2019), 127–132

DOI 10.24425/pjvs.2019.127079

Original article

Comparative ultrasonographic examination and measurements of the urethra and penis of castrated and intact male lambs

M. Sickinger[1], A. Allugami[1], K. von Pückler[2], K. Failing[3], A. Wehrend[1]

[1] Clinic for Obstetrics, Gynecology and Andrology of Large and Small Animals with Ambulatory Services, Frankfurter Str. 106, 35392 Giessen, Faculty of Veterinary Medicine, Justus-Liebig-University Giessen, Germany
[2] Clinic for Small Animals, Department of Surgery, Frankfurter Str. 108, 35392 Giessen, Faculty of Veterinary Medicine, Justus-Liebig-University Giessen, Germany
[3] Unit for Biomathematics and Data Processing, Frankfurter Str. 95, 35392 Giessen, Faculty of Veterinary Medicine, Justus-Liebig-University Giessen, Germany

Abstract

Early castration of male small ruminants is regarded as a risk factor for urolithiasis, although the underlying correlations are still unclear. One possible reason is a deferred development of the penis and the urethra after castration. Therefore, we examined the penis and urethra of castrated and intact lambs by ultrasonography to determine the correlation between urethral area and penile cross-sectional area. Ultrasonography was performed in 6-month-old Lacaune crossbred lambs (early castrated, late castrated, and intact; each group, n = 11). Sectional images at 5 locations (glans penis, penile urethra, distal and proximal sigmoid flexure, and ischial arch) were obtained to determine the urethral and penile diameters. Urethral and penile cross-sectional areas were calculated. Grey-scale analysis of ultrasound images was performed to evaluate possible differences in the penile texture between the groups. Correlation analyses between both cross-sectional areas showed a significant general correlation for location 2 in all lambs ($R = 0.52$; $P = 0.003$), for location 3 in late-castrated lambs, and for location 5 in early-castrated lambs. Statistically significant correlations between the penile and the urethral area of castrated and intact lambs were not evident. Therefore, measurement of the penile cross-sectional area alone does not allow for accurate estimation of urethral size. Statistically significant differences concerning the grey-scale analysis between the groups were also not detectable.

Thus, simplification of the formerly presented ultrasonographic examination of the urethra is not recommended. In animals at a risk of obstructive urolithiasis, complete urethral examination is essential.

Key words: lamb, morphometry, lower urogenital tract, ultrasonography, urethra

Correspondence to: M. Sickinger, e-mail: marlene.sickinger@vetmed.uni-giessen.de

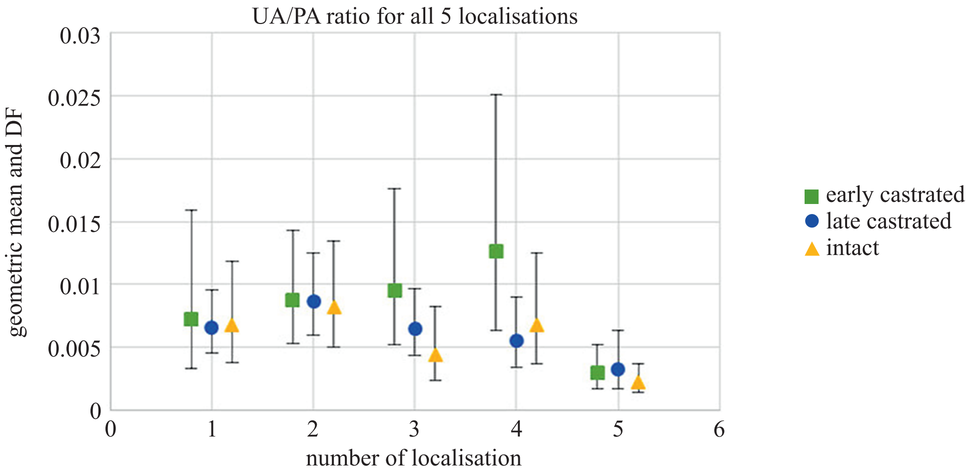

Fig. 1. Comparison of urethral area (UA) and penile area (PA) between the groups. Presentation of geometric means and dispersion factors (DF) of UA/PA ratios for the different examination groups and all 5 locations (glans penis, penile urethra, distal and proximal sigmoid flexure and ischial arch of the penis).

Introduction

Castrated ruminants are presumed to have a higher risk of obstructive urolithiasis, although mature breeding males also experience this condition (Oehme and Tillmann 1965). Obviously, the difference between castrated and intact males is the lack of testosterone, with the implicated consequences of hypoplastic development of the penis and accessory sex glands (Belonje 1965, Kumar et al. 1982). Only a few studies have investigated the postnatal development of the size, length, and diameter of the penis and urethra in ruminants (Marsh and Safford 1957, Belonje 1965, Kumar et al. 1982, Bani Ismail et al. 2007), using mainly histological preparations and measurements in tissue preparations that were harvested during slaughtering (Marsh and Safford 1957, Belonje 1965, Kumar et al. 1982, Bani Ismail et al. 2007). In contrast, the objective of our study was to determine whether the results of these *in vitro* studies can be generalized to living animals via ultrasonographic measurements of urethral and penile cross-sections. Our study investigated the possible relationship between the penis and urethral lumen size, with the ultimate goal of establishing a simplified urethral examination protocol (AlLugami et al. 2017). If a significant correlation exists between penile and urethral cross-sectional areas, the measurement of the penile cross-sectional area at different locations would suffice for an indirect estimation of the urethral size. For application in the field, this approach would be far more practical than the direct measurement of the urethra and could be used as a simple method for the evaluation of the risk to develop obstructive urolithiasis.

Materials and Methods

Sonographic cross-sectional images of the penises of 33 Lacaune-crossbred fattening lambs were acquired for morphometric analysis. Animals were castrated (n = 11; castrated at 3-4 weeks of age and n = 11 castrated at 4 months of age) or intact (n = 11). The corresponding examinations were performed in accordance with the German National Ethical Guidelines of Animal Welfare. The analysis encompassed determination of the urethral luminal area (hereafter, UA) and penile cross-sectional area (hereafter, PA). Sonographic images were obtained from a descriptive study on the performance of sonography in the lower urinary tract of lambs. The sonographic technique used here was described previously (AlLugami et al. 2017). Briefly, transverse sectional images at 5 locations - glans penis, penile urethra, distal and proximal sigmoid flexure, and ischial arch - were obtained to determine the urethral and penile diameters (transverse and sagittal), and sonographic images were obtained. To evaluate the size of the urethral lumen in relation to the size of the penis, horizontal and vertical diameters of the urethra and penis were measured, respectively. The UA and PA were calculated, and the PA/UA ratio was determined. Measurements were performed using the measuring tool of the veterinary documentation software easyVET (IFS Informationssysteme GmbH, Hannover, Germany). Grey-scale analyses were performed using the software program Adobe Photoshop (Adobe Systems Software Ireland Limited, Dublin, Ireland). Within each ultrasonographic picture two nonoverlapping regions of interest (ROI) of 0.125 cm^2 were analysed. The first ROI was chosen directly next to the urethra and the second one within the tissue of the corpus spongiosum.

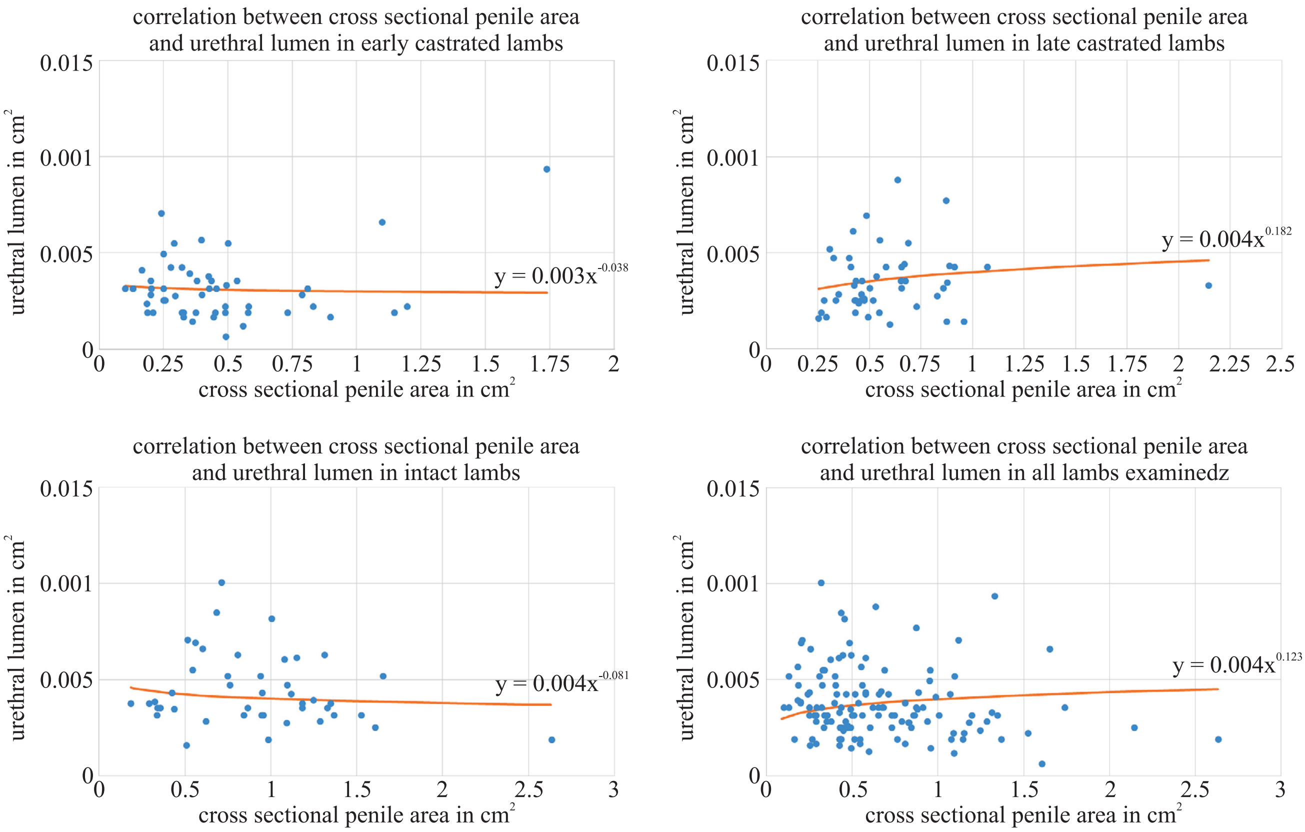

Fig. 2. Diagrams of correlation analysis. Scatter plots with regression lines for the correlation between penis size and urethra size for all groups (top left) and for individual groups (top right to bottom right).

Statistical analyses consisted of a simple data description with calculation of the ratio of UA/PA, as well as that of the mean diameters of the urethra (mean DU) and penis (mean DP). For all analyses, data were logarithmically transformed because the statistical distributions of the generated values were skewed to the right. After the transformation, the distributions were nearly normal. Subsequently, correlation analyses for UA versus PA in all lambs, all locations together, and single locations were calculated. Data from the grey-scale analysis were examined using a two-way ANOVA with repeated measures concerning the parameter localisation to determine possible differences in the grey-scales of penile tissue in respect to the castration status. Statistical analyses were performed using the BMDP Statistical Software package (University of California Los Angeles, Los Angeles, CA, USA) (Dixon 1992).

Results

Geometric means and dispersion factors (geometric standard deviations) for all examination groups and locations are shown in Fig. 1.

In general, no statistically significant correlation was observed between the PA and UA ($R = 0.16$; $P = 0.05$) in all tested lambs and locations. However, in all groups, location 2 (the penile urethra) was an exception ($R = 0.52$; $P = 0.003$).

Separate analyses of the five locations revealed two statistically significant correlations. There were significant correlations for location 3 in late-castrated lambs and for location 5 in early-castrated lambs. Details, including all P values, are shown in Tables 1 and 2, as well as in Fig. 2. An exemplary overview of the calculated mean cross sectional areas for PA and UA is given in Table 3.

The measured mean grey-scale values ranged between 57.3 ± 15.0 ($\xi \pm s$) at localisation 4 and 65.2 ± 18.0 at localisation 2. No statistically significant differences between the groups ($P = 0.53$) nor the localisations ($P = 0.06$) could be determined.

Discussion

Studies on the clinical evaluation of obstructive urolithiasis, including therapeutic measurements, have often reported an overrepresented percentage of castrated animals (Oehme and Tillmann 1965, Haven et al. 1993, Ewoldt et al. 2006, Duehlmeier et al. 2007).

Table 1. Results of the correlation analysis for penile and urethral cross-sectional areas for all locations. P values and regression coefficients (R) are shown.

Correlation PA/UA	P	R
All groups	0.05	0.16
Early	0.73	-0.05
Late	0.25	0.17
Intact	0.47	-0.11

Table 2. Results of the correlation analysis for penile and urethral cross-sectional areas following separate evaluations of individual locations (Loc.). P values and regression coefficients (R) are shown.

Correlation PA/UA	Loc. 1 P/R	Loc. 2 P/R	Loc. 3 P/R	Loc. 4 P/R	Loc. 5 P/R
All groups	0.22/0.22	0.003/0.52	0.34/0.18	0.27/0.23	0.07/0.33
Early	0.29/-0.35	0.74/0.11	0.1/-0.54	0.61/-0.19	0.04/0.62
Late	0.7/0.14	0.21/0.43	0.02/0.73	0.42/0.3	0.73/0.12
Intact	0.25/0.4	0.62/0.18	0.54/-0.25	0.46/0.38	0.22/-0.42

Table 3. Overview of the mean penile and urethral cross-sectional areas for the examination groups with separate evaluation of individual locations (Loc.).

Mean UA [cm^2]	Loc. 1	Loc. 2	Loc. 3	Loc. 4	Loc. 5
All groups	0.0036	0.0043	0.0035	0.0033	0.0034
Early	0.0031	0.0031	0.0027	0.0029	0.0032
Late	0.0034	0.0038	0.0035	0.0031	0.0035
Intact	0.0043	0.0059	0.0041	0.0040	0.0035
Mean PA [cm^2]	Loc. 1	Loc. 2	Loc. 3	Loc. 4	Loc. 5
All groups	0.53	0.50	0.54	0.49	1.13
Early	0.38	0.37	0.32	0.26	0.93
Late	0.52	0.43	0.48	0.58	0.96
Intact	0.70	0.72	0.86	0.69	1.52

In contrast, other studies have referred to urolithiasis as a disease of castrated and intact males (Van Metre et al. 1996) and assumed that anatomical factors (i.e., the sigmoid flexure and urethral process) were responsible for this disease. As the development of the urogenital tract, and particularly the penis, are testosterone-dependent (Belonje 1965), the assumption that those anatomical factors are of importance for the development of obstructive urolithiasis, especially in castrated animals, seems to be self-evident. Therefore, studies on calves, sheeps, and goats have examined the penile and urethral size in early- and late-castrated animals in comparison to uncastrated controls (Marsh and Safford 1957, Kumar et al. 1982, Bani Ismail et al. 2007). The results of these studies indicate that castrated animals have smaller penises and narrower urethras. The ultrasonographic examination of the urethra in lambs was the objective of a previous study where we demonstrated that visualization of the urethra in its entire course is possible using high-resolution ultrasound probes (AlLugami et al. 2017). Because ultrasound probes commonly used in the field use lower frequencies, the purpose of this study was to evaluate a possible simplification of this formerly presented technique for its use in the field.

Although there has been no previous correlation analysis, former studies have indicated a possible relationship between the penile and urethral sizes. Bani Ismail et al. examined urethral and penile size in sheep and found that the penis and urethra were smaller in castrated animals than in uncastrated controls (Bani Ismail et al. 2007). They also compared the diameter of the penis measured at slaughter by a digital micrometre calliper and the urethral cross-sectional area measured by histomorphometry. However, in our study, ultrasonography was performed in living animals for determining the penile and urethral size. Our evaluation supposes that the results might be somewhat less prone to artifacts because of the unavoidable shrinking effects that occur during tissue sample fixation for histomorphometry. Nevertheless, our results did not show persistent significant correlations, although we must consider that our analysis resulted in significant P values for locations 2, 3, and 5. Sonourethrography is a more precise method to measure the diameter and examine the patency of the urethra (Chen et al. 2017). This method is used in human medicine, where it is referred to as a gold standard in this respect. Because wilful voiding is necessary for this examination technique, its application is not possible in veterinary medicine. As discussed previously (AlLugami et al. 2017), some other examination techniques, including contrast ultrasonography or radiography (Palmer et al. 1998, Poulsen Nautrup 2007), are also not suited for use in small ruminants. Therefore, we opted to perform plain ultrasonography in our study.

Critical discussion of the present results must consider how testosterone influences the postnatal development of the penis and urethra. Cannizzo et al. (2007) showed no effect of boldenone, an anabolic steroid similar to testosterone, on the epithelial cells of the prostatic urethra in calves. Thus, internal narrowing of the urethra by a hypertrophic urothelium could not be seen. As the postnatal development of the penis begins at puberty, there is general truth to the fact that, in early castrated animals, the penis and urethra remain infantile (Belonje 1965). Belonje (1965) describes a testosterone-dependent release of the infantile adhesion between the prepuce and glans penis in ruminants. According to this study, the age of the animals at the time of castration is not as important as their weight and achieved maturity of the penis. This could account for the lack of significant correlations across all locations in this study. On one hand, there seems to be an allometric growth development of the penis and urethra, as has been demonstrated for the development of mammary parenchyma in prepubertal Holstein heifers (Esselburn et al. 2015). Conversely, the interindividual differences within location groups could account for the observed variable significance in correlation between the penile and urethral size. This effect might have been less prominent if the number of animals in our study was greater. Another limitation of the present study is the grouping criteria. For our previous study, animals had to be grouped by age without accounting for body weight. During the course of that study, regrouping was not performed. Considering Belonje's results (1965), the present findings might be different in terms of a far clearer correlation between urethral lumen and penis size. These considerations should be addressed in future studies.

Nevertheless, according to our data, the anatomical and histological tissue texture of the penis does not differ between castrated and intact male lambs. The grey-scale analysis in our study failed to show statistically significant differences. In contrast to this, similar examinations in dogs could show such differences even though these differences were only present at one localisation and concerning one of the examined parameters (Goericke-Pesch et al. 2013).

In conclusion and based on the present data, measurement of the PA alone is insufficient for evaluating the size or diameter of the urethra in lambs, particularly in relation to the risk of obstructive urolithiasis. A complete examination of the urethra along its entire course, as described previously (AlLugami et al. 2017), should be performed.

Grey-scale analysis of the penile tissue failed to show differences between the penile tissue texture in respect to castration status. The effect of an early castration remains to be further discussed.

Acknowledgements

The authors thank Prof. Dr. Dr. h.c. Martin Kramer for the use of the facilities in the Clinic for Small Animals (Department of Surgery) of the Justus-Liebig-University of Giessen.

References

AlLugami A, von Pückler K, Wehrend A, Sickinger M (**2017**) Sonography of the distal urethra in lambs. Acta Vet Scand 59: 16.

Bani Ismail ZA, Al-Zghoul MF, Al-Majali AM, Khraim NM (**2007**) Effects of castration on penile and urethral development in awassi lambs. Bulg J Vet Med 10: 29-34.

Belonje PC (**1965**) Observations on the post natal development of the penis in Merino ram Iambs and wethers: the possible relationship to the passage of urinary calculi. J South Afr Vet Med Assoc 36: 381-383.

Cannizzo FT, Zancanaro G, Spada F, Mulasso C, Biolatti B (**2007**) Pathology of the testicle and sex accessory glands following the administration of boldenone and boldione as growth promoters in veal calves. J Vet Med Sci 69: 1109-1116.

Chen L, Feng C, Lv XG, Fan HH, Joshi P, Barbagli G, Zhao YM, Xu YM, Li Z, Hu B, Fu Q (**2017**) Three-Dimensional Computerized Model Based on the Sonourethrogram: A Novel Technique for Evaluating the Anterior Urethral Stricture. J Urol 199: 568-575.

Dixon WJ (**1992**) BMDP Statistical Software Manual, University of California Press, Berkeley, Los Angeles, Oxford.

Dühlmeier R, Zibell G, von Altrock A, Roth C, Schroeder C, Thies K, Ganter M (**2007**) Urolithiasis in small ruminants - methods of treatment and recovery. Tierärztl Prax 35: 175-182.

Esselburn KM, Hill TM, Bateman HG, Fluharty FL, Moeller SJ, O'Diam KM, Daniels KM (**2015**) Examination of weekly mammary parenchymal area by ultrasound, mammary mass, and composition in Holstein heifers reared on 1 of 3 diets from birth to 2 months of age. J Dairy Sci 98: 5280-5293.

Ewoldt JM, Anderson DE, Miesner MD, Saville WJ (**2006**) Short- and long-term outcome and factors predicting survival after surgical tube cystostomy for treatment of obstructive urolithiasis in small ruminants. Vet Surg 35: 417-422.

Goericke-Pesch S, Hoelscher C, Failing K, Wehrend A (**2013**) Functional anatomy and ultrasound examination of the canine penis. Theriogenology 80: 24-33.

Haven ML, Bowman KF, Engelbert TA, Blikslager AT (**1993**) Surgical management of urolithiasis in small ruminants. Cornell Vet 83: 47-55.

Kumar R, Kumar A, Singh H, Singh B, Prakash P (**1982**) Effect of castration on urethra and accessory sex glands in goats. Indian Vet J 59: 304-308.

Marsh H, Safford JW (**1957**) Effect of deferred castration on urethral development in calves. J Am Vet Med Assoc 130: 342-344.

Oehme FW, Tillmann H (**1965**) Diagnosis and treatment of ruminant urolithiasis. J Am Vet Med Assoc 147: 1331-1339.

Palmer JL, Dykes NL, Love K, Fubini SL (**1998**) Contrast radiography of the lower urinary tract in the management of obstructive urolithiasis in small ruminants and swine. Vet Radiol Ultrasound 39: 175-180.

Poulsen Nautrup C (**2007**) Harnblase und Harnröhre. In: Poulsen Nautrup C, Tobias R (eds) Atlas und Lehrbuch der Ultraschalldiagnostik bei Hund und Katze, 4th ed., Schlütersche, Hannover, pp 237-239.

Van Metre DC, House JK, Smith BP, George LW, Angelos SM, Angelos JA, Fecteau G (**1996**) Obstructive urolithiasis in ruminants: Medical treatment and urethral surgery. Comp Contin Educ Pract 18: 317-327.