

## The Effect of Prepubertal Castration on Wool Diameter and Blood Testosterone in Ghezel Breed

### Research Article

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### ABSTRACT

Wool growth depends on hormones activity. Owing to hormonal status is extensively affected by surgical excision of endocrine glands, disease or severe congenital abnormalities, wool growth will be altered. This study intends to assess the effect of prepubertal castration at different ages on wool traits and testosterone with draw in male lambs. One month of age Ghezel male lambs (n=20) were selected and allotted in 5 groups (n=4 lambs per group), 4 groups (1, 2, 3, 4 month ages) were surgically castrated, while the fifth group served as control. All animal were fed with the same ration for 240 days. At 9-month of age, hair samples were taken from the mid-side and shoulder regions and analyzed by image analysis. Blood samples were taken from all animals from 1 month of age to 9 month monthly to measure plasma levels of testosterone. The average fiber diameter in castrated lambs was lower compared with intact males (35.21  $\mu\text{m}$  vs. 36.40  $\mu\text{m}$ ;  $P<0.05$ ). Also, 4-month castrated lambs showed significant differences in the average diameter of wool fiber in comparison to the other castrated groups ( $P<0.05$ ). The plasma level of testosterone in castrated lambs remained in basal levels (0.51 ng/mL), contrasting to values recorded in intact male lambs at 9-month of age (2.33 ng/mL;  $P<0.05$ ). The diameter of fibers was strongly correlated ( $P<0.05$ ) with testosterone plasma concentration ( $r=0.94$ ) in castrated and in intact lambs ( $r=0.86$ ). Therefore, it is concluded that testosterone concentration affects the diameter of wool.

**KEY WORDS** castration, sheep, testosterone, wool.

### INTRODUCTION

The Ghezel breed is a common native sheep breed in Iran. It is a medium-sized breed, is considered as the best for feeding under the traditional farm-sheep condition. Although, meat, wool, skin and milk are valuable products of this sheep, the main purpose of the Ghezel sheep is meat and milk production. The wool color of this sheep varies from light to dark brown (the wool in legs is usually darker than in the body) (Nabavi *et al.* 2014; Nazari-Zenouz *et al.* 2016). Ghezel sheep breed is double-coated, and produce carpet wool, which contains some proportion of hollow or

medullated coarse and resilient hair, and Kemp fibers; the wool is used in the manufacture of hand-made carpets (Ansari-Renani, 2013). In sheep, body weight (McGregor and Butler, 2016), nutritional supplementation (McGregor and Tucker, 2010), photoperiod (Abecia *et al.* 2005), gender (Sinha and Singh, 1997; Khan *et al.* 2012) and hormonal treatments (McDowall *et al.* 2011; McDowall *et al.* 2013) influence the quality of wool. Wool is a valuable by-product of finished lamb; consequently, it is important to know how the wool is affected by physiological and hormonal treatments used to influence body gain. It has been shown that there are significant gender and seasonal differ-

ences in the annual wool production. In the male, new fibers begin to grow one month earlier than in ewes (Kendall, 1999). Also, female lambs always produce coarser wool compared with male lambs (McGregor and Butler, 2016). These characteristics would be due to the rapider growth of males compared to females, which may be influenced by nutrition and hormones. Furthermore, intact rams produce more wool than the castrated or ewes, mainly due to their greater size and the better feeding given to breeder rams (Khan *et al.* 2012). Differences between castrated ram and ewes, probably the effects of status of reproduction are in ewes (Khan *et al.* 2012). Recent research has shown that certain hormones have an important influence on the growth of wool, although it is evident that the relationship is complex. The secretions of the anterior pituitary are essential for normal wool production, as indicated by hypophysectomy which will lead to a severe depression of wool growth. Injected pituitary growth hormone stimulated wool growth in intact sheep, but failed to restore wool growth in hypophysectomized sheep (Slen and Connell, 1960; Yeates *et al.* 2013). In male hamsters, testosterone was shown to be essential for the complete change to the winter coat, demonstrating the importance of hormonal interactions on the fiber growth process (Montenegro, 2003).

The objective of the present study was first to assess the effects of androgens on wool characteristics, comparing the wool from males castrated at different ages with the one originated from intact males, and to test an eventual correlation between the wool characteristics and the testosterone blood levels.

## MATERIALS AND METHODS

Twenty Ghezel male lambs from the experimental farm of the Agriculture Faculty at University of Tabriz (38° 05'N, 46° 28'W), in East Azerbaijan (Iran), were randomly selected. All male lambs born in winter (December-March 2016) were housed in outdoor pens, and randomly distributed in 5 groups of 4 animals each, as follows: Control group – one-month-old male lambs; Gp 1) one-month-old castrated lambs; Gp 2) two-months-old castrated lambs; Gp 3) three-months-old castrated lambs and Gp 4) four-months old castrated lambs.

The surgical castration was performed by a veterinarian following the international guidelines for the ethical treatment of animals. After disinfection of the place the lambs become sedation by injection of 0.5 xylazine and by the use of lidocaine the operation place was numbed. Then by a linear cutting scrotal layer were cut and the testis were separated from testicular sac. The cuttings were stitched carefully and general antibiotic and flunixin meglumine were injected to the lambs for five days. Before weaning,

lambs were fed a diet containing 14.7% crude protein, after weaning lambs were offered alfalfa hay and a concentrate with 14% crude protein until the age of 9 months.

## Body weight and wool production

Body weight was assessed at 9 months of age in all of the animals. In all the groups, samples of wool were taken from the mid-side and shoulder regions at 9-month of age. The wool fiber diameter was measured, at 500 × magnification, using a projection microscope, as described by (Sherlock *et al.* 2001). Wool traits measurements included fibre diameter mean (FD Mean), fibre diameter standard deviation (FD SD) and coefficient of variation of diameter (CVD). The coefficient of variation of diameter (CVD) was calculated by dividing the standard deviation (SD) of the means of fibre diameter (MFD) by the means of fibre diameter.

## Blood testosterone concentration

Blood samples were obtained monthly from the external jugular vein, to determine plasma testosterone levels from 1 month of age to adulthood. Blood samples were centrifuged 430 × g for 5 min, and serum was decanted and stored at – 20 °C until assayed (Nazari-Zenouz *et al.* 2016). Testosterone was measured by a ELISA using a commercial kit (Monobind Inc. Lake Forest, CA 92630, USA). The intra-assay CV was 4.8% and the assay sensitivity was 0.576 ng/mL.

## Statistical analysis

Data for testosterone concentration, body weight and wool diameter were analyzed by completely randomized design, using the general linear method (GLM) procedure of SAS (SAS, 2004), including in the model the age at castration groups. If the P value was significant, differences among means were evaluated by post hoc HSD (Tukey, Alpha=0.05) test. Data is expressed as mean (mean) and standard error of the mean (SEM). The coefficient of variation for the fiber diameter (CVD) was also estimated as the quotient between the standard deviation (SD) and the average fiber diameter (MFD).

## RESULTS AND DISCUSSION

Table 1 summarizes the data for the group body weight, testosterone concentration and fiber diameter. The average body weight at 9-months of age was higher in intact lambs (46.12±0.58 kg) than in castrated lambs (41.60±0.29 kg) (P<0.05). The plasma testosterone levels were very lower in 9-month-old castrated lambs (0.51±SD ng/mL), compared to age matched intact lambs (2.33 ng/mL). At 4 months of age, the testosterone level starts raising in intact lambs, reaching 1.69 ng/mL; from 8 to 9 months of age the testosterone levels increased again.

**Table 1** Descriptive statistics for body weight, testosterone concentration and wool morphometry in 9 months-old castrated and intact lambs (Mean±SEM)

Item	n	Body weight (kg)	Testosterone (ng/mL)	Wool fiber		
				Overall	Shoulder	Mid-side
Intact lambs	4	46.12±0.58 <sup>a</sup>	2.33±0.05 <sup>a</sup>	36.40±0.60 <sup>a</sup> [0.51]	34.07±0.85	38.73±0.85 <sup>a</sup>
Castrated lambs	16	41.60±0.29 <sup>b</sup>	0.4±0.04 <sup>b</sup>	35.21±0.34 <sup>b</sup> [0.50]	33.68±0.47	36.75±0.50 <sup>b</sup>

The means within the same column with at least one common letter, do not have significant difference ( $P>0.05$ ). SEM: standard error of the means.

Until the 4 months of age, there were no significant differences in plasma testosterone levels among castrated and intact lambs.

The average fiber diameter in shoulder and mid-side were lower in castrated lambs (36.75 and 33.68  $\mu\text{m}$ , respectively) than in intact lambs (38.73 and 34.07  $\mu\text{m}$ , respectively). Average body weight, testosterone concentration and wool trait in the groups of castrated lambs at 9 months of age are shown in Table 2. The body weight did not differ among all of the castrated groups ( $P<0.05$ ). Testosterone concentrations at castration were higher in 4 month-old castrated lambs than in the other castrated groups ( $P<0.05$ ). Also, the diameter of fiber, shoulder fiber diameter and mid-side fiber diameter in 4-month-old castrated lambs were higher than others castrated lambs ( $P<0.05$ ) (Figure 1).

Several centuries ago in England, castration of ram lambs to produce male ram was performed as a technique for selective breeding and as part of wool production policy (Davis, 2000). Wool is an important by-product of lamb production and it is of considerable practicality as well as fundamental interest to know what effect of the sex hormones can have on wool quality (Montenegro, 2003). In the fetus, the primary wool follicles develop at approximately pregnancy day 60, while secondary follicles develop around day 80, then start to branch from day 100 until early post-natal life.

During this period, the follicle development can be adversely affected by gender. It has been shown that the growth hormone was not the only pituitary secretion-necessary for normal wool growth (Corbett, 1979). It is well known that the parenteral or oral administration of testosterone substances stimulate body growth in sheep (Davari *et al.* 2012; Nazari Zenouz *et al.* 2014). The metabolic status – determined by hormonal dependence and experimental endocrine manipulation, causes large changes in the wool growth and wool traits. In this study the effect of the plasma testosterone levels on wool diameter in prepubertal male lambs was investigated. Rams tend to produce more wool than wethers and ewes, which might be mainly to their greater size and the better feeding given to breeder rams.

Differences between wethers and ewes, once the effects of reproduction in ewes are allowed for, are probably small (Khan *et al.* 2012). In this study the final body weight in male lambs was heavier than castrated lambs, but there was no difference among the study groups. It was tried to eliminate the effect of testosterone on fiber diameter on the effect of body weight. Accordingly, it was used the different groups of the castration lambs. The results showed that testosterone affects the diameter of the wool. In male lambs, the diameter of fibers was higher than castrated lambs at the end of 9-month of age. These results agree with previous studies showing that, in Merino wethers, testosterone implants increased the wool fiber diameter (Southcott and Royal, 1971).

The fiber diameter in 4-months-old castrated lambs was higher than other castrated groups although the body weight did not differ among animals in the castrated groups. Comparison of the wool diameter in castrated groups showed a positive correlation between testosterone and the wool diameter: whenever there was a significant increase in testosterone the wool diameter also increased. Correlations in fibre diameter with plasma testosterone were recorded in castrated ( $r=0.94$ ) and also intact male ( $r=0.86$ ) lambs. Castration of lambs in early prepubertal period could reduce the fiber diameter for 2-3  $\mu\text{m}$ . The advantage of using CVD to describe diameter distribution, rather than SD, is that to a first-order approximation, it is not as dependent on MDF (Cottle and Baxter, 2015).

Although the wool fiber CVD in 4 months of age castrated lambs was higher than other castrated lambs, this difference was not significant. The current study showed that the diameter of wool related to testosterone concentration. The functions of testosterone can be broadly classified into two groups.

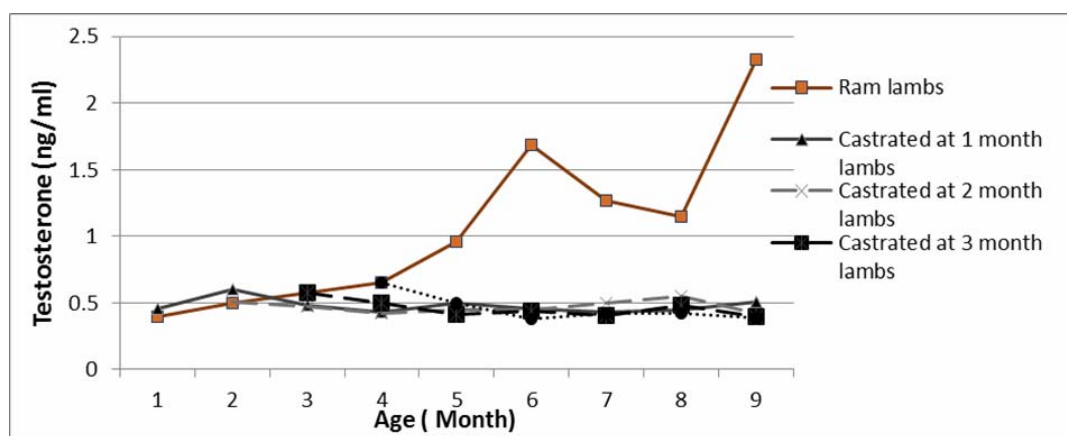
The first one is the androgenic function which includes the development of secondary male characteristics. The second one is the anabolic function which involves the promotion of protein synthesis (Sanni *et al.* 2012). All studies (Kendall, 1999; Nixon *et al.* 2002; Rogers, 2006; Yu *et al.* 2008; Galbraith, 2010) have concluded that increase in plasma insulin-like growth factor I (IGF-I) and thyroid hormone had no quantitative effect on wool traits.

**Table 2** Means of body weight, testosterone concentration and wool traits in castrated lambs at 9 months of age (Mean±SEM)

Variation	Body weight (kg)	Testosterone (ng/mL) at castration	Diameter of fibre (µm)	Coefficient of variation (CV)	Standard deviation (SD)	Shoulder fiber diameter (µm)	Mid-side fiber diameter (µm)
Castrated at 1 month of age	41.60±0.29	0.46±0.04 <sup>a</sup>	34.91±0.34 <sup>a</sup>	0.50	17.90 <sup>a</sup>	33.44±0.47 <sup>a</sup>	36.21±0.50 <sup>a</sup>
Castrated at 2 month of age	41.20±0.79	0.50±0.06 <sup>a</sup>	35.09±0.44 <sup>a</sup>	0.49	17.68 <sup>a</sup>	33.52±0.32 <sup>ab</sup>	36.44±0.19 <sup>a</sup>
Castrated at 3 month of age	40.28±0.60	0.58±0.09 <sup>a</sup>	35.17±0.21 <sup>a</sup>	0.48	17.83 <sup>a</sup>	33.61±0.26 <sup>bc</sup>	36.64±0.35 <sup>a</sup>
Castrated at 4 month of age	40.96±0.42	0.65±0.08 <sup>b</sup>	35.70±0.33 <sup>b</sup>	0.52	18.33 <sup>b</sup>	33.96±0.40 <sup>d</sup>	37.73±0.28 <sup>b</sup>

The means within the same column with at least one common letter, do not have significant difference ( $P>0.05$ ).

SEM: standard error of the means.

**Figure 1** Monthly serum testosterone levels in castrated lambs and male lambs during postnatal change

The relationship between testosterone administrations and serum IGF-I levels (Ferrando *et al.* 2002), growth hormone (Davari *et al.* 2012) and thyroid hormone (Vingren *et al.* 2010) has been shown in previous studies. Although it seems that other hormone interaction effect on the wool, but up to now, those effects on the control of wool traits has not known yet clearly.

## CONCLUSION

In conclusion, castration affects the diameter of the wool in the Ghezel male sheep. Testosterone withdrawn associated with testis removal at castration reduces the diameter of the wool.

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## REFERENCES

- Abecia J., Valares J. and Forcada F. (2005). The effect of melatonin treatment on wool growth and thyroxine secretion in sheep. *Small Rumin. Res.* **56**, 265-270.
- Ansari-Renani H. (2013). Fiber quality of Iranian carpet-wool sheep breeds. *Media Peternakan.* **35**(3), 179-184.
- Corbett J. (1979). Variation in wool growth with physiological state. Pp. 79-98 in *Physiological and Environmental Limitations to Wool Growth*. J.L. Black and P.J. Reis, Eds. University of New England Publishing Unit, Armidale, Australia.
- Cottle D. and Baxter B. (2015). Wool metrology research and development to date. *Text. Prog.* **47**, 163-315.
- Davari F., Khazali H., Rokni H. and Fatehi Z. (2012). Effects of testosterone and growth hormone on plasma concentrations of orexin in diet restricted rams. *Iranian J. Endocrinol. Metabol.* **14**, 81-87.
- Davis S.J. (2000). The effect of castration and age on the development of the Shetland sheep skeleton and a metric comparison between bones of males, females and castrates. *J. Archaeol. Sci.* **27**, 373-390.

- Ferrando A.A., Sheffield-Moore M., Yeckel C.W., Gilkison C., Jiang J., Achacosa A., Lieberman S.A., Tipton K., Wolfe R.R. and Urban R.J. (2002). Testosterone administration to older men improves muscle function: molecular and physiological mechanisms. *American J. Physiol. Endocrinol. Metabol.* **282**, 601-607.
- Galbraith H. (2010). Fundamental hair follicle biology and fine fibre production in animals. *Animal*. **4**, 1490-1509.
- Kendall P.E. (1999). Prolactin and wool growth in the Romney ewe: a thesis presented in partial fulfilment of the requirements. Ph D. Thesis. Massey Univ., Palmerston North, New Zealand.
- Khan M.J., Abbas A., Ayaz M., Naeem M., Akhter M.S. and Soomro M.H. (2012). Factors affecting wool quality and quantity in sheep. *African J. Biotechnol.* **11**, 13761-13766.
- McDowall M., Edwards N. and Hynd P. (2011). The effects of short-term manipulation of thyroid hormone status coinciding with primary wool follicle development on fleece characteristics in Merino sheep. *Animal*. **5**, 1406-1413.
- McDowall M., Watson-Haigh N., Edwards N., Kadarmideen H., Natrass G., McGrice H. and Hynd P. (2013). Transient treatment of pregnant Merino ewes with modulators of cortisol biosynthesis coinciding with primary wool follicle initiation alters lifetime wool growth. *Anim. Prod. Sci.* **53**, 1101-1111.
- McGregor B. and Butler K. (2016). Coarser wool is not a necessary consequence of sheep aging: Allometric relationship between fibre diameter and fleece-free liveweight of Saxon Merino sheep. *Animal*. **10**(12), 2051-2060.
- McGregor B. and Tucker D. (2010). Effects of nutrition and origin on the amino acid, grease, and suint composition and color of cashmere and guard hairs. *J. Appl. Polymer Sci.* **117**, 409-420.
- Montenegro R. (2003). The effects of prolactin on prolactin receptor gene expression and wool growth in Romney ewes. Ph D. Thesis. Massey Univ., Palmerston North, New Zealand.
- Nabavi R., Alijani S., Taghizadeh A., Rafat S.A. and Bohlouli M. (2014). Genetic study of reproductive traits in Iranian native Ghezel sheep using Bayesian approach. *Small Rumin. Res.* **120**, 189-195.
- Nazari-Zenouz F., Moghaddam G., Hamidiam G., Ashrafi J., Rafat S. and Qasemi-Panahi B. (2016). Postnatal testicular development and testosterone changes in Ghezel ram lambs. *Small Rumin. Res.* **141**, 70-76.
- Nazari Zenouz F., Moghaddam G. and Abdi Z. (2014). Weight changes and sexual behavior in ghezel intact and castrated male lambs. *Int. J. Adv. Biol. Biomed. Res.* **2**, 1761-1767.
- Nixon A., Ford C., Wildermoth J., Craven A., Ashby M. and Pearson A. (2002). Regulation of prolactin receptor expression in ovine skin in relation to circulating prolactin and wool follicle growth status. *J. Endocrinol.* **172**, 605-614.
- Rogers G.E. (2006). Biology of the wool follicle: An excursion into a unique tissue interaction system waiting to be re-discovered. *Exp. Dermatol.* **15**, 931-949.
- Sanni A., Arowolo R. and Olayemi F. (2012). Preliminary study on the effect of castration and testosterone replacement on testosterone level in the New Zealand male rabbit. *African J. Biotechnol.* **11**, 10146-10148.
- SAS Institute. (2004). SAS<sup>®</sup>/STAT Software, Release 9.1. SAS Institute, Inc., Cary, NC, USA.
- Sherlock R., Harris P., Lee J., Wickham G., Woods J. and McCutcheon S. (2001). Intake and long-term cysteine supplementation change wool characteristics of Romney sheep. *Australian J. Agric. Res.* **52**, 29-36.
- Sinha N. and Singh S. (1997). Genetic and phenotypic parameters of body weights, average daily gains and first shearing wool yield in Muzaffarnagri sheep. *Small Rumin. Res.* **26**, 21-29.
- Slen S. and Connell R. (1960). Effect of estradiol and testosterone injections and thyroidectomy on wool growth in Shearling Sheep. *Canadian J. Anim. Sci.* **40**, 15-22.
- Southcott W. and Royal W. (1971). Effect of implanted testosterone propionate on the growth and wool production of merino wethers. *Australian J. Agric. Res.* **22**, 271-282.
- Vingren J.L., Kraemer W.J., Ratamess N.A., Anderson J.M., Volek J.S. and Maresh C.M. (2010). Testosterone physiology in resistance exercise and training. *Sports Med.* **40**, 1037-1053.
- Yeates N., Edey T.N. and Hill M.K. (2013). *Animal Science: Reproduction, Climate, Meat, Wool*. Elsevier, Elsevier, Amsterdam, the Netherlands.
- Yu M., Kissling S., Freyschmidt-Paul P., Hoffmann R., Shapiro J. and McElwee K.J. (2008). Interleukin#6 cytokine family member oncostatin M is a hair#follicle#expressed factor with hair growth inhibitory properties. *Exp. Dermatol.* **17**, 12-19.