
SEMEN CHARACTERISTICS AND TESTICULAR MORPHOMETRY OF RABBIT BUCKS FED DIETARY LEVELS OF THYME (*THYMUS VULGARIS*)

Amaefule, B.C., Ugwuoke, K.C., Umeugokwe, P.I., Nnadozie, C.O., Ikeh, N.E., Anizoba, N.W., Ali, L.C., Ugwu, C.M., & Chukwudi, P.

Department of Animal Science, University of Nigeria, Nsukka.

Corresponding author: bright.amaefule@unn.edu.ng Tel: +2348034435785

ABSTRACT

A study was aimed at investigating the semen characteristics and testicular morphometry of rabbit bucks fed dietary levels of thyme (*Thymus vulgaris*). A total of 20 mature (seven months old) rabbit bucks were used for the study. The rabbit bucks were randomly allotted to four experimental diets (5/each) and fed for 8 weeks. The diet were formulated with the inclusion of thyme at 0, 5, 10 and 15g of thyme/kg of feed for T1, T2, T3, and T4 respectively. The Experiment was arranged in a Completely Randomized Design (CRD). Fresh experimental diets and clean water were made available *ad libitum* for the bucks throughout the experimental period. The results indicated that dietary inclusion of thyme significantly ($P<0.05$) affected all semen parameters except semen pH. Semen volume, forward motility, live sperm (%) and abnormal sperm (%) of bucks in T4 and T3 were significantly ($P<0.05$) higher than those in other treatment groups. Results of testicular morphometry revealed that dietary inclusion of thyme resulted in a significant ($P<0.05$) increase in testis weight, testis volume and testis circumference with bucks in T4 and T3 outperforming those in other treatment groups. However, no significant difference ($P>0.05$) was observed in other testicular morphometric indices studied across the treatment groups. From the result, it was concluded that dietary inclusion of thyme at 10g/kg diet improved the semen characteristics and testicular morphometry of rabbit bucks.

Keywords: Rabbits, Thyme, Testes, Semen, Morphometry.

INTRODUCTION

The most obvious limitation to rabbit production in a tropical climate is their susceptibility to heat stress that evokes a series of drastic changes in their biological functions (oxidative stress) which in turn impairs production and reproduction (Jimoh *et al.*, 2021). In recent years, studies have been carried out on the addition of natural antioxidants as nutritional supplements in animal feeding to improve reproductive performance, health and meat quality (Kandeil *et al.*, 2019). Thyme (*T. vulgaris* L.) of the *Lamiaceae* family is one of the most common herbal medicinal plants worldwide. Thyme is widely used in human food to provide a distinct flavor. Thyme contains thymol, carvacrol, steroids, saponins, flavonoids, alkaloids, polyunsaturated fatty acids, vitamins, tannins, resin potassium nitrate, aspartic acid and glutamic acid (Kandeil *et al.*, 2019). These effective ingredients can improve the reproductive and productive performance, nutrient digestibility and immunity of animals as well as decrease the problems of animal diseases and adverse effects of chemical drugs (Abd El-Hack *et al.*, 2016). Due to its antioxidant, antibacterial, antifungal, hypocholesterolemic, and therapeutic characteristics, it is usually used in livestock farms (as an alternative to antibiotics) for enhancing animal productivity and health (El-Sabrout *et al.*, 2023). Placha *et al.* (2013) reported that the supplementation of 0.5 g/kg thyme oil significantly improved the gut integrity and antioxidant status of rabbits. Most of the research on dietary thyme oil supplementation on rabbits (Placha *et al.* 2013; Abdel-Wareth *et al.* 2018) has indicated its role in stimulating feed intake, body weight gain, meat quality, antioxidant status, gut integrity and improving the overall feed conversion ratio. However, there is dearth of information on the effects of thyme leaves on the semen characteristics and testicular morphometry of rabbit bucks. Thus, the present study aimed to clarify the effect of dietary levels of thyme on the semen characteristics and testicular morphometry of rabbit bucks.

MATERIALS AND METHODS

Experimental Animals

The study was carried out at the Rabbit Unit of the Department of Animal Science Teaching and Research Farm, University of Nigeria, Nsukka. Nsukka, Enugu State, South Eastern Nigeria. A total of 20 mature (seven months old) rabbit bucks were used for the study. The rabbit bucks were

randomly allotted to four experimental diets (5/each) for 8 weeks. The diets were formulated with the inclusion of thyme at 0g inclusion, 5g of thyme/kg of feed, 10g of thyme/kg of feed and 15g of thyme/kg of feed for T1, T2, T3, and T4 respectively. The Experiment was arranged in a Completely Randomized Design (CRD). Fresh experimental diets and clean water were made available *ad libitum* for the bucks throughout the experimental period. All the management practices and ethics associated with animal use for experimentation in the University of Nigeria were strictly adhered to.

Semen collection and evaluation

The quality of the semen ejaculates of the sexually mature and healthy male rabbits were assessed as described by El-Desoky *et al.* (2017) and Abdel-Wareth *et al.* (2019). Semen was collected from twenty rabbits (one ejaculate per rabbit) per treatment at the end of 56 days. The ejaculates were collected using an artificial vagina. The volume of each ejaculate was recorded using a graduated collection tube after removal of the gel mass. The assessment of the live, dead, and abnormal sperm was performed by counting 200 sperm cells using an eosin-nigrosine blue staining mixture. The complete or partial purple-stained sperm cells were considered non-viable, whereas the non-stained cells were viable. The percentage of the motile sperm (progressive forward motility %) was performed in several microscopic fields in the semen samples under $\times 100$ magnification using a light microscope with a hot stage and subjectively assessed from 0% to 100%.

Testicular morphometry

After rabbits were euthanized, the testes were collected and immediately covered in ice and was taken to the laboratory and processed the same day. The testes were trimmed off all adhering tissues and fat. The epididymis was carefully removed. Weights of each testis and epididymis were obtained to the nearest milligram using an electronic scale (M-Metlar®). The length and width of testis and the length of the epididymis were measured using a digital Vernier caliper. The volumes of the testes were measured volumetrically using the Archimedes principle of water displacement in a measuring cylinder. Using measuring cylinder, 1000ml of water was measured and the testis was inserted into the water, and the volume of water displaced represented the volume of the testis.

RESULTS AND DISCUSSION

Table 1. Effect of dietary inclusion of thyme on semen characteristics of rabbit bucks.

Parameters	T1	T2	T3	T4	P-value
Semen volume (mL)	0.30±0.00 ^b	0.50±0.10 ^{ab}	1.25±0.15 ^{ab}	1.50±0.30 ^a	0.021*
Forward motility	56.00±1.00 ^b	65.00±0.00 ^a	65.00±0.50 ^a	72.00±2.50 ^a	0.005**
Live sperm (%)	61.50±1.50 ^c	71.00±1.00 ^b	77.00±2.00 ^{ab}	84.00±1.00 ^a	0.002**
Abnormal sperm (%)	32.50±2.50 ^a	21.50±1.50 ^b	19.00±1.00 ^b	13.00±2.00 ^c	0.007**
Semen pH	7.05±0.00	7.09±0.00	7.07±0.00	7.06±0.00	0.056 ^{NS}

*Significant (P<0.05), ** significant (P<0.01), NS – not significant.

^{abc}: means on the same column with different superscripts are significantly different (p≤0.05).

The effects of inclusion of thyme on the semen characteristics of rabbit bucks is presented in Table 1 above. The results depicts there were significant (p<0.05) differences in semen volume, forward motility, live sperm and abnormal sperm. Semen volume was highest in T4 (1.50±0.30 mL) while the control (T1) had the lowest volume (0.30±0.00mL). Bucks in T4 (72.00±2.50) recorded the highest forward motility which was similar to those in T3 and T2 while the lowest was observed in T1 (56.00±1.00). Live sperm was highest in T4 (84.00±1.00%) while the T1 (61.50±1.50) recorded the lowest. Abnormal sperm was highest in T1 (32.50±2.50%) and lowest in T4 (13.00±2.00%). There was no significant (p>0.05) difference in semen pH across the various treated group.

The results of the semen characteristics of rabbit fed diet containing thyme showed that the test ingredients have positive effect on the semen quality indices. The values obtain in this research for semen characteristics are within normal ranges of healthy rabbit as reported by (Ajayi *et al.*, 2009). The improvements that occurred in the semen volume, forward motility, and live sperms showed that with an increasing inclusion of thyme, there existed a positive effect on semen quality. Thyme contains antioxidants that can help protect cells from oxidative stress, including the cells involved in sperm production. This explains the high percentage of live sperm, low percentage of abnormal sperm

and increase massive motility of sperm in all group treated with thyme. The results of the current study support the findings of Ahmed *et al.*, (2020) and Ezzat *et al.*, (2020) who reported a significant increase in semen volume forward motility live sperm and abnormal sperm on rabbit supplemented with thyme essential oil.

Table 2. Effect of dietary inclusion of thyme on testicular morphometry of rabbit bucks

Parameters	T1	T2	T3	T4	P-value
Paired testis weight (g)	4.00±0.10 ^b	4.40±0.20 ^b	5.00±0.00 ^{ab}	5.40±0.20 ^a	0.009**
Right testis weight (g)	2.00±0.00 ^b	2.10±0.10 ^b	2.40±0.10 ^{ab}	2.65±0.05 ^a	0.011*
Left testis weight (g)	2.00±0.10 ^c	2.30±0.10 ^b	2.60±0.10 ^{ab}	2.75±0.15 ^a	0.033*
Right testis length (cm)	30.25±0.51	30.90±0.84	34.14±1.55	31.51±0.10	0.132 ^{NS}
Left testis length (cm)	29.52±0.92	29.97±0.65	34.10±1.20	32.39±0.47	0.054 ^{NS}
Paired testis volume (mL)	4.50±0.50 ^c	5.50±0.50 ^b	6.50±0.50 ^a	6.50±0.50 ^a	0.021*
Right testis circumference (cm)	2.90±0.10 ^b	2.90±0.10 ^b	3.15±0.05 ^{ab}	3.70±0.20 ^a	0.029*
Left testis circumference (cm)	2.95±0.50 ^b	3.10±0.00 ^b	3.20±0.20 ^{ab}	3.85±0.15 ^a	0.026*
Right epididymis length (cm)	6.00±1.40	6.70±1.00	6.50±0.50	7.40±0.20	0.735 ^{NS}
Left epididymis length (cm)	6.50±1.00	6.95±1.25	7.40±0.30	7.40±0.10	0.853 ^{NS}
Right epididymis weight (g)	0.60±0.10	0.85±0.50	0.80±0.00	1.60±0.70	0.33 ^{NS}
Left epididymis weight (g)	0.55±0.50	0.75±0.50	0.75±0.50	1.50±0.40	0.100 ^{NS}

*Significant (P<0.05), ** significant (P<0.01), NS – not significant.

^{a,b,c} – Means in the same row with different superscripts are significant at 5% (* P < 0.05.)

The effects of inclusion of thyme on the testicular morphometry of rabbit bucks is presented in Table 2 above. The results showed that there were significant (p<0.05) differences in testis weight and testis circumference. Paired testis weight was highest in T4 (5.40±0.20g) while the control (T1) had the lowest (4.00±0.10). The same trend was observed in right testis and left testis weights. Bucks in T4 (3.70±0.20cm) recorded the highest right testis circumference while those in T1 (2.90±0.10cm) had the lowest. The left testis circumference was highest in T4 (3.85±0.15cm) bucks while those in T1 (2.95±0.50cm) recorded the least value. Testis volume was highest in T4 (6.50±0.50mL) and lowest in T1 (4.50±0.50mL). There were no significant (p>0.05) differences in all other parameters across the various treatment groups.

The results of the testicular morphometry of rabbit fed diet containing thyme showed that the test ingredient had positive effect on the testicular morphometry indices. The values obtained in this research for testicular morphometry are within the range obtained by Olarotimi *et al.* (2015) and Ate *et al.* (2021). The improvements witnessed in testes weight, testis circumference and volume in this study agrees with the reports of Shanoon and Mahdi (2012) that reported that the aqueous extracts of either ginger or thyme produced a significant increase in testis weight. Similar results were obtained by Kandeil *et al.* (2019) on testes weight on rabbit V-line male rabbits fed oral ginger and/or thyme aqueous. Toukala *et al.* (2020) in their study also reported significant increase on paired testis weight, left testis circumference, right testes circumference and testes volume on local rabbit supplemented with thyme.

CONCLUSION

From this study, it was concluded that dietary inclusion of thyme improved the semen characteristics and testicular morphometry of rabbit bucks. The study therefore recommends that 10g thyme/kg diet should be included in the diet of rabbit bucks.

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