SEMEN CHARACTERISTICS OF RABBIT BUCKS SUPPLEMENTED WITH BITTER KOLA (*Garcinia kola*) SEED CRUDE EXTRACT IN WATER

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ABSTRACT
The semen characteristics of rabbit's bucks fed bitter kola seed crude extracts (BKSCE) in drinking water was investigated during a 24–week study conducted at the Rabbitry unit, Livestock Teaching and Research Farm, Joseph Sarwuan Tarka University, Makurdi. Twenty-seven (27) nondescript rabbit bucks, aged 9–10 weeks were assigned randomly to 3 groups of 9 bucks each that was replicated 3 times with 3 bucks per replicate in a Completely Randomized Design. Fresh bitter kola seeds were processed and extracted by macerating 1500 g powder per 10 L water for 24 hours followed by sieving and cold storage until used. The stored extract was reconstituted at 0, 25 and 50% levels respectively and offered daily at a single dose (100 ml) in drinking water. Bucks received the basal diet and 3 levels of the reconstituted extract accordingly. Semen samples were evaluated from 18 bucks (2 per replicate) using standard procedures while data were subjected to General Linear Model using IBM SPSS version 21. Creamy– white semen colour dominated followed by milk–white in accordance with the BKSCE levels. Although, semen pH, volume, mass motility, progressive motility, sperm concentration, sperm count, live sperm, morphology and acrosome integrity respectively were statistically similar (P>0.05), bucks offered 25% BKSCE recorded numerically higher values in all of these seminal traits except semen volume and normal sperm thus indicating its possible androgenic properties. In conclusion, BKSCE has potentials of improving the seminal characteristics of rabbit bucks.
Keywords: Crude extracts, *Garcinia kola*, Micro livestock, Rabbit bucks, Seminal traits

INTRODUCTION

The rabbit is a micro livestock with potentials to provide sustainable dietary animal proteins and incomes to poor households in developing countries like Nigeria with a rapidly growing population. Attributes such as its miniature size, low space requirements, short gestation and generation intervals, high fecundity etc gives the rabbit leverage for increased exploitation. In rabbit production, male rabbits form the basis of reproductive success even as profitability is not only dependent on female fertility but also on bucks (Cherfaoui *et al*., 2013). Semen quality is a major determinant of reproductive performance of adult bucks (Sikiru *et al*., 2017) and heat stress typical of the tropics, adversely affects sperm production and seminal traits due to testicular oxidative damages thus causing infertility problems. From the foregoing, improving seminal traits via dietary supplementation with antioxidants from plants such as bitter kola (*Garcinia kola*) seed is pivotal to enhancing the reproductive efficiency of rabbits. *G. kola* is a tree common to West and Central Africa (Manourova *et al*., 2019) with medicinal uses of all of its parts (Onasanwo and Rotu, 2016). The seed is the most valued (Manourova *et al*., 2019) with antioxidant properties and low anti-nutritive levels (Onyekwelu *et al*., 2015). A few studies exist on the androgenic and pro–seminal properties of bitter kola seed meal (BKSM) in rabbits while literature is too limited on the influences of bitter kola seed crude extracts (BKSCE) on the seminal traits of rabbit bucks. Thus, this study investigated the semen characteristics of rabbit bucks supplemented with BKSCE in drinking water.

MATERIALS AND METHODS

Experimental site

The study was conducted (within January–June, 2024) at the rabbitry unit of the Livestock Teaching and Research Farm, Joseph Sarwuan Tarka University, Makurdi, Benue state, Nigeria. Makurdi falls within latitude 7˚33' 00"N–7˚47' 00"N and longitude 8˚27'00" E–8˚4'00"E.

Sourcing, processing and preparation of bitter kola seed crude extract (BKSCE)

Bitter kola seeds (BKS) were sourced from a local market in Enugu state then processed (sorted, sliced, crisp–dried, dehulled and milled) and the powder stored in an air tight container. Extraction was done by macerating 1500g milled BKS per 10 L of mildly hot water (50–55°C) for 24 hours followed by sieving and refrigeration (4–5°C) until used.

Experimental animals and management

A total of 27 weaned nondescript rabbit bucks, aged 9–10 weeks were randomly assigned after weight equalization to 3 groups having 9 bucks each. Each group was replicated 3 times with 3 bucks per replicate in a Completely Randomized Design. The rabbits were housed singly in galvanized wire and mesh cages under strict management practices and acclimated for 2 weeks prior to start of trial. The BKSCE stock was reconstituted at 0, 25 and 50 %

levels respectively for inclusion in the drinking water of rabbit bucks. The bucks irrespective of their grouping received a formulated basal diet (moisture 5.34%, crude protein 17.06%, crude fibre 10.23%, crude fat 8.52%, ash 19.98%, energy 3530 kcal, lysine 0.94%, methionine 0.60%, calcium 0.52% and phosphorus 0.33%). The basal diet contained maize (25.51%), full fat soya bean (20.00%), soya bean cake (7.94%), brewers dried grain (20.00%), rice offal (22.00%), unrefined soya oil (2.70%), bone ash (0.55%), limestone (0.50%), methionine (0.30%), salt (0.25%) and vitamin premix (0.25%). The reconstituted levels of BKSCE were offered daily as a single dose (100 ml) in drinking water based on estimated drinking rate at 10 % body weight (Brewer and Cruise, 1994; Harcourt Harcourt–Brown, 2002). The study lasted for 24 weeks.

Parameters and data analysis

The seminal characteristics (semen colour, pH and volume, mass motility, progressive motility, sperm concentration, sperm count, live sperm, normal and abnormal sperm, and acrosome integrity) were evaluated from a total of 18 bucks (2 per replicate) according to standard procedures at week 24 (aged 33–34 weeks) using locally improvised artificial vagina and live teaser doe. Data were subjected to General Linear Model using IBM SPSS version 21 while significance was detected at probability of $P<0.05$.

RESULTS AND DISCUSSION

The seminal characteristics of rabbit bucks supplemented with BKSCE in water is presented in table 1. BKSCE did not ($P>0.05$) influence any of the seminal traits studied, although, the 25% BKSCE recorded numerically higher values for all the traits with the exception of semen volume and normal sperm. The semen colour of creamy–white dominated followed by milk–white. Mataveli (2008) opined that rabbit semen was mostly milky–white and creamy–white was the best quality whereas Alvarez *et al*. (2006) opined that milky–white was the best and common semen in rabbits. Semen pH and semen volume in this study did not differ ($P>0.05$) and the latter decreased numerically in order of BKSCE levels which agreed with Iwuji and Herbert (2012) who fed rabbit bucks 0–5% bitter kola seed meal (BKSM). In contrast, Ebenebe *et al*. (2017) observed significant reductions ($P<0.05$) after rabbit bucks were fed 0–7.5% BKSM. In addition, these authors reported lower semen volume values (0.60–0.76 ml and 0.47–0.75 ml respectively) compared to this study (1.00–1.27 ml). Mass motility (63.33–78.33)% and progressive motility (56.67–70.00)% in this study ($P>0.05$) disagreed with the significant ($P<0.05$) findings of Iwuji and Herbert (2012) and Ebenebe *et al*. (2017) after these authors fed varying levels of BKSM to rabbit bucks. The low statistical sensitivity due to small sampling size may have accounted for these variations. Although, these semen traits did not differ, the bucks that received the 25% BKSCE recorded values that were higher and well above the minimum requirement. According to Boitrelle *et al*. (2021) and Vasan (2011), the minimum threshold for motility and progressive motility was considered to be 40% and 50% respectively.

Similarly, sperm concentration and sperm count in this study were similar ($P>0.05$) even as higher values (323.33 and 355.33x10^6 respectively) were recorded by bucks offered 25% BKSCE. Sperm concentration

Table 1: Semen Characteristics of Rabbit Bucks Fed Bitter Kola Seed Crude Extract in Water

	BKSCE Levels				
Parameters	**0 %** (n=6)	**25 %** (n=6)	**50 %** (n=6)	**± SE**	**P-value^{LOS}**
Semen Colour:					
Milk white (%)	33.33	0.00	33.33	–	–
Creamy white (%)	66.67	100.00	66.67	–	–
Semen pH	6.67	7.00	7.00	0.24	0.549^{ns}
Semen Volume (ml)	1.27	1.13	1.00	0.17	0.547^{ns}
Mass Motility (%)	63.33	78.33	71.67	5.77	0.217^{ns}
Progressive Motility (%)	56.67	70.00	63.33	3.60	0.059^{ns}
^{1}Sperm Conc. (x10^6)	260.00	323.33	286.00	28.54	0.316^{ns}
^{2}Sperm Count (x10^6)	318.67	355.33	286.80	40.38	0. 276^{ns}
Live Sperm (%)	68.33	75.00	70.17	2.82	0.256^{ns}
Normal sperm (%)	69.50	68.67	71.83	1.81	0.459^{ns}
Abnormal sperm	30.50	31.33	28.17	1.81	0.459^{ns}
Acrosome Integrity (%)	67.67	70.00	66.00	1.95	0.369^{ns}

Means within the same row without superscripts do not differ significantly; SE = standard error; BKSCE = bitter kola seed crude extract; n = sample size; ^{LOS} = level of significance; ns = not significant ($P>0.05$); ^{1}sperm concentration/ml semen; ^{2}sperm count per ejaculate

in this result concurred with Iwuji and Herbert (2012) but contrasted with the significant findings of Ebenebe *et al*. (2017). Also, Ebenebe *et al*. (2017) observed significant differences ($P<0.05$) in sperm count which contrasted with this study, however, these authors recorded lower values (104.4–117.7x10^6) compared to this study. In addition, sperm concentration values (260–323.33x10^6) in this study far exceeded 143.7–237.19x10^6 reported by

these authors. These variations may have been accounted for by differences in sampling size. Live sperm (viability) in this study contrasted with the significant (P>0.05) findings by Iwuji and Herbert (2012) and Ebenebe *et al*. (2017) for rabbit bucks fed varying levels of BKSM, even though, values in this study (68.33–75.00%) were comparable to 81.67–93.33% and 71.01–93.30% respectively found by these authors. Sperm morphology in this study showed that normal sperms (P>0.05) ranged from 68.67–71.83% sperm while abnormal sperms (P>0.05) ranged from 28.17–31.33%. There is a serious literature gap on the reproductive characteristics of rabbit bucks fed bitter kola seed (BKS) particularly on sperm morphology and acrosome integrity that could be useful for adequate comparisons.

The normo–morphological sperm evaluation in this study (P>0.05) contrasted with the significant (P<0.05) findings by Abu *et al*. (2013) after albino Wistar rats were fed 0–400 mg/kg body weight of aqueous BKS extract. The values (39.40–68.80%) reported by these authors were lower compared to this study and also showed marked dose-dependent reductions to aqueous BKS extract. The observed variations between this study and these authors may be attributed to the dose used as well as the prominent impact on body and testes sizes. The abnormal sperms in this study concurred with the non–significant trend in rabbits fed BKSM reported by Iwuji and Herbert (2012) with comparably lower values (17.67–23.67%). The acrosome integrity in this study was similar (P<0.05) and ranged from 66.00–70.00%, though, the values from this work were comparably lower than 86.56% reported by Di Iorio *et al*. (2014) for freshly collected semen of rabbits. In summary, the improvement in the array of the seminal characteristics under the present investigation points to the possible androgenic properties of BKSCE supplementation on the overall fertility status of rabbit bucks.

CONCLUSION

In conclusion, semen characteristics of the rabbit bucks were not affected by the supplementation with BKSCE adopted in this study, which may have been occasioned by the low statistical sensitivity of the data collected due to small sampling size which stemmed from high research costs. However, there was notable concentration–dependent improvement in these seminal traits especially at 25% level of BKSCE which was also corroborated by the previous works of other authors cited on rabbit bucks fed varying levels of BKSM. Therefore, this suggests that BKSCE has potential beneficial androgenic properties that could enhance the seminal quality of rabbit bucks.

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