

Potentials of Honey: Fertility Booster and Garnish of Semen Quality in Bucks (West African Dwarf) Goat

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ABSTRACT

This study was designed to investigate the values of honey as a potential fertility booster in West African Dwarf (WAD) bucks. Twelve apparently healthy matured WAD bucks, weight 18-20 kg were selected and randomly divided into groups A (untreated) and B (treated). Group B bucks were treated daily with honey 1ml/10kg body weight administered orally for twenty eight days. Semen samples were collected from both groups weekly for four consecutive weeks using electro-ejaculator. Serum samples were obtained from both groups twice a week for period of four weeks for evaluation of testosterone, follicle stimulating hormone (FSH) and luteinizing hormone (LH) profiles. Gross morphometric and histomorphometric studies of testes and epididymides were done using standard methods. Group B showed significant ($p < 0.05$) increased on sperm motility, morphology, live-dead ratio, concentrations and mass activity compared with group A. There were significant increases in the weight and volume of the right testes of group B compared with the group A ($p < 0.05$). The caudal epididymides of the group B were more densely concentrated with sperm cells compared with those of group A. The GEH of the right and left testes of the group B were significantly higher than those of the group A ($p < 0.05$). The EDD and ELD of the tail of the right epididymides of group B were significantly larger than those of the group A ($p < 0.05$). Serum testosterone and LH concentrations increased significantly in group B while FSH was not significantly different compared with those of group A from week 1 through to week 4 ($p < 0.05$). In conclusion, these findings suggest that honey is a potential fertility booster in WAD bucks. We therefore recommend its inclusion in the diet of WAD bucks propose for breeding programme.

Key words: honey, booster, fertility, sperm quality, West African dwarf buck

INTRODUCTION

Honey is a sweet food that is produced by honey bees (genus *Apis*) by using nectar from flowers through the process of regurgitation and evaporation (Subramanian *et al.*, 2007; Muhammad *et al.*, 2012). It contains water, carbohydrates, protein, vitamins (especially vitamin C) as well as essential trace elements like calcium, magnesium, iron, phosphorus, potassium, sodium and zinc (Kumar *et al.*, 2010; Syazana *et al.*, 2011). It has a long history in human consumption. The composition and properties of honey varies with floral sources utilized by the bees as well as regional and climatic conditions (Salinas *et al.*, 1994; Perez-Arquillue *et al.*, 1994). The studies on physical and chemical properties of honey produced in different countries have been reported (Langridge, 1977). It also has many health benefits and had been reported to have been used both orally and topically to treat many ailments in the conventional and various traditional and medical practices; including rhino-sinusitis, burns dressing, skin infections, as well as immune-compromising, gastroenterological, ophthalmological, metabolic, cardiovascular and tumor related disease conditions amongst many others (Bogdanov *et al.*, 2008; Mandal and Mandal, 2011; Ajibola *et al.*, 2012; Aliyu *et al.*, 2012). However, there are very few reports on the fertility effect of honey especially on animal reproduction. West African Dwarf (WAD) goats are small ruminants with great breeding potentials (Wilson, 1989; Chukwuka *et al.*, 2010). They serve as good sources of protein as well as income especially to the low earn class

of people in the rural areas (Adebayo & Chineke, 2011). The WAD buck play significant role in goat production as it can sire many does on the farm (Nolte, 2012). Improving the fertility of these bucks will go a long way in improving the reproduction potentials of WAD bucks. However, the increased demand of animal protein by humans in developing countries for optimum human physiology cannot be overemphasize. Therefore this study was undertaken to investigate the potentials of honey in enhancing fertility that can guarantee optimum reproduction WAD bucks.

Materials and Methods

Animals and Management

Twelve matured and reproductively active WAD bucks of the average age and weight of two years and 11.4 ± 0.7 kg body weights (BW) respectively were used for this study. They were stabilized for 4 weeks during which prophylactic treatment and vaccinations were administered; and thereafter divided into groups A and B comprising of six bucks per group. Group A served as control (given water) while group B were given honey orally daily (using a sterile 10 ml syringe placed over the tongue) for twenty eight days at a dose rate of 1ml/10kg BW. Animals were dosed at 8:00 am daily.

Source of Honey

Five liters of honey was purchased from central market of Ilorin, Kwara state, Nigeria. The honey was physical examined. The honey used in present study was ripe honey with presence of nectar. Extraction of honey was carried out by filtering and squeezing the honey through muslin cloth and stored in air tight plastic jars for further analysis.

Evaluation of Honey Quality

The honey was evaluated for colour, consistency, taste, flavour and mouth feel using 9-point hedonic scale as described by (Narpinder and Parminder, 2011) for overall acceptability. A pure honey does not foam under any circumstance, is quite thick and only trickles in a stream, soft texture thin filament-like and never separates into layers with a good aroma.

Semen Collection and Analysis

Semen was collected using electro-ejaculator as described by (Iliyasu *et al.*, 2014a) from both groups of bucks weekly for the period of four consecutive weeks and the semen sampled were evaluated for color, volume, mass sperm motility, sperm morphology, sperm live-dead ratio and sperm concentration as described by Iliyasu *et al.* (2014a) and Raji and Ajala (2015).

Blood sample collection

Blood samples were collected by 8:00 am from bucks in both groups twice in a week aseptically for the period of four weeks via the jugular vein into non-heparinized sample bottles. The blood samples were allowed to clot then centrifuged at $3000 \times g$ for 30 minutes to separate serum from the plasma as described by Iliyasu *et al.* (2014b). The serum samples were carefully decanted into plain bottles and stored in a deep freezer at -40°C until required. The serum were assayed for testosterone, follicle stimulating hormone (FSH) and luteinizing hormone (LH) profiles using Enzyme-Linked Immunosorbent Assay (ELISA) kits designed for each hormone.

Gross testicular morphometric studies and Histological procedure

The parameters studied were testicular weight and volume. These were taken as described previously by Raji and Ajala, 2015 and Iliyasu *et al.*, 2014. The histology of the testes and epididymides were done as described previously by Raji *et al.*, 2012; Raji and Ajala, 2015). Selected images were captured using Moticam 2.0 digital camera attached to a computer.

Histomorphometric studies of the gonads

The parameters studied included seminiferous tubule diameter (STD), seminiferous luminal diameter (SLD) and germinal epithelial height (GEH). Ten measurements were taken per section for each parameter. Images were taken and analyzed using the Motic Images Plus (MIPlus).

Statistical Analysis

The data obtained were analyzed using Analysis of variance (ANOVA) and Student t-test at 5% level of significance ($p < 0.05$) using SPSS version 20.

Results

The results of the semen analysis for the bucks in (group A) and (group B) are presented in Table 1. There were significant increase in sperm motility; sperm morphology; sperm live-dead ratio, and sperm concentration in group B bucks compared to group A bucks. ($P = 0.03, 0.03, 0.02, 0.04$ and 0.01 respectively).

Table 1: Evaluation of semen characteristics of the untreated and treated groups of West African Dwarf bucks administered honey orally.

Semen characteristics	Groups (N=6)	Pre-experiment	Week one	Week two	Week three	Week four
Color	B Untreated A Treated with Honey (1ml/10kg)	Cream Cream	Cream Cream	Cream Cream	Cream Cream	Cream Cream
Volume (ml)	B Untreated A Treated with Honey (1ml/10kg)	0.3±0.06 0.3±0.02	0.3±0.05 0.4±0.03	0.3±0.07 0.4±0.09	0.3±0.04 0.5±0.07	0.3±0.08 0.5±0.13
Mass activity	B Untreated A Treated with Honey (1ml/10kg)	4 4	4 5	4 5	4 5	4 5
Motility (%)	B Untreated A Treated with Honey (1ml/10kg)	82 ± 2.36 81 ± 3.04	81 ± 3.09 84 ± 4.16	82±2.16 89±5.03	81±3.25 92 ± 3.18	81±2.18 94 ± 4.06
Morphology (%)	B Untreated A Treated with Honey (1ml/10kg)	88 ± 3.05 87 ± 2.65	89 ± 2.74 92 ± 3.07	88 ± 2.91 93 ± 4.25	87 ± 3.24 95 ± 3.18	89 ± 3.20 95 ± 4.23
Live-dead ratio (%)	B Untreated A Treated with Honey (1ml/10kg)	82 ± 1.94 81 ± 2.04	83±1.86 85±2.45	81 ± 1.74 88±2.36	82±2.15 90 ± 3.04	80 ± 1.92 92 ± 2.81
Concentration (×10 ⁹ sperm cells/ml)	B Untreated A Treated with Honey (1ml/10kg)	1.81± 0.16 1.80 ± 0.21	1.79±0.06 2.12±0.17	1.84±0.05 2.18±0.14	1.81±0.13 2.26±0.19	1.85±0.07 2.31±0.10

All values within the rows at P < 0.05 level were considered significance.

Table II: Dimension of testicular weight and volume of the untreated and treated West African Dwarf bucks with honey.

Parameters	N	WRT (kg)	VRT (cm ³)	WLT (kg)	VLT (cm ³)
Untreated group	6	27.6 ± 0.5 ^{ab}	27.8 ± 0.6 ^{ab}	28.1 ± 0.9	28.3 ± 1.1
Treated group	6	28.9 ± 1.3 ^{ab}	28.7 ± 1.2 ^{ab}	28.6 ± 1.2	28.8 ± 1.3

* All asterisk values with superscript are significant at P < 0.05 across the rows.

Keys: WRT – weight of right testis; VRT – volume of right testis; WLT – weight of left testis; VLT – volume of left testis

Table III: Mean and Standard deviation of the histomorphometric dimension of the testes of control and West African Dwarf bucks dosed with honey.

Parameters	Control (µm)	Right testis	Control Left testis (µm)	Honey treated Right testis	Honey treated Left testis
Seminiferous tubular diameter	584.6 ± 30.2		579.4 ± 31.45	588.6 ± 42.7	590.1 ± 41.3
Seminiferous luminal diameter	261.4 ± 31.8 ^{ac}		257.3 ± 30.9 ^{bd}	236.8±28.9 ^{ca}	239.7 ± 29.4 ^{db}
Germinal epithelial height	192.6 ± 28.1 ^{ac}		195.8 ± 29.4 ^{bd}	218.4 ± 24.6 ^{ca}	213.9 ± 25.6 ^{db}

* All asterisk values with superscript are significant at P < 0.05 across the columns.

Table IV: Mean and Standard deviation of the histomorphometric of epididymis of untreated and treated groups West African Dwarf bucks with honey.

Parameters	N	HRE (μm)	BRE (μm)	TRE (μm)	HLE (μm)	BLE (μm)	TLE (μm)
Epididymal ductal diameter	6	C- 984.8 ± 107.2	C- 951.8 ± 93.5	C- 1936.1 ^{*ab} ± 237.8	C- 979.4 ± 102.3	C- 956.4 ± 90.8	C- 1930.9 ± 228.4
	6	H- 987.3 ± 118.4	H- 964.5 ± 97.4	H- 1984.3 ^{*ab} ± 248.0	H- 981.1 ± 105.4	H- 967.2 ± 94.3	H- 1983.2 ± 241.5
Epididymal luminal diameter	6	C- 714.6 ± 62.3	C- 692.1 ± 39.2	C- 1694.7 ^{*cd} ± 241.8	C- 718.6 ± 58.1	C- 701.6 ± 39.5	C- 1701.9 ± 215.4
	6	H- 721.4 ± 73.9	H- 702.1 ± 46.3	H- 1759.2 ^{*cd} ± 274.9	H- 732.6 ± 66.3	H- 718.4 ± 52.5	H- 1748.2 ± 236.1
Epididymal epithelial height	6	C- 195.2 ± 21.4	C- 201.2 ± 22.5	C- 93.8 ± 15.1	C- 198.4 ± 22.6	C- 207.4 ± 23.5	C- 96.1 ± 16.3
		H- 193.7 ± 20.6	H- 193.5 ± 19.2	H- 87.3 ± 14.2	H- 191.6 ± 21.4	H- 196.0 ± 20.9	H- 89.6 ± 15.2

* All asterisk values with superscript are significant at $P < 0.05$ across the rows.

The results of gross weight and volume of the testes of the group A and B bucks are as presented in (Table II). There were significant increases in the weight and volume of the right testes of group B bucks compared to that of group A bucks ($p = 0.01$ and 0.02 respectively); It was also observed that testicular weight was approximately equal to the testicular volume of all the bucks in the two groups ($p < 0.05$).

The results for the histomorphometric studies are presented in (Table III). There were no significant differences in the Seminiferous tubular diameter (STD) of the untreated (group A) and treated (group B) bucks, right testes ($p = 0.68$) and left testes ($p = 0.73$) respectively. However, the Seminiferous luminal diameter (SLD) of the right and left testes of group A bucks were significantly larger than those of the group B bucks ($p = 0.03$ and 0.04 respectively).

While the Germinal epithelial height (GEH) of the right and left testes of group B bucks were significantly higher than those of the group A bucks ($p = 0.02$ and 0.03 respectively) as presented in (Table IV). It showed that when these two groups were compared, there were no significance differences in the Epididymal epithelial height (EEH) of the carpus, caput and caudal of the right epididymides ($p = 0.96$, 0.82 and 0.86 respectively) and those of the left epididymides ($p = 0.87$, 0.92 and 0.72 respectively). However, the Epididymal ductal diameter (EDD) and Epididymal luminal diameter (ELD) of the tail of the right epididymides of the group B bucks were significantly larger than those of the group A bucks ($p = 0.03$ and $p = 0.02$ respectively); and those of the left epididymides ($p = 0.01$ and $p = 0.02$ respectively).

There were significant increases in serum testosterone concentration in group B bucks compared with group A bucks from week 1 ($p = 0.03$), week 2 ($p = 0.02$), week 3 ($p = 0.01$) and week 4 ($p = 0.01$) post treatment. There were also increases in serum testosterone level in group B bucks from week 1 to 2, week 2 to 3 and week 3 to 4 post treatment but these increases were not significant ($p = 0.13$, 0.19 and 0.22 respectively).

There were increases in the serum concentration of follicular stimulating hormone (FSH) in the group B bucks compared with that of group A bucks, however, these increases were not significant at the end of week 1 ($p = 0.84$), week 2 ($p = 0.89$), week 3 ($p = 0.75$) and week 4 ($p = 0.81$) post treatment. The luteinizing hormone (LH) serum concentration was significantly higher in group B than group A bucks in week 1, 2, 3 and 4 ($p = 0.04$, 0.04 , 0.02 and 0.01 respectively). There were also increases in LH level from week 1 to week 4. Although, these increases were not significant initially from week 1 to 2 ($p = 0.13$) and from week 2 to 3 ($p = 0.09$), however the increase from week 3 to 4 was significant ($p = 0.04$). Also, LH concentration in week 4 was significantly higher than that of week 1 ($p = 0.02$) post treatment as presented in (Table V).

Table V: Profiles of some reproductive hormones of untreated and treated West African Dwarf bucks with honey.

Hormone	G	Pre-Experiment	Week one	Week two	Week three	Week four
Testosterone (ng/ml)	U	4.75±0.08	4.72±0.11 ^{*ab}	4.79±0.09 ^{*ab}	4.74±0.12 ^{*ab}	4.81±0.07 ^{*ab}
	T	4.73±0.11	5.04±0.02 ^{*ab}	5.15±0.03 ^{*ab}	5.31±0.02 ^{*ab}	5.45±0.04 ^{*ab}
FSH (mMol/ml)	U	2.81±0.07	2.78±0.12	2.77±0.15	2.79±0.13	2.80±0.06
	T	2.78±0.15	2.80±0.11	2.83±0.14	2.85±0.23	2.89±0.19
LH (mMol/ml)	U	3.07±0.21	2.96±0.15 ^{*ef}	3.04±0.09 ^{*ef}	3.00±0.17 ^{*ef}	2.94±0.24 ^{*ef}
	T	3.02±0.16	3.05±0.28 ^{*ef}	3.16±0.04 ^{*ef}	3.27±0.23 ^{*ef}	3.40±0.30 ^{*ef}

* All asterisk values with superscript are significant at $P < 0.05$ across the rows.

Keys: - LH – Luteinizing Hormone, FSH – Follicle Stimulating Hormone, U – Untreated group, T – Treated group, G- group.

Discussion

This study was conducted to evaluate honey as a potential fertility booster in the WAD goat bucks. We observed that the semen characteristics including motility, mass activity, concentration, percentage live-dead ratio and morphology increased significantly in WAD bucks dosed with honey compared with those of the control. This is similar to the report on the effects of honey on semen preservability in cattle bulls (El-Sheshtawy *et al.*, (2014) and as a supplement in tris extender on cryopreservation of bull spermatozoa (Yimer *et al.*, 2015). However, the dose and duration should be monitored as high dose and chronic use of honey could be detrimental to spermatozoa and semen characteristics (Dare *et al.*, 2013; Yimer *et al.*, 2015).

We also observed that the WAD bucks dosed with honey had heavier testicular weights and higher testicular volumes which were significant on the right testes compared with those of the control. Similarly, Osman (2011) reported a reversal to normal testicular weight with the use of honey alone and in combination with *Ferula hermonis* following an initial reduction due to the effect of gamma irradiation on rat testes. However, contrarily, Asiyah *et al.*, (2011) reported significant differences in testicular weights of the control and rats dosed with gleam honey. Although, reports on the effect of honey on testicular volume is very scarce, in this study, we observed that testicular weight was approximately equal to testicular volume similar to the report by Raji *et al.* (2016) in WAD bucks and Iliyasu *et al.* (2014a) in Yankasa rams.

The histology results in this study revealed that the testicular architecture for the control and WAD bucks dosed with honey were similar to those reported by Raji *et al.*, (2015); also for the epididymal architecture similar to the report by Raji *et al.*, (2016). However, we observed that the cauda epididymis of WAD bucks dosed with honey were more densely concentrated with sperm cells. This suggests that more spermatozoa were produced by the testes and stored at the caudal epididymis due to the effect of the honey. However, further studies should be carried out to establish this fact.

The results for the histomorphometric studies revealed that there were no significant differences in the STD of the testes of the WAD bucks compared to the control and dosed groups. The values we observed were similar for those reported for mature WAD bucks by Raji *et al.*, (2015). However, the GEH of the honey dosed WAD bucks were significantly higher than those of the control while the reverse was the case with the SLD between the two groups of WAD bucks. These findings implies that as the GEH was increasing in the honey dosed group the SLD was decreasing indicating higher spermatogenic activities within the testes of this group compared to the control group. Similarly, Shinkut (2009) reported increase in spermatogenic activities in rabbit bucks testes fed with supplements of *Allium sativum*. Histomorphometric results also showed that the EDD and ELD of the tail of the epididymis of the honey dosed WAD bucks were significantly larger than those of the control bucks. This implies that more spermatozoa are stored within the tail epididymides of the honey dosed WAD bucks. This is comparable to the report by Shinkut (2009) were more epididymal reserves were observed in rabbit bucks fed with supplements of *Allium sativum*.

From the results obtained from the hormonal analysis we also observed that there were increases in the FSH concentration of the honey dosed WAD bucks compared with the control but these increases were not significant. However, the testosterone and LH concentrations were significantly higher in the honey dosed WAD bucks compared to the control bucks. Hence, the reason for increase in spermatogenic activities evidences of more spermatozoa in the seminiferous tubules and cauda epididymides of the honey dosed WAD bucks this agreed with the findings of (Iliyasu *et al.*, 2014a) in Yankasa rams. Muhammed (2014) also observed increases in testosterone, LH and FSH concentration in male mice dosed with honey. However, Salman *et al.*, (2013) reported only increase in testosterone serum concentration but decrease in FSH and LH

concentrations in male albino rats dosed with honey. Anyakudo *et al.*, (2015) reported increase in testosterone concentration and no significant difference in FSH and LH concentration in diabetic rats dosed with honey. These varying reports may be due to species differences. We suggest that further studies should be carried out on the effects of honey on the reproductive hormones of not only WAD bucks but also on other livestock animals.

CONCLUSION

In conclusion, the findings of this study suggest that honey can be use as potential fertility booster in WAD bucks because it has shown significant increase in semen quality and spermatogenic activities in the seminiferous tubules, extragonadal sperm reserves and some reproductive hormones like testosterone and LH. We also advocate honey as fertility booster in WAD bucks while further research should be done to establish the basic mechanism of the concept. The incorporation of honey in management of fertility will go a long way in improving WAD goat production which is known to be good sources of animal protein and economic income for human.

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