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Research Article

Semen and Testicular Dynamics of Mature Nigerian Indigenous Cocks Administered Exogenous Follicle Stimulating and Luteinizing Hormone (Menotropin)

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Abstract

Background and Objective: This study was conducted to examine the synergistic effect of exogenous Follicles Stimulating Hormone (FSH) and Luteinizing Hormone (LH) on semen, spermatogenesis and testicular dynamics using Menotropin injection. **Materials and Methods:** About 72 mature Nigerian indigenous cocks, comprised of the frizzle feather, naked neck and normal feathered were randomly divided into four groups (T_1 , T_2 , T_3 and T_4) of 18 cocks each, at 3 replicated with 6 cocks per replicate. The cocks were intramuscularly injected with different doses of Menotropin at 0.0 mL, 0.10 mL, 0.20 mL and 0.30 mL, respectively. Semen samples were collected by abdominal massage and examined for macroscopic and microscopic indices. Semen was evaluated for concentration, pH, color, volume, viscosity, spermatozoa counts, morphology and many more. The various testicular dynamics were determined. **Results:** There were significantly ($p < 0.05$) improvements in semen volume (T_1 0.55 - T_4 0.83 mL), concentration (T_1 3.21 - T_4 4.40×10^9 mL⁻¹), consistency (T_1 2.67 - T_4 4.00), pH (T_1 7.40 - T_4 7.14), spermatozoa motility (T_1 60.25 - T_4 83.20%), live proportion (T_1 74.62 - T_4 89.56%), viable spermatozoa (T_1 9.80 - T_3 31.08×10^{12} mL⁻¹) and the total number of spermatozoa (T_1 1.77 - T_2 3.78×10^9 mL⁻¹), while, spermatozoa abnormalities (T_1 22.33 - T_4 10.87%) significantly reduced. **Conclusion:** The exogenous FSH (Follicle Stimulating Hormone) and LH (Luteinizing Hormone) at 0.10 and 0.20 mL had synergistic effects on the semen and testicular dynamics of the cocks as they improved the semen characteristics and had no adverse effects on the testicular dynamics. The dosage at 0.10mL produced the best results, while up to 0.30 mL Menotropin negatively impacted some semen parameters of the cocks.

Key words: Semen, menotropin, testis, follicles stimulating hormone, luteinizing hormone, Nigerian indigenous cocks

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The assessment of semen quality of cocks gives an excellent indicator of their reproductive potential. The semen quality of cocks has been reported to be one of the major determinants in the fertility and hatchability of eggs^{1,2}. Although the hens and the cocks are genetically considered to be equal partners in mating, the cock's semen has a major impact on the fertility and hatchability of the egg and on reproductive efficiency just like many other animal species^{3,4}. This variation in male reproductive potentials is attributable to semen quality, management, environment, nutrition, genetics or the combined effects of these factors^{5,6}. In view of the fundamental use of semen in animal reproduction, there is a need to improve the semen quality of indigenous cocks using exogenous gonadotrophins.

Menotropin, a gonadotrophin preparation, is a mixture of pituitary gonadotrophins, consisting of 75 International Unit (IU) Follicles Stimulating Hormone (FSH) and 75 International Unit (IU) Luteinizing Hormone (LH) in the ratio of 1:1. Menotropin is used in the treatment of infertility in both males and females. It is utilized in the treatment of hypogonadism, resulting in delayed puberty, low sperm count and increases testosterone level in males. In females, Menotropin works by stimulating follicular growth and the release of ova from the ovary. The injection can be administered intramuscularly or subcutaneously. Semen and spermatogenesis involve the activities of the Follicle Stimulating Hormones (FSH) and Luteinizing Hormone (LH) in synergy with other male reproductive hormones^{7,8}. Spermatogenesis and folliculogenesis involve cell-to-cell interactions regulated by Luteinizing Hormone (LH) and Follicle Stimulating Hormone (FSH). The FSH and LH regulate the initiation, proliferation and maturation of germ cells during spermatogenesis⁹. Spermatogenesis is completely dependent on pituitary hormones, Follicle-stimulating Hormone (FSH), Luteinizing Hormone (LH) and other androgenic hormones. The FSH and LH are critical for the initiation, stimulation and maintenance of the seminiferous epithelial cycle which is a prerequisite for spermatogenesis. The FSH acts on the Sertoli cells to stimulate germ cell proliferation as well as increase androgen production by the Leydig cells in the seminiferous tubule. The LH supports the function of Sertoli cells, which in turn support many aspects of spermatozoa maturation^{10,11}. Spermatogenesis is a concerted sequence of events during the production and maturation of spermatogonia into spermatid and spermatozoa¹². These two pituitary gonadotrophins, FSH and LH stimulates high intra-testicular testosterone concentration which is very crucial for

spermatogenesis^{13,14}. Therefore, this study aimed to evaluate the complementary and synergistic effects of exogenous follicle stimulating hormone and luteinizing hormone on the semen constituents and testicular dynamics of mature Nigerian indigenous cocks.

MATERIALS AND METHODS

The study was carried out from June to August, 2022.

Experimental location: This study was conducted at the Poultry Unit of the Teaching and Research Farm of the College of Animal Science and Animal Production, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. The area is located in the South-Eastern part of Nigeria on latitude 5°27' North, longitude 7°32' East, an altitude of 123 m above sea level with an annual rainfall of 2177 mm, temperature of 22°C-36°C and relative humidity of 50-90%. It is situated within the humidrain forest zone of West Africa, characterized by the long duration of the rainy season (March-October) and a short period of the dry season (November-February). Climatic data were collected from the Meteorological Center of the National Root Crop Research Institute, Umudike, Abia State¹⁵.

Experimental animals and management: Ninety sexually matured Nigerian indigenous cocks comprised of the frizzle feather, naked neck and normal feathered, purchased from the rural farmers in Ngwa, Ikwuano and Umuahia, Abia state, Nigeria were used for the study. The study was in a Completely Randomized Design (CRD) experiment that lasted for 2 months. Pre-experimental period of 2 weeks was used to acclimatize and stabilize the cocks to the experimental procedures. Thereafter, Seventy-two mature Nigerian indigenous cocks selected were randomly assigned to 4 treatment groups and replicated 3 times with 6 cocks per replicate. The cocks were raised in deep litter pens throughout the experimental period and fed a concentrated diet with clean drinking water supplied *ad-libitum*. All routine management practices for mature Nigerian indigenous cocks as recommended were carried out appropriately. The cocks in each treatment were fed the same experimental diet and administered different doses of Menotropin injection intramuscularly on the thigh using a 1 mL syringe with 0.1 mL graduation at 2 weeks' intervals. The semen was collected at 4 weeks intervals. The conduct of this experiment followed the procedure and ethical guidelines stipulated for the use of animals in experimentation as outlined by the Animal Welfare

Table 1: Composition of the experimental diet

Ingredients	Composition (%)
Maize	60.00
Soya bean meal	18.00
Palm kernel cake	5.00
Fish meal	2.00
Blood meal	2.00
Wheat offal	8.00
Bone meal	4.00
Common salt	0.25
Lysine	0.25
Methionine	0.25
Vit/mineral Premix*	0.25
Total	100.00
<i>Jatropha tanjorensis</i> leaf meal	0.00%
Crude protein (%)	19.00
Energy (kcal kg ⁻¹)	2920.00

*Premix composition (per kg of feed), Folic acid: 1.00 mg, Calcium pantothenate: 10 mg, Antioxidant: 200 mg, Vitamin A: 12,500 IU, Vitamin B1: 3.00 mg, Vitamin B2: 6.00 mg, Vitamin B6: 6.00 mg, Niacin: 40 mg, Biotin: 0.08 mg, Vitamin B12: 0.25 mg, Vitamin D3: 2500 IU, Vitamin E: 50.00 mg, Vitamin K3: 2.50 mg, Iodine: 1.55 mg, Iron: 50 mg, Zinc: 45 mg, Chlorine chloride: 300 mg, Manganese: 100 mg, Copper: 2.00 mg, Cobalt: 0.25 mg and Selenium: 0.10 mg

and Ethics Committee, the Michael Okpara University of Agriculture, Umudike, Nigeria. The experimental diet for the cocks was shown in Table 1.

Experimental design: The study was a Completely Randomized Design (CRD) with four treatments consisting of T₁, T₂, T₃ and T₄. The T₁ administered no FSH and LH served as the control. Eighteen sexually matured cocks were randomly assigned to each treatment and replicated 3 times with 6 cocks per replicate. Menotropin, a gonadotrophin preparation which is a mixture of pituitary gonadotrophins, consisting of 75 IU follicles stimulating hormone and 75 IU luteinizing hormone in the ratio of 1:1 were sourced from a reputable pharmacy in Umuahia, Abia State. Thereafter, the Menotropin were intramuscularly administered to the cocks between 7.00 and 8.00 a.m. local time after feeding on each day of administration at 2 weeks interval for 8 weeks. Each cock on T₁ was intramuscularly administered Menotropin at 0.0 mL, T₂ at 0.10 mL, T₃ at 0.20 mL and T₄ at 0.30 mL Menotropin, respectively. The field work lasted for 10 weeks. The model of this study is as follows:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where:

Y_{ij} = Individual observation on the broiler characteristics
 μ = Overall mean
T_i = Treatment effect
e_{ij} = Random error assumed to be identically, independently, with constant variances and normally distributed with zero means¹⁶

Data collection

Semen collection and evaluation: Semen was collected from four cocks sampled from each replicate through abdominal massage. Before semen collection, the cocks were trained to ejaculate by abdominal massage and manipulation of the cloaca. The semen was collected at 4 weeks intervals between 7 a.m. and 10 a.m. local time on each day of semen collection. Semen evaluation involved the estimation of both the macroscopic and microscopic indices. Semen volume was determined in millimeters using a calibrated glass collection tube (collected with a 0.01 mL calibrated test tube). Semen pH was determined using a pH meter (Sperm 360® by Sperm Processor Pvt. Ltd., Aurangabad MS-431005 India). Semen color and consistency were determined subjectively and scored using the scoring pattern: Color (whey = 1, creamy = 2), consistency (very thick = 4, thick = 3, fairly thick = 2, watery = 1). Spermatozoa concentration was determined using a haemocytometer as described by Jequier¹⁷. A total number of spermatozoa per ejaculate was determined by multiplying semen volume by the spermatozoa concentration. Spermatozoa motility was determined subjectively in a drop of fresh semen on a glass slide covered with slip and examined using a microscope as described by Oguike *et al.*¹⁸. Spermatozoa morphology was determined by performing differential counts of the morphologically normal and abnormal shape of the spermatozoa using eosin-nigrosin stain. Spermatozoa live proportions were determined using the methods outlined by Brazil¹⁹.

Testicular dynamics: At the end of the experiment, four cocks were sampled from each replicate and the individual testicles were collected to determine the various testicular dynamics. Weights of the testicles were recorded after the epididymis had been trimmed off. The various testicular dynamics were measured using a 5 kg digital weighing scale (Camry EK 5055 Digital Scale) of 0.01 sensitivity.

Statistical analysis: The various data collected on the different seminal and testicular parameters were subjected to analysis of variance in-line with the methods as described by Kim²⁰. All significant means were separated according to Duncan's Multiple Range Test at a 5% level of significance²¹.

RESULTS

The effects of graded dosage of intramuscular injection of Menotropin on semen characteristics of mature Nigerian indigenous cocks were presented in Table 2.

Table 2: Semen characteristics of mature Nigerian indigenous cocks administered graded dosage of Menotropin

Parameter	T ₁	T ₂	T ₃	T ₄	SEM
Semen volume (mL)	0.55 ^a	0.78 ^b	0.91 ^b	0.83 ^b	0.10
Semen color (1-2)	1.00	2.00	1.00	2.00	0.11
Semen consistency (1-4)	2.67 ^a	3.98 ^b	3.88 ^b	4.00 ^b	0.16
Semen pH (1-14)	7.40 ^c	7.22 ^b	7.25 ^b	7.14 ^a	0.02
Spermatozoa mass motility (%)	60.25 ^a	72.43 ^b	80.00 ^c	83.20 ^c	2.89
Spermatozoa live proportion (%)	74.62 ^a	78.12 ^{ab}	85.89 ^b	89.56 ^b	1.88
Sperm concentration ($\times 10^9$ mL ⁻¹)	3.21 ^a	4.20 ^b	4.11 ^b	4.40 ^b	0.20
Total number of sperm per Ejaculate ($\times 10^9$ mL ⁻¹)	1.77 ^a	3.78 ^b	3.74 ^b	3.65 ^b	0.44
Total viable sperm ($\times 10^{12}$ mL ⁻¹)	9.80 ^a	24.28 ^b	31.08 ^c	28.06 ^b	2.90
Percentage normal sperm (%)	93.23	95.00	94.03	93.80	1.00

^{abc}Means with different superscripts along rows are significantly different ($p < 0.05$) and SEM: Standard error of treatment means

Table 3: Effects of graded levels of Menotropin on the second semen collection of mature Nigerian indigenous cocks

Parameter	T ₁	T ₂	T ₃	T ₄	SEM
Semen volume (mL)	0.50 ^a	0.69 ^{ab}	1.00 ^b	0.90 ^b	0.89
Semen color (1-2)	1.00	1.80	2.00	2.00	0.14
Semen consistency (1-4)	3.00	4.00	3.60	4.00	0.20
Semen pH (1-14)	6.97	7.10	7.03	7.20	0.01
Spermatozoa mass motility (%)	72.52 ^a	82.52 ^b	85.00 ^{bc}	89.20 ^c	2.95
Spermatozoa live proportion (%)	78.00 ^a	88.90 ^b	90.10 ^b	81.20 ^a	2.00
Semen concentration ($\times 10^9$ mL ⁻¹)	4.30 ^{ab}	5.24 ^c	4.81 ^b	3.94 ^a	0.14
Total number of sperm per Ejaculate ($\times 10^9$ mL ⁻¹)	2.15 ^a	3.62 ^b	4.81 ^b	3.55 ^b	0.40
Total viable spermatozoa ($\times 10^{12}$ mL ⁻¹)	18.20 ^a	34.67 ^c	27.51 ^b	24.80 ^b	3.72
Percentage of normal sperm (%)	87.33 ^a	94.00 ^{bc}	95.03 ^c	92.90 ^b	1.00
Total abnormal spermatozoa (%)	12.67 ^c	6.00 ^{ab}	4.97 ^a	7.10 ^b	1.00

^{abc}Means with different superscripts along rows are significantly different ($p < 0.05$) and SEM: Standard error of treatment means

Table 4: Effects of graded levels of Menotropin on spermatozoa abnormalities of mature Nigerian indigenous cocks

Parameter (%)	T ₁	T ₂	T ₃	T ₄	SEM
Headless spermatozoa	4.00 ^c	1.43 ^b	1.41 ^b	0.70 ^a	0.50
Double-headed spermatozoa	2.68 ^b	2.04 ^a	2.15 ^a	2.04 ^a	0.03
Twisted tail spermatozoa	3.82 ^c	2.10 ^a	1.70 ^{ab}	1.48 ^b	0.45
Tailless spermatozoa	3.20 ^b	1.20 ^a	0.80 ^a	0.76 ^a	0.20
Broken neck spermatozoa	2.33	2.05	2.43	1.95	0.13
Cytoplasmic droplets	1.43 ^c	1.13 ^{ab}	1.20 ^b	1.08 ^a	0.21
Bent mid-piece	4.87 ^c	2.53 ^{ab}	2.27 ^a	2.86 ^b	0.20
Total abnormal spermatozoa	22.33 ^c	12.48 ^b	11.96 ^{ab}	10.87 ^a	1.10

^{abc}Means with different superscripts along rows are significantly different ($p < 0.05$) and SEM: Standard error of treatment means

Table 5: Effects of graded levels of Menotropin on testicular dynamics of mature Nigerian indigenous cocks

Parameter (g)	T ₁	T ₂	T ₃	T ₄	SEM
Right testis weight	4.50	5.14	5.75	4.21	0.13
Left testis weight	5.96	6.23	6.90	4.90	0.11
Paired testis weight	10.46	11.37	12.65	9.11	0.84
Right epididymis weight	0.28	0.42	0.48	0.36	0.43
Left epididymis weight	0.83	0.60	0.65	0.72	0.10
Paired epididymis weight	1.01 ^a	1.12 ^b	1.13 ^b	1.08 ^b	0.80
Paired vas deference weigh	1.40	1.32	1.30	1.43	0.13
Length of vas deference (cm)	10.00	12.03	12.00	9.80	0.84

^{abc}Means with different superscripts along rows are significantly different ($p < 0.05$) and SEM: Standard error of treatment means

The effects of a graded dosage of intramuscular injection of Menotropin on the second semen collection of mature Nigerian indigenous cocks were presented in Table 3.

The effects of graded dosage of intramuscular injection of Menotropin on spermatozoa abnormalities of mature Nigerian indigenous cocks were presented in Table 4.

The effects of graded dosage of intramuscular injection of Menotropin on testicular dynamics of mature Nigerian indigenous cocks were shown in Table 5.

DISCUSSION

The results of the first semen collection (Table 2) showed that semen volume, semen consistency, spermatozoa mass motility, spermatozoa live proportion, sperm concentration, the total number of spermatozoa per ejaculate and total viable spermatozoa significantly ($p < 0.05$) increased across the treatment group, while semen pH significantly reduced ($p < 0.05$) following the intramuscular injection of 0.10 mL,

0.20 mL and 0.30 mL of Menotropin on the cocks. The semen volume in T_2 (0.78 mL), T_3 (0.91 mL) and T_4 (0.83 mL) are statistically similar ($p>0.05$) and significantly higher ($p<0.05$) compared to the semen volume of the cocks in T_1 (0.55 mL). The semen consistency of the cocks in T_2 (3.98), T_3 (3.88) and T_4 (4.00) are statistically similar ($p>0.05$) and significantly better ($p<0.05$) compared to the consistency of the semen of cocks in T_1 (2.67). The semen pH of cocks in T_2 (7.22) and T_3 (7.25) are statistically similar and significantly higher than those of the cocks in T_4 (7.14). The spermatozoa motility in T_3 (80.00%) and T_4 (83.20%) are statistically similar and significantly higher than in T_1 (60.25%) and T_2 (72.43). The proportion of live spermatozoa in T_1 (74.62%) and T_2 (78.12%) was similar and significantly lower than in T_3 (85.89%) and T_4 (89.56%). The total viable sperm of T_2 ($24.28 \times 10^{12} \text{ mL}^{-1}$) and T_4 ($28.06 \times 10^{12} \text{ mL}^{-1}$) were similar and statistically lower compared to T_3 ($31.08 \times 10^{12} \text{ mL}^{-1}$).

The observed decrease in semen pH from 7.40 in T_1 to 7.14 in T_4 could be attributable to the increase in spermatozoa concentration of the semen, the increase in the total number of spermatozoa per ejaculate and as well as the accompanying increase in spermatozoa metabolic activity as the level of administration of the Menotropin increased in T_2 (0.1 mL), T_3 (0.2 mL) and T_4 (0.3 mL), which in turn increased the rate of fructolysis and fructolytic index of the collected semen. The rate of fructolysis and fructose utilization is higher in good-quality semen²². The higher the semen concentration, the higher the metabolic activities in the semen sample and the more fructose utilization in a given semen sample. A higher rate of fructose utilization and fructolytic index is an indicator of good quality semen. Fructose utilization by sperm cells produced lactic acid in semen, which in turn reduced the semen pH^{23,24}. Therefore, the observed increase in semen concentration, spermatozoa number, spermatozoa motility, spermatozoa live proportion and increased in total viable spermatozoa resultantly increased the rate of metabolic activity in the semen, increased in the rate of lactic acid production in the semen and thus decreased the pH of the collected semen sample.

The significant improvements observed in these semen characteristics could also be attributable to the mechanism of action of gonadotrophins on the gonads/testicles during spermatogenesis^{25,26}. This may have implied that the Menotropin injection enhanced spermatogenic processes in the seminiferous tubule, stimulated the activities of the Leydig cells and the Sertoli cells, improving testosterone secretion by the Sertoli cells, enhanced the activities of the testosterone during spermatogenesis, thereby improving the quality and quantity of semen production²⁷. The significant improvements observed in the spermatozoa motility, spermatozoa live

proportion, semen concentration and the total number of sperm per ejaculate and the viability of the sperm cells following the administration of Menotropin showed a good reproductive potential and fertility in either normal mating or artificial insemination.

The results of the second semen collection (Table 3) showed that significant improvements ($p<0.05$) were also observed in the semen volume, spermatozoa mass motility, spermatozoa live proportion, the total number of sperm per ejaculate, total viable sperm and percentage normal, while abnormal spermatozoa significantly reduced ($p<0.05$) compared with the control group following the intramuscular injection of different doses of Menotropin. The semen volume of cocks in T_2 (0.69 mL), T_3 (1.00 mL) and T_4 (0.90 mL) are statistically similar and significantly higher compared to that of the cocks in T_1 (0.50 mL). The spermatozoa motility of the cocks in T_2 (82.52%) and T_3 (85.00%) are statistically similar ($p>0.05$) and significantly higher ($p<0.05$) compared to those of the cocks in T_1 (72.52%). The spermatozoa live proportion of cocks in T_1 (78.00%) and T_4 (81.20%) are statistically similar ($p>0.05$) but significantly lower ($p<0.05$) compared to the cocks in T_2 (88.90%) and T_3 (90.10%). The total number of spermatozoa per ejaculate and total viable spermatozoa in T_2 ($3.62 \times 10^9 \text{ mL}^{-1}$), T_3 ($4.81 \times 10^9 \text{ mL}^{-1}$) and T_4 ($3.55 \times 10^9 \text{ mL}^{-1}$) are significantly higher compared to those of control group T_1 ($2.15 \times 10^9 \text{ mL}^{-1}$). The percentage of normal sperm increased significantly ($p<0.05$) in T_2 (94.00%), T_3 (95.03%) and T_4 (92.90%) compared to T_1 (87.33%), while the total abnormal spermatozoa significantly ($p<0.05$) reduced simultaneously in the same trend following the administration of Menotropin across the treatment groups. The semen color, consistency and pH are statistically similar to those of the control group, while the semen concentrations of the cocks in T_1 ($4.30 \times 10^9 \text{ mL}^{-1}$) are statistically similar ($p>0.05$) to those in T_3 ($4.81 \times 10^9 \text{ mL}^{-1}$) and T_4 ($3.94 \times 10^9 \text{ mL}^{-1}$). These observations are similar to what was observed in Table 2. The Menotropin administered on the cocks might have stimulated the gonads/testicles to produce more testosterone which resultantly improved the overall spermatogenic processes^{26,28}.

The significant improvement observed in spermatozoa motility might be attributable to the observed improvements in spermatozoa live proportion, total viable spermatozoa and on the total number of spermatozoa per semen ejaculate. The tail of spermatozoa is responsible for spermatozoa motility^{4,24}. This significant improvement in spermatozoa motility might also be attributed to the reduction on the tail abnormalities of the spermatozoa following the administration of Menotropin (Table 4). The reductions in tail abnormalities of the spermatozoa in effect enhanced spermatozoa motility.

The results on the effects of Menotropin on seminal abnormalities of the cocks (Table 4) showed that the headless, double head, twisted tail spermatozoa, tailless spermatozoa and cytoplasmic droplets, significantly ($p < 0.05$) reduced as well as the bent mid-piece and the total abnormal spermatozoa following the intramuscular injection of the different doses of Menotropin on the cocks. The headless spermatozoa in T_2 (1.43%) and T_3 (1.41%) are statistically similar ($p > 0.05$) and significantly reduced compared to the cocks in T_1 (4.00%). The double-headed and the tailless spermatozoa in T_2 (2.04%), T_3 (2.15%) and T_4 (2.04%) are statistically similar ($p > 0.05$) and significantly lower ($p < 0.05$) compared to the cocks in T_1 (2.68). The twisted tail spermatozoa of T_3 (1.70%) and T_4 (1.48%) are statistically ($p > 0.05$) similar, the T_2 (2.10%) and T_3 (1.70%) are also statistically similar ($p > 0.05$) and significantly lower ($p < 0.05$) compared to the in the control group T_1 (3.82%). The bent mid-piece in T_2 (2.53%) and T_3 (2.27%) are statistically similar, the T_2 (2.53%) and T_4 (2.86) are also statistically similar ($p > 0.05$) but significantly lower ($p < 0.05$) compared to those in T_1 (4.87%). Furthermore, the total abnormality of the spermatozoa significantly ($p < 0.05$) reduced in the pattern, T_1 (22.33%), T_2 (12.48%), T_3 (11.96%) and T_4 (10.87%) as the dosage of the Menotropin administration in the cocks increased. These results showed that the Menotropin administered significantly reduced the overall spermatozoa abnormalities through different mechanisms, thereby improving the semen quality parameters. The Menotropin administered might have stimulated the hypothalamus to produce gonadotrophin releasing hormone through the feedback mechanisms and adrenergic receptor mechanism. It may have also been that the GnRH secreted from the hypothalamus stimulated the anterior pituitary to produce more FSH and LH, thus enhancing spermatogenic processes. The luteinizing hormones may also have acted by stimulating the Leydig cells to secrete testosterone, thus improving spermatogenic processes which resultantly reduced some spermatozoa abnormalities²⁹⁻³¹. It has been reported that FSH and LH increased testosterone secretion, which resultantly enhanced spermatogenesis and spermatozoa morphology³². This implied that Menotropin injected intramuscularly on the cocks enhanced the activities of the Leydig cells and the Sertoli cells at the seminiferous tubules during spermatogenesis. From this observation, it could be deduced that the intramuscular injection of Menotropin at these doses improved the semen quality of the cocks by reducing spermatozoa abnormalities. These observations are in line with the findings of Waheeb *et al.*³³, who reported that oral administration of gonadotrophin significantly reduced some spermatozoa abnormalities in bulls.

The results on the graded doses of Menotropin on the testicular dynamics of the cocks (Table 5) showed that only the paired epididymis morphometric parameter was significantly heavier ($p < 0.05$) compared to the control group. The paired epididymis weights in T_2 (1.12 g), T_3 (1.13 g) and T_4 (1.08 g) are statistically similar ($p > 0.05$) and significantly heavier ($p < 0.05$) compared to the paired epididymis weights of the cocks in the control group T_1 (1.01 g). The epididymis functioned for semen storage, spermatozoa maturation and semen transport^{34,35}. The significant increase observed in the weights of the paired epididymis of the cocks administered different doses of Menotropin might have resulted in improvements in the efficiency of spermatozoa maturation at the epididymis, increased semen storage capacity and may have also enhanced the efficiency of the semen transport from the epididymis to the urethra of the cocks.

CONCLUSION

Based on the results and observations from this study, it was concluded that the intramuscular injection of Menotropin at 0.10 and 0.20 mL had synergistic effects on the semen and testicular dynamics of the mature Nigerian indigenous cocks as it improved the semen characteristics and had no adverse effects on the testicular dynamics. The intramuscular injection of Menotropin at 0.10 mL on the mature Nigerian indigenous cocks produced the best results. Moreover, increasing the dosage of Menotropin up to 0.30 mL had some deleterious effects on some semen parameters of the cocks.

SIGNIFICANCE STATEMENT

Spermatogenesis involves the activities of the follicle stimulating hormone, luteinizing hormone in synergy with other male reproductive hormones. For effective and efficient reproduction in poultry, good quality and quantity of semen are paramount. This requires that the semen quantity and quality of the cocks must be optimum. There has been a serious decline in the semen quality of tropical animal species due to extreme tropical environmental conditions, climate change and malnutrition. As such, there is an urgent need to ameliorate this condition and improve the semen quality of our tropical animals using exogenous hormones. This research has proved that Menotropin at small doses can be used to enhance spermatogenesis, stimulate spermatogenic processes and improve semen quality of mature Nigerian indigenous cocks.

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