EFFECT OF CLOMIPHENE CITRATE ADMINISTRATION ON SEMEN CHARACTRIESTICS AND BIOCHEMICAL CONSTITUENT OF SEMINAL PLASMA OF NIGERIAN INDIGENOUS COCKS

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Abstract

A 90 day trial was conducted to evaluate the effect of Clomiphene citrate (Clomid®) on semen characteristics and biochemical constituents of seminal plasma of Nigerian Indigenous cocks. The experiment involved 48 Nigerian cocks aged 26 weeks. The birds were randomly divided into 4 treatment groups of 12 birds each. Each group was further divided into 3 replicates with 4 birds per replicates. The birds were assigned to 0, 10, 20 and 30mg Clomiphene citrate (Clomid®) treatments designated as treatments T_1, T_2, T_3 and T_4 respectively. T_1 (0.00mg) served as the control. The birds were fed ad-libitum with growers mash and water was also served ad-libitum. The results showed no significant difference ($P>0.05$) between T_2 (0.10ml) and T_1 (0.09ml) in semen volume. There was also no significant differences ($P>0.05$) between T_3 (0.14ml) and T (0.14ml). There were also significant differences among the treatment groups in mass motility, individual motility, sperm concentration and in dead/abnormal sperm cells. The sperm colour ranged from creamy white to light yellow as the Clomiphene citrate (Clomid®) increased in the treatment groups. Significant differences ($P<0.05$) were observed in seminal plasma. Total protein decreased significantly from 7.20g/l in T_1 (0.0mg) to 6.60g/l in T_4 (30mg). On the other hand, biocarbonate concentration increased from 13.40mmol/l in T_1 to 15.60mmol in T_4. So also, was the concentration of citric acid which increased from 150.21mg/100ml to 450.18mg/100ml. From the results recorded in this experiment, Clomiphene citrate (Clomid®) has no negative effect on the semen characteristics and seminal plasma constituent of Nigerian indigenous chicken when 20mg dosage is used.

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Key words: Nigerian indigenous cocks, Clomiphene citrate (Clomid®), semen and seminal plasma

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Introduction

The world wide acceptance of poultry and a special preference of products from the local birds can mainly be attributed to their unchanged composition pigmentation taste and sustainability for special dishes (Horst, 1990).

Scavenge flocks of chickens are kept by 80-90% of families in Nigeria (Sonaiya, 1990). Family poultry is based largely on locally adapted chicken. The earliest effort to improve locally adapted chicken in Nigeria dates back to 1919 and used the (hybrid) cockerel exchange failed due to inability of the hybrids to adapt well to the scavenging system (Sonaiya, 1990).

In the course of enhancing the performance of existing population, there is the need to characterize the reproductive pattern of the Nigerian domestic fowl. Drugs, especially gonadotropins have been used extensively to boost reproduction in other classes of livestock as reported in literatures. A lot of these treatments have evidently produced positive results in female goats, sheep and rabbits. As reported by Iheukwumere *et al.*, (2002) and Herbert *et al.*, (2000) on the indigenous chicken with specific reference to Nigerian chicken.

The application of modern reproductive techniques, such as the artificial insemination, hormonal drug treatments from semen quality and hormonal improvement and others adopted as in large scale in Nigeria. This study as therefore developed to investigate the effect of Clomiphene citrate (Clomid®) on the semen characteristics and semen plasma of Nigerian indigenous cocks.

Material and Methods

The experiment involved 48 matured normal feathered local cocks aged 24 to 26 weeks. The 48 cocks were randomly divided into 4 treatment groups coded T_1, T_2, T_3 and T_4 of 12 cocks per treatment in a completely randomized design which were further replicated into 3 of 4 birds per replicate in each treatment group. The birds received 4 levels of Clomiphene citrate as treatments. The levels were 0mg, 10.00mg, 20.00mg and 30.00mg representing T_1, T_2, T_3 and T_4 respectively. Treatment one (T_1), which contained no Clomiphene citrate (Clomid®) served as the control. Clomiphene citrate (Clomid®) was administered orally-through water.

The birds were managed intensively and housed in deep litter system. The birds were fed ad-libitum with growers mash and water served ad-libitum as well. Clomiphene citrate (Clomid®) treatment lasted for five days.

Semen/Data Collection

Semen collection was done one week after Clomiphene citrate (Clomid®administration). The cocks ejaculated twice a week-Tuesdays and Fridays, between 8.00am and 10.00am for four weeks. This was done by abdominal massage techniques. Semen evaluation commenced immediately after semen collection for semen colour, semen volume, mass motility, individual motility, sperm concentration and live sperm percentage. While Biochemical constituents of seminal plasma was estimated by subjecting samples of semen collected to laboratory analysis for the following parameters: total protein, albumin, globin, sodium, chloride, potassium, bicarbonate, phosphate, citric acid and inositol.

statistical analysis

The data collected were subjected to analysis of variance (ANOVA) using the techniques of Steel and Torrie (1980). Significant treatment means were separated using Duncan's New Multiple Range Test as described by Obi, (1990).

Results and Discussion

The results (Table 2) revealed that there were no significant differences (P>0.05) between the control (0.09ml) and T_2 (0.10ml) in semen volume. There were also no significant difference (P>0.05) between T_3 (0.14ml) and T_4 (0.14ml) in semen volume. The result generally showed significant differences (P>0.05) in pH, mass motility, individual motility, sperm concentration, live sperm cell, dead/abnormal sperm cell and deviation/change in colour of semen as the level of clomiphene citrate increased in the treatment groups. The result of this study showed increase in semen volume as the level of Clomiphene citrate (Clomid®) increased in the treatment groups, this coincide with the report of Nwagwu *et al.*, (1995) and Keshin *et al.*, (1997) who observed increased volume of semen with increase in fertility drug. These authors reported normal range of volume of whole ejaculate for chicken as 0.08ml to 0.58 and also within the range observed by Abu *et al.*, (2006). The range of the pH was between 7.22 in T_1 and 8.19 in T_2 and T_3 which coincided with the normal range (7-8) as reported by Meecham (2002). However, the result recorded in mass motility 85.00% in T_4 which was the highest value, and values recorded in T_2 71.00% and T_3 78.00% were comparable to report of Oguike *et al.*, (2000) who reported 77-80% in mass motility, but higher than report of Abu *et al.*, (2006) and Ameh (2004) who reported values between 36-52%, while Iheukwumere *et al.*, (2008) reported values ranging between 54 and 55.5% respectively. The differences observed in sperm motility may be as a result of breed (Oguike *et al.*, 2000) or cold shock (Iheukwumere and Okere, 1990). The range of 2.09 x 10^9sperm/ml in T_1 and 2.04 x 10^9sperm/ml in T_4 recorded were similar to earlier reported sperm concentration of 1.18 to 2.13 x 10^9 sperm/ml, 1.98 to 2.10^9sperm/ml (Ezekwe *et al.*, 2000; Oguike *et al.*, 2000) and 2.01 x 2.08 x 10^9 sperm/ml (Iheukwumere *et al.*, 2008) but lower than value of 3.8 x 10^9sperm/ml reported by Charlov (1970), but higher than value of 1.80 x 10^9sperm/ml reported by Ameh (2004). The difference in sperm concentration could be as a result of breed (Oguike *et al.*, 2000) or hormonal administration (Abu *et al.*, 2006). The highest percentage of live sperm cell obtained was recorded in T_2 (10mg) 52.80% while the lowest was recorded in T_3 (20mg) 50.08%. The average ranged from 50.06 to 52.80% which were within the range of 44.2 to 59.2% reported by Abu *et al.*, (2006) and 51.3-54.4% reported by Iheukwumere *et al.*, (2008) but lower than the values 58.02 reported by Oguike *et al.*, (2002). The highest percentage (21.07%) of dead sperm cell was recorded in the control and this differed significantly (P<0.05) from T_2, T_3 and T_4 with 17.64%, 18.03 and 18.16% respectively. This results compares with findings of Iheukwemere *et al.*, (2002) who reported 17.1 to 20.8% in normal naked neck and frizzle strains of cocks, but in contrary with the findings of Ameh (2004) who reported 14.25%. Ejaculate and consistency were quite variable as the levels of Clomiphene citrate (Clomid®) increased, some deviated from milky colour to yellow as the level increased. Ezekwu *et al.*, (2008) gave credence to this findings. However, in table 2, there were significant differences (P>0.05) in Total protein, Albumin, Globulin, Sodium, Chloride, Potassium, Bicarbonate, Phosphate, Citric acid and Inositol in the seminal plasma of the birds. According to the report of Makkar and Bedcer, (1996), seminal protein is usually a reflection of the total protein quality in the feed. Values recorded indicates decrease in the protein as the level or Clomiphene citrate (Clomid®) increased in the treatment groups. The value recorded in the albumin 3.10g/l to 3.50g/l was in contrast to the value 2.06g/l to 2.88g/l recorded by Olabode and Onyimonyi, (2013). While the value recorded in albumin 2.40 to 3.90 in this work were in agreement with values of 3.53g/dl reported by Olabode and Onyimonyi,

(2013) and 3.25 reported by Alabi *et al.*, (2011) on broilers. The values in the levels of Bicarbonate increased as the levels of Clomiphene citrate increased, which means that Clomiphene citrate (Clomid®) encourages sperm motility since sodium in seminal plasma stimulates sperm motility (Okamura *et al.*, 2006). Also there was increase in citric acid as the level of Clomiphene citrate (Clomid®) increased in the treatment groups, which indicate that Clomiphene citrate (Clomid®) encourages sperm motility since vitamin c plays important role in enhancing sperm motility. The inositol followed the same trend with citric acid that is increased as the level of Clomiphene citrate (Clomid®) increased.

Conclusion

From the above findings, it implies that the secretory ability of the structure responsible for contributing to the biochemical constituent of seminal plasma were not negatively affected rather it boosted both the sperm production and enhanced motility of the sperm.

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Table 1: Effect of clomiphene citrate on semen characteristics of Nigerian indigenous chicken.

Parameters	Treatment (Clomiphene citrate)				
	$T_1$0.00 mg	$T_2$10 mg	$T_3$20 mg	$T_4$30 Mg	SEM
Semen Volume (ml)	0.09^b	0.10^b	0.14^a	0.14^a	0.01
pH	7.22^b	8.19^a	8.19^a	8.14^a	0.19
Mass motility (%)	57.00^d	71.00^c	78.00^b	85.00^a	5.98
Individual motility (%)	2.24^b	2.58^b	3.00^a	2.25^c	0.16
Sperm concentration (x 10^a sperm/ml)	2.09^a	2.07^b	2.05^c	2.04^c	0.01
Live sperm cell (%)	51.06^c	52.80^a	50.08^c	52.19^b	0.47
Dead/abnormal sperm cell	21.07^a	17.64^b	18.03^b	18.16^b	0.98
Semen colour	Creamy white	Light yellow	Light yellow	Light yellow	

a-d: Means in the same row with different superscript are significantly different (P<0.05)
SEM = Standard Error of Mean

Table 2: Seminal Plasma Constituent of Local Cocks Administering Clomiphene Citrate (Clomid $^{(R)}$)

PARAMETERS	T_1	T_2	T_3	T_4	SEM
Total Protein (g/L)	7.20^a	7.10^a	6.80^b	6.60^c	0.14
Albumin (g/L)	3.50^a	3.20^b	3.40^a	3.10^b	0.16
Globulin (g/dl)	3.70^b	3.90^a	2.40^d	3.10^c	0.24
Sodium (mmol/L)	134.00^a	133.14^a	131.16^b	131.35^b	1.20
Chloride (mmol/L)	132.40^a	130.34^b	132.10^a	130.25^b	0.57
Potassium (mmol/L)	1.43^b	1.65^a	1.42^b	1.63^a	0.06
Bicarbonate (mmol/L)	13.40^d	14.30^c	15.06^b	15.60^a	0.48
Phosphate (mmol/L)	17.25^a	16.50^b	16.30^b	16.40^b	0.34
Citric Acid (mg/100ml)	150.21^c	224.34^b	380.00^a	420.18^a	63.87
Inositol (mmol/L)	30.00^d	34.10^c	36.21^b	38.45^a	1.80

a,b,c,d values within each row, treatment with the same superscripts are not significantly different (P>0.05)