Journal of Animal and Veterinary Advances 5 (11): 946-948, 2006

Effect of Cottonseed Cake and Clomiphene Citrate Administration on the Reproductive Characteristics of Hacco Cocks

O.A. Amao, V.A. Togun, A.F. Akindele, I.O. Oladunjoye and G.O. Tona
Department of Animal Production and Health, Ladoke Akintola University of Technology,
P. M. B. 4000, Ogbomoso, Oyo State, Nigeria

Abstract: Three hundred (300) 8-weeks old cockerels were used in a 5×2 factorial experiment to determine the effect of cottonseed cake and clomiphene citrate administration on some reproductive characteristics of the cockerels. They were randomly allocated to ten treatments of 30 birds per treatment and replicated into 3, each replicate containing 10 birds. Treatments contained cottonseed cake (CSC) meal at 0, 25, 50, 75 and 100% replacement levels for groundnut cake which were either supplemented with clomiphene citrate or not. Treatments 1 to 5 (D_1, D_2, D_3, D_4 and D_5) were not supplemented with clomiphene, while treatments 6 to 10 which had the same inclusion levels of CSC (D_6, D_7, D_8, D_9 and D_{10},) were supplemented with clomiphene citrate. Results showed that treatment had significant ($p<0.05$) effect on testis weight, parenchyma weight and testis volume. Birds on treatments 2 and 7 (D_2 and D_7) with 25% CSC had similar testis weight and testis volume with those on the control diet (D_1). However, they had significantly ($p<0.05$) higher values for these two parameters than those on other treatments. Degeneration and necrosis of germcells did not follow any definite pattern but varied in degree of severity. Severity of condition appeared to be higher for dietary treatments without clomiphene citrate administration than for those with clomiphene administration. It could be concluded that cottonseed cake could replace up to 25% of GNC in the diets of growing cocks when it is supplemented with clomiphene citrate administration.

Key words: Cottonseed cake, clomiphene citrate, reproductive characteristics, cocks

INTRODUCTION

The major challenge facing the animal producers in the developing countries including Nigeria is the supply of animal protein to meet the increasing need of a growing populace. The production of animal protein is affected by the scarcity and productive cost of feed ingredients. Available foodstuffs to humans are being stiffly competed for by domestic animals. Thus the prices of conventional feedstuffs such as grains and legumes are becoming more prohibitive.

Cottonseed cake is a high-protein feedstuff that is widely used in ruminant feeding and it is relatively cheaper than the popular protein feed ingredients such as soyabean and groundnut cake. However, its use in monogastric rations is limited because of the presence of gossypol, which is a naturally occurring polyphenolic factor.

Gossypol has been reported to have adverse effects on reproductive performance of male animals[1-4]. According to Randel *et al.*[5], at effective doses, gossypol causes male infertility because of sperm immotility and depressed sperm counts. Specific mitochondrial damage in the tails of spermatozoa tends to render them immotile and extensive damage to germinal epithelium may be responsible for depressed spermatogenesis. Groundnut cake and soyabean meal are the major ingredients which cottonseed cake could replace in monogastric rations, but certain nutritional bottleneck of cottonseed cake (occasioned by gossypol) must be addressed.

The possibility of using clomiphene citrate (Clomid) for the stimulation of spermatogenesis was proposed in[6]. However, there is paucity of information on its use to counteract the effect of gossypol in poultry diets.

Clomiphene citrate is a drug popularly used to induce ovulation. However, it may be used to treat low sperm counts (oligospermia) in males[7]. According to British Pharmaceutical society[6], clomiphene citrate acts in males and females by stimulating the production of pituitary gonadotrophins. Herbert *et al.*[8] reported that clomiphene citrate induced spermatogenesis in rabbits that were so treated. They concluded that the administration of 6.25 mg of Clomid would improve the sperm production in rabbits without deleterious effects on sperm morphology and physiology.

Corresponding Author: O.A. Amao, Department of Animal Production and Health, Ladoke Akintola University of Technology, P. M. B. 4000, Ogbomoso, Oyo State, Nigeria

This study was therefore conducted to determine the effect of clomiphene intervention on the reproductive characteristic of cocks fed diets containing varying levels of cottonseed cake.

MATERIALS AND METHODS

Three hundred 8-weeks old cockerels previously maintained on a standard chick mash were used for this study. They were randomly allocated to ten treatments of five dietary allocations and two levels of clomiphene citrate (+ drug and − drug). Diets were compounded such that cottonseed cake (CSC) replaced 0, 25, 50, 75 and 100% groundnut cake (GNC). Five diets D_1 (0%CSC), D_2 (25%CSC), D_3 (50%CSC), D_4(75%CSC) and D_5(100% CSC) were not supplemented with clomiphene citrate, while the other five diets (D_6 to D_{10}) with the same levels of CSC as D_1 to D_5 were supplemented with clomiphene citrate at the rate of 5 mg 70 kg^{-1} body weight Table 1. Clomiphene citrate was administered through drinking water for the first five days of the commencement of the experiment. Second priming was administered for another 5 days beginning from 17days to the end of the experiment. Birds were managed on deep litter pens. Treatments were replicated into 3 with ten birds per treatment. Feed and water were given to birds *ad-libitum* throughout the experimental period. Birds were weighed at the beginning of the experiment and thereafter once per week. Routine health practices like deworming, vaccination and changing of litter were carried out as and when necessary. Feeding trial lasted ten weeks.

Five birds per treatment were randomly picked and slaughtered after the experiment. Testes were carefully removed and testicular parameters namely testis weight, tunica albuginea weight, parenchyma weight, testis length, testis width and testis volume were measured. Means of left and right testes parameters were computed and used for analysis. Samples of testis were also fixed for histopathological analysis. Data collected were subjected to analysis of variance (ANOVA) using a 5×2 factorial in completely randomized design of SPSS computer software[9]. Significant differences between means were separated by Duncan's option of the same software.

RESULTS AND DISCUSSION

The results of this study are summarized in Table 2 and 3. Table 2 shows the effect of cottonseed cake with or without clomiphene administration on testis characteristics of cocks. Cottonseed cake with or without clomiphene citrate administration had significant ($p<0.05$) effect on testis weight, parenchyma weight and testis volume. Diets 2 and 7 (D_2 and D_7) were not significantly different from each other and from the control diet (D_1). These two diets had CSC replacing 25% of GNC. However, D_7 which also had clomiphene citrate administration resulted in highest ($p<0.05$) testis weight. This is reflected in the testis volume which tended to follow the same pattern, suggesting ability to produce larger volume of semen. Tunica albuginea, testis length and testis width were not significantly ($p>0.05$) affected by treatments. This implies that cottonseed cake with or without clomiphene citrate seems to have more impact on

Table 1: Gross composition of experimental diets and calculated nutrients

	− Clomid					+ Clomid				
Ingredient (%)	D1 (0%CBC)	D2 (25%CSC)	D3 (50%CSC)	D4 (75%CSC)	D5 (100%CSC)	D6 (0%CSC)	D7 (25%CSC)	D8 (50%CSC)	D9 (75%CSC)	D10 (100%CSC)
Maize	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00
Wheat offal	8.00	10.00	16.00	25.00	32.00	8.00	10.00	16.00	25.00	32.00
GNC	12.00	9.00	6.00	3.00	-	12.00	9.00	6.00	3.00	-
Fishmeal	1.00	1.00	1.00	1.20	1.30	1.00	1.00	1.00	1.20	1.30
Cottonseed cake	-	3.00	6.00	9.00	12.00	-	3.00	6.00	9.00	12.00
Corn bran	32.00	30.00	24.00	15.00	8.00	32.00	30.00	24.00	15.00	3.00
PKC	12.00	12.00	12.00	12.00	12.00	12.00	12.00	12.00	12.00	12.00
Rice bran	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00	16.00
Vit. min^{-1} Premix	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Salt	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Lysine	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Methionine	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Bone meal	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Oyster shell	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Total	100.60	100.60	100.60	100.20	100.90	100.160	100.60	100.60	100.20	100.90
Calculated nutrients										
Crude protein	16.68	16.12	15.96	16.04	15.85	16.68	16.12	15.96	16.04	15.85
Metabolizable Energy (ME) (Kcal kg^{-1})	2563.04	2535.62	2483.00	2417.20	2372.28	2563.04	2535.62	2483.00	2417.20	2372.04

Table 2: Effect of cottonseed cake with or without clomiphene administration on testis characteristics of cocks

	- DRUG					+ DRUG					
Parameter	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10	SEM
Testis weight (g)	1.82^{a}	1.98^{a}	1.53^{b}	0.92^{c}	1.80^{a}	0.99^{c}	2.32^{a}	1.62^{b}	0.77^{c}	0.82^{c}	0.58
Tunica albuginea weight (g)	0.13	0.11	0.11	0.07	0.16	0.08	0.14	0.08	0.06	0.10	0.05
Parenchyma weight (g)	1.39^{b}	1.63^{b}	0.81^{c}	0.72^{c}	1.56^{b}	0.75^{c}	2.15^{a}	0.79^{c}	2.15^{a}	0.77^{c}	0.62
Testis length (mm)	2.28	2.01	2.01	1.79	1.72	1.67	2.46	1.95	1.67	1.48	0.29
Testis width (mm)	1.11	0.79	1.01	0.90	0.98	0.83	1.35	1.09	0.87	0.90	0.18
Testis volume (cm^{3})	1.15^{a}	1.47^{a}	1.10^{ab}	0.52^{c}	0.58^{c}	1.00^{b}	1.22^{b}	1.01^{b}	0.73^{c}	0.65^{c}	0.48

$^{a, b, c:}$ Means along the same row with different superscripts differ significantly ($p<0.05$)

Table 3: The degree of degeneration and necrosis of germ cells in relation to the level of cottonseed cake in diet with or without clomiphene (Clomid) administration

	- Clomid					+ Clomid				
Pathological condition (%)	0% D_1	25% D_2	50% D3	75% D_4	100% D_5	0% D_6	25% D_7	50% D_8	75% D_9	100% D_{10}
Absent	0	0	0	0	0	20	0	0	0	0
Very mild	0	0	0	0	0	0	0	0	0	0
Mild	20	0	40	20	0	60	20	20	20	20
Moderate	0	0	20	0	20	0	40	40	0	20
Severe	80	100	40	80	80	20	40	40	80	60
Total	100	100	100	100	100	100	100	100	100	100s

the parenchyma tissue than on other tissues of the testis. Thus the differences ($p<0.05$) in testis weight and volume could be largely attributed to significant differences in parenchyma weights.

Table 3 shows the degree of degeneration and necrosis of germ cells in relation to the level of cottonseed cake in diet with or without clomiphene citrate administration. All treatments resulted in one form of degeneration and necrosis of germ cells or another. However they did not follow a definite pattern. Severity of the condition seemed to be higher for dietary treatments without clomiphene citrate administration than those with clomiphene administration. D_2 (25% CSC replacing GNC) had 100% severe pathological condition. The implication of this is that the bigger size of testis does not necessarily mean a physiologically healthy testis for good semen production. This condition would definitely have adverse effect on sperm production. According to Randel *et al.*[5], extensive damage to germinal epithelium may be responsible for depressions in spermatogenesis. However, at the same CSC replacement level (25%CSC), with Clomid administration (D7), severity of pathological condition reduced to 40%. This shows that at low a replacement level of CSC for GNC, clomiphene citrate administration could be effective in counteracting the adverse effect of gossypol on the testis of cocks.

It could be concluded from this study that cottonseed cake could replace up to 25% of GNC in the diets of cocks when it is supplemented with clomiphene citrate.

REFERENCES

1. Arshami, J. and J.L. Ruttle, 1988. Effects of diets containing gossypol on spermatogenic tissue of young bulls. Theriogenol., 30: 507-516.
2. Chase, C.C.Jr., P. Bastidas, J.L. Ruttle, C. R. Long and R.D. Randel, 1989. Reproductive development and functions of Brahman bulls fed diets containing gossypol. Growth and semen parameters near puberty. J. Anim. Sci., 67: 368 (Abstr).
3. Brocas, C., R.M. Rivera, F.F. Paula-Lopex, L.R. McDowell and M.C. Calhoun, *et al*., 1997. Deleterious actions of gossypol on bovine spermatozoa, oocytes and embryos. Biol. Reprod., 57: 901-907.
4. Meyer, R.O. and L.R. McDowell, 2003. Potential for gossypol toxicity when feeding whole cottonseed to beef cattle http: // edis. fas. ufl..edu.
5. Randel, R.D., C.C.Jr. Chase and S.J. Wyse, 1992. Effects of gossypol and cottonseed products on reproduction of mammals. J. Anim. Sci., 70: 1628- 1638.
6. British Pharmaceutical Society, 1973. British Pharmaceutical Codex London. The Pharmaceutical Press, pp: 117.
7. Wael, M.B., 1999. Fertility in rams treated with clomiphene citrate. Morphology Arch. Androl., 27: 109-111.
8. Herbert, U., A.N. Ezeobi and M.U. Iloeje, 2002. Induction of Spermatogenesis in Rabbits Using the Fertility Drug, Clomiphene Citrate (Clomid®). Proc. 27th Annual Conf., Nig. Soc. for Anim. Prod. (NSAP), March 17-21, 2002. Fed. Univ. of Tech. Akure. Nigeria, pp: 25-27.
9. Field, A., 2000. Discovering statistics using SPSS. Pub. London, pp: 243-322.