



Ameliorative Potential of Coconut Water in Overfed Male Broiler Breeders

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Abstract | Male broiler breeders are prone to excessive feed consumption from overfeeding due to poor management practices. There are inconsistent reports on susceptibility of male broiler breeders to obesity which results from excessive feed consumption. Coconut water is reportedly useful in reducing weight gain and inflammation. Thirty male broiler breeders of 12 weeks old were separated into three groups A, B and C and reared to twenty weeks of age. Group A was reared on recommended feeding regimen while both groups B and C received double of the recommended feeding regimen. Birds in both groups A and B were given ordinary water while birds in group C received 100 ml of coconut water / L. Blood samples and testes were collected at the end of 20th week to evaluate serum interleukin-6, total protein, leptin and testicular interleukin-1beta levels. Mean \pm SEM values were calculated and compared for significant differences with ANOVA at $p < 0.05$. Serum interleukin-6 and total protein levels were not significantly different among the groups. Group A had the lowest leptin concentration but was not significantly different from values observed in groups B and C. Interleukin-1beta decreased significantly in groups A and C when compared with group B. The above results revealed that overfed male broiler breeders are less predisposed to obesity but have tendency to develop local inflammation which can be attenuated by coconut water supplementation.

Keywords | Male broiler breeder, Coconut water, Interleukins, Leptin and Total Protein.

Received | June 25, 2019; **Accepted** | August 19, 2019; **Published** | December 22, 2019

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Citation | Bello II, Jubril ATJ, Balogun HA, Omowon AA (2020). Ameliorative potential of coconut water in overfed male broiler breeders. *Adv. Anim. Vet. Sci.* 8(1): 41-45.

DOI | <http://dx.doi.org/10.17582/journal.aavs/2020/8.1.41.45>

ISSN (Online) | 2307-8316; **ISSN (Print)** | 2309-3331

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INTRODUCTION

Modern breeds of broilers are selected for increased growth rate, breast-meat yield and feed conversion rate (Arnould and Leterrier, 2007; Estevez, 2007). The consequences of this genetic selection for improved breed of broiler include but not limited to voracious appetite (Richard et al., 2010), cardiovascular disease (Mitchell, 1997), lameness (Knowles et al., 2008). Old report showed that broilers kept for breeding had health problems such as obesity (Dunnington and Siegel, 1985; Hocking and Bernard, 1997) but the less predisposition of modern breed of broiler breeder to obesity due to genetic improvement for increased meat yield has been demonstrated (Havenstein et al., 2003). However, there is a general consensus on the predisposition of male broiler breeders to overweight, a

condition that can result from voracious appetite or overfeeding. Presently, there is no report indicating genetic improvement on the voracious appetite of modern breed of broilers. Feed restriction is the only available tool used by poultry farmers to control voracious appetite and attendant overweight in the birds (Mench, 2002). Excessive body weight gain in broiler breeders affected reproductive performance (Goerzen et al., 1996; Romero-Sanchez et al., 2007).

Obesity is associated with a state of chronic low-grade systemic inflammation (Xu et al., 2003; Lee et al., 2013). It is associated with increased production of pro-inflammatory cytokines such as interleukin-1beta (IL1 β) and interleukin-6 (Ken et al., 2001; Vozarova et al., 2001) and other acute phase proteins (Wellen and Hotamisligil, 2003; Fain, 2006).

Reports revealed that both visceral and subcutaneous adipose tissues produce IL-6 but IL-6 level is higher in subcutaneous adipose tissue (Marta et al., 2015). The inhibitory effect and modulatory role of IL1 β on the production of testosterone by Leydig cells has been reported (Pinto-Fochi et al., 2016; Jie et al., 2017). Leptin, a polypeptide hormone produced by adipocytes, links alteration in body energy store to adaptive responses in the central regulator of energy balance (Ahima et al., 2000; Myers et al., 2009). The amount of leptin synthesized and secreted is known to increase in proportion to the accumulation of body fat mass (Liuzzi et al., 1999). Increased circulating level of leptin is seen in obese animals (Considine et al., 1996; Rosenbaum et al., 1996) and overfed broiler breeder hens (Chen et al., 2006). The total protein in serum is made of albumin, globulin and fibrinogen. Acute phase proteins such as C-reactive proteins are also found in the serum during inflammation.

Elevation in serum total protein level has been reported in obese individual with diabetes (Riaz et al., 2009). A study also observed insignificant changes in serum total protein level of obese and overweight individuals (Madhuvanthi and Lathadevi, 2016).

Coconut water is the liquid endosperm found at the centre of coconut fruit. It contains antioxidants such as vitamin C, vitamin B1, vitamin B6, amino acids, selenium, cytokines and minerals (Bhagya, 2012) and flavonoid. Flavonoids are phytochemicals with potent anti-inflammatory effect (Ilavarasan et al., 2005). Plants that possess antioxidant property have also been reported to display anti-inflammatory potential (Nguemfo et al., 2009; Rathee et al., 2009). Report has shown anti-inflammatory property of the juice (Rao and Najam, 2016). A report indicated that coconut water attenuated leptin level in high calorie fed animals (Imaga et al., 2016). There are also anecdotal reports on the usefulness of the juice in reducing weight gain and building lean muscle.

The consistent data on the predisposition of broiler breeder to overweight, susceptibility of the birds to excessive feed consumption resulting from poor management and the inconsistent data on the susceptibility of male broiler breeder to obesity spurred the interest for this research. This research was done to evaluate the predisposition of overfed male broiler breeders to obesity via assessment of obesity markers and ameliorative ability of coconut water.

MATERIAL AND METHODS

This experiment was carried out at the Poultry Unit of Teaching and Research Farm in Oyo State College of Agriculture and Technology, Igboora.

Male broiler breeders, coconut water, ELISA kits, sample bottles, blood and testes samples, graph papers, de-ionized water were used.

Equipment used for this research include scapel and blade, centrifuge (Techmel and Techmel, USA), micropipette, spectrophotometer, ELISA plate, ELISA reader, cold centrifuge (CR21D11), plate shaker, pipette tips, scissors, freezer.

EXPERIMENTAL BIRDS GROUPINGS AND MANAGEMENT

Thirty male broiler breeders of twelve weeks old were obtained from a commercial farm in Ibadan, Nigeria. The birds were randomly divided into three groups (A-C) of ten birds each. The birds in group A were raised with ordinary water and on standard feeding regimen of 87 g/bird, 92 g/bird, 97 g/bird, 102 g/bird, 107 g/bird, 112 g/bird, 117 g/bird and 122 g/bird at 13 weeks to 20 weeks respectively. The birds in group B were given double of standard ration and ordinary water while in Group C, the birds were reared on double of standard ration and inclusion of coconut water at 100ml/ L of water. All experimental protocols were in compliance with Institutional Ethics Committee on Research in Animals as well as internationally accepted principles for animal use and care.

COLLECTION OF BLOOD AND SERUM SAMPLES

At the end of 20th week, all the birds in each group were bled via wing vein puncture into plain bottles and allowed to clot in other to get sera (Mellisa, 2009). Serum was separated by centrifuge at 2000-3000 RPM for 20 minutes to assess leptin, interleukin-6 and serum total protein concentration.

COLLECTION AND HOMOGENISATION OF TESTES

All the birds in each group were euthanized by cervical dislocation. One of the testes removed from each birds were put in organ bottles containing phosphate buffer solution for homogenization (10 ml of PBS for every 1 g of testis). The homogenate was centrifuged for 5 minutes at 5000xg to get the supernatant for interleukin-1beta analysis.

ANALYSIS OF INTERLEUKINS AND LEPTIN

Serum concentration of interleukin-1beta and testicular interleukin-6 level were determined using ELISA- E-EL-ROO12 (ElabScience, USA) and E0135Ra (Bioassay Technology Laboratory, USA) respectively. The ELISA assay tests operate on the basis of competition between Avidin-Horseradish Peroxidase (HRP) conjugate and the interleukins in the sample for a limited number of binding sites on the antibody coated plate. The optical density (OD) is measured spectrophotometrically at a wavelength of 450 nm \pm 2 nm. The OD value is proportional to the concentration of IL-1 β and IL-6. The concentrations of

Table 1: Comparison between Levels of IL-6, Leptin and Total Protein in the Serum and IL-1beta level in the Testes for Overfed Groups and Control

Parameter	Group A	Group B	Group C
Serum			
IL-6 (pg/ml)	7.39 ± 0.76	7.12 ± 0.41	7.90 ± 0.25
Leptin (ng/ml)	5.44 ± 0.31	7.04 ± 0.59	6.23 ± 0.47
Total Protein (g/dl)	5.23 ± 0.34	4.78 ± 0.14	4.89 ± 0.25
Testes			
IL-1beta (pg/ml)	131.81 ± 66.23*	641.98 ± 172.98	122.07 ± 44.98*

* indicates significant difference when groups A and C were compared with group B

both IL-1 β and IL-6 in the samples were calculated by comparing the OD of the samples to the standard curve. Serum concentration of leptin was analysed using ELISA- E0561Ra (Bioassay Technology Laboratory, USA). The ELISA tests operate on the basis of competition between Streptavidin- Horseradish Peroxidase (HRP) conjugate and the leptin in the sample for a limited number of binding sites on the antibody coated plate. The optical density was measured using microplate reader set to 450 nm within 10 minutes after adding the stop solution. The concentration of leptin in the samples was calculated by comparing the OD of the samples to the standard curve.

ANALYSIS OF SERUM TOTAL PROTEIN

Biuret method was used to determine total protein. The test principle is based on copper ion reacting in alkaline, with protein peptide bonds to give purple coloured biuret complex. The amount of complex formed is directly proportional to the amount of protein in the specimen or sample.

DATA ANALYSIS

Quantitative data collected from this experiment was statistically analyzed with SPSS (Version 16.0, 2007). Differences between the means were tested with independent ANOVA. The results from each group are expressed as mean \pm standard error of mean (S.E.M) and means were compared for significant differences at $P < 0.05$.

RESULT AND DISCUSSION

As shown in Table 1 the serum interleukin-6 concentration recorded in group A was not significantly different from values recorded in groups B and C. There was a decrease in the leptin level in Group A and C, which were not significantly different from leptin value observed in group B. The highest level of total protein level was recorded in group A but this was not significantly different when compared with the total protein levels observed in groups B and C. The testicular interleukin-1beta concentration, the highest value was observed in group B, decreased significantly in group A and C when compared

with value observed in group B.

The insignificant difference in the serum IL-6 level of group A when compared with groups B and C showed that the three groups had similar adiposity especially subcutaneous form. This proposition is premised on a finding that revealed high level of IL-6 in subcutaneous adipose tissue relative to visceral adipose tissue (Marta et al., 2015).

The serum leptin concentrations that were not significantly different in the three groups might have resulted from the conversion of excessive feed to meat rather than fat in overfed groups. The amount of leptin synthesised and secreted is known to increase in proportion to the accumulation of body fat mass (Liuzzi et al., 1999). The finding in this present study is corroborated by the report on the less predisposition of modern breed of broiler breeder to fat pad due to genetic improvement for increased meat yield corroborates (Havenstein et al., 2003).

The absence of significant difference in serum total protein level among the three groups could be a pointer to absence of obesity and associated inflammation in the birds even in face of overfeeding. Obesity is associated with increased serum level of proteins such as interleukin-6 and other acute phase proteins (Wellen and Hotamisligil, 2003; Fain, 2006). A study has also demonstrated insignificant changes in serum total protein of overweight and obese individuals (Madhuvanthi and Lathadevi, 2016). The result in this study further lends support to the report on the low predisposition of modern broiler breeders to obesity.

The significantly higher level of testicular interleukin-1beta in group B could have resulted from their accessibility to higher ration of feed and its attendant local inflammation in the testis. Interleukin-1beta has been shown to mediate leptin-induced local inflammation (Santana et al., 1996; Luheshi et al., 1999). Study revealed elevated IL-1 β level in ovary of *ad-libitum* fed broiler (Zu-Chen et al., 2014) and testis of obese mice (Jie et al., 2017). The significant decrease in testicular IL-1 β level observed in group C relative to group B suggests less local inflammation seemingly due to coconut water supplementation in the group. This

result aligns with the report on anti-inflammatory property of coconut water (Rao and Najam, 2016).

Going by the above results, it is plausible to conclude that overfed male broiler breeders are less predisposed to obesity but have tendency to develop local inflammation which can be attenuated by coconut water supplementation.

CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest.

ETHICAL APPROVAL

All experimental protocols were in compliance with internationally accepted principles for animal use and care.

AUTHORS CONTRIBUTION

Idris I. Bello conceptualized this work. Idris I. Bello and Afusat T. Jagun Jubril were actively involved in the execution of the project and writing of manuscript. Halimah A. Balogun carried out the laboratory analysis. Olubukola D. Adedipe helped with statistical analysis.

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