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National Animal Production Research Institute
Ahmadu Bello University
P.M.B 1096, Shika-Zaria,
Kaduna State, Nigeria.
Email: japr@napri.gov.ng Url:journals.napri.gov.ng

CARCASS, VISCERAL AND IMMUNE ORGAN RESPONSES TO DEXAMETHASONE-INDUCED STRESS IN BROILER CHICKENS

[*1]Ademu, L. A., [2]Daudu, O. M., [3]Barje, P. P., [3]Iyeghe-Erakpotobor, G. T.
*[*1]Department of Animal Production and Health, Federal University Wukari, Wukari, Nigeria*
[2]Department of Animal Science, Ahmadu Bello University, Zaria, Nigeria
[3]National Animal Production and Research Institute, Shika-Zaria, Nigeria
**Corresponding Author: lawrenceademu@gmail.com*

ABSTRACT

An experiment was conducted to determine the response of broiler chickens under dexamethasone induced stress conditions. The experiment was conducted using two hundred and forty day old *Arbor acre* broiler chickens. There were four treatments each having three replicates with twenty birds per replicate. Concentrations of dexamethasone at 0, 1, 2 and 3 mg/ L of water were supplied daily. The experiment lasted 56 days. Birds receiving 0 mg dexamethasone had higher ($P<0.05$) final live weight. Carcass cut weight of thigh and drumstick were significantly ($P<0.05$) lower with increasing dose of dexamethasone. Liver weights were higher ($P<0.05$) in the dexamethasone groups. Villus height was higher ($P<0.05$) in the groups containing dexamethasone compared with the control. It is concluded that dexamethasone induced stress had negative effects on broilers carcass and visceral organs.

Keywords: Dexamethasone, carcass, stress, immune, broilers, visceral

INTRODUCTION

Physiological stress is one of many concerns facing the modern broiler producer. When a stressor is actually causing a negative impact on the well-being of an animal, this can be defined as distress (Moberg, 2000). When the hypothalamic-pituitary-adrenocortical axis is activated, the hypothalamus produces corticotrophin-releasing factor, which in turn stimulates the pituitary to release adrenocorticotropic hormone (ACTH). Secretion of ACTH causes the cells of the adrenal cortical tissue to proliferate and to secrete corticosteroids by elevating circulatory corticosteroids and decreasing thyroid activity, stress impairs broiler performance, especially adult birds, because the ability to dissipate heat decreases with age (Mahmoud *et al.*, 2014). Dexamethasone is a synthetic glucocorticoid that has been widely used for in vitro and in vivo studies of the glucocorticoid effects on a number of different cellular and physiological responses (Bertolo *et al.*, 2011). Aengwanich (2007) demonstrated that administration of synthetic glucocorticoid, dexamethasone in doses of up to 6mg/kg had adverse effects on broilers similar to internal glucocorticoid.

Lower breast-meat yield and higher carcass-fat deposition are the other deleterious effects of heat stress that lower the economic value of broiler carcasses (Ain-Baziz *et al.*, 1996). Liver weight has been consistently shown to increase in broilers treated with coticosterone or ACTH (Malheiros, *et al.*, 2003). A common validation procedure for the occurrence of physiological stress after corticosterone or ACTH treatment involves the measurement of immune system parameters. One such example is to measure the lymphoid organs (i.e., bursa, thymus, and spleen) because regression of these organs has been shown repeatedly to occur as a result of corticosterone or ACTH treatment (Post *et al.*, 2003). It has been shown that heat stress had immunosuppressant effects on birds and

resulted in decreasing weights of lymphoid organs (Quinteiro-Filho *et al.*, 2012). If researchers have more information on the detrimental effects of physiological stress, then future research involving nutrition and more specific stressors could be conducted more efficiently. The two major criteria of an acceptable stress model are treatments that are exact and highly repeatable and a predictable set of stress responses that occur in a known temporal pattern. Therefore the objective of the experiment was to determine the carcass, visceral and organ responses of broiler chickens under dexamethasone induced stress.

MATERIALS AND METHODS

Location of the Study

The experiment was conducted at the Teaching and Research farm of the Department of Animal Science, Ahmadu Bello University, Samaru, Zaria. The farm is located on Latitude 11º 9' 45'' N and Longitude 7º 38' 8'' E, at an altitude of 610 m above sea level. The area has an average relative humidity of 36.0% during the dry season and 78.5% for the wet season. The average minimum and maximum temperatures recorded in the area are 15.6 and 38.5°C respectively (NCAT, 2008). The study occurred between the months of May and July.

Experimental Animals, Design, Diets and Management

Two hundred and forty day old *Arbor acre* broiler chicks were used in this experiment. They were randomly allotted to four experimental treatments of 0, 1, 2 and 3 mg dexamethasone inclusion. Each treatment was replicated three times with twenty birds per replicate in a completely randomized design (CRD). A maize/soybean meal based broiler starter and finisher diet was formulated according to NRC (1994) nutrient requirement for broiler chickens (Tables 1 and 2) and fed to all birds. Daily doses of dexamethasone (1, 2 and 3 mg) was administered by dissolving in a litre of water and supplied for twenty-eight days beginning from when the birds attained 14 days of age. Dexamethasone administration was terminated 14 days prior to termination of the experiment at day 56. Birds receiving no dexamethasone (0 mg) in their drinking water served as the control. The birds were raised on deep litter and housed in 2.5 m x 1.96 m bird pens for each replicate with feed and water provided *ad libitum*. All routine vaccination and management practices were strictly adhered to.

Table 1: Ingredient Composition and Calculated Analysis of the Diet for Starter Broiler Chickens

Ingredients	
Maize	55.00
Soyabean cake	24.00
Groundnut cake	14.00
Maize offal	2.00
Limestone	1.00
Bone meal	3.00
Common salt	0.30
Vitamin premix	0.30
Lysine	0.20
Methionine	0.20
Total	100.00
Calculated analysis	
ME KCal/kg	2860
Crude protein	23.06
Ether extract	4.62
Crude fibre	4.25
Calcium	1.22
Available phosphorus	0.52
Lysine	1.24
Methionine	0.60

Nutrivitas broiler premix provided per 1 kg of diet Vitamin A, 4,000,000 I.U; Vitamin D_3, 800,000 I.U; Vitamin E 16,000 mg; Vitamin K_3, 800 mg; Vitamin B_1, 600 mg; Vitamin B_2, 2,000 mg; Vitamin B_6, 1,600 mg; VitaminB_{12}, 8 mg; Niacin, 16,000; Calpan, 4,000; Folic acid, 400 mg; Biotin, 40 mg; Choline chloride, 120,000 mg; Manganese, 32,000 mg; Iron, 16,000 mg; Zinc, 24,000 mg; Copper, 3,200 mg; Iodine, 320 mg; Cobalt, 120 mg; Selenium, 80 mg.

Table 2: Ingredient Composition and Calculated Analysis of the Diet for Finisher Broiler Chickens

Ingredients	
Maize	62.5
Soya cake	18.0
Groundnut cake	15.0
Limestone	0.50
Bone meal	3.00
Common salt	0.30
Vitamin premix	0.30
Lysine	0.20
Methionine	0.20
Total	100.00
Calculated analysis	
ME KCal/kg	2950
Crude protein	21.05
Ether extract	4.48
Crude fibre	3.77
Calcium	1.03
Av. Phosphorus	0.51
Lysine	1.09
Methionine	0.58

Nutrivitas broiler premix provided per 1 kg of diet Vitamin A, 4,000,000 I.U; Vitamin D_3, 800,000 I.U; Vitamin E 16,000 mg; Vitamin K_3, 800 mg; Vitamin B_1, 600 mg; Vitamin B_2, 2,000 mg; Vitamin B_6, 1,600 mg; VitaminB_{12}, 8 mg; Niacin, 16,000; Calpan, 4,000; Folic acid, 400 mg; Biotin, 40 mg; Choline chloride, 120,000 mg; Manganese, 32,000 mg; Iron, 16,000 mg; Zinc, 24,000 mg; Copper, 3,200 mg; Iodine, 320 mg; Cobalt, 120 mg; Selenium, 80 mg.

Organ Collection and Examination

On day 56 of the experiment, three birds per replicate were weighed, slaughtered and eviscerated. Body weights, as well as weight of organs including liver, kidneys, bursa of fabricius, spleen, and the thymus were measured using a sensitive digital scale. Relative weights of each organ expressed as percentage of live weight were determined. For small intestine histomorphometric analysis, samples of the jejenum were harvested and fixed in 10% formal saline. Fixed tissues were histologically processed according to the method of Bancroft and Stevens (2008). The histomorphometric analysis was carried out at the histopathology laboratory of the Faculty of Medicine, Ahmadu Bello University, Zaria.

Data Analysis

All data collected from the experiment was subjected to analysis of Variance (ANOVA) using the general linear model procedure of JMP (2012). Where the result of ANOVA was statistically significant, Tukey (1949) test for multiple comparisons was performed to compare results of all groups.

RESULTS AND DISCUSSION

The effect of varying doses of dexamethasone on carcass cut parts and organ weights is shown on Table 3. The average live weight ranged from 1896.28 to 2765.59 g with chickens fed 0 mg dexamethasone having higher ($P<0.05$) final weights compared to other dietary treatments. Birds administered 3 mg dexamethasone had the least ($P<0.05$) final weight. The decreasing trend observed for final weight could be as a result of the attempts by the birds to combat the additional stress caused by dexamethasone which resulted in reduced feed intake and weight loss. This is in agreement with a study by Vahdatpour *et al*. (2009) that studied chronic stress conditions in broilers, from day 1 to 49 of age. Results had shown that the greater the concentration of corticosterone intake by the birds, the lower their final body weight. Aengwanich (2007) and Li *et al*. (2009) also reported that the body weight of broilers receiving up to 6 mg kg^{-1} dexamethasone in diets or by injection into the abdomen was significantly lower than control group and this decrease was consistent with increasing levels of dexamethasone.

Result on breast weight was not significant ($P>0.05$). Wang *et al*. (2012) who studied dexamethasone-induced intramuscular lipid accumulation in chickens reported no significant ($P>0.05$) effect of dexamethasone on breast muscle mass. The effect of dexamethasone on thigh weights (9.77 to 10.84) was however significant ($P<0.05$). Birds on 0 and 1 mg had higher ($P>0.05$) thigh and drumstick weights compared to those on 2 and 3 mg. Dong *et al*. (2007) and Virden *et al*. (2007) reported that thigh cuts were significantly suppressed by corticosterone treatments. From a meat production standpoint, this points to the detrimental effect of dexamethasone in catabolising structural protein to free amino acids such as glutamine and alanine for use as gluconeogenic substrates (Puvadolpirod and Thaxton, 2000a).

Table 3: Effect of Varying Doses of Dexamethasone on Carcass Cut parts and Organ Weights of Broiler Chickens

	Dexamethasone levels (mg/l)					
Parameters (%LW)	**0**	**1**	**2**	**3**	**SEM**	**P value**
Live Weight (g)	2765.59[a]	2212.72[b]	2178.10[b]	1896.28[c]	51.63	<.0001
Breast	22.71	20.32	22.24	21.25	0.86	0.2393
Thigh	10.84[a]	10.47[ab]	9.77[b]	9.79[b]	0.22	0.0044
Drumstick	5.15[a]	5.12[a]	4.59[b]	4.77[b]	0.10	0.0019
Organs						
Liver	1.72[b]	2.07[ab]	2.13[ab]	2.27[a]	0.13	0.0389
Gizzard	1.94	2.08	2.12	2.09	0.12	0.7232
Heart	0.39	0.46	0.50	0.52	0.04	0.1007
Kidney	0.41	0.45	0.50	0.47	0.04	0.4630
Intestine	4.68	4.60	5.39	5.91	0.39	0.0838

[a, b]Means with different superscript on the same row differ significantly ($P<0.05$)

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Liver weights ranged from 1.72 to 2.27% of live weight and was significant (P<0.05). Birds in the 1, 2 and 3 mg dexamethasone group had higher (P<0.05) liver weights. Liver weights were observed to increase with increasing levels of dexamethasone. Increased liver weight is always regarded as one index of stress condition (Puvadolpirod and Thaxton, 2000b). Liver result from the dexamethasone study agrees with previous studies where liver weight has been consistently shown to increase in broilers treated with corticosterone or ACTH (Puvadolpirod and Thaxton, 2000b; Malheiros *et al*., 2003). In this study an increase in liver weight of up to 32% was observed relative to the control. This is probably due to an increase in liver lipid due to the process of gluconeogenesis, because liver lipids have been shown to increase significantly in broilers treated with ACTH (Puvadolpirod and Thaxton, 2000b). This may also be partially attributable to liver tissue enlargement because corticosteroids have been shown to stimulate protein synthesis in the liver (Baxter and *Rousseau, 1979).* Gizzard, heart, kidney and intestine weights were not significant (P>0.05). The non-significant differences in visceral organ (gizzard, kidney and intestine) weights observed is in agreement with Vahdatpour *et al*. (2009) who observed no significant (P<0.05) effects on visceral organ weights with elevated corticosterone intake.

Result showing the effect of varying doses of dexamethasone on immune organ weights of broiler chickens is presented in Table 4. Dexamethasone effect on spleen, thymus and bursa was not significant (P>0.05). Regression of the thymus, bursa, and spleen has been demonstrated in chickens after corticosterone or ACTH administration (Post *et al*., 2003). Spleen, thymus and bursa weights were also contrary to reports by Yang *et al*. (2015) who reported that the relative weight of the spleen was reduced in broiler chickens treated with corticosterone in comparison with corn oil-exposed counterparts. This was not observed in this study where dexamethasone was used. Dexamethasone treatment was terminated two weeks prior to the end of the study and this may have allowed the broiler chickens sufficient time to recover from the stress caused by dexamethasone.

Table 4: Effect of Varying Doses of Dexamethasone on Immune Organ Weights of Broiler Chickens

	Dexamethasone levels (mg/l)					
Parameters (%LW)	**0**	**1**	**2**	**3**	**SEM**	**P value**
Spleen	0.07	0.07	0.07	0.06	0.01	0.9590
Thymus	0.09	0.19	0.20	0.20	0.03	0.0674
Bursa of Fabricius	0.07	0.09	0.07	0.06	0.01	0.4586

Result showing the effect of varying dexamethasone doses on jejunum mucosal morphology in stress induced broilers is presented in Table 5. Villus height was significant (P<0.05), ranging from 19.10 to 30.42µm with birds receiving 2 mg dexamethasone having higher (P<0.05) villus height compared to other treatment groups. Villus height increased with increasing levels of dexamethasone. Villus width, crypt depth and absorption area were not significant (P>0.05) for all treatment groups. Stress could affect the

intestinal function of animals (Olsen *et al*., 2005) and further disturb the absorption of nutrients (Garriga *et al*., 2006; Albin *et al*., 2007). Results on villus height, absorption area and crypt depth which was similar among treatments for the study disagrees with Li *et al*. (2009) who reported that dexamethasone decreased villus height and absorption area but increased the crypt depth significantly. Terminating dexamethasone administration 14 days before the end of the experiment may be responsible. This gave ample time for intestine villi to recover once the stressor (dexamethasone) was removed. Villus height and the absorption area are apparent indicators of the ability of the intestine to absorb nutrients and crypt depth is an indicator of the maturation of intestinal epithelium: a deeper crypt indicates a more mature intestinal epithelium (Li *et al*., 2009). The result on crypt depth and absorption area indicates that dexamethasone did not negatively affect nutrient absorption.

It can be concluded that dexamethasone had a negative effect on final live weight, carcass, visceral and immune traits of broiler chickens. Furthermore dexamethasone is a useful tool in studying the effect of induced stress in broiler chickens providing the opportunity for researchers to observe responses from a stressor that is easily measurable and controllable.

Table 5: Effect of Varying Dexamethasone Doses on Jejunum Mucosal Morphology in Stress Induced Broilers

	Dexamethasone levels (mg/l)					
Parameters (%LW)	0	1	2	3	SEM	P value
Villus height (μm)	19.10^{b}	24.13^{b}	30.42^{a}	24.01^{b}	1.34	0.0024
Villus width(μm)	3.10	2.69	2.67	2.90	0.66	0.9624
Crypt depth (μm)	14.23	16.29	12.09	14.33	1.93	0.5302
Absorption area (μm^2)	348.79	409.03	511.50	437.58	70.40	0.4734

$^{a, b}$Means with different superscript on the same row differ significantly (P<0.05)

REFERENCES

Aengwanich, W. (2007). Effects of Dexamethasone on Physiological Changes and Productive Performance in Broilers . *Asian Journal of Animal and Veterinary Advances*, 2: 157-161.

Ain-Baziz, H., Geraert, P.A., Padilha, J.C. and Guillaumin, S. (1996). Chronic heat exposure enhances fat deposition and modifies muscle and fat partition in broiler carcasses. *Poultry Science*, 75: 505–513.

Bancroft, J. D. and Stevens, A. (2008). Theory and Practice of Histological Techniques, 7th Edition, Churchill Livingstone. Edinburg, London, Melbourne and New York, Page 725.

Baxter, J. D. and Rousseau, G.G. (1979). Monographs on endocrinology (Baxter, J. D., and Rousseau, G. G., eds) 12: 1-24.

Bertolo, A., Ettinger, L., Aebli, N., Haschtmann, D., Baur, M., Berlemann, U., Ferguson, S.J. and Stoyanov, J.V. (2011). The In Vitro Effects of Dexamethasone, Insulin and Triiodothyronine on Degenerative Human Intervertebral Disc Cells under Normoxic and Hypoxic

Conditions. *European Cells and Materials*, 21: 221-229.

Dong, H., H. Lin, H.C. Jiao, Z.G. Song, J.P. Zhao and K.J. Jiang, (2007). Altered development and protein metabolism in skeletal muscles of broiler chickens (*Gallus gallus domesticus*) by corticosterone. *Comparative Biochemistry and Physiology – Part A: Molecular and Integrated Physiology*, 147: 189-195.

Garriga, C., Hunter, R. R., Amat, C., Planas, J. M., Mitchell, M. A. and Moreto, M. (2006). Heat stress increases apical glucose transport in the chicken jejunum. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology*, 290: 195 – 201.

JMP® (2012) Software Version 6.12 of the SAS System for Windows. SAS Institute, Inc., Cary, NC, USA.

Li, Y., Cai, H.Y., Liu, G.H., Dong, X.L. and Chang, W.H. (2009). Effects of stress simulated by dexamethasone on jejunal glucose transport in broilers. *Poultry Science*, 88: 330-337.

Mahmoud, U.T., Abdel-Rahman, M.A. and Darwish, M.H. (2014). Effects of propolis, ascorbic acid and vitamin E on thyroid and corticosterone hormones in heat stressed broilers. *Journal of Advanced Veterinary Resource*. 4:18–27.

Malheiros, R.D., *V.M.B. Moraes, A. Collin, E. Decuypere, and J. Buyse (2003). Free diet selection by broilers as influenced by dietary macronutrient ratio and corticosterone supplementation. 1. Diet selection, organ weights, and plasma metabolites. Journal of Poultry Science,* 82*:*123*–131.*

Moberg, G. P. (2000). *Biological response to stress: Implications for animal welfare. Pages 1–21 in The Biology of Animal Stress. G. P. Moberg and J. A. Mench, ed. CABI Publishing, UK.*

NRC (1994). Nutrient Requirements of Poultry (9th Ed.). National Academy Press, Washington, DC.

NCAT (2008). Nigeria college of Aviation technology, Zaria meteorological station data.

Olsen, R.E., K. Sundell, T.M. Mayhew, R. Myklebust and E. Ringo (2005). Acute stress alters intestinal function of rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Aquaculture,* 250: 480 –495.

*Post, J., Rebel, J.M.J. and ter Huurne, A.A.H.M. (2003). P*hysiological effects of elevated plasma corticosterone concentrations in broiler chickens. An alternative means to assess the physiological effects of stress. *Poultry Science, 82:1313-1318.*

Puvadolpirod, S. *and Thaxton, J.P. (2000a). Model of physiological stress in chickens: 4. Digestion and Metabolism. Poultry Science,* 79*:*383*–390.*

Puvadolpirod, S. and Thaxton, J.P. (2000b). Model of physiological stress in chickens: 1. Response parameters. *Poultry Science*, 79:363–369.

Quinteiro-Filho, W.M., Rodrigues, M. V., Ribeiro, A., Ferraz-de-Paula, V., Pinheiro, M.L., Sa, L.R.M., Ferreira, A.J.P. and Palermo-Neto, J. (2012). 'Acute heat stress impairs performance parameters and induces mild intestinal enteritis in broiler chickens: Role of acute hypothalamic-pituitary-adrenal axis activation'. *Journal of Animal Science,* 90(6): 1986-1994.

Tukey, J. (1949). Comparing Individual Means in the Analysis of Variance. Biometrics, 5 (2): 99–114.

Vahdatpour, T., Nazer Adl, K., Ebrahim Nezhad, Y., Mahery Sis, N., Riyazi, S.R. and Vahdatpour, S. (2009). Effects of corticosterone intake as stress-alternative hormone on broiler chickens: Performance

and blood parameters. *Asian Journal of Animal and Veterinary Advance,* 4(1): 16-21.

Wang X.J., Wei D.L., Song Z.G., Jiao H.C. and Lin H. (2012). Effects of Fatty Acid Treatments on the Dexamethasone-Induced Intramuscular Lipid Accumulation in Chickens. *PLoS ONE,* 7(5): e36663.

Virden, W. S., M. S. Lilburn, J. P. Thaxton, A. Corzo, D. Hoehler, and M. T. Kidd (2007). The effect of corticosterone-induced stress on amino acid digestibility in Ross broilers. *Poultry Science*, 86:338–342.

Yang, J., Liu, L., Sheikhahmadi, A., Wang, Y., Li, C., Jiao, H, *et al.* (2015). Effects of Corticosterone and Dietary Energy on Immune Function of Broiler Chickens. *PLoS ONE* 10(3): e0119750.

Albin, D. M., J. E. Wubben, J. M. Rowlett, K. A. Tappenden and R. A. Nowak (2007). Changes in small intestinal nutrient transport and barrier function after lipopolysaccharide exposure in two pig breeds. *Journal of Animal Science*, 85: 2517 – 2523.