Bulletin of Environment, Pharmacology and Life Sciences
Bull. Env. Pharmacol. Life Sci., Vol 2 (10) September 2013: 14-18

Online ISSN 2277-1808
Journal's URL:http://www.bepls.com

ORIGINAL ARTICLE

Changes in Metabolic Nutrients Utilization and Alterations in Biochemical and Hematological indices in Broilers Fed Graded Levels of Dietary *Moringa oleifera*

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ABSTRACT

Graded levels of full-fat undecorticated Moringa oleifera seed meal, MOSM were evaluated in diets of 96-day old broilers at 2.5, 5.0 and 7.5% while a maize-soybean diet was used as a reference diet. Nutrients utilization by the broilers and effects of the virgin dietary MOSM on serum chemistry, haematology and some bio-data-PER, protein efficiency ratio and NM, nitrogen metabolism were used as response criteria for the evaluation. The metabolic utilization of nutrients, most of the serum chemistry and haematological parameters decreased in response to increasing dietary levels of MOSM except values on products of metabolism like creatinine, uric acid, bilirubin and conjugated bilirubin that tended to elevate with increasing levels of dietary unprocessed MOSM. Since no mortality was recorded in this experiment even at 7.5% MOSM inclusion, further research is on-going todetermine the lethal level to poultry including other monogastric animals.

***Keywords**: MOSM, broilers, nutrient utilization, serum and haematological indices.*

Received 30.07.2013 Accepted 12.08.2013

INTRODUCTION

The demand for *Moringa oleifera* products is on the increase due to the consideration of the tree as one of the world most useful plants known for its nutritional, medicinal and significant economic importance. The tree, native only to the foothills of the Himalayas of the Indian sub-continent, is now widely distributed over the world where its nutritional, medicinal and industrial qualities have been discovered and are being utilize. Research works on *Moringa oleifera* are on-going in developing countries like Nigeria which are witnessing daily increase in human population with attendant malnutrition, infectious diseases and excruciating poverty, to search for alternatives for the scarce conventional foods, feed and industrial materials. The application of *M. oleifera* seed meal, MOSM as novel feedstuff to substitute for soybeans as a protein source in this study is a sequel to look inward for the alternatives of energy, protein and other sources of nutrients which are staple food/feedstuffs for human and especially monogastric animal consumption like corn, fish/fish meal, guinea corn, groundnuts/cake and soybeans/cake and which are competed for. Past works [1-4]using Moringa tree with its products warranted it as a healthy food/feed source for Man and animals. Despite Moringa's numerous economic uses, the tree is implicated in harboring anti-physiological and toxic factors such as haemagglutinins, alkaloids, glucosinolate, nitrate, oxalate, phytate, phenols etc [5-7]. These reports stimulated this research to evaluate the toxicological aspects of Moringa seeds in nutrition of poultry.

MATERIALS AND METHODS

Moringa oleifera seeds obtained from a region in Nigeria (Benue state) were sun-dried to ease grinding. Given kilograms of the seeds were ground into meal to obtain *Moringa oleifera* seed meal (MOSM) which was stored for subsequent inclusion into diet mixtures. Four iso-energy, iso-nitrogenous diets were formulated to meet NRC [8] requirements for day-old broiler chicks and made of a corn-soybean

reference diet and the other test diets containing MOSM at graded levels, 2,5, 5.0 and 7.5% for diets 2, 3 and 4 respectively. The composition of the experimental diets on as fed basis is shown on Table 1.

96-day old Hubbard breed of broiler chicks were used and housed in an electrically heated battery brooder cage designed for the purpose. Electrical bulbs, 60-watts were installed in the cage units to supply heat for brooding for temperature between 32-35°C [9]. 24-chicks were allotted to a dietary treatment replicated into A, B, C and each replicate contained 8-birds. The experiment was designed as a one-wy classification. The chicks were fed twice daily, 8.00am and 2.00pm. Both feed and drinking water were supplied to appetite during the trial

Half way into the experiment, a nutrient retention trial was conducted. Weighed quantities of feed were fed for 72-hours to the chicks and fecal samples collected and sundried and ground for the determination of nutrient retention. Percent nutrient retention was computed using the formula: nutrient in feed less nutrient in feces/nutrient in feed x 100%. Analyses were carried out on crude protein, ether extract, fiber, mineral matter while metabolizable energy was determined by calculation [109Carpenter and Clegg, 1956). Soluble carbohydrate, NFE was determined by difference. Biodata like PER, protein efficiency ratio, NM, nitrogen metabolism were estimated using appropriate formulae. At the end of the trial, one broiler per replicate was randomly picked and whole and blood sera samples were collected by head decapitation of the bird. A set of blood samples was collected into bottles containing EDTA for haematological evaluation while another set of blood samples was collected without anticoagulant for blood chemistry determination. The sample bottles were stored in the refrigerator at -20^{0}C prior to biochemical analyses. Haematological parameters , packed cell volume (PCV), red blood cells (RBC), white blood cells (WBC) and haemoglobin (HB) were determined using Wintrobe's microhaematocrit improved Neubauer haemocytometer and cyanomethaemoglobin respectively [11-12]. Total and conjugated bilirubin was determined by Jendrassik Grof method [13]. Plasma protein, albumin, globulin and the blood metabolites- creatinine, uric acid were analyzed using Sigma kits according to Feteris (1965). Serum enzyme activities, aspartate aminotransferase (AST/GOT), alanine aminotransferase (ALT/GPT) and alkaline phosphatase (ALP) were estimated by colometric method [14].

All data were subjected to analyses of variance, ANOVA using the model for a single-factor design [15]. Significance of differences between treatment means was determined using Duncan multiple range test [16].

RESULTS

Table 2 presents data on the influence of dietary graded levels of MOSM on nutrient retention and energy utilization in broilers. All the parameters of nutrients retained (dry organic matter, protein, fiber, fat, mineral matter) except soluble carbohydrate decreased with increasing dietary level of the test feedstuff. Energy utilization measured by gross energy, daily absorbed and metabolizable energy similarly decreased in response of increasing dietary MOSM ($p < 0.05$). Ingestion of MOSM in diets and its effects on protein nitrogen intake and utilization as well as some biodata is shown on Table 3. Daily retained nitrogen and nitrogen retention coefficient decreased in values following increase in MOSM inclusion in diets. PER and NM followed similar pattern as that on nitrogen utilization, decreasing in values with increasing level of moringa meal in diets ($p < 0.05$). Results on blood chemistry evaluation and enzyme activities influenced by consumption of dietary MOSM are given on Table 4. Results obtained on the biochemical indices as well as activities of enzymes were inferior to the result obtained on the control diet since values of the determinants decreased on moringa based diets ($p < 0.05$) except those of the metabolites, creatinine and uric acid levels which increased with increasing dietary level of MOSM ($p < 0.05$). Table 5 with haematological and metabolite parameters of birds maintained on moringa diets compared with the control diet showed numerical but insignificant decrease in haematocrit ($p > 0.05$) and statistical significant decrease in WBC, MCV, MCH ($p < 0.05$) while values on RBC, Hb indicated increase with increasing moringa meal in diets ($p < 0.05$). Bilirubin and conjugated bilirubin showed increase in values following increase in dietary MOSM ($p < 0.05$).

Table 1: Composition of the experimental diets on as fed basis (%)

Diets	1	2	3	4
Ingredients				
Maize	58.25	57.75	60.75	59.75
Soy meal	38.50	36.50	31.00	29.50
Moringa meal	0.00	2.50	5.00	7.50
Bone meal	1.50	1.50	1.50	1.50
Oyster shell	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
DL-methionine	0.25	0.25	0.25	0.25
Vitamin=premix	0.50	0.50	0.50	0.50
Total	100	100	100	100

Table 2: Influence of dietary graded levels of MOSM on nutrients retention and energy utilization in broilers (%).

Diets	1	2	3	4
Parameters				
Dry matter	90.76^{b}	91.23^{b}	90.86^{b}	89.70^{a*}
Protein	18.62^{c}	11.18^{b}	13.02^{b}	7.77^{a*}
Fiber	2.47	1.65	2.61	2.97 NS
Ether extract	2.57^{b}	1.39^{a}	1.13^{a}	1.13^{a*}
Mineral matter	2.13^{b}	2.11^{b}	1.97^{a}	1.15^{a*}
N. F. E.	22.47^{a}	29.69^{b}	30.10^{b}	30.81^{b*}
G. E., kcal/g	2.32^{b}	2.29^{b}	2.23b	1.99^{a*}
Dailyabsorbed energy, kca/g	0.31^{b}	0.09^{a}	0.09^{a}	0.08^{a*}
M.E., kcal/g	31.85^{c}	26.93^{b}	24.93^{b}	16.07^{a*}

*a-b-c treatment means in rows not sharing common superscripts differed significantly ($p < 0.05$).
NS, not significant ($p > 0.05$).

Table 3: Effects of dietary graded levels of MOSM intake on nitrogen utilization and some biodata in birds (%).

Diets	1	2	3	4
Indices				
Nitrogen intake	18.11	14.12	23.19	22.03 NS
Daily absorbed nitrogen	0.96^{b}	0.75^{a}	0.67^{a}	0.57^{a*}
Nitrogen retention coefficient	1.30^{b}	1.30^{b}	1.20^{a}	1.20^{a*}
PER	2.86^{b}	2.86^{b}	2.78^{a}	2.78^{a*}
NM	817^{b}	729^{ab}	608^{ab}	482^{a*}

PER, protein efficiency ratio; NM, nitrogen metabolism.

Table 4: Plasma chemistry values and enzyme activities in broilers fed dietary graded levels of MOSM.

Diets	1	2	3	4
Parameters				
Glucose,mmol/L	22.90^{b}	22.63^{b}	20.33^{a}	18.40^{a*}
Protein,mmol/L	40.33^{b}	37.00^{b}	36.00^{b}	33.00^{a*}
Albumin,mmol/L	26.00^{b}	25.00^{b}	24.33^{a}	23.00^{a*}
Globulin,mmol/L	14.33^{b}	12.00^{b}	11.33^{b}	4.39^{a*}
Creatinine,mmo/L	71.00^{a}	127.00^{b}	129.00^{b}	171.00c*
Uric acid,mmol/L	4.10^{a}	5.03^{a}	6.37^{a}	10.13^{b*}
AST, IU/L	143.00^{c}	111.33^{b}	63.00^{a}	62.50^{a*}
ALT, IU/L	281.00^{d}	75.67^{c}	61.00^{b}	54.33^{a*}
ALP, IU/L	229.00^{d}	75.67^{c}	53.00^{b}	37.00^{a*}

Table 5: Dietary effects of graded levels of MOSM on blood composition in broilers.

Diets	1	2	3	4
PCV (%)	28.33	28.00	24.00	17.67 NS
RBC ($x10^{12}$/L	2.50^{a}	2.73^{a}	2.93^{b}	3.47^{c*}
WBC ($x10^{9}$/L	18.77^{c}	15.53^{b}	11.63^{a}	11.43^{a*}
Hb (g/dl)	5.97^{a}	8.13^{b}	9.63^{c}	9.50^{c*}
MCV (fl)	104.33^{c}	81.67^{b}	80.67^{b}	68.67^{a*}
MCH (pg)	35.33^{c}	27.67^{b}	25.67^{ab}	23.00^{a*}
MCHC (%)	33.67	33.67	33.67	33.33 NS
Bilirubin (%)	2.67^{a}	3.52^{b}	3.60^{b}	3.77^{b*}
Conjugated bilirubin	0.23^{a}	0.23^{a}	0.28^{a}	0.30^{b}

DISCUSSION

Nutrients retention including energy utilization suggested that the anti-nutritional factors of *Moringa oleifera* a significant contribution to the decreasing indices measured in response to the increasing levels of the seed meal in diets. The anti-nutritional factors might also be responsible for lowering nitrogen utilization with the related parameters of PER and NM. Available literature [17, 18, 1]reported that phytochemicals in moringa are capable if reducing nutrient availability and/or utilization. The reduction

in values on nutrient retention and utilization became possible in this study since the seed meal was fed to the animals direct without processing.

Ingestion of untreated MOSM in diets at graded levels had depressive effects on blood glucose level, serum total protein, albumin and globulin. The depressive effect of the unprocessed dietary MOSM could also be attributed to the adverse influence of the moringa phytotoxins namely lecthins, alkaloids like moringin, moringinine, glucosinolates, phenols including tannins, nitrite, oxalate, phytate which acted in concert with higher levels of the meal in diets to reduce the biochemical indices [4,5,7]. Consumption of MOSM in diets at varying levels increased the concentration or amounts of creatinine and uric acid in the blood of the birds fed the test feedstuff in diets compared with the control. Elevated levels of creatinine and uric acid in response to increasing quantity of the seed meal indicated that the fed animals had problem with the quality of the protein in the test feedstuff. These findings agreed with the works of Aregheore [19] who fed animals with high levels of moringa product and reported poor results due to anti-nutrients and excessive protein in moringa compared with those animals receiving low levels of *Moringa oleifera.* Increasing the level of dietary MOSM deceased concomitantly the activities of AST, ALT ALP. These enzymes located in the liver, muscles or bones aid chemical activities within the organs in question. Reduced activities of the enzymes would adversely affect the organs or tissues where they are found [20,21].

Increasing levels of MOSM in diets decreased WBC, MCV, MCH while increasing the blood catabolic products of bilirubin and conjugated bilirubin. Decrease in WBC suggests that the fowls could not withstand disease attack due to low immunity. The significant increase in values on bilirubin and conjugated bilirubin may be linked to the toxic effects of moringa phytochemicals. On the other hand, RBC and Hb values presented increase with dietary increase in level of MOSM indicating that the birds did not experience any type of anaemia.

In conclusion, increasing dietary levels of untreated Moringa oleifera seed meal decreased progressively nutrients retention and utilization and affected adversely most of the biochemical and haematological parameters investigated. However, since survival was 1005 in this experiment even at 7.55 inclusion of the raw seed meal. Research is on-going to determine the lethal level to poultry and other monogastric animals.

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How to cite this article
A.A. Annongu A. A. Toye[1], O. R. Karim, F. E. Sola-Ojo, S. Ashi, K. J. Olasehinde, O. A. Adeyina and J. O. Aremu. Changes in Metabolic Nutrients Utilization and Alterations in Biochemical and Hematological indices in Broilers Fed Graded Levels of Dietary *Moringa oleifera*. Bull. Env. Pharmacol. Life Sci., Vol 2 (10) September 2013: 14-18