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## Nutritional Potentials of Siam Weed (*Chromolaena odorata*) Leaf Meal (SWLM) on Laying Hens: Biochemical and Haematological Implications

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**Abstract:** The biochemical and haematological indices of birds fed with varying dietary inclusions of siam weed (*Chromolaena odorata*) leaf meal (SWLM) were determined using 24 laying hens in their eighth month of lay in an eight week trial. Four diets were formulated for the purpose of this study. Diet 1 served as the control diet and had no SWLM inclusion. SWLM was introduced at 2.5%, 5.0% and 7.5% in diets 2, 3 and 4 respectively. The haematological and biochemical investigations revealed no statistical differences ( $P=0.05$ ) among the mean values of treatments 1, 2 and 3. However the mean value of treatment 4 (7.5% SWLM inclusion level) was statistically different ( $P<0.05$ ) from the others. The numerical values of most haematological indices showed an initial increase up to treatment 3 followed by a decrease in treatment 4. Almost all haematological indices studied (PCV, RBC count, Hbc, MCHC, MCH, MCV and ESR) progressively increased up to diet 3 (5% SWLM inclusion) after which there was a decline indicating a probable acceptance limit of 5% SWLM dietary inclusion in layers diets without any serious health implication.

**Key words:** Siam weed leaf meal (SWLM), laying hens, haematological indices, health status

### Introduction

Concerned professionals in the field of animal nutrition particularly in the developing countries had for long identified the cost of finished livestock feed as the most economically limiting factor in the industry (Ademosun, 1973; Obioha, 1976; Balogun, 1988; Fasuyi and Aletor, 2005a). Far more pronounced and of utmost concern are the protein sources in monogastric diets (Fasuyi, 2005). The *greens* (green plants of various sources) have long been recognized as the cheapest and most abundant potential source of proteins because of their ability to synthesize amino acids from a wide range of virtually unlimited and readily available primary materials such as water, CO<sub>2</sub>, atmospheric N<sub>2</sub> (as in legumes) (Byers, 1961; Oke, 1973; Fasuyi and Aletor, 2005b). Information on siam weed (*Chromolaena odorata*) leaf meal in livestock feedings is very scanty and its utilization by livestock in the fresh form is hampered by its offensive odour and speculation about its toxicity (Madrid, 1974). Comparison of its leaf meal with that of cassava in terms of mineral composition however revealed a higher nutritional potential for siam weed leaf meal (Nwokolo, 1987). The availability (true digestibility) of minerals and proteins in the leaf meals of both cassava and siam weed were also assayed by Nwokolo (1987) where the average to low availability for these leaf meals was said to be as a result of the antinutritional factors in both leaves. The presence of saponins as a major antinutrient cannot be ruled out since they occur in large number of plant materials where they are associated with poor growth rate and depressed dietary nutrient utilization in monogastric animals (Checke and Myer, 1975). A personal observation in the course of this

study revealed that the offensive odour of the fresh leaves is only given off when the plant is disturbed by mere touching or bruising of the leaves and that this odour is greatly reduced/eliminated when the leaves are sun dried for subsequent milling. Not much has also been written on the biochemical and haematological implications of siam weed leaf meal in livestock rations. Sajise *et al.* (1974) reported that the importance of *C. odorata* as a cause of poisoning and death in livestock is due to its conversion either in the food stuff or within the alimentary canal into nitrite. Nitrite converts haemoglobin of the blood to methaemoglobin which is unable to act as an oxygen carrier and this might possibly lead to the death of the animals through tissue anoxia. Another biochemical implication which the utilization of *C. odorata* leaf may have especially when used in combination with cassava by-products is the affinity which methaemoglobin has for cyanide leading to the formation of cyanmethaemoglobin which is another oxygen depleting compound in the blood (Datta and Ottaway, 1976). There have also been reports that in certain parts of Philippines, farmers claimed that the ingestion of *C. odorata* causes diarrhoea and consequent death among cattle (Madrid, 1974). This present study therefore explores the haematological and biochemical indices of layers fed with varying levels of the leaf meal of *C. odorata* in their diets in order to elucidate more on its nutritional potentialities vis a vis its health status implication on laying hens.

### Materials and Methods

**Collection and preparation of SWLM:** The leaf meal used for this study was prepared by cutting the stems of

Table 1: Proximate composition (g/100g) and amino acid content (g/16gN) of Siam weed (*Chromolaena odorata*) leaf meal (SWLM)

Nutrients	SWLM
Dry matter (%)	87.40
Crude protein	18.67
Ether extracts	1.01
Crude fibre	11.67
Ash	3.63
Nitrogen free extract	65.03
Amino acids:	
Alanine	4.03
Aspartic acid	6.12
Arginine	4.96
Glycine	4.61
Glutamic acid	9.38
Histidine	2.63
Isoleucine	5.52
Lysine	2.01
Methionine	1.58
Cystine	1.30
Meth. + Cys.	2.88
Leucine	7.01
Serine	3.81
Threonine	4.90
Phenylalanine	4.30
Valine	6.20
Tyrosine	4.71
Tryptophan	2.36

Means are for duplicate determinations

nearly matured and just maturing *Chromolaena odorata* plants with the leaves intact and sundrying these for 3-4 days. The dried leaves were hand picked directly into jute sacks. The entire collection period lasted 15 days. The sun-dried leaves were later milled and the leaf meal obtained there-from was used in combination with other feed ingredients to compound the treatment diets used in this study.

**Proximate and amino acids composition:** Proximate composition of the SWLM was determined by AOAC (1995) method, while the amino acids were determined using amino acid analyzer. The gross and digestible energy values were computed by method of Ng and Wee (1989).

**Experimental diets:** The SWLM processed as earlier discussed was one of the major ingredients used in the diet formulation. Other feed ingredients were purchased from reputable sources in Akure, Ondo State, Nigeria. The results of the proximate compositions earlier determined were used in the eventual formulation of the different diets. Four isonitrogenous (15% crude protein) and nearly isocaloric layers diets were formulated with the feed ingredients shown in Table 1. Diet 1 was the control diet without the test SWLM in the diet. Diets 2, 3,

and 4 were formulated such that SWLM was introduced into the diets at graduated levels of 2.5, 5.0 and 7.5% respectively. Other fixed protein sources in all the diets were groundnut cake, fish meal and brewers' dried grain. All diets were also supplemented with feed-grade methionine and lysine.

**Management of experimental layer birds and experimental design:** Twenty four layer birds were randomly selected from a stock of laying birds in their eighth month of lay. They were randomly allotted into individual cages, replicated thrice under four dietary treatments. Two layer birds were used per replicate making six birds per treatment. The birds were given feed and water *ad libitum* throughout the entire experimental period of 8 weeks.

**Blood collection for analysis:** At the end of the feeding trial, a male chick per replicate was randomly selected, weighed and scarified by severing the jugular vein and blood allowed to flow freely into labeled bottles one of which contained a speck of EDTA while the other without EDTA was processed for serum. The serum was kept deep frozen prior to analysis. The packed cell volume (PCV%) was estimated by spinning about 75:1 of each blood sample in heparinized capillary tubes in an haematocrit micro centrifuge for 5 minutes while the total red blood cell (RBC) count was determined using normal saline as the diluting fluid. The haemoglobin concentration (Hbc) was estimated using cyanomethaemoglobin method while the mean corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin (MCH) and the mean corpuscular volume (MCV) were calculated.

**Total serum and liver protein determination:** After the separation of the liver, 0.5g of each liver sample was homogenized in 5ml of ice cold distilled water using a Ystral-GmbH D-7801 homogeniser (Dottingen Type x 1020). Then 1ml was taken from this and diluted with 19ml of cold distilled water. Thus making the concentration of homogenate to be 0.5g/100ml or 0.5%. The homogenate was then put in sample bottles and kept frozen prior to analysis. The serum and the liver homogenate were thawed before the total protein as well as the albumin and globulin of serum and liver were determined.

**Statistical analysis:** Data obtained on each parameter were subjected to either coefficient of variation analysis and analysis of variance (ANOVA) (Steel and Torrie 1980) and differences between treatment means were compared using Duncan's Multiple Range Test (DMRT) (Duncan, 1955).

## Results

Table 4 shows the haematological and biochemical

Table 2: Composition of Experimental Diet (g/100g)

Ingredients	Diets			
	1	2	3	4
Maize (11.0% CP)	56.29	56.25	56.23	56.20
Groundnut cake (45.0% CP)	10.51	10.55	10.57	10.60
Fish meal (68.0% CP)	2.50	2.50	2.50	2.50
SWLM (18.67% CP)	0.00	2.50	5.00	7.50
Bone meal	6.00	6.00	6.00	6.00
Oyster shell	4.00	4.00	4.00	4.00
Premix*	0.50	0.50	0.50	0.50
Nacl	0.50	0.50	0.50	0.50
Total	100.0	100.0	100.0	100.0
Calculated crude protein %	15.00	15.00	15.00	15.00
GE**(kcal/100g) DM	263.2	263.2	263.2	263.2*

Contained Vitamins A 800 I.U.; D<sub>3</sub> (1,4731.C.U); Riboflavin 4.20mg; Pantothenic acid 5.0mg; Nicotinic acid 20.0mg; Folic acid 0.5mg; Choline 300mg; Vitamin K, 2.0mg; Vitamin B<sub>12</sub>, 0.01mg; Vitamin E, 2.5I.U; Manganese, 56.0mg; Iodine, 1.0mg; Iron 20.0mg; Copper 10.0mg; Zinc 50.0mg and Cobalt 1.25mg. GE\*\* (kcal/100g) DM calculated based on 5.7kcal/g protein; 9.5kcal/g lipid; 4.0kcal/g carbohydrate (Ng and Wee, 1989).

parameters measured in the cause of the experiment. The haematocrit values (PCV) appreciated from the control up to treatment 3 from where it showed a decline in diet 4. The PCV values were not significantly different ( $P = 0.05$ ) for birds in diets 1, 2 and 3 but all 3 (diets 1, 2 and 3) were significantly different ( $P < 0.05$ ) from the value obtained for birds on diet 4. The values for hemoglobin concentration (Hbc) and white blood cell also followed the same trend with diets 1 to 3 showing no significant difference ( $P = 0.05$ ) but all the 3 diets were significantly different from 4 ( $P < 0.05$ ). The red blood cells (RBC) and total serum protein gave a progressive increase up to treatment 3 before declining in birds in diet 4 with a significant statistical difference ( $P < 0.05$ ) between diet 4 and other diets (1, 2 and 3) combined. The erythrocyte sedimentation rate (ESR) values were not significantly different ( $P = 0.05$ ) although the numerical value of ESR in diet 4 was highest at 4.5. The albumin component of the total protein gave a higher level for SWLM diets to that of the control even though there was no significant difference among all the treatment means ( $P = 0.05$ ). There was also an increase in serum albumin as a result of the dietary treatments but this was not statistically different ( $P = 0.05$ ) in the diets. The lowest values for serum globulin was obtained for birds from the diet with the highest level of inclusion of SWLM (diet 4) and this was significantly different ( $P = 0.05$ ) from other diets (1, 2 and 3) thus implicating the high inclusion level of SWLM as the probable cause of variation. The lowest value for serum creatinine was also obtained from birds on the highest inclusion of SWLM while the highest level was obtained in treatment 2 even though these values were not significantly different ( $P = 0.05$ ). The values for uric acid and urea nitrogen rose steadily from the control diet 1 to diet 4 with the highest SWLM inclusion at 7.5% diet but with no significant difference ( $P = 0.05$ ).

## Discussion

The result of the study also showed that the inclusion of SWLM was beneficial up to 5% of inclusion without any statistically significant difference until the maximum inclusion level of 7.5% used in this trial. This result is in concert with studies earlier conducted on other leaf meals (Checke and Myer, 1975; Onwudike and Oke, 1986; Horton and Christensen, 1981; Adekanye and Sonaiya, 1992). Hutagalung (1981) also suggested an inclusion level of between 2-5% for *leucaena* leaf meal. No mortality was recorded throughout the duration of the experiment and this should allay the fear of death following the consumption of SWLM by livestock as reported in the work of Madrid (1974). This might not be unconnected with the reduction of the nitrate content in the leaves by the harvesting procedure and sun-drying. The high nitrate content of the fresh leaves has been implicated as the probable cause of death of livestock through tissue anoxia (Sajise *et al.*, 1974). The result however showed that this advantage might be wiped off at higher level of inclusion of the leaf meal. This study also revealed an initial increase in the level of haemoglobin up to 5% inclusion level after which there was a sharp and significant decrease in hemoglobin concentration. The use of SWLM in layers ration may probably lead to reduction in the oxygen carrying capacity of the blood in this species of livestock with concomitant reduction in performance. The work earlier reported by Sajise *et al.* (1974) that the conversion of oxyhaemoglobin to methaemoglobin as being the root cause of sudden death of livestock that accidentally fed on SWLM corroborated the findings of this trial. The white blood cells were lower in birds fed with SWLM than in the control suggesting a probable reduction in white blood cell count at higher SWLM inclusion levels. This may engender a higher susceptibility of the birds to diseases as a result of reduced white blood cell

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Table 3: Proximate composition (g/100g) and Amino Acid content (g/16gN) of Experimental Diets

Composition	Diets			
	1	2	3	4
Dry matter	90.10±0.1	90.05±0.1	89.46±0.3	89.3±0.4
Crude protein	15.14±0.2	15.23±0.8	15.18±0.1	15.16±0.4
Ether extracts	4.23±0.4	4.38±0.1	3.91±0.1	4.19±0.3
Crude fibre	5.38±0.4	5.72±0.7	5.60±0.8	5.23±0.4
Ash	5.32±0.1	5.78±0.1	6.12±0.1	5.82±0.4
Nitrogen free extract	69.93±0.5	68.89±0.2	69.19±0.2	69.60±0.2
Amino acids				
Arginine	5.96	6.18	6.38	6.01
Histidine	2.63	2.85	2.90	2.78
Isoleucine	5.52	5.50	5.74	5.66
Leucine	9.56	9.60	9.20	9.51
Lysine	6.69	5.18	5.12	4.92
Methionine	2.48	2.51	2.01	2.01
Cystine	1.33	1.30	1.21	1.19
Methionine + Cystine	3.81	3.81	3.22	3.20
Phenylalanine	6.30	6.52	6.22	6.01
Threonine	4.90	4.81	4.52	4.42
Thyrosine	4.71	4.91	5.00	4.73
Valine	6.20	6.35	5.98	6.49
Glycine	5.61	5.81	6.10	5.85
Tryptophan	2.36	2.30	2.37	2.42
Alanine	6.12	6.34	6.51	6.40
Aspartic acid	9.98	7.10	7.31	7.12
Glutamic acid	11.38	11.52	10.85	11.62
Serine	4.87	5.00	5.30	5.14

Means are for duplicate determinations

Table 4: Haematological Indices of Laying Hens fed varying levels of SWLM Based

Parameters	Diets			
	0.00% SWLM	2.50% SWLM	5.00%SWLM	7.50% SWLM
PCV %	25.3±1.5 <sup>a</sup>	27.7±2.1 <sup>a</sup>	28.3±1.5 <sup>a</sup>	21.0±2.0 <sup>b</sup>
RBC count (x 106/mm <sup>3</sup> )	2.1±0.2 <sup>a</sup>	2.3±0.3 <sup>a</sup>	2.4±0.1 <sup>a</sup>	1.9±0.3 <sup>b</sup>
Hbc (g/100ml)	2.1±0.2 <sup>a</sup>	2.3±0.1 <sup>a</sup>	2.3±0.3 <sup>a</sup>	1.7±0.2 <sup>b</sup>
MCHC (%)	7.4±0.9	7.4±2.9	9.2±0.8	8.3±1.5
MCH (pg)	9.5±0.3	9.0±2.9	8.7±1.5	8.2±0.7
MCV (µm <sup>3</sup> )	127.2±13.8 <sup>a</sup>	125.6±10.5 <sup>a</sup>	122.4±8.9 <sup>ab</sup>	119.5±25.7 <sup>b</sup>
ESR (mm)	4.1±0.6	4.1±0.6	3.8±0.5	4.5±1.0

Means with different superscripts in the same horizontal row are significantly different

(P < 0.05); FM = Fish Meal; SWLM = siam weed leaf meal.

PCV = Packed Cell Volume; RBC = Red Blood Cell; WBC = White Blood Cell; Hbc = Haemoglobin Concentration; MCHC = Mean Cell Haemoglobin Concentration; MCH = Mean Cell Haemoglobin, MCV = Mean Cell Volume; ESR = Erythrocyte Sedimentation Rate.

concentration which could predispose birds to leucopaenia. The red blood cell counts showed that SWLM was most beneficial at 5% level, even better than the control diets. This threshold of tolerance with a corresponding increase in haemoglobin concentration is however substained at 5% SWLM inclusion level. Values obtained on this parameter for diets 2 and 3 were all in the neighbourhood of those given in the control treatment (Maxwell, 1982). The values for total

serum protein followed similar statistic. The reason for the reduced value of total serum protein might not be unconnected with reduced feed intakes (unpublished work, Fasuyi *et al.*), and subsequently reduced nutrient digestibility and other antinutritional factors inherent in most leaf meals as earlier discussed. The values obtained in this trial were higher than those quoted in literature and might be as a result of age difference of the birds which the various researchers experimented

Table 5: Some Serum Metabolites of Laying Hens Fed Varying levels of SWLM Based

Parameters	Diets			
	0.00 SWLM	2.50 SWLM	5.00 SWLM	7.50 SWLM
Total serum protein(g/100g)	5.3±0.4	5.3±1.4	6.1± 0.6	6.1± 0.7
Albumin (g/100g)	3.2±0.1	3.5±0.2	4.2±0.1	3.5± 0.1
Globulin (g/100g)	2.1±0.5	2.2±1.5	1.8±0.5	1.6±0.7
Albumin/Globulin Ratio	1.5±0.1	1.6±0.1	2.3±0.1	2.2± 0.1

Means with different superscripts in the same horizontal row are significantly different (P < 0.05); FM = Fish Meal; SWLM = siam weed leaf meal.

with (Narcim, *et al.*, 1961; Ross *et al.*, 1976; Rivetz *et al.*, 1977). The results obtained in this trial revealed that SWLM has no deleterious effect on serum albumin and globulin. The values for serum creatinine revealed no statistical difference just like in the some of hematological and biochemical parameters already discussed. Serum creatine is a measure of muscle mass. It gives the extent of catabolism of the muscular tissue and the rate of depletion of tissue creatine-phosphate (Alleyne *et al.*, 1970). The comparison of the value obtained with that of control showed that the inclusion of SWLM up to 7.5% level had no catabolic effect on the muscular tissue. Serum uric acid concentration followed the trends given by red blood cell count and total feed consumed and hence total protein intake. It is therefore in consonance with literature that uric acid excretion is directly proportional to protein intake (Waterlow and Stephen, 1981). The result obtained in this study compared with that obtained by Paul (1961) but deviated from that given by Ross *et al.* (1976). The serum urea nitrogen gave the same pattern like the one given by the serum uric acid. These parameters are measures of the nitrogen balance of the body and they are all pointers to the fact that the utilization of SWLM had no significant effect on nitrogen balance at these dietary levels.

**Conclusion:** The preparation of SWLM by harvesting the twigs and stems of the plants with minimum disturbance or bruising to the leaves and eventual sundrying had positive effects on the palatability and acceptability of SWLM as a protein source in layers diets. The dietary inclusion of SWLM even up to 7.5% does not result in mortality among the birds but an inclusion level of 5% seemed to support a desirable health status as indicated in the haematological and biochemical parameters studied.

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