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Influence of Dietary Solid-state Fermented Wheat Offal on Haematological and Serum Biochemical Indices of Broiler Chickens

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ABSTRACT

This study was conducted to determine the effect of dietary solid-state fermented wheat offal (SOSFWO) on haematological and serum biochemical indices of broiler chickens. A total of one hundred and eighty (180) broiler chickens (Cobb 500) aged 1- 7 weeks were distributed to six experimental units of 30 birds. Each unit was subdivided into three subunits of ten birds each in a completely randomized design. The SOSFWO was used to formulate six diets, it was included at 0, 5, 10, 15, 20 and 25% of the diets. Blood samples were collected and analysed for haematological and serum biochemical parameters. Data generated was subjected to one way analysis of variance significant differences were separated using Duncan's multiple range test. The findings showed significant ($p \leq 0.05$) increase in white blood corpuscles and lymphocyte as the level of SOSFWO increases. Similarly, serum albumin was significantly ($p \leq 0.05$) higher in the groups fed 5-25% SOSFWO compared to the control. Globulin was lower ($p \leq 0.05$) in chickens fed 20 and 25% SOSFWO. Alanine-amino transferase (ALAT) was lowest ($p \leq 0.05$) in the 25% SOSFWO group. It was concluded from this study that dietary SOSFWO had no negative effect on haematological and serum biochemical indices of broiler chickens.

Keywords: Broiler; Fermented wheat offal; Haematology; Serum biochemistry.

INTRODUCTION

The increased use of agro-industrial by-products (AIBs) that are abundant, low-cost and of low value in human nutrition have been recommended as one of the strategies to address the problem of perpetual high cost of major feed ingredients for poultry. This problem is caused to some extent by competition for the available grains between the poultry sector and man as sources of food and fuel. Wheat offal (WO) is one of the most preferred and utilized AIBs used in poultry feed formulations as conventional source of dietary fibre in livestock feeds in Nigeria (Makinde, *et al.*, 2017). However, the use of wheat offal in chicken feeding is limited because of its bulkiness, low nutritional value and anti-nutritional factors called non-starch polysaccharides (NSPs). NSPs have been implicated as a factor depressing nutrient digestibility, absorption, availability and efficient utilization in monogastric animals (Alayande *et al.*, 2016). Nutritionists have identified various processing methods to enhance utilization of AIBs like WO; they include the use of exogenous enzymes and more recently, application of bio-technological methods. The use of bio technological methods has made it possible to explore feed stuff that were hitherto neglected as valuable feed ingredient for livestock especially poultry. Fermentation is an effective means of breaking down anti-nutrients and increasing the nutritive value of AIBs (Elmasry *et al.*, 2017). It is projected that solid state

fermentation of wheat offal with rumen liquor will perhaps reduce its ANFs content and increase its nutritional worth such that some percentage of the costly ingredients (especially maize and soya beans) which accounts for about 70% of the total feed ingredients can be cheaply replaced by nutrient enriched wheat offal. Blood analysis is used to determine the responsiveness of an animal to its internal and external environment including feeds and feeding. It can provide vital information on the safety and efficacy of feed ingredient. The objective of the study was to determine the effect of solid-state fermented wheat offal on the haematological and serum biochemical indices of broiler chickens.

MATERIALS AND METHODS

Study Area

The experiment was conducted at the Department of Animal Science Teaching and Research Farm, University of Maiduguri, Maiduguri, Nigeria

Experimental Birds and Design

Total of one hundred and eighty (180) broiler chicks (Cobb 500) were sourced from Zartech hatchery were used for the experiment. The chickens were brooded for one week on commercial diet after which they were divided into six experimental units of thirty birds each. Each unit was

further divided into three subunits of ten birds each in a completely randomized design. The chickens were fed with the experimental starter diet for three weeks and the finisher diets for four weeks. Feed and water were given ad-libitum. All other routine husbandry practices were duly observed throughout the experimental period...The study was conducted according to local research guidelines of the Department of Animal Science University of Maiduguri.

Experimental Diets

Six experimental diets were formulated to meet the nutritional requirement of broiler chickens at the starter and finisher phases. At each phase, solid-state fermented wheat offal (SOSFWO) was included in the diets at 0, 5, 10, 15, 20 and 25 %. These were designated as T₁ (0%SOSFWO), T₂ (5% SOSFWO), T₃ (10%SOSFWO), T₄ (15% SOSFWO), T₅ (20%) and T₆ (25% SOSFWO), respectively. Composition of the experimental diet is presented in Table 1.

Measurement of Response Criteria

At the end of the experiment, two blood samples were collected from two birds that were randomly selected from each replicate after overnight fasting to avoid elevation of metabolites by feeding. The samples were collected in well labelled plain and anticoagulant containing bottles. The samples in the bottles containing anticoagulant were processed for haematological parameters. The samples in the plain bottles were centrifuged for five minutes to separate the serum from the blood for serum biochemical indices. Haematological parameters such as Red Blood Cells (RBC) count, White Blood Cell (WBC) count, Packed Cell Volume (PCV), Haemoglobin (Hb) concentration together with differential counts of Neutrophils, Lymphocytes and Eosinophils were determined according to Jain (1986). Erythrocyte indices such as the Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC) were in according to Jain (1986). Serum biochemical indices such as the total serum protein and serum albumin and urea and cholesterol were determined according to Bush, (1991).

Table 1: Composition and Calculated Analysis of Experimental Broiler Finisher Diets

Level of solid-state fermented wheat offal (SOSFWO) inclusion (%)						
Ingredients (%)	T ₁ (0)	T ₂ (5)	T ₃ (10)	T ₄ (15)	T ₅ (20)	T ₆ (25)
Maize	55.00	54.00	53.00	52.00	49.00	46.00
Groundnut cake	26.00	22.00	20.00	18.00	15.00	12.00
RUFFWO	00.00	05.00	10.00	15.00	20.00	25.00
Wheat offal	08.00	06.00	03.00	01.00	00.00	00.00
Fish meal	03.00	04.00	05.00	05.00	06.00	06.00
Blood meal	02.00	02.00	02.00	02.00	02.00	02.00
Bone meal	04.00	04.00	04.00	04.00	04.00	04.00
Common salt	00.30	00.30	00.30	00.30	00.30	00.30
Min- Vit premix	00.40	00.40	00.40	0.400	00.40	0.400
Lysine	00.10	00.10	00.10	00.10	00.10	00.10
Methionine	00.20	00.20	00.20	00.20	00.20	00.20
Palm oil	01.00	02.00	02.00	02.00	03.00	04.00
Total (%)	100.00	100.00	100.00	100.00	100.00	100.00
Calculated Analysis (%)						
Crude Protein	20.30	20.05	20.07	20.00	19.98	20.00
Fibre	03.21	03.36	03.34	03.34	03.35	03.35
Calcium	01.19	01.24	01.26	01.26	01.28	01.30
Phosphorus	00.85	00.76	00.77	00.77	00.61	00.73
Lysine	01.14	01.13	01.14	01.11	01.06	01.08
Methionine	00.53	00.57	00.55	00.58	00.61	00.60
ME (kcal/kg)	2926.33	2934.60	2952.17	2929.10	2926.93	2967.80

ME = Metabolizable energy *1kg of premix contains: Vitamins A (5, 000, 000 I.U), Vitamin D₃ (1000000 I.U), Vitamin E (16000mg), Vitamin K₃ (800mg), Vitamin B₁ (1200mg), Vitamin B₂ (22000mg), Niacin (22000mg), Calcium pantothenate (4600mg), Vitamin B₆ (200mg), Vitamin B₁₂ (10mg), Folic acid (400mg), Biotin (32mg), Choline chloride (200000mg), Manganese (948000mg), Iron (40000mg), Zinc (32000mg), Copper(3400mg), Iodine (600mg), Cobalt (120mg), selenium (48mg), Anti-Oxidant (48000mg)

RESULTS

Effect of dietary SOSFWO on Haematological Indices of Broiler Chickens

The effect of dietary SOSFWO on haematological indices of broiler chickens is presented in Table 2. Significant ($P \leq 0.05$) effects were found among the treatment groups for red blood cells (RBC), white blood cells (WBC), mean corpuscular volume (MCV) mean corpuscular haemoglobin (MCH), lymphocytes and monocytes. No significant ($P > 0.05$) differences were observed for packed cell volume (PCV), Haemoglobin concentration (Hb), neutrophils, eosinophils and basophils and mean corpuscular haemoglobin concentration (MCHC).

The lowest RBC value was observed in chickens on the T₁ and T₄ (control and 15% SOSFWO) groups. The value was similar to those values obtained for groups on T₃ and T₅ (10 and 20% SOSFWO) diets. Chickens on T₆ (25% SOSFWO) diets had a value similar to the values obtained for chickens on T₂, T₃ and T₅ (5.10 and 20% SOSFWO) groups. MCV values recorded for chickens on the control group was higher and statistically ($p < 0.05$) similar to the values found in chickens on T₂, T₃, T₄ and T₆ (5, 10, 15, 20 and 25% SOSFWO) diets. Similarly, MCH was significantly ($p < 0.05$) higher in groups fed the T₁ and T₄ (control and 15% SOSFWO) diets. Groups on T₂, T₃, T₄ and T₆ (5, 10, 20 and 25%) SOSFWO diets had similar values with the T₁ and T₄ (control and 15% SOSFWO)

groups. The least value (6.08pg) was found in group on T₂ (5% SOSFWO) diet which was also similar to the value found in chickens on T₁, T₃, T₄ and T₆ (0, 10, 20 and 25% SOSFWO) diets.

Findings of this study revealed significantly ($P \leq 0.05$) higher WBC value in chickens on T₆ (25% SOSFWO) than other groups. Chickens on T₅ (20% SOSFWO) had a value that was statistically similar to the value obtained for chickens on T₂, T₃ and T₆ (5, 15 and 25% SOSFWO) groups. Group on T₄ (15% SOSFWO) diet had value that was similar to the values obtained for chickens on T₁, T₂, T₃ and T₅ (0, 5, 10 and 20% SOSFWO) diets. The lowest WBC value was found in the control group. The value is

also similar to value recorded for chickens on T₃ and T₄ (10 and 15% SOSFWO) groups.

The highest ($p \leq 0.05$) lymphocytes value was recorded for chickens on T₆ (25% SOSFWO) diets. The value was similar to values found in chickens on 5 and 15 SOSFWO groups. Chickens on the T₁, T₃ and T₅ (control, 10 and 20% SOSFWO) groups had similar values that were lower than values obtained for chickens on T₂, T₄ and T₆ (5, 15 and 25% SOSFWO) diets. Chickens on T₅ (20% SOSFWO) diet had the highest monocytes value (0.67%), The value is similar to values obtained for chickens on the control (0.33%). No monocytes were observed in the remaining groups.

Table 2: Effect of Solid-state Fermented Wheat Offal (SOSFWO) on Haematological Indices of Broiler Chickens

Parameters	Treatments /level of SOSFWO inclusion						SEM	Normal range
	T ₁ (0)	T ₂ (5)	T ₃ (10)	T ₄ (15)	T ₅ (20)	T ₆ (26)		
PCV (%)	29.67	27.67	26.67	31.33	29.00	29.00	0.69 ^{ns}	25-45 ¹
RBC($\times 10^6$ /L)	3.73 ^a	3.47 ^c	3.33 ^{bc}	3.73 ^a	3.60 ^{abc}	3.63 ^{ab}	0.14 [*]	2-4 ¹
Hb (g/dl)	9.87	9.20	8.87	10.40	9.63	9.63	0.28 ^{ns}	7-13 ¹
MCV (fl)	79.5 ^{ab}	79.7 ^{ab}	80.0 ^a	83.9 ^a	79.8 ^{ab}	79.8 ^{ab}	0.41 [*]	90 - 140 ²
MCH (pg)	26.5 ^{ab}	26.5 ^{ab}	26.6 ^{ab}	27.9 ^a	26.5 ^{ab}	26.5 ^{ab}	0.13 [*]	33- 47 ²
MCHC g/dl	33.26	33.25	33.25	33.19	33.22	33.21	0.02 ^{ns}	26 -35 ²
WBC ($\times 10^3$ /L)	11.47 ^d	12.67 ^{bc}	12.27 ^{cd}	12.57 ^{bcd}	13.53 ^{ab}	14.20 ^a	0.14 [*]	9- 31 ¹
Neutrophils (%)	29.33	27.33	34.33	26.00	29.67	18.00	2.47 ^{ns}	15 - 40 ¹
Basophils (%)	1.67 ^a	1.33 ^a	0.67 ^{ab}	0.00 ^b	0.00 ^b	1.00 ^{ab}	0.33 ^{ns}	0-2 ³
Lymphocyte (%)	68.67 ^b	71.33 ^a	65.00 ^a	74.00 ^a	69.67 ^b	81.00 ^a	2.43 [*]	45.00-70.00 ²
Monocytes (%)	0.33 ^b	0.00 ^b	0.00 ^b	0.00 ^b	0.67 ^a	0.00 ^b	0.07 [*]	5- 10 ²

RBC= Red Blood Cells, WBC=White Blood Cell, PCV=Packed Cell Volume, Hb=Haemoglobin, MCV mean corpuscular volume, MCH=mean corpuscular haemoglobin; MCHC=mean corpuscular haemoglobin concentration a,b,c values bearing different superscript within rows show significant difference ($P \leq 0.05$)

1 = Banerjee (2006); 2=Jain (1986); 3 = Nanbol et al. (2016)

a, b, c Means on the same row having different superscripts are significantly different ($P \leq 0.05$)

SEM: Standard Error of Mean, ns = not significantly different ($p > 0.05$)

Effect of dietary solid-state fermented wheat offal (SOSFWO) on serum biochemical indices of broiler chickens

The effect of dietary solid-state fermented wheat offal (SOSFWO) on serum biochemical indices of broiler chickens is presented in Table 3. Significant ($P \leq 0.05$) differences were found between the treatment groups for

albumin, globulin, alanine amino transferase (ALAT) and creatinine values. There was however, no significant difference ($P > 0.05$) in total protien, aspartate amino transferase (ASAT), glucose, urea and electrolytes (Ca, Na, K and Cl). However, all values fall within normal range reported for healthy chickens (Table 3).

Table 3: Serum Biochemical Indices of Broiler Chickens fed Solid-state Fermented Wheat Offal (SOSFWO)

Parameters/Treatments	Level of SOSFWO inclusion (%)						SEM	Normal range
	T ₁ (0)	T ₂ (5)	T ₃ (10)	T ₄ (15)	T ₅ (20)	T ₆ (25)		
Protien (g/L)	46.33	44.33	48.33	48.33	41.67	42.67	0.87 ^{ns}	40-62 ¹
Albumin g/l	24.71 ^b	31.32 ^a	30.00 ^a	31.00 ^a	30.70 ^a	31.07 ^a	0.56 [*]	17- 30 ¹
Globulin (g/l)	21.74 ^a	13.00 ^{ab}	18.30 ^{ab}	17.30 ^{ab}	11.0 ^b	11.00 ^b	1.07 [*]	23 - 32 ¹
ASAT IU/L	39.67	31.33	29.00	24.33	33.33	53.33	3.94 ^{ns}	88 - 208. ²
ALAT IU/L	25.67 ^a	23.00 ^{bc}	30.33 ^b	22.67 ^{bc}	19.33 ^{bc}	10.00 ^c	1.70 [*]	10-37 ²
Glucose (mmol/L)	5.87	5.40	5.70	5.73	5.97	6.33	0.16 ^{ns}	4.0-10.0 ¹
Urea (g/L)	4.30	5.17	5.33	5.53	4.50	4.93	0.15 ^{ns}	0.5 - 6 ²
Creatinine(mg/l)	55.7 ^{bc}	57.0 ^{bc}	62.3 ^a	53.3 ^{bc}	45.3 ^c	59.0 ^{ab}	0.97 [*]	4.00 - 7.50 ³
Na (mmol/L)	138.33	140.67	136.67	140.00	138.00	141.00	139.11	129-142 ³
K (mmol/L)	3.50	3.40	3.37	4.13	3.57	3.40	3.56	3.0-3.97 ³
CL (mmol/L)	98.67	99.33	100.67	99.67	101.33	101.33	100.17	54-70 ¹
Ca (mmol/dl)	2.30	2.30	2.33	2.33	2.40	2.30	2.33	2 -4 ³
Cholesterol (mmol/l)	3.73 ^{ab}	3.13 ^{bc}	4.30 ^a	3.67 ^{ab}	2.57 ^c	3.50 ^{ab}	0.24	4.5 - 7.0 ³

1 = Nanbol; 2=Bernajee; 3=Anon (1980.) a, b, c Means on the same row having different superscripts are significantly different ($P \leq 0.05$), ns = not significantly different ($p > 0.05$) SEM: Standard Error of Mean, ALAT=Alanine amino transferase; ASAT =Aspartate amino transferase

The result of this study revealed significantly ($P \leq 0.05$) higher albumin values in chickens on the SOSFWO groups (30.00 – 31.67 g/dl) compared to the control which recorded the lowest value (24.67 g/dl). Globulin was significantly ($P \leq 0.05$) higher in chickens on the T₁ (control group) (2.17g/dl). The value was, statistically similar to values obtained for chickens on T₂, T₃ and T₄ (5, 10 and 15% SOSFWO) levels. Chickens on T₅ and T₆ (20 and 25% SOSFWO) levels had statistically similar and lowest value (1.10 and 1.10 g/dl respectively).

Creatinine values observed in this study were highest ($P \leq 0.05$) in chickens on T₃ (10% SOSFWO) level. The lowest level was found in chickens on T₅ (20% SOSFWO diet). Chickens on the T₁, T₂, and T₆ (control 5 and 25% SOSFWO) groups had similar values that were also similar to values obtained for groups on T₃ and T₄ (10 and 15% SOSFWO) groups.

Alkaline amino transference (ALAT) values were highest ($p \leq 0.05$) in chicken on T₃ 10% SOSFWO (30.33IU/l). The value is however, similar to value obtained for chickens on T₁, T₂, T₄ and T₅ (0, 5, 15 and 20% SOSFWO) levels. The lowest value was recorded for chickens on T₆ (25% SOSFWO) groups (10.00).

Cholesterol values for chickens on the T₁, T₄ and T₆ (control, 15 and 25% SOSFWO diets) were similar and higher ($P < 0.05$) than values obtained for chickens on T₅ (20% SOSFWO diets). The highest value (4.30mmol/l) was found in chickens on 10% SOSFWO diet. Chickens on T₂ (5% SOSFWO diet) had values that were similar to values obtained in chickens on T₁, T₄, T₅ and T₆ (0, 15, 20 and 25% SOSFWO).

DISCUSSION

The RBC values found in this study are similar to the values ($3.56 - 3.83 \times 10^6/L$) reported by Abdulazeez *et al.* (2022) in egg type chickens fed with SOSFWO diets. They are within the normal range reported for healthy chickens by Bernarjee (2006). The trend in WBC observed in this study showed relatively higher WBC in the SOSFWO groups than the control. The findings of this study is in consonance with the report of Kang *et al.* (2015) who found significant differences in white blood cell (WBC) in chickens fed fermented rice bran. Since WBC are known to fight against diseases, animals with low white blood cells are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the process of phagocytosis and have higher degree of resistance to diseases (Soetan *et al.*, 2013). This report, coupled with high serum globulin and higher mortality observed in the study in chickens on control, may indicate that feeding SOSFWO had conferred some immunological advantage the chickens against the control group.

Lymphocytes (T, B and killer cells) are responsible for antibody production, mediation of immune response and killing viral infections. An increase in lymphocyte count is a sign of viral infection Yousaf *et al.* (2024). The result of this study indicates that chickens on the SOSFWO groups had generated higher number of lymphocytes for immunological defence against new castle disease infection observed during the study than the other groups.

Monocytes are macrophages that engulf and ingest pathogenic materials. High monocyte count occurs as a result of chronic or sub-acute infection or during recovery from acute infection (Web-md, 2021). The findings of this study is in consonance with the report of Kang *et al.* (2015) who found significant differences in, monocytes, eosinophils and basophils levels with trend showing better immune status in chickens fed fermented rice bran.

For the serum biochemical indices, the relatively higher albumin observed in chickens on SOSFWO groups may perhaps be to better crude protein (CP) content (21%) of the test material. Globulin is responsible for antibodies (immunoglobulin) production they are elevated in inflammatory condition and infection. Values obtained in this study are lower than normal range reported by Nanbol *et al.* (2016). The findings of this study are consistent with the reports of Abdulazeez *et al.* (2022) who showed significant effect of SOSFWO on serum albumin, globulin and creatinine in pullets. Creatinine test measures how well kidneys are performing their jobs of filtering waste from the blood. High levels of creatinine are as a result of kidneys malfunction, and exposure to toxic substance while low levels of creatinine might be due to muscle degeneration, liver disease, and significant fluid overload (Champe *et al.*, 2008). Values obtained in this study are within normal range of 4.00 - 7.50 mg/dl reported by Jain (1986).

Elevated level of ALAT is a specific indication of liver damage while low ALAT level results in a normal healthy liver. The findings of this study are in consonance with the reports of Elmasry *et al.* (2017) who showed significant effect of fermented wheat bran on plasma levels of serum glutamic oxaloacetic transaminase (SGOT) and serum glutamic pyruvic transaminase (SGPT). Values obtained in this study are similar to range of 7- 35 IU/l reported by Majekodunmi *et al.* (2013). With the exception of 2.57mmol/l value found in groups fed 20% SOSFWO diet, cholesterol values found in this study were within normal range reported for healthy chickens.

Blood parameters are good indicators of physiological, pathological and nutritional status of an animal. The values for haematological and serum biochemical indices observed in this study indicate that there is nutritional adequacy and safety of the SOSFWO. Thus, the test material had, no adverse effect on nutritional and health status of the broiler chickens.

Conclusion

It was concluded from this study that solid-state fermented wheat offal can be included up-to 25% in the diet of broiler chickens with no adverse effect on the haematological and serum biochemical indices.

Conflict of interest

The authors declare that there is no conflict of interest

Author Contribution

H.A, conceived and designed the experiments, reviewed the final manuscript ASS conducted the experiment, RMS performed the experiment, IL supervised the work, analyzed the data; wrote the article.

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