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Research Article
Hematological and Biochemical Indices of West African Dwarf Sheep Fed Diets Containing Yeast (*Saccharomyces cerevisiae*), Grass, Grass/Legume (50:50) and Legume

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Abstract

Background and Objective: The European Union banned the use of antibiotics for non-therapeutic purposes because of the possibility of the transfer of antibiotic resistance to pathogenic bacteria in humans. It is therefore imperative to find safe alternatives to the use of antibiotics. The aim of this study was to determine the effects of the dietary inclusion of yeast (*Saccharomyces cerevisiae*) on hematological and biochemical indices of West African dwarf sheep. **Materials and Methods:** A total of twenty four (24) lambs (12 males and 12 females) with an average weight of 10.30 kg were randomly allotted to six treatment diets in a 3×2 factorial arrangement involving grass (*Panicum maximum*) hay, grass-legume mixture (50:50) hay and legume (C*entrosema pubescens*) hay, as well as with two yeast levels (0 and 1.5 g per kg of basal diet). The six diets were abbreviated as G0, G1.5, G/L0, G/L1.5, L0 and L1.5 (G: grass, L: Legume, G/L: Grass/legume (50:50) mixture, 0: 0 g of *S. cerevisiae* per kg of diet and 1.5:1.5 g of *S. cerevisiae* per kg of diet). **Results:** The results showed that the packed cell volume, hemoglobin concentration and white blood cell count were significantly ($p<0.05$) higher for sheep fed a legume diet supplemented with *S. cerevisiae* compared to that for sheep fed other diets. Sheep fed the grass and legume mixture and the legume diets supplemented with *S. cerevisiae* had significantly ($p<0.05$) higher albumin values than those of sheep fed other diets. Sheep fed the legume diet without *S. cerevisiae* supplementation had the highest calcium values of all sheep diet groups tested. **Conclusion:** Based on the results obtained, the addition of 1.5g of *S. cerevisiae* per kg of legume diet is recommended.

Key words: Biochemistry, grass, hematology, legume diets, *S. cerevisiae*, sheep

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

There is a public and scientific concern about the widespread use of antibiotics and the possibility for the transfer of antibiotic resistance to pathogenic bacteria in humans[1]. In addition, the presence of antibiotic residues in the meat may have deleterious effects on human consumers. For these reasons the European Union banned the use of antibiotics for non-therapeutic purposes in January 01, 2006. It is therefore imperative to find safe alternatives to the use of antibiotics. Yeast and fungal probiotics, such as *Saccharomyces cerevisiae* and Amaferm (*Aspergillus oryzae*), have yielded better results in adult ruminants[2]. The most common marketed products for ruminants contain live yeast (*S. cerevisiae*), which is widely used as a feed additive because of its beneficial effects on animal performance[3]. The cells are dried to preserve viability and metabolic activity. The effectiveness of fungal probiotics stems from their influence on rumen fermentation and they fall into the category of ruminal modifiers. Yeasts are most efficient when the rumen is not functioning optimally and when diets are overloaded with easily fermentable energy components or are poor in nutrients. The selection of a suitable microorganism strain is a primary requirement for its use as a probiotic.

The inclusion of probiotics in feeds is designed to encourage the growth of certain strains of microbes in the gut at the expense of less desirable ones. Unlike the destructive action of antibiotics, S. *cerevisiae* is able to grow rapidly in the rumen and facilitate fiber digestion. Micro-nutrients found in *S. cerevisiae* also stimulate cellulolytic bacteria growth. *S. cerevisiae* in the rumen can utilize the remaining dissolved oxygen and save anaerobic microorganisms from the toxic effect of oxygen. Live yeasts are also able to improve the rumen maturity and stabilize the ruminal pH, thus reducing the risk of acidosis by competing with lactic acid- producing bacteria[4,5].

The supplementation of yeast in the ruminant diet is known to improve feed intake[6], milk production[7], weight gain[8], digestion[9], the numbers of anaerobic and cellulolytic bacteria[10] and alter the patterns of volatile fatty acids[11] or even supply the animal with unknown growth factors[12]. Yeast have positive effects on blood hematology resulting in the improvement in the health status of animals[13]. The addition of yeast culture to feed has many positive effects on the absorption of some minerals and improves the metabolic health of animals[14]. Live yeast, cultures were reported to influence blood constituents through the remodeling of ruminal microbial populations. *Saccharomyces cerevisiae* were found to produce vitamins B, positively affecting blood-cell forming processes[15]. Nevertheless, the results of these studies have been variable and are strongly influenced by feed composition. Taking these findings into consideration, the present study was conducted to determine the effects of the dietary inclusion of yeast (*S. cerevisiae*) in animal feed on the hematological and biochemical indices of West African dwarf sheep fed diets based on grass, grass/legume and legume.

MATERIALS AND METHODS

The study was carried out at the Sheep and Goat Unit of the Department of Animal Science Teaching and Research Farm, University of Nigeria, Nsukka, Enugu State, Nigeria. The yeast (*Saccharomyces cerevisiae*) was procured from B.F.P. Dock Road, Felixstowe, U.K.

Experimental animals and management: Twenty four lambs (12 males and 12 females) with an average weight of 10.30±.079 kg were used for the study. The animals were randomly divided into six treatment groups of four sheep each and assigned to six diets in a 3×2 factorial arrangement involving grass (*Panicum maximum*) hay, grass-legume mixture (50:50) hay and legume (C*entrosema pubescens*) hay and the animals were supplemented with two yeast levels (0 and 1.5 g per kg of basal diet). The six dietary treatments were as follows: treatment 1 was grass hay alone with no inclusion of *S. Cerevisiae*, treatment 2 was grass hay alone with 1.5 g of *S. cerevisiae* per kg of diet; treatment 3 was grass/legume mixture (50:50) hay with no inclusion of *S. Cerevisiae*, treatment 4 was grass/legume mixture (50:50) hay with 1.5 g of *S. cerevisiae* per kg of diet; treatment 5 was legume hay alone with no inclusion of *S. Cerevisiae* and treatment 6 was legume hay alone with 1.5 g of *S. cerevisiae* per kg of diet. Each group was made up of four replicates with each sheep serving as a single replicate. Approximately 500 g of each diet was given to each animal daily in the morning and the left over feed was weighed the following morning to determine the average daily feed intake (ADFI). Water was provided to the animals *ad libitum*. The animals were housed individually in pens and the initial weights of the animals were measured. Twenty-one days prior to the start of the experiment, all the animals were allowed to acclimate and the experimental diets were gradually introduced. The animals were vaccinated with the PPR vaccine, dewormed with Albendazole and injected with Oxytetracycline LA to prevent bacterial infections. Chemical analysis of the diets for dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE) and crude fiber (CF) were determined according to

the AOAC method[16]. The neutral detergent fiber (NDF) and acid detergent fiber (ADF) compositions were determined according to the method of Goering and Van Soest[17].

Blood collection: At the 8th and 12th weeks of the experimental period blood was collected in the morning from each sheep. Ten milliliters of blood were collected from the jugular vein of each animal using a sterile disposable syringe. Five mL were emptied into sterile sample bottles containing the anti-coagulant Ethylene Diamine tetra acetic acid (EDTA) for laboratory analysis to determine hematological indices. The remaining 5 mL of blood were emptied into sample bottles without EDTA for serum extraction and biochemical analysis.

The packed cell volume (PCV) was determined by the microhematocrit method[18]. The hemoglobin concentration (HbC) was determined by the cyanmethemoglobin method[19]. The red blood cell (RBC) and the total white blood cell (WBC) counts were determined by the hemocytometer method[18]. The Differential White Blood Cell (Leukocyte) Count was determined by Leishman Technique[18]. The mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated using the standard formula[20].

Serum total protein (TP) concentration was determined using the Tietz[21] method. Serum albumin (ALB) concentration was determined using the method of Grant *et al.*[22]. The determination of the plasma globulin level was by the following formula:

$$\text{Plasma globulin} = \text{Total protein (TP)} - \text{Plasma albumin (ALB)}$$

The phosphorus (P) concentration was determined using the phosphomolybdate method as described by Pearson[23]. Sodium (Na), potassium (K) and calcium (Ca) concentration were determined by the Flame photometric method as described by Pearson[23].

Statistical analysis: The collected data were subjected to two way analysis of variance (ANOVA) for factorial arrangement in a completely randomized design (CRD). Significantly different means were separated using Duncan's New Multiple Range Test[24]. The treatment effects were considered significant at $p<0.05$.

RESULTS

Chemical composition of the experimental diets: The chemical composition of the experimental diets is presented in Table 1.

There were significant ($p<0.05$) differences among treatments in the DM, OM, CP, CF, EE, ash, nitrogen free extract (NFE), NDF and ADF percentages, in the grass, a combination of grass and legume or legume diets. The composition percentages of DM (65.23), CF (33.80), NDF (62.42) and ADF (42.24) were highest in the grass diet, followed by the grass/legume diet, while the composition percentages of DM, CF, NDF and ADF were lowest in the legume diet. The composition percentages of OM (56.45), CP (22.42) and NFE (55.50) were highest in the legume diet. The grass and grass/legume diets had similar and higher EE and ash percentages, respectively, compared with those of the legume diet.

Effects of diet types with or without *Saccharomyces cerevisiae* supplementation, on the hematology of West African dwarf sheep: The main effects of diet types (DT) and *S. cerevisiae* supplementation levels (CL), as well DT×CL interactions on the hematology of West African dwarf sheep are presented in Table 2.

There were no significant effects ($p>0.05$) due to the DT on MCV, neutrophil, lymphocytes, monocytes and eosinophil counts of sheep fed grass, a mixture of grass and legume, or legume. The PCV, RBC, MCHC, HbC and WBC counts, as well as

Table 1: Chemical composition of the experimental diets

Chemical composition (%)	G	GL	L	SEM
DM	65.23^{a}	60.23^{b}	58.72^{c}	0.26
OM	52.84^{b}	51.68^{b}	56.45^{a}	0.36
CP	11.90^{c}	16.82^{b}	22.42^{a}	0.10
CF	33.80^{a}	30.54^{b}	23.17^{c}	0.27
EE	2.65^{a}	2.58^{a}	1.80^{b}	0.10
Ash	6.64^{a}	6.55^{a}	6.23^{b}	0.03
NFE	38.40^{c}	44.66^{b}	55.50^{a}	0.69
NDF	62.42^{a}	58.56^{b}	53.26^{c}	0.15
ADF	42.24^{a}	39.67^{b}	36.21^{c}	0.21

G: Grass; G-L: Grass and Legume (50:50), L: Legume, CP: Crude protein, CF: Crude fiber, EE: Ether extract, NFE: Nitrogen free extract, NDF: Neutral detergent fiber, ADF: Acid detergent fiber, a,b: Means on the same row with different superscripts are significantly ($p<0.05$) different, SEM: Standard error of the mean

Table 2: Effect of diet type with or without *Saccharomyces cerevisiae* supplementation on the hematology of West African dwarf sheep

Items	PCV (%)	RBC ($\times10^6$ μ^{-1})	MCHC (g dL^{-1})	HbC (g dL^{-1})	MCV (fl)	MCH (pg)	WBC ($\times10^3$ μ^{-1})	Neutrophil ($\times10^3$ μ^{-1})	Lymphocytes ($\times10^3$ μ^{-1})	Monocytes ($\times10^3$ μ^{-1})	Eosinophil ($\times10^3$ μ^{-1})
Main effect of feed type											
G	33.81^{c}	9.38^{c}	33.75^{a}	9.43^{c}	26.66	9.69^{b}	5.29^{c}	3.15	2.37	0.46	0.06
G-L	37.75^{b}	12.65^{b}	31.23^{c}	11.31^{b}	26.04	10.37^{b}	7.26^{b}	3.42	2.30	0.51	0.05
L	39.38^{a}	13.60^{a}	32.53^{b}	13.34^{a}	26.65	10.98^{a}	8.86^{a}	3.14	2.62	0.65	0.05
SEM	0.33	0.18	0.41	0.20	1.32	0.35	0.16	0.29	0.18	0.08	0.01
Main effect of SC supplementation											
0 g kg^{-1}	35.42^{b}	11.27^{b}	34.13^{a}	10.63^{b}	22.93^{b}	9.38^{b}	6.63^{b}	3.26	1.73^{b}	0.50	0.06
1.5 g kg^{-1}	38.54^{a}	12.49^{a}	30.87^{b}	12.1^{a}	29.98^{a}	11.31^{a}	7.65^{a}	3.21	3.14^{a}	0.58	0.05
SEM	0.27	0.15	0.33	0.16	1.08	0.28	0.13	0.23	0.15	0.06	0.01
Interaction (feed type with SC supplementation											
G (SC-0 g kg^{-1})	30.75^{d}	8.54	36.41^{a}	8.98^{d}	23.85	8.55	4.35^{d}	3.06	1.70	0.36	0.06
G (SC-1.5 g kg^{-1})	36.88^{c}	10.23	31.08^{c}	9.89^{c}	29.48	10.83	6.23^{c}	3.24	3.04	0.56	0.06
G-L (SC-0 g kg^{-1})	37.00^{c}	11.00	32.38^{b}	10.06^{c}	22.10	8.26	6.29^{c}	3.51	1.49	0.50	0.06
G-L (SC-1.5 g kg^{-1})	38.50^{b}	13.30	30.09^{d}	12.56^{b}	29.99	11.48	7.54^{b}	3.33	3.12	0.53	0.05
L (SC-0. g kg^{-1})	38.50^{b}	12.16	33.60^{b}	12.85^{b}	22.83	10.33	7.55^{b}	3.21	1.99	0.65	0.06
L (SC-1.5 g kg^{-1})	40.25^{a}	13.95	31.45^{c}	13.84^{a}	30.48	11.64	9.18^{a}	3.08	3.26	0.65	0.05
SEM	0.47	0.26	0.58	0.28	1.87	0.49	0.22	0.41	0.26	0.11	0.01

G: Grass, G-L: Grass and Legume, L: Legume, G (SC-0 g kg^{-1}): Grass with *S. cerevisiae* (SC) supplementation at 0 g kg^{-1} feed, G (SC-1.5 g kg^{-1}): Grass with SC supplementation at 1.5 g kg^{-1} feed, G-L (SC-0 g kg^{-1}): Grass and Legume with SC supplementation at 0 g kg^{-1} feed, G-L (SC-1.5 g kg^{-1}): Grass and Legume with SC supplementation at 1.5 g kg^{-1} feed, L (SC-0 g kg^{-1}): Legume with SC supplementation at 0 g kg^{-1} feed, L (SC-1.5 g kg^{-1}): Legume with SC supplementation at 1.5 g kg^{-1} feed, PCV: Packed cell volume, RBC: Red blood cell count, MCHC: Mean corpuscular hemoglobin concentration, HbC: Hemoglobin concentration, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, WBC: White blood cell count, SEM: Standard error of the means, a,b,c,d: Means on the same column with different superscripts are significantly ($p<0.05$) different

MCH were significantly ($p<0.05$) affected by diet types. The PCV (39.38%), RBC (13.60×10^6 μ^{-1}), HbC (13.34 g dL^{-1}) and WBC count (8.86×10^3 μ^{-1}) values of sheep fed the legume diet were the highest ($p<0.05$), followed by those of sheep fed diets containing a mixture of legume and grass while the PCV (33.81%), RBC (9.38×10^6 μ^{-1}), HbC (9.43 g dL^{-1}) and WBC count (5.29×10^3 μ^{-1}) values of sheep fed the grass diet were the lowest ($p<0.05$). The MCHC value (33.75 g dL^{-1}) of sheep fed the grass diet was the highest ($p<0.05$), followed by that of sheep fed the legume diet (32.53 g dL^{-1}) while the MCHC value (31.23 g dL^{-1}) of sheep fed the mixture of legume and grass was the lowest ($p<0.05$). The MCH value (10.37pg) of sheep fed the legume diet was higher ($p<0.05$) than those of sheep fed the diet containing a mixture of legume and grass (10.37pg) or grass diets (9.69 pg), whose values were similar ($p>0.05$). There were no significant effects ($p>0.05$) due to the CL on neutrophil, monocytes and eosinophil counts of sheep fed diets with or without *S. cerevisiae* supplementation. However, there were significant effects ($p<0.05$) due to CL on PCV, RBC, MCHC, HbC, MCV, MCH and WBC counts and on the lymphocytes counts of sheep fed diets with or without *S. cerevisiae* supplementation. All the sheep fed diets with *S. cerevisiae* had higher ($p<0.05$) PCV (38.54%), RBC (12.49×10^6 μ^{-1}), HbC (12.1 g dL^{-1}), MCV (29.98fl), MCH (11.31 pg) and WBC (7.65×10^3 μ^{-1}) and lymphocytes counts (3.14×10^3 μ^{-1}) as well as lower MCHC (30.87 g dL^{-1}) values than those of their counterparts fed diets without *S. cerevisiae* supplementation whose values were as follows: PCV (35.42%), RBC (11.27×10^6 μ^{-1}), HbC (10.63 g dL^{-1}), MCV (22.93 fl), MCH (9.38 pg), WBC (6.63×10^3 μ^{-1}), lymphocytes (1.73×10^3 μ^{-1}) and higher MCHC (34.13 g dL^{-1}).

Significant ($p<0.05$) DT×CL interactions in PCV, MCHC, HbC and WBC count values existed. However, there were no significant ($p>0.05$) DT X CL interactions in RBC, MCV, MCH, neutrophils, lymphocytes, monocytes and eosinophil values. Sheep fed the legume diet with the *S. cerevisiae* additive had the highest ($p<0.05$) PCV (40.25%), HbC (13.84 g dL^{-1}) and WBC count (9.18×10^3 μ^{-1}) values, while PCV (30.75%), HbC (8.98 g dL^{-1}) and WBC count (4.35×10^3 μ^{-1}) values of sheep fed the grass diet without *SC* were the lowest ($p<0.05$). Sheep fed the grass diet without the *S. cerevisiae* additive had the highest ($p<0.05$) MCHC (36.41%) value while the MCHC (30.09%) value of sheep fed the grass:legume mixture diet with *S. cerevisiae* was the lowest ($p<0.05$).

Effects of feed type and *S. cerevisiae* supplementation and their interactions on the blood biochemistry of West African dwarf sheep: The main effect of diet type (DT) and *S. cerevisiae* supplementation levels (CL) and the DT×CL interactions on the blood biochemistry of West African dwarf sheep are presented in Table 3.

There were no significant effects ($p>0.05$) due to the DT on Na and K values of sheep fed the grass, mixture of grass

Table 3: Effects of diet type with or without *Saccharomyces cerevisiae* supplementation on the blood biochemistry of West African dwarf sheep

Items	TPP (g dL^{-1})	Albumin (g dL^{-1})	Globulin (g dL^{-1})	Ca (mg dL^{-1})	P (mg dL^{-1})	Na (mmol L^{-1})	K (mmol L^{-1})
Main effect of feed type							
G	6.91^{b}	4.31^{b}	2.66^{c}	8.17^{c}	5.76^{b}	143.56	4.82
G-L	7.98^{a}	4.50^{a}	3.55^{b}	9.34^{b}	6.14^{b}	143.62	4.91
L	8.38^{a}	4.58^{a}	4.36^{a}	11.32^{a}	6.81^{a}	144.00	4.94
SEM	0.15	0.03	0.11	0.12	0.14	0.42	0.16
Main Effect of SC supplementation							
0 g kg^{-1}	7.24^{b}	4.22^{b}	3.21^{b}	10.31^{a}	7.02^{a}	143.85	4.94
1.5 g kg^{-1}	8.28^{a}	4.70^{a}	3.83^{a}	8.91^{b}	5.46^{b}	143.60	4.84
SEM	0.12	0.03	0.09	0.10	0.12	0.35	0.13
Interaction (feed type with SC supplementation)							
G (SC-0 g kg^{-1})	6.37	4.23^{cd}	2.30	8.40^{c}	6.41	143.94	4.71
G (SC-1.5 g kg^{-1})	7.45	4.39^{b}	3.02	7.95^{c}	5.10	143.18	4.93
G-L (SC-0 g kg^{-1})	7.58	4.15^{d}	3.31	10.24^{b}	7.05	143.56	5.13
G-L (SC-1.5 g kg^{-1})	8.38	4.85^{a}	3.78	8.44^{c}	5.23	143.68	4.69
L (SC-0 g kg^{-1})	7.76	4.30^{bc}	4.03	12.30^{a}	7.59	144.06	4.99
L (SC-1.5 g kg^{-1})	9.01	4.86^{a}	4.68	10.34^{b}	6.04	143.94	4.90
SEM	0.21	0.05	0.15	0.17	0.20	0.60	0.23

G: Grass, G-L: Grass and Legume, L: Legume, G (SC-0 g kg^{-1}): Grass with S. cerevisiae supplementation at 0 g kg feed, G (SC-1.5 g kg^{-1}): Grass with SC supplementation at 1.5 g kg^{-1} feed, G-L (SC-0 g kg^{-1}): Grass and Legume with SC supplementation at 0 g kg^{-1} feed, G-L (SC-1.5 g kg^{-1}): Grass and Legume with SC supplementation at 1.5 g kg^{-1} feed; L (SC-0 g kg^{-1}): Legume with SC supplementation at 0 g kg^{-1} feed, L (SC-1.5 g kg^{-1}): Legume with SC supplementation at 1.5 g k^{-1} g feed, TPP: Total plasma proteins (g dL^{-1}); Albumin (g dL^{-1}); Globulin (g dL^{-1}), Ca: Calcium, P: Phosphorous, Na: Sodium, K: Potassium, SEM: Standard error of the means, a,b,c,d: Means on the same column with different superscripts are significantly ($p < 0.05$) different

and legume or legume diets. The total plasma proteins (TPP), albumin, globulin, Ca and P values were significantly ($p<0.05$) affected by diet types.

The TPP and albumin values of sheep fed the legume diet and those of sheep fed a mixture of legume and grass diet were similar ($p>0.05$) but were significantly ($p<0.05$) higher than the TPP and albumin values of sheep fed the grass diet. The globulin (4.36 g dL^{-1}) and Ca (11.32 mg dL^{-1}) values of sheep fed the legume diet were the highest ($p<0.05$), followed by those of sheep fed the diet containing a mixture of legume and grass, while the globulin (2.66 g dL^{-1}) and Ca (8.17 mg dL^{-1}) values of sheep fed the grass diet were the lowest ($p<0.05$).

The phosphorous value (5.76 mg dL^{-1}) of sheep fed the grass diet and that (6.14 mg dL^{-1}) of sheep fed the mixture of legume and grass diet were similar ($p>0.05$) but were significantly ($p<0.05$) lower than the phosphorous value (6.81 mg dL^{-1}) of sheep fed the legume diet. There were no significant effects ($p>0.05$) of CL on Na and K levels of sheep fed diets with or without *S. cerevisiae* supplementation. However, there were significant effects ($p<0.05$) of CL on TPP, albumin, globulin, Ca and P levels of sheep fed diets with or without *S. cerevisiae* supplementation. All the sheep fed the diet with the *S. cerevisiae* additive had higher ($p<0.05$) TPP (8.28 g dL^{-1}), albumin (4.70 g dL^{-1}) and globulin (3.83 g dL^{-1}) and lower Ca (8.91 mg dL^{-1}) and P (8.91 mg dL^{-1}) values than those of their counterparts fed diets without the *S. cerevisiae* additive.

Significant effects ($p<0.05$) due to DT×CL on albumin and Ca values existed. However, there were no significant ($p<0.05$) DT×CL effects on TPP, globulin, P, Na and K values. The albumin values of sheep fed the legume diet with *S. cerevisiae* (4.86 g dL^{-1}) and the grass:legume mixture diet with *S. cerevisiae* (4.85 g dL^{-1}) were similar ($p>0.05$) but were higher than those of sheep fed other diets. The Ca value of sheep fed the legume diet without the *S. cerevisiae* additive was significantly ($p<0.05$) higher than the Ca values of sheep fed other diets.

DISCUSSION

Chemical composition of the experimental diets: As shown in Table 1 there were significant ($p<0.05$) differences among treatments in the DM, OM, CP, CF, EE, ash, nitrogen free extract (NFE), NDF and ADF percentages for the grass, a combination of grass and legume, or legume diets. The high contents of DM, CF, NDF and ADF in the grass based diet could be attributed to the high content of roughage in the diet. Protein content is often considered a good determinant of diet quality. The highest crude protein content tends to suggest that a legume based diet has the highest nutritional value.

Hematology: Present study showed (Table 2) that there were no significant ($p>0.05$) differences among diet types on the MCV, neutrophil, lymphocytes, monocytes and eosinophil values of sheep fed the grass, mixture of grass and legume,

or legume diets, while the PCV, RBC, MCHC, HbC, MCH and WBC count were significantly ($p<0.05$) affected. However, supplementation of some of the diets with *S. cerevisiae* had a significant ($p<0.05$) effect on the PCV, RBC, MCHC, HbC, MCV, MCH, WBC count and lymphocytes values, while neutrophil, monocytes and eosinophil counts were not significantly ($p>0.05$) affected. Significant ($p<0.05$) DT×CL interactions in the PCV, MCHC, HbC and WBC count values were also observed. *Saccharomyces cerevisiae* supplementation have been shown to have significant ($p<0.05$) effects on the hematological parameters such as HbC, PCV and RBC's counts in weaned Najdi ram lambs[25]. It does seem that the supplementation of some of the diets with *Saccharomyces cerevisiae* enhanced iron and salt absorption from the small intestine. Kander[15] had also shown that dietary *Saccharomyces cerevisiae* supplementation had the ability to produce vitamins B, which could positively affect blood-cell forming processes. Milewski[26] reported that feeding lambs with diets containing *Saccharomyces cerevisiae* had a significant ($p<0.05$) effect on the blood's WBC count and contributed to higher lymphocyte percentages in the leukogram. Increased WBC counts might be related to the production of more immune cells (and thus antibodies) that play an important role in defending the biological system against different diseases[20]. Dietary supplementation with yeast caused an increase in the counts of erythrocytes and leukocytes and in the levels of hemoglobin and hematocrit in ewes which resulted in a significantly lower mean corpuscular hemoglobin concentration (MCHC)[27]. Dietary supplementation with yeast significantly increased the values of the Mean Corpuscular Volume (MCV) and the Mean Corpuscular Hemoglobin (MCH)[27]. The observed changes in the blood hematological indices of ewes suggest an improvement in their body condition. According to Milewski *et al.*[27], the immunostimulatory effect of *Saccharomyces cerevisiae* can be ascribed to the activity of β-1,3/1,6-D-glucans and mannan-oligosaccharides present in the yeast cell walls. This mechanism involves the stimulation of immunocompetent cells, mainly by β-1, 3/1,6-D-glucans. In contrast to the result obtained in the present study Ghazanfer *et al.*[28] reported that lymphocytes were not significantly ($p>0.05$) affected by *Saccharomyces cerevisiae* supplementation while eosinophils were significantly ($p<0.05$) increased.

Biochemistry: Present study showed (Table 3) that there were significant ($p<0.05$) differences among diet type on the TPP, albumin, globulin, Ca and P values of sheep fed the grass, mixture of grass and legume, or legume diets, while Na and K values were not significantly ($p>0.05$) affected. There were no significant ($p>0.05$) differences due to the CL in sheep fed diets with or without *Saccharomyces cerevisiae* in their Na and K values, while the TPP, albumin, globulin, Ca and P values were significantly ($p<0.05$) affected.

The results of the present study are in agreement with those reported by Abdel Rahman *et al.*[29] who showed that the concentration of albumin was significantly increased by *S. cerevisiae* supplementation in the diets of growing lambs. In contrast, Galip[30] had shown that serum albumin, Na and K levels were not significantly ($p>0.05$) affected in the serum of rams that received a dietary supplemental yeast. A similar report by Shehu *et al.*[31] had shown that *S. cerevisiae* supplementation caused no significant ($p>0.05$) increase in the serum levels of Na^+, K^+ and HCO_{-3} in rabbits. Therefore, dietary *Saccharomyces cerevisiae* may be able to enhance the activities of hormones, involved in the maintenance of normal mineral balance.

However, the biochemical results of the present study differ with those reported by Abdel Rahman *et al.*[29] which indicated that blood total protein or globulins levels were not significantly ($p>0.05$) affected by *S. cerevisiae* supplementation. El-Ashry *et al.*[32] who researched Barki lambs reported that yeast supplementation significantly ($p<0.05$) increased plasma globulin values of the animals. They were of the opinion that such increase could have helped to confer immunity to the animals. Regarding total serum protein, Abu El-Ella and Kommonna[33] reported that Damascus goat fed a diet supplemented with 2.5 g *S. cerevisiae*/head/day had the highest ($p<0.05$) value of total serum protein followed by those supplemented with a high level of *S. cerevisiae* (5.0 g *S. cerevisiae*/head/day) while the control group had the least amount of protein. Galip[30] had also shown that total protein was increased in the serum of rams that received dietary supplemental yeast ($p<0.01$) in comparison to control animals. The observed enhancement in total serum protein may be attributed to the beneficial effect of *S. cerevisiae* supplementation on increasing protein digestibility through the enzymatic effect of protease and through an alteration of the amino acid profile of the digested food due to an increase in microbial protein synthesis[34]. Yeast cultures have been found to stimulate microbial activity and increase the incorporation of nitrogen into microbial protein, which confirmed the suggestion of Erasmus *et al.*[35] that yeast cultures may exert an effect on the flow of protein, which is also related to the changes in the number and activity of rumen microorganisms. The efficiency of feed nitrogen utilization in ruminants supplied with yeast culture involved

not only the increase of ammonia incorporation into microbial protein and a higher flow and absorption of amino acids but also an altered endogenous nitrogen metabolism.

The present results on serum calcium concentration are in agreement with those of Galip[30] who showed that phosphorus concentrations tended to diminish significantly when *S. cerevisiae* was added to diets and also, Ca^{2+} concentrations and calcium/creatinine ratio were significantly lowered ($p<0.01$) in assay groups. In addition, Onifade *et al.*[36] reported significant decreases of Ca^{2+} and phosphorus concentrations in rabbits supplemented with *S. cerevisiae* and suggested that these variations would be related to the enhancement of bone mineralization.

CONCLUSION

Significant ($p<0.05$) improvement in the PCV, HbC and WBC counts, as well as serum albumin content in sheep fed a legume diet supplemented with *S. cerevisiae* suggest that the addition of 1.5g of *S. cerevisiae* per kg of legume diet is feasible.

SIGNIFICANCE STATEMENT

The study discovers that the dietary supplementation of *S. cerevisiae* can be beneficial to West African dwarf sheep. This study will enable researchers to further investigate the effects of other levels of *S. cerevisiae* which had been previously unexplored. Thus, a new theory on the optimum level of *S. cerevisiae* supplementation may elucidated.

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