Growth Performance And Blood Profile Of Clarias Gariepinus Fed Processed Soybean Based Diets Supplemented With Amino Acids

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Abstract: This study was aimed at assessing the effect of supplemental lysine and DL-methionine in a solvent extracted soybean based diets on performance and blood profile of Clarias gariepinus. Six diets containing varied combinations of lysine+DL-methionine (g/100g) in solvent extracted soybean based diets (SESBD) were formulated; S_1 (without lysine and DL-methionine), S_2 (0+1), S_3 (0.25+0.75), S_4 (0.5+0.5), S_5 (0.75+0.25), S_6 (1+0) lysine and DL-methionine, respectively. Clarias gariepinus (n=360) weighing 17.77±0.27g were fed to satiation with the diets for 12 weeks. Each treatment was in triplicate. Significantly (P<0.05) higher final weight value were observed in control (43.35±4.03) but this was not significant from those fed diets S3 and S5, however, these were superior to other diets. Supplementation of lysine and DL-methionine had no significant effect (P>0.05) on Feed Conversion Ratio (FCR). White blood cell (x 10^3/µL) of Clarias gariepinus on S4 (17.48±0.60) was significantly higher (P<0.05) than those on S3 (13.78±1.26). A significantly higher (P<0.05) globulin (g/L) were observed in S5 (5.03±0.21) and least in S3 (4.70±0.20). Optimal weight gain occurred with SESBD of 0.6+0.4g/100g dietary inclusion (R^2= 0.61, 0.97) of lysine and DL-methionine, respectively. Lysine and DL-methionine supplementation in solvent extracted soybean based diets improved growth performance and had no negative effect on blood profile of Clarias gariepinus.

Keywords: lysine, DL-methionine, solvent extracted soybean, performance, blood profile

1. Introduction

Soybean meal is the most widely used plant protein source in animal feeds globally especially in Nigeria. However, the presence of anti-nutritional factors (trypsin) make soybean not easily digested and utilised by fish when compared with animal protein sources [1]. Researches are ongoing on methods to improve digestion, nutritional quality and enhance performance of fish species by ascertaining the appropriate processing methods. These include solvent extraction [2], roasting [3], and micronisation [4]. Solvent extraction of soybean has been shown to inactivate anti-nutritional factors, improves protein and energy digestibility of plant protein sources [5]. However, digestion of solvent extracted soybean remains low compared to fishmeal [5], [6]. Processed soybean was considered to be one of the best available soybean meal sources in terms of its protein quality and amino acid profile among plant protein feedstuff used as alternative protein sources in fish diets [7], [8]. Despite the noted favorable amino acid profile [9] of solvent extracted soybean meal, it is deficient in DL-methionine, lysine and have low cysteine [10], [11]. However, growth depressions caused by soybean inclusions in Clarias gariepinus diets have been attributed to amino acid deficiencies, especially lysine and/or DL-methionine [7]. The limiting amino acids in soybean cake limits their inclusion in fish diet to about 45% [12], [13], despite its availability. Nwanna et al. [14] ascertained that lysine and DL-methionine are generally the first limiting amino acids in fish feeds that needed attention. Therefore, dietary supplementation of amino acids in soybean based diet provides new strategies to develop amino acid balanced feeds that can offset environmental impacts on aquaculture animals. Also, this is to restore the amino acid profile to a level which matches the requirement of the target species, improve growth performance, and profitability of the aquaculture industry [15]. Research have proven that supplementation of lysine and DL-methionine in the diet of Clarias gariepinus improved the feed quality, growth performance, digestibility and profitability of the aquaculture industry [15], [16]. Fagbenro et al. [16] observed that inclusion of DL-methionine and lysine could enhance weight gain of Clarias gariepinus. Ochang et al. [17] reported that amino acid requirement of fish diet could be improved in the plant protein based diet when it is supplemented with dietary lysine and DL-methionine. Ajani et al. [18] affirmed that total replacement of fish meal with soybean meal is achievable when DL-methionine is supplemented in Oreochromis niloticus diet. However, there is scanty information on the performance of Clarias gariepinus fed plant protein based diet, most especially, solvent extracted soybean based diet. There is need to evaluate the effect of supplemental limiting dietary amino acid (lysine and DL-methionine) in solvent extracted soybean based diet for Clarias gariepinus. Therefore, the present study was aimed at assessing the growth performance and blood profile of Clarias gariepinus fed solvent extracted soybean based diet supplemented with graded inclusion levels of DL-methionine and lysine.

2. Materials and Methods

The feeding trial was conducted at the Aquaculture Research Laboratory, Aquatech College of Aquaculture, Fodacis, Ibadan, Nigeria between February and April, 2017. Clarias gariepinus juveniles (n=360) aged eight weeks, weighing 17.77±0.27grams were purchased from a reputable fish farm in Ibadan, Nigeria. Six isonitrogenous diets were formulated with varying levels of lysine and DL-methionine (Table 1). The dietary protein level was fixed at 40% crude protein (Table 2), reported optimum [19] for the

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growth of Clarias gariepinus and mainly based on soybean, yellow maize, vitamin/mineral premix, soya oil and salt. The ingredients were thoroughly mixed together and each diet mixture was pelleted at 60 ^{o}C, using 2mm pellet die to form noodle-like strands, which were manually crumbled into a suitable size for the Clarias gariepinus juveniles. The pellets were sundried, packed into transparent bags and stored in a cool dry place to prevent fungal growth. The six dietary treatments were as follows: S1 (Control) = no supplemental lysine and DL-methionine; S2 = 0g lysine+ 10g DL-methionine; S3 = 2.5g lysine + 7.5g DL-methionine; S4 = 5.0g lysine +5.0g DL-methionine; S5 = 7.5g lysine + 2.5g DL-methionine; S6= 10g lysine + 0g DL-methionine

2.1. Fish feeding trial

The experiment was conducted at the Research Laboratory with eighteen plastic tanks of dimension measuring 60 cm × 45 cm × 30 cm for 12 weeks (February-April, 2017). Every three days, each tank were supplied with well water up to 80% capacity. The water were replaced to prevent fouling from feed residues and maintain relatively uniform physico-chemical parameters. The dietary treatments were six and each were on triplicates with 20 fish each. The fish were weighed and randomly allocated to experimental tanks. They were thereafter acclimatize for 14 days with experimental diets. Weight changes were documented weekly and feeding rate properly regulated to the new body weight. The fish were fed to satiation and the experiment lasted for 84 days.

Table 1: Gross composition of solvent extracted soybean based diets.

Ingredient (g/100g)	Control	S2	S3	S4	S5	S6
Soybean meal	70	70	70	70	70	70
Yellow maize	26	26	26	26	26	26
*Vit/min premix	0.25	0.25	0.25	0.25	0.25	0.25
Soybean oil	1	1	1	1	1	1
$CaCO_3$	0.5	0.5	0.5	0.5	0.5	0.5
Salt	0.5	0.5	0.5	0.5	0.5	0.5
Chromic Oxide	0.5	0.5	0.5	0.5	0.5	0.5
Lysine	0	0	0.25	0.5	0.75	1
DL-methionine	0	1	0.75	0.5	0.25	0
Total (g/100g)	99	100	100	100	100	100

**1kg of premix contains Vitamin A-22,000I.U; Vitamin D3-5,000I.U, Vitamin E-300mg; Vitamin k3-10mg; Vitamin B1-20mg; Vitamin B2-25mg; Vitamin C-300mg; Niacin-120mg; Calcium Pantothenate-60mg; Vitamin B6-10mg; Vitamin B12-0.05mg; Folic Acid-5mg; Biotin-1mg; Choline Chloride-500mg; Inositol-50mg; Manganese-30mg; Iron-35mg; Zinc-45mg; Copper-3mg; Iodine-5mg; Cobalt-2mg; Lysine-85mg; selenium-0.15mg; Anti-Oxidant-80mg; DL-methionine-100mg*

Table 2: Chemical composition of test diets (%DM) fed to fish

Parameter (%)	Control	S2	S3	S4	S5	S6
Dry matter	92.29	92.25	92.09	92.31	92.09	92.11
Crude protein	41.59	40.98	41.08	40.75	40.83	40.78
Ash	9.05	7.95	9.30	8.15	7.20	7.80
Ether extract	6.75	6.35	6.55	6.55	7.05	6.70
Crude fibre	5.85	5.15	6.05	5.25	6.70	5.00
Gross energy (kcal/g^{-1})	3.99	4.06	3.98	4.11	4.02	3.98

S2- 1gram (g) Methionine; S3- 0.75g Methionine, 0.25g Lysine; S4- 0.5g Methionine, 0.5g Lysine; S5- 0.25 Methionine, 0.75 Lysine; S6 - 1g Lysine.

2.2. Proximate composition

Proximate composition of the diets was determined according to AOAC [20].

2.3. Growth studies

Calculations on growth performance and feed utilization were according to Falayi [21]. Data on weight gain (WG), feed conversion ratio (FCR), protein intake (PI), feed intake (FI), protein efficiency ratio (PER), specific growth rate (SGR), gross protein retention (GPR), nitrogen retention efficiency (NRE) and survival rate (SR) were taken bi-weekly throughout the duration of the study.

2.4. Blood sampling and analysis

Blood (5 mL) was sampled from three randomly selected C. gariepinus per replicate group into bottles containing ethylene dia-amine tetra acetic acid (EDTA) in treated heparinized plastic bottles for haematological assay. The blood samples were analysed for packed cell volume (PCV), haemoglobin concentration, red blood cell (RBC), white blood cell (WBC) as outlined by Blaxhall and Daisley [22].

Mean Corpuscular Volume (MCV)

MCV was estimated using the model described by Feldman et al. [23]:

$$\text{MCV} = \frac{\text{volume of red blood cell (in mL per 100mL blood)}}{\text{Number of red blood cells oer 100mL blood}} \times 100$$

Mean Corpuscular Haemoglobin (MCH)

MCH was estimated using the model described by Stoskopf [24]

$$\text{MCH} = \frac{\text{Haemoglobin } (\frac{\text{g}}{\text{100ml}})}{\text{Number of red blood cells } (\frac{\text{millions}}{\text{L}}\text{blood})} \times 100$$

Mean Corpuscular Haemoglobin Concentration (MCHC)

MCHC was estimated using the model as described by Stoskopf [24]

$$\text{MCHC} = \frac{\text{Haemoglobin concentration}}{\text{Packed cell volume}} \times 100$$

2.5. Serum Biochemical Analysis

Blood (2 mL) was sampled from three randomly selected C. gariepinus per replicate group into heparinized flask without anticoagulant for serum biochemical analysis. The blood was allowed to clot and samples was then centrifuged at 3000rpm for five minutes with Hawsley minor bench

centrifuge (P spectra, Centromix no 231254 CD7000549, Spain). The blood serum was harvested and stored at -20 ^{o}C. Total protein and albumin were determined by Biuret method according to Reinhold [25], globulin was estimated as the difference between total protein and albumin. Albumin:globulin ratio, serum enzymes: aspartate amino transaminase, alanine amino transaminase and alkaline phosphatase activities were determined spectrophotometrically according to Henry et al. [26]. Blood urea nitrogen and creatinine were determined by the techniques of Harrison [27].

Statistical Analysis

Data were analyzed using descriptive statistics, polynomial regression and analysis of variance [28]. Means were separated using Duncan multiple range test option of the same software at $\alpha_{0.05}$.

3. Results

Supplemental lysine and DL-methionine in solvent extracted soyabean based diets had no significant influence (P>0.05) on weight gain of C. gariepinus on diets S1, S3 and S5 and these were significantly higher than other values (Table 3). No significant difference (P>0.05) was observed in FCR of fish fed diets supplemented with lysine and DL-methionine. The obtained value ranged from 2.35±0.15 (control) to 3.12±0.31 (S2). Protein efficiency ratio was significantly (P<0.05) higher in fish on control diets but similar (P>0.05) to fish on diet S3 and S5. However, fish on diets without lysine and DL-methionine (control) had a significantly higher specific growth rate value but similar to other diets except fish on diet S2 with the least value. Lysine and DL-methionine supplementation significantly (P<0.05) improved gross protein retention in fish on diet S5. Nitrogen retention efficiency was significantly higher (P<0.05) in fish on control diet (49.49±6.19) but similar (P>0.05) to those on S3 (47.64±2.10). Fish on S5 had higher survival rate. The relationship between dietary supplement of lysine and DL-methionine in solvent extracted soyabean based diets and weight gain is shown by equations 1 and 2 From the graphs in Figure 1, it was deduced that 0.6 and 0.4 g/100g levels of supplemental lysine and DL-methionine were respectively, required for optimal weight gain of fish fed solvent extracted soybean based diets. Amino acid supplementation in solvent extracted soybean based diet had no influence (P<0.05) on packed cell volume (%), hemoglobin (g/dL) and red blood cell (×10^{12}/L) (Table 4). White blood cell (x109/L) was significantly higher (P<0.05) in SS4 (17.48±0.60) and least value in S3 (13.78±1.26) while fish fed diet S2, S5, S6 and control were intermediate to them. MCV (fl) and MCH (pg) of fish fed diet SS4 and SS5 were not significantly different from each other, but these were all significantly lower than other diets. MCHC (g/dL) values were not significantly (P<0.05) different among the treatment. Platelet (x10^{9}/L) significantly ranged from S2 (10.83±0.97) to S4 (17.53±1.29), however, fish fed diet S5 and S6 were similar to S4 diet. Lymphocytes (%) value significantly decreased with amino acid supplementation in C. gariepinus diet. Moreover, heterocytes (%) value significantly (P<0.05) increased with amino acid supplementation in fish fed diet S4 (30.67±4.04) but differ significantly from fish fed diet S2, S3, S5 and S6. Supplementation of amino acid influenced (P<0.05) lymphocytes: heterocytes ratio with higher value of 0.50±0.10 in S4 and control diet (0.34±0.08) had the least value while, fish fed diet S2, S3, S5 and S6 were similar to each other. Supplementation had no significant influence on Monocytes (%), Eosinophils (%) and Basophiles Dietary amino acid supplementation had no significant effect (P<0.05) on total protein (g/L), Albumin (g/L), A-G ratio, aspartate amino transferase (IU/L), alanine amino transferase (IU/L) and alkaline phosphatase (IU/L) (Table 5). Significantly higher (P<0.05) globulin (g/L) was observed in C. gariepinus fed diet S5 and least in those on S3 (4.70±0.20). Increased blood urea nitrogen (µmol/L) was observed in fish on S5 (10.00±0.20) compared with those on control diets. Also, significantly higher (P<0.05) creatinine (µmol/L) was observed in fish on S3 (0.76±0.06) while those on control diet (0.57±0.06) was least.

Table 3: Growth performance and feed utilisation of C. gariepinus fed solvent extracted soybean based diets supplemented with lysine and DL-methionine

		Solvent Extracted (S)				
Parameter	Control	2	3	4	5	6
IW(g)	17.85±0.49	18.03±0.38	17.50±0.10	17.77±0.32	17.50±0.00	17.80±0.17
WG (g)	43.35±4.03^{b}	34.77±1.17^{a}	40.80±1.74^{ab}	36.13±1.27^{a}	36.70±5.11^{ab}	35.40±4.99^{a}
FCR	2.35±0.15	3.12±0.31	2.27±0.32	2.88±0.26	2.95±0.60	2.88±0.62
GEFC	72.96±1.52	67.25±4.32	77.97±8.93	68.66±4.85	66.97±7.03	72.53±7.51
PI	13.73±1.56^{b}	11.95±0.67^{ab}	12.15±1.05^{ab}	12.17±0.47^{ab}	12.65±1.14^{ab}	11.24±0.72^{a}
FI	0.92±0.10^{b}	0.80±0.04^{ab}	0.81±0.07^{ab}	0.81±0.03^{ab}	0.84±0.08^{ab}	0.75±0.05^{a}
PER	14.45±1.34^{b}	11.59±0.39^{a}	13.60±0.58^{ab}	12.04±0.42^{a}	12.23±1.70^{ab}	11.80±1.66^{a}
SGR	0.59±0.08^{c}	0.44±0.02^{a}	0.57±0.03^{bc}	0.47±0.01^{abc}	0.49±0.09^{abc}	0.46±0.09^{ab}

GPR	0.72±0.02^{b}	0.66±0.02^{a}	0.75±0.04^{bc}	0.77±0.03^{cd}	0.79±0.02^{d}	0.78±0.03^{cd}
NRE	49.49±6.19^{b}	35.15±1.42^{a}	47.64±2.10^{b}	41.27±1.66^{ab}	43.17±7.76^{ab}	40.74±7.38^{ab}
SR %	87.70±0.20^{d}	87.70±0.20^{d}	82.20±0.20^{b}	85.60±0.20^{c}	88.90±0.10^{e}	77.80±0.20^{a}

Means with different superscripts on the same row are significantly different (P<0.05)
IW = Initial Weight, WG= Final Weight, FCR= Feed Conversion Ratio, GEFC= Gross Efficiency Feed Conversion, PI= Protein Intake, FI= Feed Intake, PER= Protein Efficiency Ratio, SGR= Specific Growth Rate, GPR= Gross Protein Retention, NRE= Nitrogen Retention Efficiency, SR= Survival Rate.

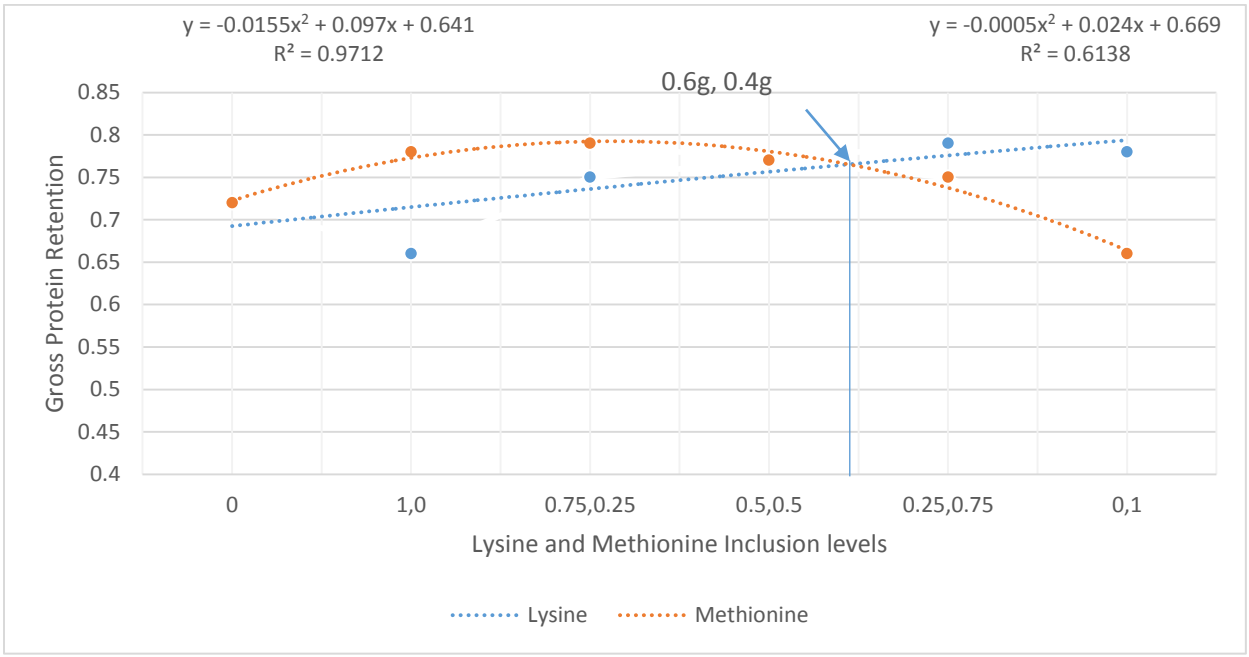

Figure. 1. *Relationship between dietary supplemental of lysine and DL- methionine in a solvent extracted soybean based diet and gross protein retention of Clarias gariepinus.*

Table 4: *Haematology of C. gariepinus fed solvent extracted soybean based diets supplemented with lysine and DL-methionine*

`		Solvent Extracted (S)				
Parameter	Control	2	3	4	5	6
PCV (%)	22.67±3.79	25.67±0.58	23.33±0.58	26.33±1.53	25.67±1.53	24.67±1.53
HB (g/dL)	7.57±1.44	8.37±0.12	7.90±0.61	8.63±0.71	8.77±0.67	8.30±0.78
RBC(x10^{12}/L)	2.01±1.05	2.17±0.15	1.5±0.04	2.74±0.65	2.58±0.92	2.08±0.41
WBC(x10^{9}/L)	14.82±2.47^{ab}	16.27±0.38^{ab}	13.78±1.26^{a}	17.48±0.60^{b}	15.78±2.93^{ab}	14.6±1.98^{ab}
MCV (fl)	125.59±36.44^{ab}	118.39±6.32^{ab}	154.32±7.88^{b}	98.64±16.18^{a}	106.68±31.36^{a}	120.79±18.40^{ab}
MCH (pg)	41.68±11.55^{ab}	38.60±2.19^{ab}	52.29±5.21^{b}	32.24±4.77^{a}	36.33±10.40^{a}	40.46±4.75^{ab}
MCHC(g/dL)	33.30±0.79	32.60±0.64	33.84±2.27	32.75±0.79	34.15±1.06	33.61±1.52
Platelet (x 10^{9}/L)	12.53±2.64^{ab}	10.83±0.97^{a}	14.40±0.46^{bc}	17.53±1.29^{d}	15.63±0.91^{cd}	16.90±1.45^{cd}
Lym (%)	69.33±5.13^{b}	65.67±4.61^{ab}	63.33±1.52^{ab}	61.67±4.51^{a}	67.00±1.73^{ab}	66.67±4.04^{ab}
Het (%)	23.33±3.51^{a}	27.00±5.29^{ab}	30.00±2.00^{ab}	30.67±4.04^{b}	26.33±3.21^{ab}	27.67±3.06^{ab}
Lym:Het (%)	0.34±0.08^{a}	0.41±0.10^{ab}	0.48±0.05^{ab}	0.50±0.10^{b}	0.39±0.06^{ab}	0.42±0.07^{ab}

Means with different superscripts on the same row are significantly different (P<0.05)
PCV= Packed Cell Volume, HB= Hemoglobin, RBC= Red Blood Cell, WBC= White Blood Cell, MCV= Mean Cell Volume, MCH= Mean Cell Hemoglobin, MCHC= Mean Cell Hemoglobin Concentration, Lym= Lymphocytes, Het= Heterocytes, Mono= Monocytes, Eos = Eosinophils, Baso = Basophils.

Table 5: *Serum biochemical indices of C. gariepinus fed solvent extracted soybean based diets supplemented with lysine and DL-methionine*

		Solvent Extracted (S)				
Parameter	Control	2	3	4	5	6
Total protein(g/L)	6.50±0.50	6.83±0.76	6.80±0.20	7.00±1.32	7.70±0.17	7.43±0.40
Albumin(g/L)	1.63±0.49	2.00±0.87	2.10±0.40	1.97±1.24	2.53±0.21	2.33±0.21
Globulin(g/L)	4.86±0.21^{ab}	4.83±0.21^{ab}	4.70±0.20^{a}	5.03±0.21^{ab}	5.17±0.06^{b}	5.10±0.20^{b}
A-G ratio	0.27±0.27	0.40±0.17	0.43±0.15	0.43±0.23	0.47±0.06	0.40±0.00
AST (IU/L)	188.67±2.52	206.00±37.04	184.00±4.00	187.00±7.21	190.00±3.00	181.33±3.21
ALT (IU/L)	29.00±7.55	23.00±2.00	31.00±1.73	24.00±7.94	31.00±3.61	23.67±7.77
ALP (IU/L)	309.00±43.51	266.66±3.51	336.00±13.75	270.33±11.02	326.67±76.14	336.67±48.81
BUN(µmol/L)	8.60±0.40^{a}	8.67±0.83^{a}	8.90±0.56^{ab}	8.87±0.90^{ab}	10.00±0.20^{b}	9.37±0.85^{ab}
Creatinine(µmol/L)	0.57±0.06^{a}	0.73±0.06^{bc}	0.76±0.06^{c}	0.73±0.06^{bc}	0.77±0.06^{c}	0.63±0.06^{ab}

Means with different superscripts on the same row are significantly different (P<0.05)
A-G Ratio- Albumin-Globulin Ratio, AST = Aspartate Transaminase, ALT = Alanine Transaminase, ALP = Alkaline Phosphatase, BUN = Blood Urea Nitrogen.

4. Discussion

Findings from Table 3 indicated that C. gariepinus fed soybean based diet when supplemented with lysine and DL-methionine was a better diet for the fish growth performance. Lu et al. [29] attributed it to the fact that, amino acid has a powerful feeding stimulant and it has been good in reducing feed intake while improving the body weights of fish. There have been similar reports of improved performance of fish fed diets supplemented with lysine and DL-methionine [30], [31]. The authors suggested that supplemental amino acid improved amino acids utilization for protein synthesis which shows that lysine and DL-methionine are essential for the growth of C. gariepinus. Also, Nwanna et al. [14] reported that supplementation of DL-methionine in DL-methionine deficient diet significantly increased feed utilization with better feed conversion ratio which resulted in significant improvements in weight gain and feed efficiency of common carp. Similar trends were noted by Gao et al. [32]; Ren et al. [33] and Sardar et al. [34] in Juvenile grass carp (Cyprinus carpio), juvenile blunt snout bream (Megalobrama amblycephala) and Rohu (Labeorohita) fingerlings when fed increasing levels of dietary histidine, isoleusine and Met+Cystine, respectively. Also, the supplementation of lysine and DL-methionine improved feed efficiency and decreased the amount of feed consumption necessary for fish growth which could result in production cost reduction. These observations conformed to those of Yuan et al. [35] Nwanna et al. [14] and Wang et al. [36] that reported lower FCR due to lysine and DL-methionine supplementation for Myxocyprinus asiaticus, Cyprinus carpio and Pseudobagrus ussuriensis, respectively. Zhou et al. [37] observed reduced growth, feed utilization and PER when lysine was supplemented to diets of Sparus macrocephalus. It may be due to the negative effects of excessive or insufficient amount of free lysine and DL-methionine and poor palatability. Wang et al. [36] suggested that DL-methionine levels used in the diet were not high enough to induce toxic effect. However, the relationship between solvent extracted soybean based diet and WG of C. gariepinus fed dietary amino acid was shown by the regression model in Figure 1. From the growth response curve, the estimated optimum level of 0.6g/100g lysine and 0.4g/100g DL-methionine for weight gain.The low WBC recorded could be attributed to the fish exposure to different pollutants and could also reduce the amount of circulating lymphocytes and thrombocytes [38]. Reduction in MCV observed in this study was in agreement with the findings of Alwan et al. [39]; Adesina [40]; Adeyemo [41] who ascribed reduction in MCV to the shrinking of RBCs which could be due either to hypoxia or balanced water condition or microcytic anaemia. The decreased values observed was contrary to the report of Anyanwu et al. [42] who attributed it to the swelling of erythrocytes which could be macrocytic. The reduction observed in MCH could be ascribed to reduced iron deficiency and microcytic anemia, which is a condition where RBC is abnormally small and carrying less haemoglobin. The result of this finding was in agreement with Anyanwu et al. [42] who fed diets incorporated with Carica papaya leaf meal to Heteroclarias. Significantly lower lymphocytes were observed in C. gariepinus fed solvent extracted soybean based diet. Lymphocytes are the defense cells of the body and help to determine the immune response and ability of fish to fight infection. Though, the values obtained were within the reported recommended ranges [22], for fish, lysine and DL-methionine inclusion in the diet of C. gariepinus had significant effect on the immune system. Increased globulin level is an indicator of proper body defense mechanism [43]. The increased level of globulin obtained in fish on S5 was an indication of improved immune response which could be attributed to the presence of adequate supplementation of dietary lysine and DL-methionine in the diet as earlier reported by Abdo [44] and Ogunwole et al. [45]. Elevated levels of BUN and creatinine indicated that,

there was no renal damage or muscle wastage that could be attributed to dietary inclusion of lysine and DL-methionine in soybean based diets. This observation was corroborated by the report of Azza and Naela [46]. Therefore, the serum biochemical parameters obtained from this study suggested that dietary lysine and DL-methionine supplementation had no adverse effects on the physiological indices of C. gariepinus since the serum protein values were also within the normal range for fish.

5. Conclusion

Supplementation of lysine and DL-methionine at 0.6g/100g and 0.4g/100g respectively in solvent extracted soybean based diet fed to C. gariepinus improved growth and nutrient utilization. Also, C. gariepinus fed soybean based diet with supplemental lysine and DL-methionine had no adverse effect on fish health status.

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