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Effect of Low Protein Diets with Amino Acids Supplementation on Growth Performance, Carcass Traits, Blood Parameters and Muscle Amino Acids Profile in Broiler Chickens under High Ambient Temperature

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Abstract: This study evaluates low protein diets with amino acid supplement on growth, biochemical markers and muscle amino acids profile in broilers under high ambient temperature. A total of 480 one-day-old chicks were allocated into three treatments with four replicates (n = 40). Control fed optimal protein and optimal amino acids which contains 23% and 21% crude protein (CP) with 65% methionine + cysteine/ lysine (Met + Cys/Lys) and 55% threonine/lysine (Thr/Lys), LPOA (low protein and optimal amino acids) which contains 21% and 19% CP with 65% Met + Cys/Lys and 55% Thr/Lys and 3. LPHA (low protein and high amino acids) which contains 21 and 19% CP with 74% Met + Cys/Lys and 67% Thr/Lys, respectively during the summer months (The temperatures 32 ± 3 °C, and the relative humidity 72.5% ± 4). Birds fed LPOA diets significantly highest body weight, while those fed LPHA recorded significantly the lowest body weight (BW). Dressing percentages not revealed significantly affected by reducing dietary protein levels, while the blood plasma total protein, albumin, and globulin were not significant differences due to dietary low protein. Chicks fed LPHA diets recorded the highest liver content of malonaldehyde. It could be concluded that feeding the Cobb 500 broilers on low protein diets with the same amino acid levels had no adverse effect on growth, carcass markers, and liver function, however increased amino acids levels to low protein diets may led negative impacts for the broiler performance under high ambient temperature.

Keywords: broilers; amino acids; growth performance; carcass traits; blood parameters

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1. Introduction

The most challenges facing the broiler production in Middle East countries is the raised ambient temperature especially in open systems farms. In fact, the optimum performance temperature for growing broilers is ranged from 18 to 24 °C [1]. When thermos requirements of birds are not met (average high temperature of 30 °C), thermal anxiety can occur. Heat stress trigger considerable reduces in the broilers production owing to adversarial behavioral [2], physiological, and immunological impacts [3–5]. In addition, the major problem of production the in progressive poultry enterprise is to decrease feed cost that is represents about 70% of the total cost production. On the economic prospects, the lowering of crude dietary protein with supplementation of amino acids in broiler diets were the effective strategies for reducing the feed cost. Moreover, on the environmental prospects, feeding low protein diets are an effective strategy for reducing the contamination by reduces the nitrogen and ammonia exit from the farms. Therefore, focusing mainly on crude protein (CP) while formulating the broiler diets, is the first step to solving the problem, because raw protein is important and cost-effective elements of the poultry diet. Therefore, it is proposed to lessen the level of dietary protein and use synthetic amino acids to decrease the cost of feed. On the other hand, and as we mentioned previously additionally to include environmental pollution of nitrogen [6,7]. The feeding cost for broiler chicks, especially dietary protein sources is the most expensive component. Therefore, more studies have been made to reduce feeding costs to the minimum level by reducing the level of CP or reducing CP with supplementation of purified amino acids [8]. However, Awad et al. reported that the decreasing dietary CP is suggested when birds are subjected to moderate heat anxiety circumstances [9]. Neto et al. found that feeding 17 vs. 24% CP in broiler diet for 21 days showed a significant decreased in body weight gain as well as increased both feed consumption and abdominal fat bad weight [10]. The birds have not maintained any of the dietary nitrogen intake but was excreted into the environment. Reducing the CP percentage of the diet might be the best method to lower the excretion of nitrogen and ammonia from broiler farms [11,12]. In addition, lowering the CP substance of broiler chickens' fed could be reduce the water intake due to the reduced need for excess nitrogen excretion [13]. In addition, the reduced consumption of less water may be also reduced the problems in the litter moisture, which in turn leads to the risk of impaired welfare because the presence of a wet litter is the major reason of dermatitis diseases for example foot-pad, and breast sores [14]. Furthermore, the decline of CP of the dietary broiler chickens can promote to decreasing the importing and usage of the soybean meal. Nevertheless, decreasing the CP component of broiler diets might create a danger of decreased growth efficiency, precisely when essential amino acids becomes restricting in the diets. It is crucial to understand that dietary amount of CP could be decreased if the minimum amino acid requirements were adequate to sustain broiler growth and muscle [15].

Furlan et al. reported that among dietary nutrients, protein has the greatest heat increase; hence, through many years, low-protein diets were advised to minimize heat output in broiler under heat stress. Nevertheless, studies have demonstrated that low protein diets can reduce performance during heat stress for broilers, because of insufficient protein intake during periods of high environmental temperature provoke amino acid inadequacy [16].

Nevertheless, threonine has become one of the main restricting elements for animal performance over the past ten years as the inclusion of crystalline lysine and methionine in full diets has been increased. Threonine amino acid is the third highly limiting amino acid, particularly in a diet with reduced CP [17]. It is well known, threonine is involved in protein creation, and its catabolism produces many metabolism-important products. Threonine is involved in immune responses as a part of body protein and a precursor of glycine and serine [15]. In mammals, threonine is a significant component of intestinal mucin and alfa-globulin plasma [18].

Maharjan et al. noted that broilers fed low protein diets with the optimal amino acid's levels may be good goal to save the broiler performance under heat stress [19]. Conversely, Zulkifli et al. documented that feeding broilers low CP diets negatively impacted growth output under heat impact state. On the opposite, low-CP diets did enhance longevity. As acute phase proteins participate in homeostasis stabilization, it may be an unfavorable impact of low-CP on the creation of these proteins [20].

Therefore, we performed this bioassay was presented to appraise the impacts of amino acid supplements, low protein diets on growth efficiency, carcass traits, and blood parameters in mixed-sex Cobb 500 broiler under hot weather condition.

2. Material and Methods

The current experiment operated by the department of poultry production, faculty of agriculture, Kafrelsheikh university from July to August period of the summer season. This research was accepted by the ethics committee of the Kafrelsheikh University, Egypt, and carried out following the guidelines of the Kafrelsheikh University (Number 4/2016EC).

2.1. Birds, Experimental Design and Husbandry

A total of 480 one-day-old mixed-sex Cobb broiler grown over a 35-days period. The chicks were wing-banded, weighed individually and arbitrarily divided into three equally major groups, each contained four replicates of 40 birds each. Chicks in the control group (optimal protein and optimal amino acids) were fed the starter and grower diets (23% and 21%), which contains 65% Met + Cys/Lys and 55% Thr/Lys (Table 1). The chicks of the LPOA group (low protein and optimal amino acids) were fed the control starter and grower diets low 2% CP (21 and 19%) with the same content of amino acids (65% Met + Cys/Lys and 55% Thr/Lys). The chicks of the LPHA group (low protein and high amino acids) were fed the control starter and grower diets low 2% CP (21% and 19%) with the higher content of amino acids (74% Met + Cys/Lys and 67% Thr/Lys). All diets were prepared to provide the recommended supplies for broilers following the Cobb broiler chick's requirements with a chain bird feeder and automated nipple cup drinker in a totally randomized layout fresh water and feed were offered ad libitum. Chicks were housed in open-sided housing within the same sanitary, environmental, and managerial conditions throughout the experimental period. We used the Newcastle disease and vaccine for avian influenza (Vol-vac® B.E.S.T. AI + ND, Boehringer Ingelheim Co., Ingelheim am Rhein (Rheinland-Pfalz, Germany) for vaccinating birds in the neck at 7 d S/C injection. At day 14, birds were vaccinated by eye-droppers (Nobilis® GUMBORO D78 (Intervet, The Netherlands) and (Nobilis® ND LaSota) (Intervet). The lighting system was kept at 23:1 h light/dark for the first 3 days, followed by 20:4 h light/dark till the end of the study. The average temperature was 32 ± 3 °C, the relative humidity (RH) 72.5% ± 4, and the temperature-humidity index (THI) was 85.3 ± 6.

For ascertaining Amino acids in protein of the diets, about 100 mg samples were hydrolyzed in 10 mL of 6 N HCl at 110 °C for 24 h under N2. Rates of AA recoveries through acid or base hydrolysis were assessed using purified bovine insulin with known AA alignment and AA standard. Then the AA samples calculated on the base of identified quantities of standards (Sigma Chemicals, St. Louis, MO, USA) utilizing Millenium-32 Software (Waters, Milford, MA, USA).

2.2. Growth Effieciency

Initial and final body weight, body weight gain (BWG) and feed intake (FI) were determined, and a weekly calculation of the feed conversion ratio (FCR). Throughout the experimental phase the health status and mortalities were measured daily on a regular basis.

2.3. Carcass Traits

Five birds (male) from each replicate (20 birds/treatment) At the end of the experiment were arbitrarily chosen and slaughtered. Eviscerated weight, giblets (liver, gizzard, and heart), total edible parts (carcass and giblets), and abdominal fat were weighted as carcass traits.

2.4. Blood Parameters and Lipid Peroxidation

At slaughtering time, the blood samples (5 mL) were collected then centrifuged at 3000 rpm for 20 min then, the plasma kept at −20 °C until examination. Blood parameters including total protein, albumin, glucose, cholesterol, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were assessed colorimetrically using commercial kits (Diamond Diagnostics, Egypt) following the manufacturer outlines.

Table 1. The ingredients and nutrient analysis of the experimental starter and grower diets.

Ingredients, %	Experimental Diets					
	Control		LPOA		LPHA	
	Starter	Grower	Starter	Grower	Starter	Grower
Yellow corn	59.0	64.1	60.0	65.1	60.8	65.1
Soybean meal, (47%)	28.1	22.5	28.1	22.5	27.3	22.5
Corn gluten meal, 60%	6.20	6.50	1.20	1.50	2.20	1.50
DL-Methionine, 99%	0.02	-	0.19	0.08	0.31	0.23
L-Lysine HCl, 98%	0.15	0.23	0.24	0.14	0.32	0.22
L-Threonine, 99%	0.06	0.08	0.12	0.09	0.32	0.25
Full fat soybean	2.31	2.52	6.00	6.59	4.60	6.20
Dicalcium phosphate	2.21	2.10	2.20	2.05	2.20	2.10
Calcium carbonate	1.10	1.07	1.10	1.05	1.10	1.00
Sodium chloride	0.35	0.35	0.35	0.35	0.35	0.35
Sodium bicarbonate	0.15	0.13	0.15	0.13	0.15	0.13
Potassium carbonate	0.05	0.12	0.05	0.12	0.05	0.12
Premix *	0.30	0.30	0.30	0.30	0.30	0.30
Total	100	100	100	100	100	100
Nutrient Analysis, on DM Basis						
Crude Protein, %	23	21	21	19	21	19
AME, kcal/kg	2974	3033	2974	3034	2979	3033
Calcium, %	1.00	0.95	1.01	0.94	1.00	0.93
Available P, %	0.52	0.49	0.52	0.49	0.52	0.50
Fat, %	3.16	3.32	3.75	3.99	3.54	3.92
Crude fiber, %	2.88	2.77	3.03	2.49	2.95	2.91
Sodium, %	0.21	0.20	0.21	0.20	0.21	0.20
Chloride, %	0.25	0.25	0.25	0.25	0.25	0.25
Potassium, %	0.79	0.73	0.77	0.72	0.76	0.72
Lys, %	1.25	1.15	1.35	1.13	1.37	1.18
Met, %	0.45	0.41	0.55	0.42	0.68	0.57
Met + Cys, %	0.81	0.74	0.89	0.73	1.01	0.88
Thr, %	0.69	0.64	0.74	0.64	0.93	0.80
Met + Cys/Lys	0.65	0.65	0.65	0.65	0.74	0.74
Thr/Lys	0.56	0.55	0.55	0.56	0.68	0.67

* Premix produced by Pestar company, China. each 3 Kg vitamin and mineral mixture contain: Vitamin A 12,000,000 IU, Vitamin D3 2,200,000 IU, Vitamin E 10,000 mg, Vitamin K 2000 mg, Vitamin B1 1000 mg, Vitamin B2 4000 mg, Vitamin B6 1500 mg, Vitamin B 1210 mg, pantothenic acid 10,000 mg, niacin 20,000 mg, biotin 50 mg, folic acid 1000 mg, choline chloride 500 gm, selenium 100 mg, manganese 55,000 mg, zinc 50,000 mg, iodine 1000 mg and carrier $CaCO_3$, to 3000 gm. Abbreviations: control (OPOA): optimum protein and optimum amino acids; LPOA: low protein and optimum amino acids, LPHA: low protein and high amino acids. Apparent Metabolizable Energy (AME).

2.5. *Assessments of Antioxidant Status*

Lipid peroxidation was assessed by analyzing the malondialdehyde (MDA) level in the liver by using kits from Cell Biolabs Inc. (San Diego, CA, USA), according to Richard et al. [21]. Vitamin E quantity in liver using HPLC, following Surai et al. [22]. In short, the liver tissues (200 to 500 mg) were mixed with a 5% (w/v) aqueous solution of NaCl (0.7 mL) and ethanol (1 mL) and vortexed for 2 s, then add hexane (2 mL) homogenized for 2 min. The hexane layer, having the Vitamin E split by centrifugation and accumulated. The isolation was reiterated twice more with 3 mL hexane. Hexane extracts were blended, and re-dispersed in a mix of methanol/ dichloromethane (1:1, v/v). Samples were injected into an HPLC system (Shimadzu Liquid Chromatograph, LC-10AD, Hachioji, Tokyo, Japan Spectroscopic Co. Ltd. with JASCO Intelligent Spectrofluorometer 821-FP) equipped with a Spherisorb, type S30DS2, 3 m C18 reverse phase HPLC column, 15 cm46 mm (PhaseSeparations Limited, UK). Chromatograph was achieved utilizing a mobile phase of methanol/ water (97:3, v/v) at a flow speed of 105 mL/min. Fluorescence analysis of Vitamin E using excitation at 295 nm and emission at 330 nm. Typical solutions of α-tocopherol in methanol were utilized for calibration.

2.6. Muscle Amino Acids Profile

Breast muscle amino acids contents including Lys, Met and Thr were analysis according to the method of Ceylan and Aksu [23]. A total of 10 g of breast meat with 40 mL of 0.1 N HCl homogenized for 45 s at 4 °C subsequently centrifuged 15,000× *g* for 50 min at 4 °C. The supernatants were filtered and were evaluated utilizing (GC-4 CM-PFE, Shimadzu gas chromatograph, Tokyo, Japan) outfitted with a flame ionization detector (FID). The amino acids assessed were lysine, methionine, and threonine. The values were expressed in grams of amino acid per 100 g of breast meat.

2.7. Data Analysis

The collected data were assessed utilizing the generalized linear model (GLM) function of the SAS software package (Version 9.4, trademarks of SAS Institute Inc., Cary, NC, USA, 2016). The significance of means' differences was examined utilizing the Duncan analysis and all variations were judged significant at $p < 0.05$.

3. Results

3.1. Growth Effeicienc

Table 2 illustrates the impact of dietary therapies on growth efficiency, including BW, BWG, FI, FCR, and mortality. It is clear that no substantial differences in the initial BW in Cobb 500 chicks. Chicks fed a diet containing LPOA and LPHA didn't register differences compared to C and but differed among them. Chicks fed a diet containing low protein with optimum amino acid diet (LPOA) recorded significantly the highest BW, while those fed low protein with high amino acid diet (LPHA) recorded significantly the lowest BW (1940.0 vs. 1737.5 g, $p < 0.05$). Likewise, birds receiving LPOA diet had the maximum WG, while those fed LPHA diets recorded the lowest WG (54.29 vs. 48.50 g, $p < 0.05$). As for FI, chicks fed LPOA diet had the highest amount, while those fed LPHA diets had the lowest value, in relation to those fed control diet (96.77 and 87.83 vs. 92.97 g, $p < 0.05$, respectively). No substantial differences could be noticed among treatments in the FCR. Mortality rate (%) was significantly higher with chicks fed LPOA diet (10.0%), as linked with those control and LPHA diets (4.38 and 3.75%, respectively).

Table 2. Effect of experimental diets on growth efficiency in Cobb 500 broiler chickens.

Item	Experimental Diets			*p*-Duncan
	Control	LPOA	LPHA	
Initial body weight, g	40.0 ± 0.51	40.0 ± 0.64	40.0 ± 0.56	0.95
Final body weight, g/35 d	1929.0 ± 93.60 ^{ab}	1940.0 ± 47.08 ^{a}	1737.5 ± 34.97 ^{b}	0.05
Body weight gain, g/d	53.97 ± 2.674 ^{ab}	54.29 ± 1.345 ^{a}	48.50 ± 0.999 ^{b}	0.05
Feed intake, g/d	92.97 ± 3.19 ^{ab}	96.77 ± 2.442 ^{a}	87.83 ± 0.731 ^{b}	0.05
Feed conversion ratio, g/g	1.727 ± 0.04	1.784 ± 0.04	1.813 ± 0.03	0.32
Mortality *, %	4.38 ^{b}	4.42.00 ^{a}	3.75 ^{b}	-

^{a,b} The means values placed at the rows by different superscript letters are significantly different (p <0.05). Values are expressed as means ± standard error. Abbreviations: control (OPOA), optimum protein and optimum amino acids; LPOA, low protein and optimum amino acids, LPHA, low protein and high amino acids. * Chi-square test.

3.2. Carcass Traits

Table 3 demonstrates the findings related to the impact of dietary treatments on certain carcass traits. There were no substantial effects on the carcass and dressing percentage by reduction dietary CP in compared to control diet. Chicks fed with diet of LPHA showed the highest significantly liver and giblets percentages, whereas there were no major variations between those fed the control and the LPOA diets. Gizzard and heart percentages did not significantly affect by reducing dietary protein levels. The rate of abdominal fat in chicks fed LPHA was increased ($p < 0.05$) in relation to LPOA and control diets (1.277 vs. 0.939

and 1.010%, respectively). In addition, the percentages of the breast and thigh muscle did not considerably affect by reduced CP in the diet.

Table 3. Effect of experimental diets on carcass traits in Cobb 500 broiler chickens.

Item	Experimental Diets			*p*-Duncan
	Control	LPOA	LPHA	
Carcass weight, g/100 g BW	66.5 ± 0.80	66.3 ± 0.24	67.0 ± 0.43	0.61
Dressing weight, g/100 g BW	70.6 ± 0.79	70.2 ± 0.29	71.5 ± 0.35	0.22
Breast muscle weight, g/100 g BW	15.33 ± 0.36	15.95 ± 0.44	15.77 ± 0.27	0.48
Thigh muscle weight, g/100 g BW	15.68 ± 0.27	15.63 ± 0.30	15.81 ± 0.27	0.90
Gizzard weight, g/100 g BW	1.57 ± 0.09	1.45 ± 0.09	1.67 ± 0.05	0.19
Heart weight, g/100 g BW	0.455 ± 0.02	0.469 ± 0.02	0.462 ± 0.04	0.94
Liver weight, g/100 g BW	2.02 ± 0.06 [b]	2.03 ± 0.11 [b]	2.37 ± 0.11 [a]	0.04
Giblets weight, g/100 g BW	4.04 ± 0.11 [b]	3.96 ± 0.16 [b]	4.50 ± 0.13 [a]	0.02
Abdominal fat weight, g/100 g BW	1.010 ± 0.08 [b]	0.939 ± 0.08 [b]	1.277 ± 0.06 [a]	0.02

[a,b] The means values placed at the rows by different superscript letters are significantly different (p <0.05). Values are expressed as means ± standard error. Abbreviations: control (OPOA), optimum protein and optimum amino acids; LPOA, low protein and optimum amino acids, LPHA, low protein and high amino acids. Body weight (BW).

3.3. Blood Parameters and Lipid Peroxidation

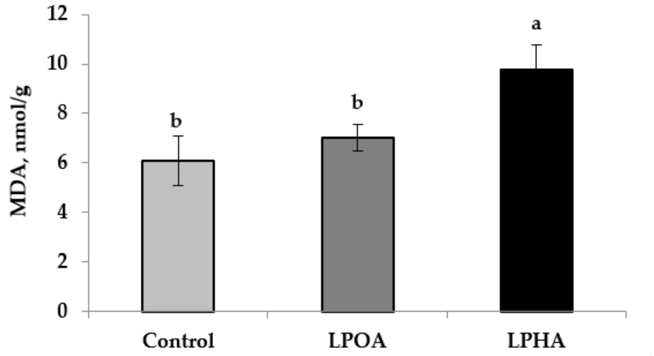
Results concerning the effects of different experimental treatments on blood parameters including albumin, globulin and glucose, cholesterol, AST and ALT concentration in broiler chickens are displayed in Table 4. Plasma total protein, albumin, and globulin were not significant differences due to dietary low protein. Total protein concentration in the LPOA treatment was 12.6% higher than the control treatment. Nevertheless, these differences were not statistically meaningful.

Table 4. Effect of experimental diets on blood parameters in Cobb 500 broiler chickens.

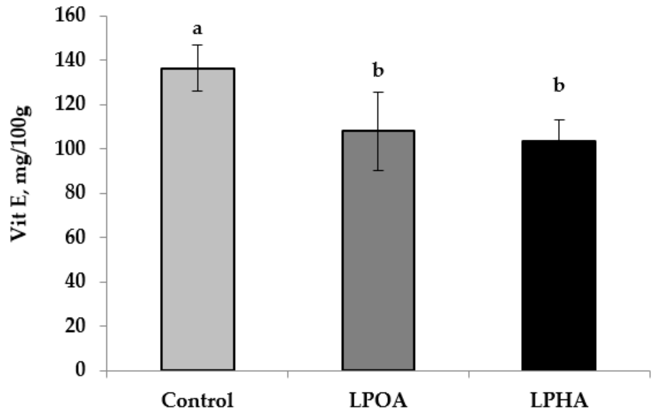
Item	Experimental Diets			*p*-Duncan
	Control	LPOA	LPHA	
Total protein, mg/dL	3.41 ± 0.35	3.84 ± 0.43	3.15 ± 0.17	0.37
Albumin, mg/dL	1.00 ± 0.04	1.15 ± 0.10	1.05 ± 0.07	0.38
Globulin, mg/dL	2.50 ± 0.26	2.88 ± 0.53	2.10 ± 0.10	0.32
Glucose, mg/dL	238.3 ± 12.73	210.8 ± 18.06	209.6 ± 9.65	0.29
Total cholesterol, mg/dL	125.9 ± 5.99	115.7 ± 1.62	131.9 ± 10.51	0.27
AST, I/U	288.6 ± 21.29	276.4 ± 32.76	297.9 ± 33.87	0.88
ALT, I/U	5.64 ± 2.27	5.59 ± 1.33	6.51 ± 1.25	0.91

Values are expressed as means ± standard error. Abbreviations: control (OPOA), optimum protein and optimum amino acids; LPOA, low protein and optimum amino acids; LPHA, low protein and high amino acids; AST, aspartate aminotransferase; ALT, alanine aminotransferase. Eight samples were analysis per treatment.

The same trend was observed for plasma glucose and cholesterol, which did not significant differences by decreasing dietary protein. There were no substantial differences in liver function enzymes amongst treatments (AST, aspartate aminotransferase and ALT, alanine aminotransferase). While the results of liver MDA and Vitamin E contents are shown in Figure 1a,b. Chicks fed low CP with high amino acid diet (LPHA) recorded the highest ($p < 0.05$) liver content of malonaldehyde (MDA), while there were no substantial differences found between control and LPOA treatment diets (9.77 vs. 6.08 and 7.02 mg/mL, respectively). The reduction of dietary protein levels significantly decreased the liver content of Vitamin E by 18.75%, as compared with the control group treatment.



(**a**)



(**b**)

Figure 1. Effect of dietary treatments on liver Malondialdehyde (MDA) (**a**) and Vitamin E (**b**). Values are means represented by vertical bar. ^{a,b} Mean values with unlike letters were significantly different ($p < 0.05$). Abbreviations: control (OPOA), optimum protein and optimum amino acids; LPOA, low protein and optimum amino acids; LPHA, low protein and high amino acids.

3.4. Muscle Amino Acids Profile

Respective to the impact of dietary treatments on muscle amino acids profile including Lys, Met and Thr levels (Figure 2a–c), no significant differences could be observed among treatments due to the muscle content of Lys, Met, and Thr.

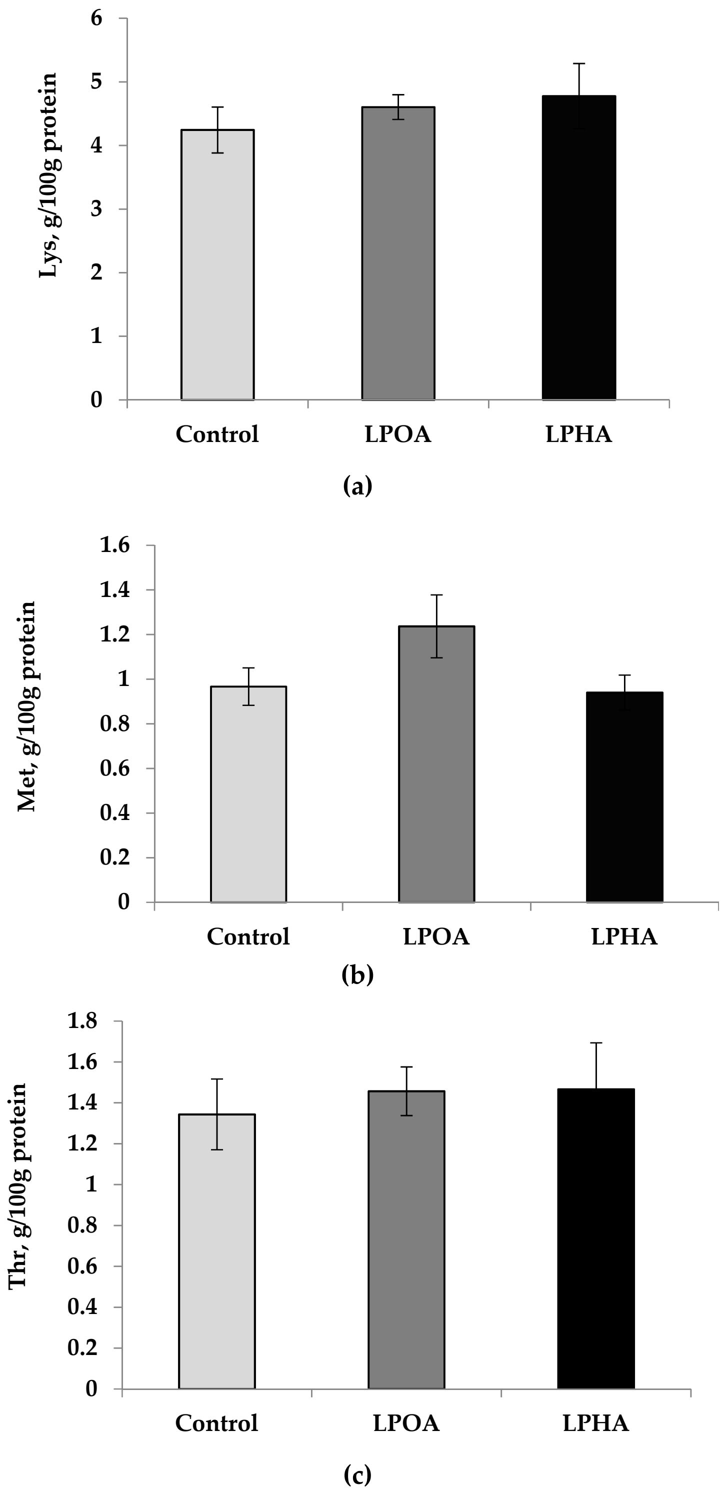


Figure 2. Effect of dietary treatments on Lysine (Lys) (**a**), Methionine (Met) (**b**) and Threonine (Thr) (**c**) breast muscle amino acids. Values are means represented by vertical bar. Abbreviations: control (OPOA), optimum protein and optimum amino acids; LPOA, low protein and optimum amino acids.

4. Discussion

4.1. Growth Performance

Our findings correspond to earlier research, which showed that by using synthetic glutamic amino acids and essential amino acid reinforcement, the CP can be successfully reduced to 25% and thus achieve the same result for higher CP [8,24]. Besides, CP of the starter feed for broilers (up to three weeks) can be reduced to 19.20% with no unfavorable impact on efficient efficiency under the high temperature and wet tropics based on the essential amino acid supplement [25]. In addition, Cobb Avian 48 broilers fed a 1% lower protein diet at constant ME with identical levels of amino acid did not adversely affect the performance of growth, liver function, and carcass parameter; although; dietary CP and crude fiber consumption were improved [26]. Kobayashi et al. [27] reported similar findings, it was found that the BWG in the low CP with EAA (ELCP) treatment was 130 g/10 days smaller than that of the control treatment, but the differences were not considerably significant. Lowering the protein content in a diet was presumed to provide less substrates from which putrefactive bacteria at the gastrointestinal tract (GIT) would generate poisonous substances (i.e., ammonia, amines, indoles, phenols) that might trigger large intestine disruption and impaired growth [28–30]. Interestingly, Ospina-Rojas et al. [31] recorded that the diet that supplemented with synthetic amino acids has no any harmful impacts on the growth efficiency of broilers on day 42, the CP level might be decreased by 3%, in accordance with our finding for the grower and finisher periods. Additionally, broilers in the finisher period demonstrated the reduced non-essential amino acids requirements that illuminates the similar results achieved in the current study for broilers fed with control and LPOA diets [32]. Previous studies [33,34] indicated that the unfavorable impact of the low CP diet on intestinal morphology may be due to a decrease in quantities of non-essential amino acids for instance glycine, glutamine and proline required to grow the GIT epithelium and to create digestive secretions and mucin. Badawi et al. [35] reported that when broiler chicks fed 2, 4 and 6% of standard CP control diet, there was no significant change in BW, BWG and FI concerning the control treatment ($p < 0.05$). In addition, Lemme et al. [36] studied the impact of reducing the CP quantities with ideal supplementation amino acid on the growth efficiency of broiler chickens and found that final BW was comparable among Treatments 1, 2, and 3 but marginally reduced in Treatment 4 ($p < 0.05$) even though FCR was not altered. They also observed that the yield of carcass and breast meat in male broilers has no substantially among treatments ($p < 0.05$). Recently, Macelline et al. [2] observed that there were no any adverse effects in broiler that fed with low CP diet with essential purified amino acid supplied under deprived sanitary circumstances for 14 days after hatching whereas resultant in reduce of the nitrogen excretion in any environmental circumstances during the experiment. They also observed that the feeding low protein diet with supplemented of an artificial amino acid caused expression of tight junction genes (ZO1 and claudin 1) that are necessary for the maintaining gut health. There was also a marked increase in weights of liver of chickens fed with scanty CP diets relative to those fed with higher CP diets [37] that could be correlated with increased ME: CP ratio in low CP fed (31.5 vs. 16.5) due to rise ME: CP ratio will efficiently improve body lipogenesis [38]. The increased concentration of blood plasma cholesterol in the chickens fed low CP diets supports this assumption.

4.2. Carcass Traits

Our results on carcass traits are consistent with the trial stated that there are no alterations in the carcass, breast yield, and body fat in birds fed low CP together with an ideal dietary ratio of ME:CP [39]. Moreover, the essential amino acid level of a diet affects the carcass components of broiler chickens, and sufficient essential amino acids have no impact on the carcass' protein and fat quantity even if the CP is lowered [40]. In addition, Badawi et al. [35] studied the effect of broiler chicks fed reduced 2%, 4% and 6% of the standard CP control diet, and reveals no considerable variation in the weight percentage of dressing, and visceral organs in relation to live body weight in all experimental birds ($p < 0.05$). Concerning abdominal fat, our data was in the line with the report noted that abdominal fat weight was risen by 104% when the broilers had a 17% CP diet in relation to those fed 24% CP Van Harn et al. [7]. In comparison, broilers fed on low CP diets kept high fat in their bodies than the those fed the diet on power. The increased ME:CP is one key element accountable for enhanced abdominal fat with low CP fed [8,41]. This induced increased energy availability surplus, which boosted body lipogenesis of chickens fed low CP diets, which increased abdominal fat content. Moreover, Kobayashi et al. [27] stated that in the low CP treatment, the breast yield was smaller ($p < 0.05$) in relation to the control one, with no substantial differences between low CP complemented with essential amino acids (ELCP) and control. In the low CP treatment, the mean abdominal fat yield was 36.6% greater than in the control one. Nevertheless, due to the large differences, the difference was not statistically significant.

4.3. Blood Parameters and Lipid Peroxidation

Dietary protein reduction did not appear any significant differences in blood parameters. This finding in harmony with that of Dairo et al. who stated that the blood plasma GOT and GPT were not affected by decreasing dietary protein by 1% [42]. Moreover, Badawi et al. [35] no noticeable changes have been observed among different treatments based on low CP diets in total cholesterol, LDL, total protein, albumin, globulin, ALT, AST, creatinine, uric acid, urea, sodium and potassium (2, 4 and 6% of the standard CP control diet), and complemented with synthetic essential, non-essential amino acids and L-carnitine ($p < 0.05$). In addition, Ndazigaruye et al. [43] showed that the blood serum triglyceride, albumin, total cholesterol, high-density lipoprotein, globulin, GOT and GPT were not affected in the diets that supplemented with exogenous protease enzyme and low 1% CP and 8–12 amino acid. Low protein diet with high amino acids increased liver peroxidation, this finding was not identical with Elize et al. who reported that while the liver of animals fed low protein diet had lower levels of glutathione (GSH) and Vitamin E, it was proposed that GSH and Vitamin E played a crucial role in removing damaging free radicals created during low protein diets. These findings show that hyperoxia and a protein deficit potentiates lipid peroxidation [44].

4.4. Muscle Amino Acids Profile

There were insignificant differences between breast muscle Lys, Met and Thr amino acids profile values observed in Cobb 500 broiler chickens received the various experimental diets. There is no information available to explain these results, but perhaps these results are positive and logical because it is difficult to modify the components of the levels of amino acids in the meat because they are among the components of protein. This is considered a positive result, meaning that changing the composition of protein and amino acids is difficult [45]. Otherwise, the percentages of fats, fatty acids and vitamins in the meat can be changed by increasing or decreasing the content of fats and fatty acids in the diet [8,38,46]. On the other hand, the bird tries to obtain its nutritional requirements of protein or amino acids only, but in the event that the added percentages of these nutrients exceed the need, the bird gets rid of the excess from them, therefore the data of this study displayed that the increase in the amino acids ratios in the diet did not give any positive effects, on the contrary, were negative [9].

5. Conclusions

In conclusion, feeding Cobb 500 broiler on low protein diets at persistent ME with the same concentrations of amino acid does not adversely affect growth performance, carcass traits, blood parameters and liver functions, However, increased amino acids levels to scanty protein diets led negative effects on growth efficiency under high ambient temperature.

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M.H.A.; visualization, A.A.S., K.A.A., M.M.S., M.Y.S., W.A.M., M.S., M.H.A.; supervision, M.H.A.; project administration, A.A.S., M.S.; funding acquisition, M.M.S., W.A.M., M.H.A. All authors have read and agreed to the published version of the manuscript.

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