



Effect of dietary amino acid regimens on growth performance and body conformation and immune responses in Aseel chicken

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ABSTRACT

The present study was designed to evaluate the effect of different levels of Lysine (Lys) on early growth, body conformation and immune response of three varieties of Aseel chicken. Five hundred and forty day old chicks were used in this experiment. Experimental treatments were three varieties i.e Mianwali (MW), Peshawari (PW) and Lakha (LK) and three level of Lys i.e L1 (1.35), L2 (1.30) and L3 (1.25%). Treatments were arranged in factorial arrangement under randomized complete block design (RCBD) with sex as block. Each experimental group was replicated 6 times with 10 birds in each. Results indicated that PW variety showed a greater feed intake (FI) throughout the experiment, however, body weight gain (BWG) and feed conversion ratio (FCR) were remained unchanged within varieties. Lower FI was observed in birds fed diets containing 1.35, 1.30% digestible Lys. Birds fed diets containing 1.35, 1.30% digestible Lys showed higher (BWG ($P < 0.05$) and improved FCR ($P < 0.05$). Mianwali variety with 1.35, 1.30% digestible Lys diets, showed (252.0±17.4 and 251.4±7.15; 2.37±0.17 and 2.35±0.06) an interaction for BWG and FCR. Birds fed amino acid dense diets depicted improved body conformation traits (length of body, drum stick, shank keel bone, circumference of drumstick and shank, wing spread and breast width), whereas immune responses (were not influenced by the dietary treatments. In conclusion, it was found that 1.30% digestible Lys level with ideal amino acid ratio was better for improving early BWG and FCR in Mianwali Aseel.

Key words: Amino acids, Aseel chicken, Body conformation, Immune responses.

INTRODUCTION

Rural poultry is an important sub sector of poultry production in developing countries. Indigenous breeds of poultry have better disease resistance and adaptability to the local harsh environmental (Khan *et al.*, 2017). Moreover, local breeds have additional attraction to local folks as a meat source (Barua and Howlader, 1990). Aseel is a prominent indigenous breed of Indo-Pak sub-continent having higher final body weight (4 to 7 kg) (Babar *et al.*, 2012; Usman *et al.*, 2014) and superior quality of meat in (Dohner, 2001). Problem related with Aseel birds are poor early growth, poor FCR, higher cost of production and late maturity (Jatoi *et al.*, 2014).

Aseel can be used as a meat type bird and its growth potentials can be enhanced through improved nutrition using modern nutritional tools. Recent study reported organic selenium supplement (improve the growth performance of Aseel (Zia *et al.*, 2017). Similarly, increasing the level of energy and protein enhance the FCR of Aseel Haunshi *et al.* (2012). Baker and Han (1994) found that specific amount of individual amino acids is necessary for proper growth performance, termed as ideal amino acid ratio. Increasing the amount of amino acids (AA) improve the growth and breast meat yield because of increase in protein synthesis (Dozier *et al.*, 2008). Lysine (Lys) is one of critical AA which contribute in increase in breast yield and it is estimated that breast meat contains 7% Lys 7% (Dozier *et al.*, 2007; Mahdavi *et al.*, 2012). Therefore, increasing the level of Lys in the diet could increase breast yield and ultimately growth performance of the Aseel.

During last few years, ideal protein or ideal amino acid

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(IAA) and tissue accretion methods are used to check retention efficiency of AA (Tillman and Dozier, 2013). These methods have beneficial effects by improving protein efficiency and decrease N excretion. Numerous studies conducted on total and digestible AA of different ingredients revealed better growth performance in birds (Lemme *et al.*, 2004). With this background, the present study was designed to investigate the effect of different levels of digestible amino acids in the diets formulated based on ideal amino acid concept using digestible amino acids on early growth performance, body conformation and immune response among three varieties of indigenous Aseel chicken of Pakistan.

MATERIALS AND METHODS

The study was conducted at Indigenous Chicken Genetic Resource Center (ICGRC), Department of Poultry Production, University of Veterinary and Animal Sciences, Lahore-Pakistan. In total, 540 day old chicks, 180 from each variety i-e Mianwali (MW), Peshawari (PW), and Lakha (LK) were randomly assigned 3 Lys regimens i-e L1 (1.35), L2 (1.3) and L3 (1.25) by using factorial arrangement under randomized complete block design (RCBD) with sex as block. Each experimental group was replicated 6 times with 10 birds in each with average weight of 29 gram.

Six different diets with variable Lys levels formulated. Experimental diets were formulated by using local grain data on the basis of ideal digestible amino acid ratios by Tillman and Dozier (2013). Control dietary treatment was formulated in such a way that the allowance of Lys 1.25% throughout the experimental period (0 to 28 days) was maintained. Whereas for high and medium dietary treatments Lys levels

were decreased by 0.15% to 1.35 to 1.20% for high dense diet and 1.30 to 1.15% for medium dense diets. All diets were iso-nitrogenous and iso-caloric to avoid any other variable (Table 1). Before the start of trial, experimental diets were analyzed for dry matter (DM), crude protein (CP), crude fiber, ether extract (EE), ash, calcium and phosphorus by following AOAC (2005). Amino acid analysis was executed using Biochrome 30+ Series amino acid analyzer resulting in $\pm 2\%$ from the calculated values and metabolizable energy was calculated through regression equation as described by NRC (1994). Linear formulation method was used for diets formulation. Experimental diets were offered for the duration of 4week.

Parameter studied

Data were collected (weekly and pen wised) to evaluate the growth performance, immune response and body conformation. The following parameters were studied;

Table 1: Feed formulation and nutrients calculations.

Ingredients (%)	Treatments ¹					
	1 to 14 day			15 to 28 day		
	L1	L2	L3	L1	L2	L3
Corn	61.97	62.44	62.66	65.28	64.03	65.78
Soybean Meal	23.05	19.94	21.97	20.93	23.72	20.20
Fish meal	8.00	11.13	9.04	6.06	5.46	6.09
Corn Gluten 60%	3.00	3.00	3.00	3.00	2.00	3.00
CaCO ₃	0.97	0.97	0.99	0.95	0.80	0.95
DCP	0.20	0.20	-	-	0.10	-
Common Salt	0.30	0.30	0.30	0.30	0.30	0.30
NaHCO ₃	0.20	0.34	0.20	0.24	0.26	0.24
Vegetable Oil	0.20	-	-	1.43	1.94	1.26
Vit-Min Premix	0.50	0.50	0.50	0.50	0.50	0.50
Lysine Sulphate	0.61	0.46	0.40	0.54	0.37	0.67
DL-Methionine	0.35	0.29	0.26	0.30	0.26	0.34
L-Threonine	0.25	0.21	0.18	0.19	0.14	0.23
L-Valine	0.09	0.03	0.38	0.06	0.01	0.11
L-Isoleucine	0.11	0.04	0.01	0.06	0.00	0.10
L-Tryptophan	-	-	-	0.00	-	0.01
L-Arginine	0.19	0.16	0.10	0.16	0.11	0.23
Nutrients						
CP (%)	23	23	23	21	21	21
ME (Kcal/kg)	3000	3000	3000	3100	3100	3100
Calcium	1.00	1.00	1.00	0.80	0.80	0.80
Available Phosphorus	0.40	0.40	0.40	0.30	0.30	0.30
Digestible Lysine	1.35	1.30	1.25	1.20	1.15	1.25
Digestible TSAA	1.02	0.98	0.94	0.92	0.88	0.95
Digestible Threonine	0.95	0.91	0.88	0.83	0.79	0.86
Digestible Tryptophan	0.22	0.21	0.20	0.20	0.19	0.20
Digestible Arginine	1.42	1.37	1.32	1.28	1.22	1.33
Digestible Valine	1.02	0.98	0.94	0.92	0.88	0.95
Digestible Isoleucine	0.89	0.86	0.83	0.81	0.77	0.84
Digestible Phenylalanine	0.85	0.82	0.79	0.76	0.72	0.79

¹L1=high d-lysine L2=Medium d-lysine L3= Low d-lysine

Body weight gain (BWG) (g):

Final weight – Day Old Chick Weight

Feed intake (FI) (g): Feed Allowances – Feed Residue

Feed conversion ratio (FCR): Feed Intake / Weight Gain

Protein efficiency ratio (PER): Weight Gain / Protein Intake

Lysine efficiency ratio (LER): Weight gain / Lysine Intake

At the end of experiment, blood samples were collected and serum was separated. The titer of Newcastle Disease (ND) using hemagglutination inhibition assay, infectious bronchitis (IB) and Infectious Bursal Disease (IBD) using ELISA test were performed according to the methods described by Chang *et al.* (2011). Body, drumstick, shank, keel bone length were measured with the help of measuring scale capable of measuring 0.05 mm. Drumstick, shank circumference and breast width were measured with the help of Vernier calliper capable of measuring up to 0.05 mm.

Statistical analysis

Before analysis, data were checked for uniformity and homogeneity of variance and verified for the normality. After that, the data were analyzed through ANOVA technique in factorial arrangement under RCBD by using GLM procedure of SAS assuming following mathematical model:

$$Y_{ijk} = \mu + \beta_i + V_j + L_k + (VL)_{jk} + \varepsilon_{ijk}$$

Where, Y_{ijk} is Dependent variables, μ is overall population mean, β_i are the blocks that is sex, V_j is effect of i th treatment

($i=3$; Varieties), L_k is effect of j th treatment ($j=3$; Lysine Regimens), $(VL)_{jk}$ is interaction effect and ε_{ijk} is residual error. Aseel variety and Lys regimens were taken as main effects. Interaction of these were also tested. Treatment means were compared through Tukeys HSD test at 5% probability level. Each replicate was considered as an experimental unit.

RESULTS AND DISCUSSION

The greatest feed intake (FI) was observed in Peshawari variety followed by Mianwali and Lakha Table (2). With respect to dietary treatments, reduced ($P<0.05$) FI was observed with high dense diets. In interaction of varieties and different dense diets, minimum FI (569.8±4.4) was observed in Lakha variety fed high dense diet, whereas higher FI (640.7±3.6, 640.0±3.4 and 639.0±3.9) was found in low Lys diets fed birds of all varieties. Greater FI in Peshawari variety may be attributed to the genetic variation among varieties. It has already been reported Peshawari variety has higher FI as compare to Lakha and Mianwali (Jatoi *et al.*, 2014). High dense diets in birds reduced FI because dense diet full fill the nutritional requirement of the birds and satiated them (Corzo *et al.*, 2005). In current study, decrease in FI might be attributed to satiated effect of dense diet on intake of birds. Similar finding of lower FI due to high dense diets has also been reported in broilers (Corzo *et al.*, 2005; Sklan and Plavnik, 2002). Other possible reason

Table 2: Growth performance of indigenous chicken in response to dietary amino acid regimens.

Variables ¹	Parameters ²					
	FI	BW	FCR	LER	PER	
Varieties						
LK	600.4±7.5 ^b	230.4±7.1	2.66±0.10	28.61±1.2	1.57±0.06	
MW	604.4±7.3 ^b	240.6±7.3	2.56±0.09	29.59±1.3	1.63±0.07	
PW	610.8±6.0 ^a	230.1±4.6	2.68±0.07	27.59±1.2	1.53±0.04	
AA Densities						
L1	578.1±2.2 ^c	241.8±8.2 ^a	2.44±0.08 ^b	22.94±0.9 ^b	1.71±0.06 ^a	
L2	597.6±2.9 ^b	244.3±4.2 ^a	2.46±0.05 ^b	31.96±0.6 ^a	1.67±0.04 ^a	
L3	639.9±2.0 ^a	214.9±3.6 ^b	2.99±0.05 ^a	30.88±0.6 ^a	1.35±0.03 ^b	
Varieties × AA Densities						
LK	L1	569.8±4.4 ^e	239.2±15.5 ^{abcd}	2.43±0.16 ^b	23.01±1.5 ^b	1.71±0.12 ^a
	L2	590.5±1.8 ^c	244.7±8.03 ^{abc}	2.43±0.08 ^b	32.61±1.0 ^a	1.71±0.06 ^a
	L3	640.7±3.6 ^a	207.4±6.42 ^d	3.11±0.10 ^a	30.20±1.3 ^a	1.30±0.05 ^c
MW	L1	583.7±2.6 ^{cd}	252.0±17.4 ^a	2.37±0.17 ^b	23.86±1.93 ^b	1.77±0.14 ^a
	L2	589.4±2.5 ^{cd}	251.4±7.15 ^{ab}	2.35±0.06 ^b	33.57±0.61 ^a	1.75±0.05 ^a
	L3	640.0±3.4 ^a	218.4±6.49 ^{cd}	2.95±0.09 ^a	31.34±1.15 ^a	1.38±0.05 ^{bc}
PW	L1	580.7±1.0 ^d	234.4±9.95 ^{abcd}	2.50±0.12 ^b	21.95±1.06 ^b	1.64±0.08 ^a
	L2	612.8±2.3 ^b	236.7±6.44 ^{abcd}	2.60±0.08 ^b	29.72±1.08 ^a	1.57±0.05 ^{ab}
	L3	639.0±3.9 ^a	219.2±5.73 ^{bcd}	2.92±0.07 ^a	31.11±0.96 ^a	1.38±0.04 ^{bc}
Linear and Quadratic Responses Due to Dietary Treatments						
L	*	NS	NS	NS	NS	
Q	NS	*	*	*	*	

¹LK=Lakha, MW=Mainwali, PW=Pashwari, L1=high d-lysine, L2=Medium d-lysine, L3= Low d-lysine diet

²FI=feed intake, BW=body weight, FCR= feed conversion ratio, PER= protein efficiency ratio, LER= lysine efficiency ratio. Value with different superscript within column differ significantly ($P<0.05$).

to decrease the feed intake might be due branched chain AA which results in a decrease of FI (Trottier and Easter, 1995).

Body weight gain (230.4±7.1, 230.1±4.6 and 240.6±7.3) was similar (P>0.05) in all varieties. Dietary treatments resulted in a higher (P<0.05) BWG in birds fed high and medium dense diets (244.3±4.2 and 241.8±8.2) is higher in comparison to low Lys diets (214.9±3.6). Interaction between varieties and AA densities displayed improved (P<0.05) BWG in Mianwali with high dense (252.0±17.4), followed by Mianwali with medium dense (251.4±7.15), Lakha with medium (244.7±8.03) and high dense (239.2±15.5) diets. Higher BWG (240.6±7.3) in MW Aseel reared under dense diets could be due to increased protein synthesis which involves the role of dietary amino acids (Dozier *et al.*, 2008). Dense diet improve the nutritional status of the feed and recent study has reported improved body weight gain and FCR in males Aseel in nutritionally improved diets (Zia *et al.*, 2017). Interactions of amino acids, play a vital role in the protein synthesis like Lys, methionine and arginine interaction involves in the synthesis of creatine that improves the muscle growth (Chamruspollert *et al.*, 2002). Similar with our findings Ngambi *et al.*, 2017 also reported that diet dense with threonine improve performance of slow-growing Venda chickens (Ngambi *et al.*, 2017).

Varieties showed similar (P>0.05) FCR. The dietary treatment improved (P<0.05) FCR, where linearly improved (P<0.05) FCR was observed by increasing diet densities from lower to higher dense diets. Dietary treatments demonstrated improved (P<0.05) FCR in all varieties fed high and medium dense diet and Mianwali variety (2.35±0.06 and 2.37±0.17) in particular compared to diet containing control level of AA. Improved FCR of Mianwali Aseel might be attributed to the lower FI and higher weight gain, since birds perform better with increasing AA densities (Zhai *et al.*, 2013, 2014). Similar findings have also been reported in broiler fed AA supplemented diets (Quentin *et al.*, 2003). Furthermore, Li *et al.*, 2013 reported increasing Lys level enhance FCR in local Chinese chicken. Phase wise WG and FCR also showed significant differences (P<0.05).

Where for 0-14 WG for all dietary treatments were 87.5±3.5, 87.5±2.3, 64.9±4.2 and FCR were 2.04±0.04, 2.14±0.03, 2.84±0.06 for L1, L2 and L3 respectively. For 15-28 WG were 154.3±6.4, 156.8±4.9, 150±4.8 and FCR 2.67±0.09, 2.64±0.04, 3.08±0.07 for L1, L2 and L3 Lys regimens (Fig 1).

Protein efficiency ratio (PER) shows similar results like FCR, whereas varieties showed similar (P<0.05) PER. The dietary treatment improved (P<0.05) PER, where linearly improved (P>0.05) PER was observed by increasing diet densities from low to high dense diets. Dietary treatments demonstrated improved (P<0.05) PER in all variety fed on high and medium dense diets. Better (P>0.05) LER reflected in birds fed high dense diets.

Dietary treatments had no significant effect (P>0.05) on immune status. Overall immune responses were much protective against ND, IB and IBD diseases (Table 3). Higher growth rate in MW chicken did not compromised immune response which might be attributed to the result of amino acids relation with immune system in terms of lymphocyte proliferation and activation (Tsiagbe *et al.*, 1987), improve cellular redox status, cytotoxic cell-mediated immunity, serum blocking activity, and haemagglutinin titre (Konashi *et al.*, 2000). Better immune response against different diseases due to Lys and methionine supplementation

Table 3: Effect of dietary amino acid densities on immune responses of birds.

Parameters	Treatments ¹			SE
	L1	L2	L3	
NDV	4.67	4.67	5.0	0.69
IB	4491.7	4459.1	4790	690.7
IBD	1409.5	1339.7	1420.9	143.8
SP	N/D ²	N/D	N/D	-
MG	N/D	N/D	N/D	-

¹L1=high d-lysine L2=Medium d-lysine L3= Low d-lysine diet² Not Detected

Value with different superscript within row differ significantly (P<0.05).

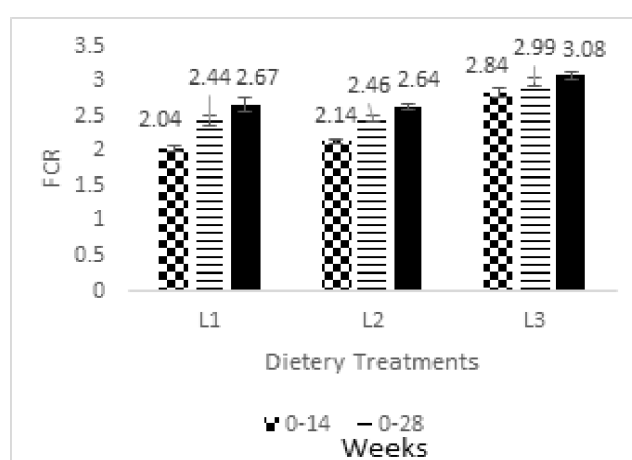
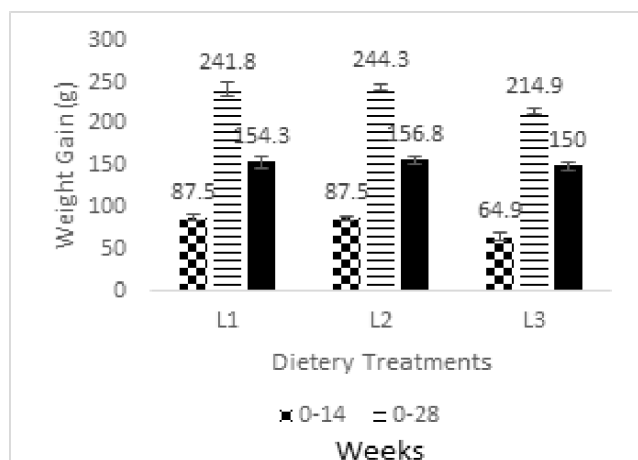


Fig 1: Phase wise weight gain and feed conversion ratio in varying lysine diets.

Table 4: Effect of dietary amino acid densities on body confirmation traits of birds.

Variables ¹	Traits ²								
	BL	DSL	DSC	SL	SC	WS	WSPN	KBL	BW
L1	12.8±0.13 ^{ab}	2.52±0.06 ^b	2.21±0.09 ^a	1.83±0.05 ^a	1.00±0.03 ^{ab}	7.58±0.08 ^a	15.9±0.15 ^b	2.09±0.03 ^{ab}	24.4±0.53 ^a
L2	13.08±0.17 ^a	2.66±0.06 ^a	2.09±0.06 ^a	1.91±0.03 ^a	1.04±0.03 ^a	7.61±0.10 ^a	16.3±0.10 ^a	2.15±0.03 ^a	24.9±0.42 ^a
L3	12.5±0.07 ^b	2.52±0.01 ^b	1.84±0.05 ^b	1.67±0.03 ^b	0.94±0.02 ^b	7.22±0.05 ^b	15.7±0.16 ^b	2.03±0.06 ^b	21.9±0.49 ^b

¹L1=high d-lysine L2=Medium d-lysine L3= Low d- lysine diet. ²BL=body length, DSL=drum stick length, DSC=drum stick circumference, SL=shank length, SC=shank circumference, WS=wing spread, WSPN=wing span, KBL=keel bone length and BW=breast width. Value with different superscript within column differ significantly (P≤0.05).

especially against ND has already been highlighted (Bouyeh 2012). Recently a study with Aseel concluded that nutritional manipulation can increase glutathione peroxidase and ultimate superior performance with improve blood biochemical profile (Zia *et al.*, 2016)

Dietary treatments had a significant effect (P<0.05) on all body conformation traits except keel length and shank circumference. Overall medium dense diets showed better body conformation traits (5%) followed by high dense (2.5%) then control diets (Table 4). Length of feathers decreased in response to decreased dietary protein intake (Wylie and Hocking, 1999). Effect on body confirmation traits might be the effect of differential nutrient partitioning due to increase nutrient densities, which resulted in better body frame development (Sikur *et al.*, 2004).

CONCLUSION

It can be concluded that diet containing 1.30 % digestible Lys level with ideal amino acid ratios, is better to improve early weight gain, FCR and body conformation in Mianwali Aseel chicken.

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