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Effects of Reduced- Protein Diets at Constant Total Sulfur Amino Acids: Lysine Ratio on Pullet Development and Subsequent Laying Hen Performance

[1]Fariborz Khajali, [1]M. Faraji, and [2]Saeid Karimi Dehkordi
[1]Department of Animal Science, Shahrekord University, Shahrekord Iran, postal code: 88186-34141
[2]Laboratory for Animal Nutrition, University of Ghent, Ghent, Belgium

Abstract: The effect of using a low protein regime in pullet development and subsequent layer performance was studied. The experiment consisted of two periods of rearing and laying. Two hundred and sixty Hy-line W36 chicks were used. In the rearing period, dietary protein level and ME content series of the control group used during starter (18-42), grower (43-63) and developer (64-119d) phases were 20, 2930, 18, 2930, and 16, 3025, respectively. Dietary protein sequence of the low protein group was 18.5, 16.5, and 14.6% during the respective periods. In the laying period lasted from week18 to week32 of age, CP content of the control and reduced-protein diets were 17.8 and 16.3%, respectively. Reduced-protein diets were kept isoenergetic with their corresponding controls in each period and phase and they balanced to keep the same total sulfur amino acid to lysine ratio as well. Results indicated that birds on reduced-protein diet during starter phase of the rearing period consumed less feed ($p = 0.003$) and as a result they had significantly ($p = 0.012$) better feed conversion ratio (FCR) compared to the control. However, dietary protein regime had no significant effect on weight gain, feed intake and FCR during grower and developer phases. In the laying period, there was no significant difference in terms of layer performance and egg quality criteria. In conclusion, reduced-CP diets can be satisfactory used for rearing pullets and laying hens up to 32 weeks of age.

Key words: Laying hen, lysine, methionine, protein, pullet

INTRODUCTION

Current standard practice to feed Hy-line W 36 is allocation of a series of diets termed starter, grower and developer, during 0-6, 6-9 and 9-17 wk of age, respectively[7]. The current feeding standard follows a step-down protein regime. Nevertheless, several researchers have examined the step- up and constant dietary protein regimes on pullet development and subsequent laying performance. Leeson and summers[10] reported that pullets given the step-up protein regime had significantly lower body weight at 20 wk of age than those on step-down protein system. Moreover, Cantor and Johnson[4] reported that increasing protein levels sequence (13-16-19%) during the rearing period decreased pullet body weight up to 20 wk of age and reduced egg production, whereas feeding a constant protein sequence (16-16-16%) had no effect on 20-wk body weight or egg production, when both compared to the conventional decreasing protein level sequence. In contrast, using a step-up protein system increased body weight of pullets at 14wk of age[6]. In addition, Hussein[5] developed a modified constant protein feeding system as effective as step-down protein system for pullet growth and development.

Effects of different protein levels in the rearing and laying periods on performance of laying hens have also evaluated by several researchers. Keshavarz[8] observed lower body weight at 20wk and decreased performance during the early phase of egg production cycle when pullets were given low-protein diets during the rearing period. Blair *et al*[3] reported that performance of laying hens on a low protein diet (13.5% CP) even supplemented with essential amino acids at 90% of NRC specifications was as comparable as that of hens on a 17%CP diet. Balance of essential amino acids, especially in terms of total sulfur amino acids and lysine which are two first limiting amino acids is highly important when low-protein diets are fed. Except for few studies taking the total sulfur amino acids: lysine ratio into account conducted with commercial laying hens[13], little attempts have been made in pullet production. The present study used a low-protein diet with similar total sulfur amino acids: lysine ratio as the control and effects of feeding these diets on pullet development and subsequent laying performance were studied.

Corresponding Author: Saeid Karimi Dehkordi, Laboratory for Animal Nutrition, University of Ghent, Ghent, Belgium Proefhoevestraat 10, 9090 Melle Belgium Tel: +32 9 264 90 06

Table1: Dietary composition of the dietary treatments (air dry basis)

18 to 32week of age		64-119day		43-63day		18-42day		
Low-protein	control	Low-protein	control	Low-protein	control	Low-protein	control	Feed ingredients
64.2	60	76.6	74.5	71.5	69.5	65	62	corn
17.2	21.5	14.5	19	20	24.6	28.3	32.8	soybean meal (44%)
4.5	4.5	2	2	2	2	0	0	fish meal (64%)
0.4	0	2.2	0	2.3	0	0.8	0	wheat bran
2.7	3.2	0.5	0.5	0	0	0.9	1	sunflower oil
1.1	1.1	1.7	1.8	1.8	1.8	2.2	2.2	dicalcium phosphate
8.7	8.6	1.4	1.3	1.3	1.2	1.3	1.3	oster shell
0.3	0.3	0.3	0.3	0.35	0.35	0.4	0.4	Salt
0.25	0.2	0.13	0.1	0.15	0.1	0.17	0.1	DL-Met
0.1	0	0.05	0	0.7	0	0.05	0.05	L- HCl-Lysine
0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	Vitamin premix
0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	Mineral premix
2911	2905	3025	3025	2930	2930	2930	2930	(kcal/kg) ME
16.3	17.8	14.6	16	16.5	18	18.5	20	CP(%)
0.41	0.4	0.36	0.35	0.42	0.4	0.44	0.44	Met(%)
0.82	0.82	0.65	0.65	0.75	0.75	0.81	0.81	Met+Cys(%)
0.95	0.95	0.8	0.8	0.94	0.94	1.1	1.1	Lys(%)
0.69	0.72	0.68	0.75	0.76	0.84	0.86	0.93	Thr(%)
0.86	0.86	0.83	0.83	0.8	0.8	0.74	0.74	TSAA:Lys ratio

MATERIALS AND METHODS

Pullet study: Two hundreds and sixty day-old Hy-line W36 pullets were raised and fed for 17 days according to Hy-line management guide[7]. Following a 6h fasting, chicks were randomly assigned to 26 groups of 10 birds. Each group was a floor pen (1.2*1.5 m²) and considered as a replicate. Thirteen such replicates were randomly assigned to each treatment. There were two dietary protein regimes designated as treatments: a step-down protein series of 20, 18 and 16% as specified by Hy-line W36 management guide, fed during starter (18-42d), grower (42-63d) and developer (64-119d) periods, respectively. This treatment was regarded as the control. The second step-down protein series of 18.5, 16.5 and 14.6% in respective periods mentioned above was considered as reduced- protein treatment. The proportion of total sulfur amino acids: lysine and dietary metabolizable energy content were kept similar between the control and the reduced-protein groups. Dietary composition of the treatments is shown in Table 1.

Feed and water were offered at free access throughout the study. Lighting program was followed recommendations of Hy-line W36 management guide[7]. Records of body weight and feed consumption were recorded weekly during the study (18 to 119d of age). ANOVA procedure of SAS[15] software was used to analyze data of pullet period.

Layer study: The pullets from previous experiment were transferred to an adjacent caged house on 120 days of age. Each cage (40*50cm) accommodated four hens and two adjacent cages were considered as a replicate. Thirteen such double cages were randomly allotted to each treatment group (dietary protein regime). Composition of experimental diets during laying period is presented in Table 2. Dietary specifications and feed allocation were followed as recommended by Hy-line W36 management guide[5]. Feed and water were provided at *ad libitum* access. Sixteen hours light a day was provided throughout the laying period.

Records of egg production and feed consumption were kept daily. Records of egg weight were kept weekly. The results of laying performance have been presented on a monthly basis. Egg quality measures were recorded on two eggs randomly collected from each replicate during the week of peak production. Layer study was started from 18wk of age and continued up to 32wk of age. ANOVA procedure of SAS[15] software was used to analyze data of laying period.

Table 2: Effect of dietary protein regime on body weight during pullet rearing

Body weight(gr)	Control	Reduced -protein	SEM	P- value
18 d	111.3	111.7	0.19	0.202
25 d	168.3	169.7	0.6	0.09
32 d	236.1	239.6	1.33	0.064
42 d	369.7	368.2	0.93	0.278
49 d	463.6	460.9	1.89	0.331
56 d	564.9	561	2.27	0.334
63 d	651.9	646.8	2.78	0.199
70 d	754.2	746.7	3.69	0.165
77 d	850.4	841.8	4.49	0.186
84 d	940.4	933.2	5.64	0.381
91 d	1013.1	1009	6.29	0.646
98 d	1091.5	1086.7	7.14	0.638
105 d	1143.8	1131	9.04	0.326
112 d	1156.7	1145.1	8.65	0.352
119 d	1242.7	1236	8.85	0.595

Feed analysis: Samples taken from feed ingredients were taken to analyze for CP and amino acids. Experimental diets were also analyzed for CP and amino acids. Crude protein was determined according to AOAC[2]. For the determination of amino acids, feed samples were subjected to 6N HCl in duplicate and hydrolyzed for 24h at 110C[1]. Acid hydrolyzed were subsequently analyzed for amino acid contents using an ion exchange chromatography (LKB Biochrom 4141). Performic acid oxidation was done for the determination of sulfur amino acids[12].

RESULTS AND DISCUSSION

Pullet study: There was no significant effect of protein regime on body weight throughout the rearing period (Table 3). Target body weight on 119days of age is 1250gr as recommended by Hy-line management guide[7]. Birds on the control had body weight very close to target weight and those on reduced-protein regime were 14gr lighter. However, the difference was not significant between treatments. The effect of dietary protein regime on body weight gain, feed intake and feed conversion ratio (FCR) are shown in Table 4. The data have been summarized in three distinct feeding phases: starter (18-42d), grower (43-63d) and developer (64-119d). Dietary protein regime had no significant effect on weight gain, feed intake and FCR during grower and developer phases. However, birds on reduced-protein diet during starter phase consumed less feed (p = 0.003) and as a result they had significantly (p = 0.012) better FCR compared to the control.

Table 3: Effect of dietary protein regime on body weight gain, feed intake and feed conversion ratio of pullets in the rearing periods

	control	Low-protein	SEM	p-value
Weight gain (g r)				
18-42 d	258.5	256.8	0.86	0.158
43-63 d	282.2	278.5	2.65	0.329
64-119 d	590.8	589.2	7.09	0.877
Feed intake(g r)				
18-42 d	794.3^a	762.7b	6.74	0.003
43-63 d	1101.8	1064.6	15.33	0.098
64-119 d	3811.8	3703.5	15.67	0.176
Feed conversion ratio				
18-42 d	3.07^a	2.97b	0.026	0.012
43-63 d	3.9	3.83	0.05	0.303
64-119 d	6.45	6.29	0.08	0.152

Table 4: Effect of dietary protein regime on laying hen performance from 18 to 32wk of age

variable	Control	Reduced-CP	SEM	P- value
HDEP* (%)	74.49	74.13	1.32	0.747
Egg mass (g)	8217	8077	119.4	0.12
Egg weight (g)	47.9	47.6	0.7	0.67
Feed conversion ratio	2.49	2.42	0.089	0.469
Albumin height (mm)	4.64	5.25	0.377	0.26
Shell thickness (mm)	0.382	0.393	0.006	0.28
Shell strength (kg m^{-2})	3.39	3.5	0.16	0.62

*Hen-day egg production from week18 through week32

Layer study: Table 4 depicts laying hen performance and egg quality measures for the control and reduced-CP groups. Hen day egg production was comparable between the control and reduced-CP group throughout the laying period (18-32 wk of age). Egg weight was not influenced by feeding the low-protein diet as well. In addition, egg mass and feed conversion ratio were not significantly affected by feeding the reduced-CP diet.

Results of egg quality traits pertaining peak period were also shown in Table 4. There were no significant difference between the control and reduced-CP diet with respect to albumin height, shell thickness and shell strength against breaking.

Pullet study: Significant reduction in feed intake of pullets during the starter phase as a result of feeding reduced-CP diet was consistent with report of Hussein *et al.*,[6]. They reported that pullets fed a reduced-CP diet (16%CP) consumed significantly less feed than the control (19%CP) and as a result they had significantly lower weight gain. Insignificant difference of weight gain between the control and reduced-CP group in the present experiment could account for the lower CP level reduction (1.5 *vs* 3%) as well as similar TSAA: lysine ratio. No attempt was made to keep the same ratio of TSAA: lysine by Hussein *et al.*,[6]. There is another report quite reverse to these observations. Hussein[3] reported that under hot climates, pullets fed a reduced-CP diet (16%CP) consumed significantly more feed than the control (19%CP). It seems, therefore, that the response of pullets to reduced-CP diets depends on the ambient temperature. Few studies have been reported on this subject.

Layer study: Results of this experiment with respect to egg production was in agreement with those reported in previous studies[3,9], indicating egg production can be well maintained on reduced-CP diet.

Egg weight was not influenced by feeding the reduced-CP diet. In contrast, Leeson and Caston[11] reported that egg weight was lower with diets containing 14.4 than 16.8% CP, although both diets had equal levels of methionine and lysine. They attributed the lower egg weight to an inadequate level of total nitrogen. In their study, low-protein diet had 2.4 percentage units CP lower than the control. In the present study, however, the difference between treatments in terms of CP content is 1.5 percentage units. Comparable egg weight between the low-protein and the control groups of the present study suggests that low-protein is well fortified with essential amino acids and have adequate level of total nitrogen. In a study with similar percentage unit reduction in CP level conducted by Roberts *et al*[14] egg weight was not affected by reduced-CP diet but hens fed the reduced-CP diet produced less number of eggs and as a result had lower egg mass compared to their control. This discrepancy with present findings can be accounted for difference in essential amino acid balance between treatments as Roberts *et al* did not maintain similar TSAA: Lys between their treatments. Sohail *et al*.,[16] demonstrated that essential amino acids had significant influence on egg weight so that removing an indispensable amino acid resulted in reduced egg weight within 2 weeks.

Egg mass was not significantly affected by feeding the reduced-CP diet. Egg mass is determined by two components: egg weight and egg production. As egg production and egg weight were not affected by dietary treatment, insignificant results with regard to egg mass would be expected.

Feed conversion ratio was comparable between the control and reduced-CP diet throughout the trial. Wu *et al*.,[17] indicated that hens fed a 16% CP diet consumed less feed compared to those fed diets containing 15.5 or 14.9% CP. Surprisingly, egg mass was similar between these groups.

There were no significant difference between the control and low-protein with respect to albumin height, shell thickness and shell strength against breaking. This is in agreement with the results of Novak *et al*.,[13] who showed that feeding reduced-CP diets to laying hens did not influence shell and internal egg quality measures.

In general, it can be concluded that reduced-protein diets can be satisfactory used for rearing pullets and laying hens up to 32 weeks of age when TSAA: Lysine kept similar to standard norm.

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