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Decreasing Broiler Crude Protein Requirement by Methionine Supplementation

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Abstract: The experiment was conducted with two levels of protein (23 and 20% in starter, 20 and 18% in grower) and three levels of methionine (NRC, 110 of NRC and 120% of NRC in starter and grower stages). No significant differences were found for protein, methionine or their interaction on Feed Intake (FI) and Feed Conversion Ratio (FCR) for starter and grower, with the exception of an interaction of 20% CP and 0.46% (120% of NRC) methionine which significantly improved ($p<0.05$) the FCR compared with 18% CP and 0.38% (NRC) level of methionine, at the grower stage. Carcass yield and breast meat were significantly ($p<0.05$) increased by 23% Crude Protein (CP) compared to 20% in starter. In contrast, abdominal fat was significantly decreased ($p<0.05$) by 23 and 20% CP diet in starter and grower, respectively compared to the other levels of CP as well as by high levels of methionine. In the interaction of 20% CP and high level of methionine (0.46%) on carcass yield, a significant increase ($p<0.05$) was found in grower stage compared with 18% CP and both 0.38% and 0.42% (110% of NRC) of methionine levels. A similar effect was found on breast meat yield compared with interaction of both 0.5% (NRC) and 0.55% (110% of NRC) of methionine and 20% CP at 21 days, as well as at 42 days of age with 0.38% methionine and 18% CP. Finally, the results of this study, have suggested that methionine/CP ratio could be adjusted at 0.024% (0.024 g methionine/per g crude protein) in starter and 0.023% for grower. Decreased CP in the diet, increased breast meat yield and reduction of abdominal fat were achieved by these ratios of methionine/protein.

Key words: Methionine, protein, breast meat, broiler

INTRODUCTION

The carcass composition of broilers is receiving considerable attention, due to industry processing (Garcia Neto *et al.*, 2000). There is an emphasis on increasing the meat yield, particularly breast meat and decreasing the fat content of carcass. An important aspect of the protein and methionine interrelationship is the ability of both to act as lipotropic agents (Hueghebaert *et al.*, 1994). Requirement for methionine can largely be related to existing dietary protein (Baker *et al.*, 1993; Edwin and Moran, 1994; Deschepper and De Grootem, 1995). After a series of experiments in which protein and methionine levels were altered for broilers, Morris *et al.* (1992) have concluded that the amount of methionine need to be optimized since performance was increased by protein and methionine in amounts which were excessive for maximizing growth. However, the reduction in abdominal fat was occurred in response to supplemental methionine and excessive protein diet (Mendonca and Jensen, 1989).

Morris *et al.* (1987) have further shown that the lysine requirement for maximum growth or maximum efficiency of feed utilization continues to increase in direct proportion to the dietary protein concentration even in the range where no response was seen for additional growth. Waldroup *et al.* (1997) found that percentage of breast meat yield in large white toms increased by diets formulated to provide 85 to 120% of NRC amino acid recommendations. Morris *et al.* (1992) have suggested that for the purpose of formulation practical diets, minimum methionine concentration should be specified as a proportion of the protein rather than as a proportion of the diet. They have also indicated that for an estimate of maximum growth or feed efficiency, 0.025 g methionine/g CP could be useful. Kidd *et al.* (1997) have noted that similar trends were found in growth and feed conversion ratio by decreasing CP to 92% of NRC and supplementation of lysine, methionine, threonine and tryptophan (at 105% NRC level) compared with control diet (including reducing CP to 84% and extra threonine). Sell (1993) has found that reduction in breast meat

yield occurred by reducing CP protein diet (to 93% of NRC 1984) and supplementing with lysine and methionine. Body weight and feed conversion ratio were not affected by these diets. Otherwise, in a number of other studies, breast meat yield was not affected by protein level (Sell *et al.*, 1985, 1989 and 1994).

Based on these conflicting results, the purpose of this study was to investigate methionine and protein interrelationships by decreasing CP level during the starter and grower stage and to measure treatment effects on breast meat yield as well as on broiler performance.

MATERIALS AND METHODS

Four hundred, day-old unsexed Ross chickens were used. The birds were grown in floor pens 1.5×2 m in size. Pre-starter period (7 days) was used as a pre-experimentally to achieve uniformity and stable weight in chickens. At 8 day of age, birds were weighed and randomly divided into 18 pens by 23 birds each. Experimental diets were based on corn and soybean meal (Table 1 and 2). The experimental design was a Completely Randomized Design (CRD) with 2x3 factorial arrangement. Two levels of protein: (23 and 20% in starter and 20% and 18% in grower) and three levels of methionine (NRC (0.5%), 110% of NRC (0.55%) and 120% of NRC (0.6% in starter) and (NRC (0.38%) 110% of NRC (0.42%) and 120% of NRC (0.46%) in grower) were optimized as the treatments. Diets were available as a mash, *ad libitum*. Chicken in all treatments were reared under the similar conditions such as drinking water, temperature and lighting regime (the lighting program consisted of 24 h of light during the first 3 days and 23 h of light until slaughter). Chemical composition of ingredients including corn, soybean meal, wheat, fish meal and mixed diet were determined for formulation ration based on AOAC (1990) method (Table 1 and 2).

Amino acids content of diets were determined by HPLC (AOAC, 1990) (Table 3). Samples were digested in 0.6 N hydrochloric acid and proportion of amino acids was indicated by peaks of the graphs. Performic acid oxidation was performed on the samples prior to hydrolysis to allow quantitative analyses of methionine and cystine. The birds were weighed at 8, 21 and 42 days. Feed consumption and feed to gain ratio were measured for the period 8-21 and 22-42 days of age. Two birds randomly selected in each pen, were individually weighed, slaughtered and commercially processed at 21 and 42 days of age. After processing, oven ready carcass was weighed. The breast meat (pectorals major and minor muscles) was excised and weighed carcass yield, breast meat as a fraction of live body weight and abdominal fat

deposition were also measured. Organs were removed, washed and weighed. All data was analyzed by analysis of variance using the GLM procedure of SAS (SAS Institute, 1996). The experiment was approved by the animal care committee of Bu-Ali Sina University in Iran.

Table 1: Ingredient and composition of rations in starter broiler feed with 23 and 20% protein (%)

Ingredients/CP	Starter rations (%)					
	23	23	23	20	20	20
	48.00	48.00	48.00	56.50	56.50	56.50
Soybean meal	35.00	35.00	35.00	29.60	29.60	29.60
Fish meal	5.60	5.60	5.60	4.00	4.00	4.00
Wheat	5.00	5.00	5.00	5.00	5.00	5.00
Vegetable oil	2.80	2.80	2.80	1.60	1.60	1.60
DCP	1.31	1.21	1.21	1.20	1.20	1.20
Oyster shell	1.50	1.55	1.50	1.20	1.20	1.20
Salt	0.22	0.22	0.22	0.27	0.22	0.17
DL-Methionine	0.07	0.12	0.17	0.13	0.18	0.23
Mineral pre mix ¹	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin per mix ²	0.25	0.25	0.25	0.25	0.25	0.25
Calculated composition						
Protein (%)	23.00	23.00	23.00	20.00	20.00	20.00
ME (KJ/Kg)	12.18	12.18	12.18	12.18	12.18	12.18
Calcium (%)	1.00	1.00	1.00	1.00	1.00	1.00
Available						
Phosphorus (%)	0.50	0.50	0.50	0.50	0.50	0.50
Lysine (%)	1.46	1.46	1.46	1.21	1.21	1.21
Methionine (%)	0.50	0.55	0.60	0.50	0.55	0.60
Methionine + Cystine (%)	0.87	0.87	0.87	0.84	0.84	0.84

CP, Crude Protein; 23 and 20. Crude protein levels. 1, Mn 64 g, Zn 44 g, Fe 100 g, Cu 16 g, I 10.64 g and S 8 g 9 (per 1 kg premix; 2, A 7.7 g, D, 7g, E 14.4 g, B₁₂ 14.4 g, K, 1.6 g, B₂ 3.3 g, B₆ 12 g and colin chloride 440 mg (per 1 kg premix)

Table 2: Ingredient and composition of rations in grower broiler feed with 20 and 18% protein (%)

Ingredients/CP	Gower rations (%)					
	20	20	20	18	18	18
	55.00	55.00	55.00	58.50	58.50	58.50
Soybean meal	31.00	31.00	31.00	24.60	24.60	24.60
Fish meal	3.50	3.50	3.50	3.50	3.50	3.50
Wheat	4.30	4.30	4.30	8.00	8.00	8.00
Vegetable oil	3.50	3.50	3.50	2.50	2.50	2.50
DCP	1.00	1.00	1.00	1.10	1.10	1.10
Oyster shell	1.00	1.00	1.00	1.00	1.00	1.00
Salt	0.19	0.15	0.11	0.26	0.22	0.18
DL-Methionine	0.01	0.05	0.09	0.04	0.08	0.12
Mineral pre mix ¹	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin per mix ²	0.25	0.25	0.25	0.25	0.25	0.25
Calculated composition						
Protein (%)	20.00	20.00	20.00	18.00	18.00	18.00
ME (KJ/Kg)	12.97	12.97	12.97	12.97	12.97	12.97
Calcium (%)	0.90	0.90	0.90	0.90	0.90	0.90
Available Phosphorus (%)	0.45	0.45	0.45	0.45	0.45	0.45
Lysine (%)	1.20	1.20	1.20	1.20	1.20	1.20
Methionine (%)	0.38	0.42	0.46	0.38	0.42	0.46
Methionine + Cystine (%)	0.72	0.72	0.72	0.74	0.74	0.74

CP, Crude Protein; 20 and 18. Crude protein levels. 1, Mn 64 g, Zn 44 g, Fe 100 g, Cu 16 g, I 10.64 g and S 8 g 9 (per 1 kg premix; 2, A 7.7 g, D, 7g, E 14.4 g, B₁₂ 14.4 g, K, 1.6 g, B₂ 3.3 g, B₆ 12 g and Colin chloride 440 mg (per 1 kg premix)

Table 3: Protein and amino acid measurement in starter and grower broiler feed (%)

Nutrients	Starter		Grower	
	23% CP	20% CP	20% CP	18% CP
Protein	22.10	19.50	19.70	17.80
Methionine	0.47	0.35	0.35	0.34
Methionine + Cystine	0.89	0.75	0.79	0.69
Lysine	1.86	1.65	1.16	1.04
Tryptophan	0.73	0.71	0.83	0.60
Proline	1.44	1.38	1.65	1.19
Valine	1.23	1.02	1.12	0.84
Glycine	1.13	0.98	1.03	0.78
Serine	1.42	1.23	1.30	0.97
Histidine	0.83	0.79	0.78	0.56
Arginine	1.46	1.33	1.40	1.02
Threonine	0.73	0.71	0.83	0.60
Leucine	1.36	1.20	1.13	1.48
Asparagine	2.17	1.69	2.04	1.27
Alanine	1.15	1.15	1.21	0.90
Tyrosine	0.43	0.33	0.24	0.22
Isoleucine	1.77	1.56	1.48	0.83
Phenyl alanine	1.61	1.25	1.10	0.90

CP, Crude Protein; 23, 20 and 18% are crude protein levels

RESULTS

Mortality was very low, being 0.5% in all treatments and no appreciable differences were found in mortality among the treatments. No significant differences were shown in Feed Intake (FI) and Feed Conversion Ratio (FCR) at 21 and 42 days of age for the different level of protein, methionine and their interactions, with the exception of 20% CP and the high level of methionine (0.46%) on FCR which was significantly ($p < 0.05$) improved compared with 18% CP and 0.38% of methionine at 42 days of age (Table 4). There were no significant differences in Body Weight (BW) and Leg Weight (LW), ($p > 0.05$) among treatments, although a significant decrease in carcass yield ($p < 0.05$) was caused by reducing CP from 23 to 20% at 21 days of age. No response was shown in carcass yield to different levels of methionine. Carcass yield was significantly ($p < 0.05$) increased by interaction between 20% CP and 0.46% of methionine compared with interaction of 18% CP and both 0.38 and 0.42% of methionine at 42 days of age (Table 5).

Breast meat yield was increased significantly ($p < 0.05$) at both 21 and 42 days of age by 23 and 20% of CP, respectively compared to other levels of protein (Table 5). This parameter was dramatically increased ($p < 0.05$) by high level of methionine (0.6%) at 21 days compared to other levels particularly at 42 days of age with 0.46% methionine. Breast meat yield was increased significantly ($p < 0.05$) by interaction of high levels of protein (23 and 20%) and high levels of methionine (0.6 and 0.46%) for 21 and 42 days of age, respectively. A similar trend was observed with the interaction of 18% CP with both 0.42 and 0.46% of methionine at 42 days of age (Table 5). Abdominal fat significantly ($p < 0.05$) declined with high level of CP (23 and 20%) as well as with both 0.6% and 0.46% of methionine compared with 0.5 and 0.38% methionine at 21 and 42 days of age respectively. As can be seen in Table 5, abdominal fat was significantly ($p < 0.05$) reduced with interaction of 23% CP and 0.6% methionine compared with 20%, CP with (0.5 and 0.55% methionine). Similar effects were found in interaction of 20% of CP with 0.46% methionine compared with 18% CP and 0.38 and 0.42% methionine at 42 days of age. No response was shown in organ weight to protein, methionine and their interaction. In contrast,

Table 4: Effect of different levels of protein and methionine on feed intake and feed conversion ratio in broiler feeding

Treatments	FI (g)		FCR	
	21 days	42 days	21 days	42 days
CP	23.0	20.0	789.92	2751.77
	20.0	18.0	790.37	2835.43
Met.	0.50	0.38	736.56	2810.34
	0.55	0.42	806.91	2805.52
	0.60	0.46	797.81	2761.94
CP×Met.	23×0.5	20×0.38	789.92	2762.1
	23×0.55	20×0.42	790.37	2927.8
	23×0.60	20×0.46	736.56	2864.9
	20×0.5	18×0.38	806.91	2858.0
	20×0.55	18×0.42	797.81	2689.3
	20×0.60	18×0.46	766.75	2659.0

CP, Crude Protein; Met. Methionine; CP×Met., interaction of crude protein and methionine; FI, Feed intake; FCR, Feed Conversion Ratio; a, b- means in columns with no common superscript are significantly different ($p < 0.05$)

Table 5: Effect of different levels of protein and methionine on broiler performance

	T (%)		BW		CY		BM		LG		AF	
	21	42	21	42	21	42	21	42	21	42	21	42
CP	23.0	20.0	485.94	2197.4	62.78 ^a	47.28	15.7 ^a	19.82 ^a	15.55	17.88	0.64 ^b	1.67 ^b
	20.0	18.0	479.33	2064.8	60.19 ^b	65.51	14.7 ^b	17.88 ^b	15.33	18.96	1.18 ^a	2.12 ^a
Met.	0.50	0.38	496.83	1976.6	61.64	65.56	14.77 ^b	18.15 ^b	15.29	18.94	1.12 ^a	2.06 ^a
	0.55	0.42	477.00	2032.5	60.68	67.09	14.96 ^{ab}	18.17 ^b	15.48	19.26	0.98 ^{ab}	1.95 ^{ab}
	0.60	0.46	501.08	2163.8	60.94	66.54	15.95 ^a	19.76 ^a	15.55	19.14	0.87 ^b	1.68 ^b
CP×Mt.	23×0.5	20×0.38	489.17	2185.0	60.92	66.32 ^{ab}	15.62 ^{ab}	19.52 ^b	15.09	19.29	0.74 ^{ab}	1.74 ^b
	23×0.55	20×0.42	490.13	2166.2	61.54	66.99 ^{ab}	15.12 ^{ab}	19.42 ^b	15.35	19.08	0.72 ^{ab}	1.64 ^b
	23×0.6	20×0.46	491.64	2161.3	62.88	68.54 ^a	16.25 ^a	20.51 ^a	15.56	19.63	0.53 ^b	1.63 ^b
	20×0.5	18×0.38	464.83	1880.0	60.4	64.81 ^b	14.31 ^b	17.78 ^b	14.49	19.42	1.24 ^a	2.37 ^a
	20×0.55	18×0.42	462.67	1925.0	59.83	64.96 ^b	14.33 ^b	19.36 ^a	15.62	19.8	1.25 ^a	2.27 ^a
	20×0.6	18×0.46	510.50	2028.2	60.95	66.08 ^{ab}	15.64 ^b	18.8 ^{ab}	15.54	19.64	1.04 ^{ab}	1.73 ^b

T, Treatments; BW, Body Weight; CY, Carcass Yield; BM, Breast Meat; LG, Leg Weight; AF, Abdominal Fat; CP, Crude Protein; Met. Methionine. a, b- means in columns with no common superscript are significantly different ($p < 0.05$)

Table 6: Effect of different levels of protein and methionine on whole and different segments of gastrointestinal tract weight (g) of broiler

	(%)		GIT		PV		GZ		LV		PAN	
	21	42	21	42	21	42	21	42	21	42	21	42
CP	23.0	20.0	18.60	9.99	0.79	0.44	2.73	1.73	3.30	2.26	0.49	0.23
	20.0	18.0	18.03	9.37	0.83	0.44	3.00	1.69	3.38	2.29	0.47	0.24
Met.	0.50	0.38	17.59	10.05	0.79	0.47	2.76	1.73	3.33	2.31	0.48	0.22
	0.55	0.42	18.78	9.28	0.85	0.43	2.95	1.73	3.39	2.33	0.50	0.23
	0.60	0.46	17.90	9.71	0.79	0.42	2.96	1.63	3.29	2.19	0.45	0.24
CP×Mt.	23×0.50	20×0.38	17.21	10.16	0.72 ^b	0.46	2.83	1.66	3.27	2.21	0.51	0.21
	23×0.55	20×0.42	18.68	9.45	0.83 ^{ab}	0.43	2.78	1.72	3.25	2.35	0.51	0.25
	23×0.6	20×0.46	18.59	10.36	0.83 ^{ab}	0.43	2.74	1.82	3.37	2.22	0.45	0.22
	20×0.50	18×0.38	17.79	9.94	0.81 ^{ab}	0.47	2.69	1.80	3.39	2.41	0.46	0.23
	20×0.55	18×0.42	18.88	9.80	0.89 ^a	0.43	0.12	1.73	3.54	2.30	0.48	0.23
	20×0.6	18×0.46	17.07	9.05	0.79 ^{ab}	0.41	3.19	1.55	3.20	2.16	0.46	0.25

GIT, Gastrointestinal Tract; PV, Proventriculus; GZ, Gizzard; LV, Liver weight; PAN, Pancreas. Met, Methionine; a, b - means in columns with no common superscript are significantly different (p<0.05)

proventriculus weight was significantly (p<0.05) higher with interaction of 20% CP and 0.55% of methionine compared with 23% CP and 0.50% methionine at 21 days of age (Table 6).

DISCUSSION

Methionine and protein interrelationship may play major roles in poultry nutrition (Ohta, 1999). Methionine consideration has shown that it is possible to be adjusted as a part of CP diets instead of the whole diet (Morris, 2004). In low CP diets, methionine improved feed conversion ratio as shown by the results of the current study. Summers *et al.* (1988) also noted this point. Results of the present study have shown that decreased CP was compensated by methionine addition at 120% of NRC (0.6% and 0.46% at 21 and 42 days of age, respectively) which was supported by Schutte and Pack (1995b). Present results suggest that NRC (1994) methionine recommendation at (0.5%) at 21 days and 0.38% at 42 days for obtaining high breast meat yield is low. Graber and Baker (1971) also reported similar results. Methionine in this case could be offered at 120% of NRC (0.6 and 0.46% for 21 and 42 days of age respectively). If methionine was added at 120% of NRC (0.6 and 0.46%), CP diet could be reduced to 20% (3% less than NRC recommendation in starter) and 18% (2% less than NRC offered in grower). These diets are economic and approach desirable performance, in addition there is decreased nitrogen excreta. This observation is in agreement with Scutte and Pack (1995a) and Abebe and Morris (1990) for 21 days of age and with Fanher and Jensen (1989) at growing stage. Breast meat yield is affected by diet methionine rather than diet CP, since 3% reduction of diet CP was compensated by only 0.1% of methionine (120% of NRC at 21 day of age) and 2% reduction (from 20 to 18% CP) by only 0.08 of methionine addition at 42 days of age. Waible *et al.* (1995) found similar results. NRC methionine recommendations (0.5 and 0.38%, at 21 and 42 days of age) are sufficient for obtaining satisfactory weight gain,

carcass yield and desirable FCR, but are inadequate for obtaining high breast meat yield and low abdominal fat. Current results have shown that methionine requirement for increasing carcass quality was higher than the requirement for obtaining high growth and improved FCR. These results were in agreement with the observations of Scutte and Pack (1995a) and Bedford and Summers (1985) but not with Esteven-Garcia and Laurado (1997). Since their experiment was carried out by male broiler, in contrast, current study was conducted by unsexed broiler chickens. Methionine requirement for obtaining high breast meat yield and low abdominal fat was 0.1% and 0.08% in excess of the requirement for weight gain and carcass yield at 21 and 42 days of age, respectively. These results were supported by Hickling *et al.* (1990). Total digestive system, liver, pancreas and gizzard weight were not affected by CP or by dietary methionine. This may relate to independent effects on these organs by CP diet and methionine supplementation. These achievements are inconsistent with Garcia Neto *et al.* (2000). Proventriculus weight was increased by 20 CP and 110% of NRC (0.55%) methionine level at 21 days of age, this is a predominantly a response of proventriculus to methionine level. With supplementation of methionine it is possible to reduce dietary CP, this is one of the most important ways to reduce feed expenses. Also with reduction of diet CP, environmental pollution is reduced because of a decrease nitrogen excretion (Ishibashi and Yonemochi, 2002). Morris *et al.* (1992) have shown that 0.025 is suitable for methionine/CP ratio. In conclusion, the results of this experiment suggest that dietary methionine could be adjusted as a proportion of diet CP instead of whole diet. These results have suggested that methionine/CP ratio can be adjusted at 0.024% (0.024 g methionine/per g crude protein) and 0.023% at 21 and 42 days of age, respectively. These ratios could lead to obtaining higher breast meat yield, lower abdominal fat, desirable FCR and declining CP requirement in the diet. Therefore these findings could lead to financial advantages in the poultry industry.

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