# Effects of Additives on Laying Performance, Metabolic Profile, and Egg Quality of Hens Fed a High Level of Sorghum (Sorghum vulgare) during the Peak Laying Period

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**ABSTRACT**: This experiment was conducted to determine the effects of supplemental methionine, lysine, choline, and sulfur on laying performance, metabolic parameters, and egg quality of hens fed diets containing sorghum (Sorghum vulgare) during the peak laying period. Lohman layers (n = 144), 30-wk of age as 6 replicate cages of 4 hens, were allocated randomly to receive basal diets containing either 22% corn (B) or 22% sorghum (BS) and diets BS plus 0.57% methionine, 0.66% lysine, 0.47% choline, or 0.05% sulfur for 98 d. Feed intake (FI) and egg production (EP) were recorded daily, egg weight (EW) was measured bi-weekly, and body weight (BW) was measured monthly. A sample of 12 eggs from each experimental group was collected every month to evaluate egg quality. At the end of the experiment, blood samples were collected for metabolite concentrations. Data were analyzed using one-way ANOVA as repeated measures and significant differences between the experimental groups were assessed using Duncan's Multiple Range test. Partial replacement of corn with sorghum in the basal diet did not affect BW, EP, and FCR but increased FI by 5.7% and EW by 2.4%. The effects of additives on laying performance were variable. Except for serum total protein (STP) concentration, other metabolic parameters were not affected by partial replacement of corn with sorghum in the basal diet. Hens fed diet BS had lower SPT concentration than hens fed diet B. Except for methionine supplementation, other supplements ameliorated depression in STP concentration. The additives did not affect other metabolic parameters. Egg quality responses to the experimental diets were also variable. Partial replacement of corn with sorghum in the basal diet did not affect eggshell characteristics (both thickness and stiffness), whereas it had variable effects on inner egg quality parameters (increased yolk index, depressed yolk color, and unaltered albumen index and Haugh unit). In conclusion, laying hen diets could include low-tannin sorghum (0.26%) up to 22% without necessitating extra supplements to overcome compromised performance. (Asian-Aust. J. Anim. Sci. 2006. Vol 19, No. 4: 573-581)

Key Words: Laying Hen, Sorghum, Methionine, Lysine, Choline, Sulfur

## INTRODUCTION

Sorghum (Sorghum vulgare) is well adapted to arid and semi-arid regions and could be used as corn substitute due to its energy content in poultry feeding. Sorghum ranks the fifth most produced crop worldwide after wheat, corn, rice, and barley (NRC, 1996). The chemical composition of sorghum is similar to corn, except for being deficient in lysine, methionine, and threonine (Sikka and Johari, 1979). However, due to presence of tannin predominantly in bran, its usage is limited (Nelson et al., 1975). Tannins are divided into two groups. The condensed tannins consist of

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flavan-3-ols (oligoflavanols made up catechin), also called proanthocyanidins, and predominantly are present in leaves and roughages. The hydrolysable tannins are polyesters of phenolic acids such as gallic acid, hexahydrocydipenic acid, and quinic acid and present predominantly in cereal grains. Presence of these compounds between 1 and 5% depresses performance. Growth-inhibiting effect of gallic acid is reported to be one-third of tannic acid (Harvey and McAllan, 1992). Excessive tannin consumption results in depressed growth rate, reduced feed intake, poor feed efficiency, decreased nutrient digestibility, compromised bone development (Nyachoti et al., 1997). Tannins bind protein, nucleic acids, polysaccharides, and steroids. They presumably impair digestion and absorption of nutrients (Lasheras et al., 1980a, b), alter nutrient partitioning and tissue composition (Cherian et al., 2002; Du et al., 2002), and cause cell degeneration in the liver and kidneys (Harvey and McAllan, 1992). Maximum tannin tolerance level is reported to be 9, 3, and 1% for goats, cows, and chickens, respectively (Begovic et al., 1978).

Numerous articles have focused on improving nutritional quality of high-tannin cereal grains. Physical (Teeter et al., 1986; Elkin et al., 1990) and chemical (Mitaru

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Table 1. Ingredients and amino acid profiles of the experimental diets

Item			Experime	ental diets <sup>1</sup>		
	В	BS	BSM	BSL	BSC	BSS
Corn	53.00	31.00	31.00	31.00	31.00	31.00
Sorghum	-	22.00	22.00	22.00	22.00	22.00
Soybean meal	16.00	16.00	16.00	16.00	16.00	16.00
Wheat	6.00	5.50	5.50	5.50	5.50	5.50
Barley	2.50	2.50	2.50	2.50	2.50	2.50
Wheat bran	10.30	11.00	10.43	10.34	10.53	10.95
Molasses	1.80	1.60	1.60	1.60	1.60	1.60
Limestone	8.00	8.00	8.00	8.00	8.00	8.00
Salt	0.30	0.30	0.30	0.30	0.30	0.30
Dicalcium phosphate <sup>2</sup>	1.50	1.50	1.50	1.50	1.50	1.50
Vitamin-mineral premix <sup>3</sup>	0.30	0.30	0.30	0.30	0.30	0.30
Antioxidant <sup>4</sup>	0.10	0.10	0.10	0.10	0.10	0.10
Methionine	0.10	0.10	0.67	0.10	0.10	0.10
Lysine	0.10	0.10	0.10	0.76	0.10	0.10
Choline	-	-	-	-	0.47	-
Sulfur	-	-	-	-	-	0.05
Amino acid profie <sup>5</sup>						
Methionine (%)	0.36	0.38	0.95	0.38	0.38	0.38
Lysine (%)	0.80	0.80	0.80	1.46	0.80	0.80
Choline (g/kg)	0.90	0.89	0.88	0.88	1.35	0.89
Sulfur (%)	0.16	0.16	0.16	0.16	0.16	0.21

Diets: B = basal diets containing additional 22% corn; BS = basal diet containing 22% sorghum; BSM = diet BS plus 0.57% methionine as DL-methionine; BSL = diet BS plus 0.66% lysine as L-lysine monohydrochloride; BSC = diet BS plus 0.47% choline as choline chloride; BSS = diet BS plus 0.05% sulfur as inorganic sulfur.

et al., 1983) treatments and dietary nutrient fortification (Nyachoti et al., 1997; Kumarl et al., 2005) have been employed to reduce extractable tannin content and improve sorghum utilization. Sell et al. (1983) increased dietary protein level and Pour-Reza and Edriss (1997) increased dietary energy content to support compromised nutrient utilization due to high-tannin sorghum. Both nutrition partially ameliorated regimes laving performance. Results of biotechnological products to detoxify tannin are inconclusive. Feed additives such as polyethylene glycol (Ford and Heweitt, 1977, 1979) succeeded to enhance availability of methionine and lysine, whereas polyvinylpyrrolidone (Ambula et al., 2001) and bentonite (Ambula et al., 2003) supplementations failed to bind tannin. It was hypothesized that supplementation of methionine would ameliorate the adverse effects of tannin in sorghum through either being an essential nutrient, playing a role in methyl donor metabolism, containing sulfur, or all of them (Jurgens, 1996). The objective of this experiment was therefore to evaluate the effects of methionine, lysine, choline, or sulfur supplementations on laying performance and egg quality of peak producing hens fed diet containing a high level of sorghum.

#### **MATERIALS AND METHODS**

## Animal, diet and management

The Research Animal Ethic Committee of Atatürk University approved this experimental protocol. Lohman layers (n = 144), 30-wk of age with uniformity of 91% (the number of hens weighing between 0.9-1.1% of the mean body weight), were selected from the University Research Farm. Hens were blocked according to the location of cages (50×46×46 cm) and then assigned randomly to receive one of six isolocaloric and isonitrogenous experimental diets: basal diets containing additional 22% corn (B) or 22% sorghum (BS) and diets BS plus 0.57% methionine as DLmethionine (BSM), 0.66% lysine as L-lysine monohydrochloride (BSL), 0.47% choline as choline chloride (BSC), or 0.05% sulfur as inorganic sulfur (BSS) for 14 wks. Each diet was offered to 6 cages, each containing 4 hens.

The experimental diets (Table 1) were formulated to meet or exceed the NRC recommendations (NRC, 1994). The experimental diets contained an average of 89.9% DM and 2,573±12 kcal/kg apparent ME, 15.3±0.7% CP, 2.6±1.2% crude fiber, 3.1±2.3% ether extract, 10.3±1.1% ash, 2.15±0.08% Ca, and 0.56±0.01% P (mean±SD).

<sup>&</sup>lt;sup>2</sup> Per kg contains: Ca, 24% and P, 17.5%.

<sup>&</sup>lt;sup>3</sup> Per kg contains: Vitamin A, 15.000 IU; cholecalciferol, 1,500 ICU, vitamin E (DL-α-tocopheryl acetate), 30 IU; menadione, 5.0 mg; thiamine, 3.0 mg; riboflavin, 6.0 mg; niacin, 20.0 mg; panthotenic acid, 8.0 mg; pyridoxine, 5.0 mg; folic acid, 1.0 mg; vitamin B<sub>12</sub>, 15 mcg; Mn, 80.0 mg; Zn, 60.0 mg; Fe, 30.0 mg; Cu, 5.0 mg; I, 2.0 mg; and Se, 0.15 mg.

<sup>&</sup>lt;sup>4</sup> Per kg contains: Ethoxyquin, 50 g. <sup>5</sup> Calculated values (Jurgens, 1996).

Table 2. The effects of supplemental methionine, lysine, choline, and sulfur on laying performance of hens fed sorghum during the peak production period

Experimental diets<sup>1</sup>

SEM

Parameter	Experimental diets <sup>1</sup>						
	В	BS	BSM	BSL	BSC	BSS	SEM
Initial BW (kg)	1.60	1.60	1.59	1.60	1.66	1.60	0.05
Final BW (kg)	1.57	1.57	1.56	1.58	1.62	1.58	0.04
BW change (%) <sup>3</sup>	-2.20	-2.58	-2.02	-1.01	-2.37	-1.30	1.64
Feed intake (g) <sup>4,5</sup>	128.8°	136.2 <sup>b</sup>	138.5 <sup>b</sup>	142.2 <sup>a</sup>	145.8 <sup>a</sup>	142.7 <sup>a</sup>	1.3
Egg production (%) <sup>4</sup>	92.3	92.4	93.0	91.3	90.8	91.5	0.9
Defective egg (%) <sup>6</sup>	0.20	0.18	0.18	0.36	0.14	0.44	0.15
Egg weight (g) <sup>4</sup>	63.2 <sup>b</sup>	64.7 <sup>a</sup>	61.2 <sup>c</sup>	64.1 <sup>ab</sup>	$63.7^{b}$	63.5 <sup>b</sup>	0.3
Feed:egg ratio (kg:kg) <sup>4</sup>	2.29 <sup>a</sup>	$2.30^{a}$	$2.46^{b}$	$2.48^{b}$	2.55 <sup>b</sup>	$2.50^{\rm b}$	0.05

The least square means. Different superscripts within the same rows differ (p<0.05). Diets: B = basal diets containing additional 22% corn; BS = basal diet containing 22% sorghum; BSM = diet BS plus 0.57% methionine as DL-methionine; BSL = diet BS plus 0.66% lysine as L-lysine monohydrochloride; BSC = diet BS plus 0.47% choline as choline chloride; BSS = diet BS plus 0.05% sulfur as inorganic sulfur.

During the experimental period, feed and water were provided *ad libitum* and eggs were collected once daily at 08:30 h. Hens were subjected to a 17L:7D cycle.

#### Sample collection and analytical procedure

Feed samples were collected monthly and analyzed for DM, CP, crude fiber, and ash (AOAC, 1990). Metabolizable energy, Ca, and P contents and amino acid profile of the experimental diets were calculated from tabular values of feedstuffs for chickens reported by Jurgens (1996). Tannin content was measured using Folin-Denis solutions (AOAC, 1990). Feed intake and egg production were recorded daily, egg weight was measured bi-weekly, and body weight was measured monthly. Before determination of egg weight, a sample of 12 eggs was stored for 24 h in room temperature. Feed conversion ratio (FCR) was expressed as kilogram of feed consumed per kilogram of egg produced.

Another sample of 12 eggs was randomly collected from each experimental group every month to assess egg quality parameters as described by Ergün et al. (1987) using the following formulas: shape index (%) = ((egg width (cm)/egg length (cm))× 100; shell strength (kg/cm·cm) was determined by using machine with "spiral pressure system"; shell thickness (mm) was determined in 3 different parts (upper and lower ends and middle) by using "micrometer"; albumen index (%) = ((albumen height (mm)/average of albumen length (mm) and albumen width (mm))×100; yolk index (%) = ((yolk height (mm)/yolk diameter (mm))×100; yolk color was determined by using commercially available "yolk color fan" according to the CIE standard colorimetric system; Haugh unit =  $100 \times \log (H+7.57-1.7 \times W^{0.37})$ , where H = albumen height (mm) and W = egg weight (g).

At the end of the experiment, blood samples were collected from brachial vein for analyses of serum glucose, creatine, total protein, albumin, triglyceride, cholesterol, VLDL, Ca, and P by spectrophotometric method using

commercial kits (DDS, Diasis Diagnostic Systems Co., İstanbul, Turkey) in auto-analyzer (Hitachi, Boehringer-Mannheim 717, Germany).

#### **Statistics**

In a randomized block design experiment, the location of cages was considered as a blocking factor. Feed intake and egg production data were reduced to bi-weekly means before statistical analyses. Data were subjected to one-way ANOVA using the Mixed Procedure as repeated measures with time being a subplot (SAS, 1998). The linear model to test the effects of the experimental diets on laying performance and egg quality parameters was as follows:

$$Y_{ijk} = \mu + B_i + TRT_j + T_k + Error A + (TRT*t)_{jk} + Error B$$

where  $Y_{ijk}$  = response variable,  $\mu$  = population mean,  $B_i$  = block (i = 1 cage at lower level by corridor side to 6 cage at upper level by window side),  $TRT_j$  = experimental diet (j = diet B to diet BSS),  $T_k$  = time (k = wk relative to initiation of the experiment), error A = whole-plot error,  $(TRT \times t)_{jk}$  = experimental diet j and time k interaction, and error B = subplot error. Time and experimental diet by time interaction were omitted from this linear model to evaluate the effect of the experimental diets on metabolic profile parameters because blood samples were collected only at the end of the experiment.

In this experiment, diet B served as positive control, whereas diet BS served as negative control for determining the effects of methionine, lysine, choline, and sulfur supplementations on response variables. Significant differences (p<0.05) among the experimental groups were attained using Duncan's Multiple Range test. The experimental diet by time interaction term was dropped from the linear model when its probability of significance was greater than 0.25.

<sup>&</sup>lt;sup>3</sup> Relative change = (final body weight-initial body weight)×100/initial body weight.

<sup>&</sup>lt;sup>4</sup> Time effect (p<0.0001). <sup>5</sup> Experimental diet by time interaction effect (p<0.0001). <sup>6</sup> Time effect (p<0.02).

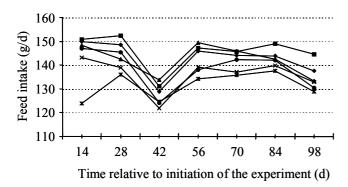


Figure 1. The effects of supplemental methionine, lysine, choline, and sulfur on feed intake of hens fed sorghum during the peak production period. Hens fed basal diets containing additional 22% corn (--\*--) or 22% sorghum (--×--) and diets containing sorghum plus supplemental methionine (0.57%) as DL-methionine (--•-), lysine (0.66%) as L-lysine monohydrochloride (--•-), choline (0.47%) as choline chloride (--•-), and sulfur (0.05%) as inorganic sulfur (--•-). Probability for the effect of experimental diet by time interaction was less than 0.0001, with SEM of 3.5.

# **RESULTS**

# Laying performance

Table 2 summarizes the effects of the experimental diets on laying performance parameters. Initial and final body weights of laying hens did not differ across the  $(1.61\pm0.05)$ experimental diets and  $1.58\pm0.04$ kg, respectively [mean±SE]). All hens lost weight an average of 1.91% relative to initial body weight, which was also independent from the experimental diets. replacement of corn with sorghum in the basal diet caused 5.7% increase in feed intake. Moreover, except for methionine, other supplements increased feed intake further (5.4%). Feed intake changed as the experiment continued (p<0.0001); the mean feed intake was 140.7, 144.0, 127.3, 142.3, 141.8, 142.5, and 134.6 g/d on wk 2, 4, 6, 8, 10, 12, and 14 relative to initiation of the experiment, respectively. Moreover, feed intake response was in different fashion depending upon the type of supplementation during the experimental period (p<0.0001; Figure 1).

The experimental diets affected neither hen-day egg production (91.9±0.9%) nor defective egg percentage (0.25±0.15). However, both parameters varied as the experiment continued; the mean values were 79.9, 94.6, 93.6, 94.0, 95.8, 87.8, and 97.4% for hen-day egg production (p<0.0001) and 0.08, 0.10, 0.11, 0.16, 0.16, 0.66, and 0.57 for defective egg percentage (p<0.02) on wk 2, 4, 6, 8, 10, 12 and 14 relative to initiation of the experiment, respectively. Eggs for hens fed diet BS were 2.4% heavier than for hens fed diet B. Only lysine supplementation maintained this increase in egg weight. Choline and sulfur supplementations did not affect egg weight, whereas methionine supplementation decreased egg weight by 3.4% as compared with diet B. Egg weight also changed as the experiment continued (p<0.0001); the mean egg weight was 61.4, 62.8, 62.9, 64.3, 64.3, 63.6, and 64.3 g on wk 2, 4, 6, 8, 10, 12, and 14 relative to initiation of the experiment, respectively. Partial replacement of corn with sorghum in the basal diet did not alter FCR. All of the supplements, however, increased FCR by an average of 8.8%. As the experiment continued, FCR varied (p<0.0001); the mean FCR was 3.02, 2.43, 2.17, 2.36, 2.31, 2.57, and 2.16 on wk 2, 4, 6, 8, 10, 12, and 14 relative to initiation of the experiment, respectively.

### Metabolic parameters

Serum glucose (244.3±12.7), creatine (0.36±0.06), albumin (1.80±0.11), triglyceride (730.9±70.4), cholesterol (136.3±20.2), VLDL (146.1±14.1), and Ca (15.0±0.6) concentrations (mg/dL, mean±SE) were not different across the experimental diets (Table 3). Partial replacement of corn with sorghum in the basal diet decreased serum total protein concentration by 11.5%. Except for methionine, other supplements ameliorated depression in serum protein concentration. Serum P concentrations of hens fed diets B

**Table 3.** The effects of supplemental methionine, lysine, choline, and sulfur on metabolic profile of hens fed sorghum during the peak production period

Parameter <sup>2</sup> -		Experimental diets <sup>1</sup>						
	В	BS	BSM	BSL	BSC	BSS	SEM	
Glucose	249.3	239.7	235.5	242.7	249.7	248.7	12.7	
Creatine	0.38	0.37	0.37	0.27	0.40	0.38	0.06	
Total protein	5.80 <sup>a</sup>	$5.20^{b}$	$4.60^{b}$	5.35 <sup>ab</sup>	5.32 <sup>ab</sup>	5.50 <sup>ab</sup>	0.28	
Albumin	1.80	1.83	1.64	1.80	1.87	1.85	0.11	
Triglyceride	766.8	639.5	758.2	697.0	726.0	797.8	70.4	
Cholesterol	157.3	122.2	123.8	158.5	112.5	143.7	20.2	
VLDL	153.2	127.7	151.7	139.3	145.2	159.5	14.1	
Ca	15.4	14.5	14.7	15.3	14.7	15.4	0.6	
P	5.78 <sup>b</sup>	5.88 <sup>b</sup>	$4.80^{b}$	$6.05^{ab}$	5.87 <sup>b</sup>	7.25 <sup>a</sup>	0.61	

The least square means. Different superscripts within the same rows differ (p<0.05). Diets: B = basal diets containing additional 22% corn; BS = basal diet containing 22% sorghum; BSM = diet BS plus 0.57% methionine as DL-methionine; BSL = diet BS plus 0.60% lysine as L-lysine monohydrochloride; BSC = diet BS plus 0.47% choline as choline chloride; BSS = diet BS plus 0.05% sulfur as inorganic sulfur.

<sup>&</sup>lt;sup>2</sup> Unit for metabolic parameters is mg per deciliter. VLDL = very low-density lipoprotein.

Table 4. The effects of supplemental methionine, lysine, choline, and sulfur on egg quality of hens fed sorghum during the peak production period

Parameter		SEM					
	В	BS	BSM	BSL	BSC	BSS	SEM
Shape index (%) <sup>2</sup>	74.5	75.4	74.7	74.4	75.7	75.1	0.5
Shell stiffness (kg/cm <sup>2</sup> ) <sup>3</sup>	2.06	2.23	2.11	1.84	1.92	1.97	0.13
Shell thickness (mm×10 <sup>-2</sup> ) <sup>4</sup>	1.153 <sup>ab</sup>	1.166 <sup>a</sup>	1.165 <sup>a</sup>	1.114 <sup>c</sup>	$1.100^{c}$	1.124 <sup>bc</sup>	0.019
Yolk color <sup>5</sup>	9.83 <sup>a</sup>	8.78°	8.72°	9.36 <sup>b</sup>	8.96 <sup>bc</sup>	8.72°	0.20
Yolk index (%)	41.6 <sup>b</sup>	44.1 <sup>a</sup>	43.5 <sup>ab</sup>	$41.7^{b}$	43.5 <sup>ab</sup>	44.3 <sup>a</sup>	0.5
Albumen index (%) <sup>6</sup>	9.42 <sup>bc</sup>	$9.89^{ab}$	9.82 <sup>ab</sup>	9.88 <sup>ab</sup>	10.25 <sup>a</sup>	10.32 <sup>a</sup>	0.35
Haugh unit <sup>4</sup>	85.2	86.9	87.2	87.4	88.1	88.0	1.3

The least square means. Different superscripts within the same rows differ (p<0.05). Diets: B = basal diets containing additional 22% corn; BS = basal diet containing 22% sorghum; BSM = diet BS plus 0.57% methionine as DL-methionine; BSL = diet BS plus 0.66% lysine as L-lysine monohydrochloride; BSC = diet BS plus 0.47% choline as choline chloride; BSS = diet BS plus 0.05% sulfur as inorganic sulfur.

<sup>&</sup>lt;sup>2</sup> Experimental diet by time interaction effect (p<0.02). <sup>3</sup> Time effect (p<0.002). <sup>4</sup> Time effect (p<0.03). <sup>5</sup> Time effect (p<0.0001). <sup>6</sup> Time effect (p<0.02).

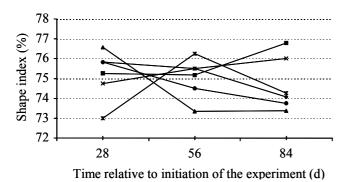


Figure 2. The effects of supplemental methionine, lysine, choline, and sulfur on egg shape index of hens fed sorghum during the peak production period. Hens fed basal diets containing additional 22% corn (--\*--) or 22% sorghum (--×--) and diets containing sorghum plus supplemental methionine (0.57%) as DL-methionine (--●--), lysine (0.66%) as L-lysine monohydrochloride (--▲--), choline (0.47%) as choline chloride (--■--), and sulfur (0.05%) as inorganic sulfur (--◆--). Probability for the effect of experimental diet by time interaction was 0.02, with SEM of 0.8.

and BS were not different. Lysine and sulfur supplementations increased serum P concentration, whereas methionine and choline supplementations did not affect serum P concentration.

# Egg quality

The effects of experimental diets on egg quality parameters are summarized in Table 4. Shape index (75.0±0.5%), eggshell stiffness (2.02±0.13 cm²), and Haugh unit (87.1±1.3) were independent from the experimental diets (mean±SE). Eggshell stiffness and Haugh unit, however, varied as the experiment continued and their mean values were 2.24, 2.05 and 1.78 kg/cm² (p<0.002) and 88.8, 87.3, and 85.3 (p<0.03) on mo 1, 2, and 3 relative to initiation of the experiment, respectively. During the experimental period, shape index continuously increased for hens fed diet BS and continuously decreased for hens fed diets BSM, BSL, and

BSS (p<0.02; Figure 2). Partial replacement of corn with sorghum in the basal diet did not affect eggshell thickness. Except for methionine supplementation, other supplements decreased eggshell thickness by 4.8% as compared with diet BS. As the experiment continued, eggshell thickness quadratically increased (p<0.03); the mean eggshell thickness was 1.139, 1.163, 1.109 mm×10<sup>-2</sup> on mo 1, 2, and 3 relative to initiation of the experiment, respectively.

Yolk color was depressed by 11.9% due to inclusion of sorghum into the basal diet. Lysine and choline supplementations partially alleviated depression in yolk color, whereas methionine and sulfur supplementations failed to alleviate depression in yolk color. As the experiment continued, yolk color linearly decreased (p<0.0001); the mean yolk color was 9.9, 9.1, and 8.2 on mo 1, 2, and 3 relative to initiation of the experiment, respectively. Partial replacement of corn with sorghum in the basal diet increased yolk index by 6.0%. Except for lysine supplementation, other supplements maintained this increase in yolk index. As the experiment continued, yolk index did not change. Albumen indexes for hens fed diets B and BS were not different. As compared with diet BS, choline and sulfur supplementations increased albumen index, whereas methionine and lysine supplementations did not alter albumen index. Change in albumen index was a linear fashion (p<0.02), as the experiment continued. The mean albumen index was 10.4, 10.1, and 9.3% 9.9, 9.1, and 8.2 on mo 1, 2, and 3 relative to initiation of the experiment, respectively.

# **GENERAL DISCUSSION**

Because of its high soluble carbohydrate content, sorghum can be used alternative to corn (Sauer et al., 1978) if its nutritive value is improved or adverse effects of tannins are eliminated (Dale et al., 1980; Nyachoti et al., 1997; Kumarl et al., 2005). The effects of sorghum on growth and laying performance vary highly depending upon

the variety, feeding level, processing method, and poultry species because of great variation in tannin contents (Firdous and Gilani, 2001; Kim et al., 2000; Singh et al., 2003). Also, reports on minimum level of tannin without adversely affecting performance are variable. After physical treatment, tannin level was reduced from 2.3 to 1.6% without causing toxicity when to feed broilers up to 33% (Kumarl et al., 2005). Despite a lack of difference in mortality rate of chicks, feeding high-tannin sorghum with 1.36% catechin equivalent had significantly depressed feed intake and daily gain and increased feed conversion as compared with feeding low-tannin sorghum with 0.28% catechin equivalent (Hassan et al., 2003). After extraction of tannin, increased protein digestibility of high-tannin sorghum may suggest that low tannin can be tolerated (Armstrong et al., 1974). Based on regressing tannin level on apparent nutrient absorption rate, chicks are estimated to tolerate low-tannin (2.6 g/kg). Armonious et al. (1973) reported that 1% tannin level did not adversely affect laying performance in hens. Moreover, depending upon dietary protein content, sorghum with catechin equivalent of 2.7% at 20% CP and of 3.2% at 23%CP can be substitute corn (Ambula et al., 2001). However, Potter et al. (1967) reported decreased egg production in response to additions of 2 and 4% tannic acid. In the present experiment, replacing corn with sorghum did not affect body weight, egg production, and FCR but increased feed intake and egg weight (Table 2 and Figure 1), which could be related to low-tannin content (0.26%), agreeing with other studies in which low-tannin sorghum (0.33%) was fed (Armstrong et al., 1973a; Nyachoti et al., 1997).

The effects of additives on laying performance are inconsistent, and this could be related to amino acid imbalance. Sohail et al. (2002) also supplemented laying hens fed lysine deficient-diet with 0.65% lysine and reported increases in feed intake and egg weight. In a similar study, supplementation of various levels of methionine was shown to increase egg production, egg weight, and feed intake and decrease feed efficiency (Harms and Miles, 1988). Although 0.1% choline supplementation did not affect growth performance during the growing period, continuing supplementation during the laying period increased egg production and egg size in chickens, suggesting that choline requirement may increase when diet is methionine deficient (Tsiagbe et al., 1982).

Tannin interferes with protein metabolism, compromises starch digestibility (Streeter et al., 1990), and activities of pancreatic and intestinal enzymes (Lizardo et al., 1995), and consequently may adversely affect metabolic profile. Thus, increasing nutrient density and dietary fortification with amino acids may eliminate adverse effects of tannin. However, replacing corn with sorghum and including

additives did not affect metabolic profile in the present study (Table 3).

Partial replacement of corn with sorghum (22%) had no adverse effect on egg quality parameters, except for yolk discoloration and shape index (Table 4 and Figure 2). This could be related to low xantrophyll content in sorghum, not low tannin (Potter et al., 1967) because adding 40% hightannin sorghum plus xantrophyll did not result in yolk discoloration (Fry et al., 1972). Reddy and Rao (2000) summarized numerous research articles about feeding sorghum with various tannin contents. In these studies, sorghum was substituted corn up to 60% in layers and broilers. In layers, inclusion of sorghum up to 60% decreased egg weight, eggshell thickness, and yolk color. Inconsistent effects feed additives on egg quality parameters in the present study are ambiguous. Prochaska et al. (1996) investigated egg quality responses to increasing lysine supplementation in hens fed sorghum-soybean based diet and reported that increasing lysine intake from 677 to 1.613 mg per hen increased albumen weight and egg weight.

#### SPECIFIC DISCUSSION

Sorghum is deficient in lysine and methionine as compared with corn (Sikka and Johari, 1979). Moreover, availability of methionine and lysine in sorghum depends on its tannin content (Ford, 1977), suggesting that either feeding sorghum may increase demand for methionine and lysine or their supplementation could alleviate the adverse effects of tannin. Sohail et al. (2002) showed that absence of sulfur amino acids in the diet caused depression in body weight, feed intake, and egg production; inclusion of sulfur amino acids alleviated the depression within 1 week, suggesting that adverse effects of sorghum could be linked to low methionine and cysteine levels (Sikka and Johari, 1979). Methionine, via S-adenosylmethionine, alleviates the adverse effects of tannins by acting a methyl donor for hydrolysis of tannins into gallic acid, which is excreted via urine as 4-O-methyl gallic acid (Potter and Fuller, 1968). Thus, methionine supplementation may be necessary for 0methylation (Armonious et al., 1973).

Armstrong et al. (1973) fed laying hens 1 and 2% tannic acid. Both levels decreased egg production, egg weight, and body weight and increased egg mottling. Methionine (0.24%)alleviated supplementation partially parameters. In following experiment, addition methionine (0.4%) to diet containing 50% sorghum with contributing an average of 0.4% tannin was investigated. Due to tannin, sorghum depressed laying parameters and methionine ameliorated these parameters. In a 2x2 factorial arrangement, Sell and Rogler (1984) added 0.2% methionine or glutamic acid to diets containing low- and high-tannin sorghum for Leghorn layers for a period of 44 days. In both types of sorghum, addition of methionine increased egg production, egg weight, feed intake and decreased body weight loss, which were more pronounced for high-tannin sorghum than for low-tannin sorghum. In chicks fed diet containing 70% sorghum, supplementation of methionine alone (0.31%) or choline (0.20%) and lysine (0.20%) in combination with methionine (0.15%), but not alone, were shown to alleviate adverse the effects of tannins on feed intake and daily gain (Armstrong et al., 1973b). Similar results were also reported in other studies (Chang and Fuller, 1964; Elkin et al., 1978).

In other experiment involving chicks, Elkin et al. (1978) showed that as compared with low-tannin sorghum, hightannin sorghum depressed growth and feed conversion and the adverse effect of high-tannin sorghum was overcome by 0.15% methionine supplementation. Jacob et al. (1996a) replaced corn with sorghum for broilers (55-65%) and layers (60-70%), contributing an average of 2.3% tannin. In broilers, overall body weight and FCR were not different. In layers, however, egg production decreased, FCR increased, egg weight did not change. In another study, Jacob et al. (1996b) supplemented broilers with methionine and fed diet containing a mixture of Serena and white sorghums with 1.3% catechin equivalent. Methionine supplementation did not differ body weight and FCR, possibly due to low tannin content. In broilers, it was shown that supplementing 0.12% methionine and 0.20% lysine alleviated deterioration in growth performance resulting from feeding sorghum (Bornstein and Lipstein, 1975; Lipstein and Bornstein, 1975). Finally, although increased incidence of leg abnormality in response to tannic acid addition was reported (Armstrong et al., 1973b), this was not observed in the present study and others (Jacob et al., 1996a; Mitaru et al., 1983).

# CONCLUSION

This experiment failed to demonstrate the adverse effect of partially replacing corn with sorghum (22%) in the diet on laying performance, metabolic parameters, and egg quality during the peak laying period. Supplementations of methionine (0.57%), lysine (0.66%), choline (0.47%), and sulfur (0.05%) had variable effects on laying performance, metabolic profile, and egg quality parameters. Lack of adverse effect of sorghum inclusion was attributed to low-tannin content (0.26%). Results of this experiment suggest that laying hen can be fed low-tannin sorghum up to 22% without compromising laying performance and requiring additives. Further studies should test the effect of these supplements into diets containing high-tannin sorghum or low-tannin sorghum more than 22%.

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