



## The Use of Sweet Almond Meal as a Protein Source in Japanese Quails Diets

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### Abstract

In the first experiment, the chemical composition, apparent metabolizable energy (AME), AME corrected for nitrogen (AME<sub>n</sub>), true metabolizable energy (TME), TME corrected for nitrogen (TME<sub>n</sub>) values of the sweet almond meal were determined in adult Leghorn cockerels. The second experiment was performed to evaluate the effects of different levels of sweet almond meal at 0, 100, 200 and 300 g/kg on Japanese quail's growth performance, some blood metabolites, relative weight of different organs, meat quality and egg yolk cholesterol in a completely randomized design with 288 Japanese quails including 4 treatments, 4 replicates and 18 birds per replicate. The metabolizable energy values of sweet almond meal were following: AME = 3734, AME<sub>n</sub> = 3648, TME = 3908, TME<sub>n</sub> = 3746 kcal/kg as fed basis. Feed intake, feed conversion ratio and live weight gain and relative weight of different organs in the birds fed diets with different levels of the sweet almond meal were not statistically different from control. A sweet almond meal at 300 g/kg level showed the lower serum total cholesterol and low-density lipoprotein ( $P < 0.05$ ) compared to control and 100 g/kg sweet almond meal. Dietary treatments had no significant effect on the total cholesterol content of quail's eggs. Malondialdehyde concentration in breast meat samples after 40 days freezing decreased, whereas the level of sweet almond meal increased ( $P < 0.01$ ). In general, a sweet almond meal without any adverse effect on growth performance is a good source of energy and protein and can be used up to 300 g/kg of the Japanese quail diets.

### Introduction

Almond nuts (*Prunus amygdalus*) contain two types: 1) sweet almonds (*Prunus amygdalus dulcis*) 2) bitter almonds (*Prunus amygdalus amara*) (Monaghan, 2008). According to FAO (2012), the top of the four almond producers was the United States, followed by Spain, Australia and Iran. The almond production in the world

has a growing rate of 5 percent annually. Almond with 87708 hectares cultivated area is one of the most important horticulture products in Iran (FAO, 2012). According to USDA (2013), dry matter, ether extract, crude protein, ash and crude fiber content of almond nut are 953, 494.2, 212.2, 29.9 and 122 g/kg, respectively. Oil

extraction of almond is done by cold pressing methods, so sweet almond meal (SAM) contains considerable amounts of oil.

There are a large number of reports regarding the effects of almond consumption on blood biochemical parameters (Jenkins *et al.*, 2002; Hyson *et al.*, 2002) and preventing and treating diseases (Prior and Cao, 2000) in human. Different phenolic compounds were characterized and identified in almond nut extract and its skin, shell and hull as almond by-products (Wijeratne *et al.*, 2006; Jahanban-Esfahlan *et al.*, 2010). Whole almond seed, brown skin, shell and hull possess potent free radical scavenging capacities (Subhashinee *et al.*, 2002; Pinelo *et al.*, 2004; Amarowicz *et al.*, 2005; Mour *et al.*, 2007; Jahanban-Esfahlan *et al.*, 2009) and brown skin and green shell cover are more effective than whole seed (Subhashinee *et al.*, 2002). It seems almond meal should have prebiotic properties in poultry because fiber from almond skins can alter the composition of gut bacteria and could be used as potential prebiotics in human diets (Mandalari *et al.*, 2008).

There is no available report regarding the use of SAM in poultry diets. So, in the present study metabolizable energy values of the almond meal were determined and then the effects of using different levels of SAM on Japanese quail's growth performance, blood parameters, the relative weight of different organs, meat quality and egg yolk cholesterol were evaluated.

## Materials and Methods

### Metabolizable energy assay (first experiment)

In the first trial, the apparent metabolizable energy (AME and AME<sub>n</sub>) content of SAM was determined using total collection method (Macleod, 2002). SAM was substituted with a corn-soybean meal basal diet (corn 68.38%, fish meal 2 %, soybean meal 26.8%, oyster shell 1.2%, dicalcium phosphate 0.8%, salt 0.28 %, DL-methionine 0.04 % and vitamin-mineral premix 0.5%) at 40% level, and then AME and AME<sub>n</sub> of this experimental diet and basal diet were determined. Twelve adult leghorn cockerels (165 d old with mean body weight of 1645±30 g) were housed in individual cages, with 6 pens per treatment. Feed (as mash) and water were provided *ad libitum*. After 4 days adaptation to experimental diets, excreta were collected and corresponding feed intake recorded for

subsequent three days. Feed was removed overnight at the start and termination of the excreta collection period. The collected excreta were dried at 65°C for 48 hrs in a conventional oven. Then samples were placed in the lab environment for 24 hrs to equilibrate with ambient humidity and then ground before DM, gross energy, and nitrogen determinations. All samples were analyzed in duplicates. The ME value of almond was determined according to the formula:  $ED = (P \times EF) + (1 - P) EB$ , Where ED is the ME of the experimental diet, P is the level of almond in the experimental diet, EF is the ME of almond, (1 - P) is the level of basal diet in experimental diet and EB is the ME of basal diet.

In the second trial, twelve adult leghorn cockerels with nearly same weights were used to determine the true metabolizable energy using Sibbald procedure method (Sibbald, 1986). Birds fasted for 48 hrs, and then 6 birds received 25 g SAM using the force-feeding procedure. Also, six cockerels were fasted to determine the endogenous urinary energy and fecal metabolic energy losses. Finally, TME and TME<sub>n</sub> were calculated. Chemical compositions of SAM and excreta samples were measured according to the prevalent methods (AOAC, 2005).

### Quail assay (second experiment)

A total of 288 day-old quail chicks were randomly allocated to four experimental groups with four replicates and 18 chicks per replicate. The diets were fed to quail chicks for six weeks. Four experimental diets were formulated to meet the NRC requirements (NRC, 1994) of quails as shown in Table 1.

All experimental diets were formulated and adjusted to be isonitrogenous and isocaloric. Diets contained four levels of SAM (0, 100, 200 and 300 g/kg). Feed and water were provided on *ad libitum* basis. Body weight gain, feed intake and feed conversion were determined on a weekly basis. To measure the weight of different organs, two chicks from each replication were selected randomly and sacrificed at 42 days of age. At 42 days, blood samples were also taken from the neck vein for analyses of total cholesterol, glucose, triglyceride, serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities, uric acid, high density lipoprotein (HDL) and low-density lipoprotein (LDL), using Pars-Azmoon kits and

an autoanalyzer (model: Cobas-Mira).

Malondialdehyde (MDA) concentration in breast meat (fresh and/or frozen at -20°C for 40 days) was also determined (Tarladgis *et al.*, 1960). Breast meat pH was also determined by blending 10-gram sample in 100 mL distilled water for one minute and pH was measured using a pH meter (model AZ. 86502) (Ensoy *et al.*, 2004). According to the method of Pasin *et al.* (1998), three eggs per replication were selected

at the beginning of laying (at 50 d of age) and total cholesterol content in yolks determined.

#### Statistical analysis

This experiment was performed as completely randomized design. The data were analyzed using the general linear (GLM) procedure of SAS (2003) and Duncan's Multiple Range test was used to detect ( $P < 0.05$ ) differences among treatment means.

**Table 1.** Diet formulation and calculated chemical composition of the rations (g/kg as fed)

Ingredients	Control	SAM (100 g/kg)	SAM (200 g/kg)	SAM (300 g/kg)
Corn	433.0	411.9	392.0	374.5
Barley grain	2.0	28.0	54.0	80.0
Soybean meal (44%)	477.9	373.0	266.5	159.0
SAM	-----	100.0	200.0	300.0
Soybean oil	54.4	40.0	24.8	10.0
Calcium carbonate	13.5	12.0	11.0	9.0
Dicalcium phosphate	8.0	7.5	7.0	6.6
Salt	3.7	3.7	3.6	3.7
DL-Methionine	1.5	1.9	2.4	3.1
L-Lysine-HCl	-----	2.0	4.8	7.6
Premix <sup>1</sup>	5.0	5.0	5.0	5.0
Grit	1.0	15.0	28.9	41.5
<i>Chemical composition</i>				
ME (kcal/kg)	3000	3000	3000	3000
Crude protein (g/kg)	248.2	248.2	248.2	248.2
Lysine (g/kg)	13.9	13.4	13.4	13.5
Methionine (g/kg)	5.2	5.2	5.2	5.2
Methionine + Cystine (g/kg)	9.2	8.6	8.1	7.8
Calcium (g/kg)	8.4	8.3	8.4	8.3
Available phosphorus (g/kg)	3.1	3.1	3.1	3.1
Sodium (g/kg)	1.5	1.5	1.5	1.5
Crude fiber (g/kg)	4.3	4.6	4.9	5.2

<sup>1</sup>The vitamin-mineral premix provided the following per kg of diet: retinol acetate 3.78 mg, cholecalciferol 0.055 mg, dl- $\alpha$ -tocopheryl acetate 30 mg, menadione 2 mg, pyridoxine 4.5 mg, thiamin 2.5 mg, riboflavin 6 mg, pantothenic acid 10 mg, niacin 60 mg, folic acid 0.6 mg, biotin 0.15 mg, cyanocobalamin 0.02 mg, choline chloride 400 mg, Zn 80mg, Cu 10 mg, Mn 80 mg, Se 0.3 mg.

#### Results and Discussion

Dry matter, ether extract, crude fiber, crude protein, ash, calcium, total phosphorus and sodium content of SAM were 950, 220, 95, 430, 60, 9.5, 9.9 and 0.01 g/kg, respectively. AME, AME<sub>n</sub>, TME and TME<sub>n</sub> of SAM were 3734  $\pm$  102, 3648  $\pm$  90, 3908  $\pm$  147, 3746  $\pm$  92 (kcal/kg as fed basis), respectively. According to Monaghan (2008), moisture, lipid, protein, ash, soluble sugars, and tannins in almond nuts ranged from 29-56, 509-667, 157-267, 26-35, 30-65 and 0.7-3.6 g/kg, respectively.

The acidic amino acids (glutamic acid and aspartic acid) were the dominant amino acids in almond accounting for 24.4-43.3% of total amino acids and the majority of lipids in almond seeds

are monounsaturated (~67%) and polyunsaturated (~25%) fatty acids. We could not find any report regarding the chemical composition and ME content of SAM. It can be concluded that SAM is an appropriate source of energy and protein.

#### Performance traits

The effects of different levels of SAM on feed intake, weight gain and feed conversion ratio (FCR) of Japanese quails are shown in Table 2. Growth performances of the birds fed diets with different levels of SAM were not statistically different compared to control. Although the birds fed with the diet containing 300 g/kg SAM showed lower feed intake and weight gain and

higher feed conversion ratio. Thus, it seems using SAM up to 300 g/kg of the diet had no adverse effect on quail's growth performance. There is, so far, no report regarding the effect of

SAM on poultry growth performance. There was no significant treatment effect for the relative weight of different internal organs and carcass traits at 42 d of age (Table 3).

**Table 2.** The effects of different levels of SAM on feed intake (FI, g/b/d), body weight gain (WG, g/b/d) and feed conversion ratio (FCR) of Japanese quails

Treatments	7-21 d			21-42 d			7-42 d		
	FI	WG	FCR	FI	WG	FCR	FI	WG	FCR
Control	14.7	6.8	2.2	26.5	5.0	5.3	21.8	5.7	3.8
SAM 100 g/kg	14.6	6.9	2.1	26.2	5.4	4.9	21.6	6.0	3.6
SAM 200 g/kg	14.5	6.7	2.2	25.7	5.2	5.0	21.2	5.8	3.7
SAM 300 g/kg	14.6	6.6	2.2	26.0	4.9	5.3	21.4	5.6	3.8
SEM	0.31	0.12	0.02	0.63	0.15	0.16	0.46	0.10	0.08
P-values	NS	NS	NS	NS	NS	NS	NS	NS	NS

No significant difference was observed between treatments ( $P > 0.05$ ).

**Table 3.** The Effects of dietary levels of SAM on the relative weight of different organs and carcass traits in Japanese quails aged 42 d (% of live weight)

Treatments	Heart	Liver	Spleen	Large intestine	Small intestine	Ceca	Bursa of Fabricius	Pancreas	Thighs	Breast	Abdominal fat
Control	0.82	1.39	0.038	0.58	1.25	0.45	0.085	0.18	13.89	25.31	1.37
SAM 100 g/kg	0.91	1.48	0.055	0.57	1.34	0.47	0.055	0.19	13.36	24.28	1.06
SAM 200 g/kg	0.82	1.47	0.043	0.61	1.43	0.43	0.055	0.18	13.18	24.49	1.29
SAM 300 g/kg	0.79	1.51	0.046	0.57	1.36	0.55	0.06	0.22	13.76	25.13	1.14
SEM	0.049	0.10	0.005	0.043	0.101	0.058	0.013	0.025	0.623	0.495	0.217
P-values	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

No significant difference was observed between treatments ( $P > 0.05$ ).

### Blood analysis

The effect of different dietary treatments on blood parameters at 42 days of age is presented in table 4. There were no significant differences in serum triglyceride, glucose, AST and ALT levels between different dietary treatments. The blood cholesterol level in quails fed diets containing 200 and 300 g/kg SAM, were significantly lower than quails fed diets containing 100 g/kg SAM and control ( $P < 0.05$ ). The uric acid level in quails fed diets containing 300 g/kg SAM was significantly lower than quails fed control diet ( $P < 0.05$ ). LDL level in quails fed diets containing 200 and 300 g/kg

SAM, was significantly lower than quails fed control diet ( $P < 0.05$ ).

The findings presented here are in agreement with previous reports which showed the application of almond nut in human diet can reduce blood total cholesterol and LDL and also can increase blood HDL (Fulgoni *et al.*, 2002; Hyson *et al.*, 2002; Jenkins *et al.*, 2002). These effects of almonds are mediated by components in the oil fraction of this nut (Hyson *et al.*, 2002) or probably in part because of the non-fat (protein and fiber) and monounsaturated fatty acid components of the nut (Jenkins *et al.*, 2002).

**Table 4.** Effects of dietary treatments on blood serum parameters in Japanese quails aged 42 d

Treatments	LDL (mg/dL)	HDL (mg/dL)	AST (IU/L)	ALT (IU/L)	Uric acid (mg/dL)	Triglyceride (mg/dL)	Cholesterol (mg/dL)	Glucose (mg/dL)
Control	63.6 <sup>a</sup>	60.1	7.67	199.0	10.2 <sup>a</sup>	61.2	136 <sup>a</sup>	275
SAM 100 g/kg	57.4 <sup>ab</sup>	67.7	8.75	218.7	9.6 <sup>ab</sup>	59.0	137 <sup>a</sup>	270
SAM 200 g/kg	34.6 <sup>bc</sup>	65.3	8.75	213.5	9.4 <sup>ab</sup>	57.2	115 <sup>b</sup>	240
SAM 300 g/kg	27.9 <sup>c</sup>	78.3	6.75	210.7	8.1 <sup>b</sup>	47.2	113 <sup>b</sup>	249
SEM	8.09	4.43	0.69	6.87	0.462	4.36	6.3	13.4
P-values	0.02	0.07	NS	NS	0.04	NS	0.03	NS

<sup>a-c</sup> Means within a column with no common superscripts differ significantly ( $P < 0.05$ ).

### Meat quality

The effects of SAM on fresh and frozen breast meat quality (pH, CP and MDA values) are given in table 5. There were no significant differences in fresh breast meat pH, CP and MDA concentration between different dietary treatments. But SAM significantly decreased MDA concentration in breast meat kept frozen for 40 d ( $P < 0.01$ ). Subhashinee *et al.* (2002) showed the scavenging activity of superoxide and hydroxyl radicals by different almond extracts. Wijeratne *et al.* (2006) indicated that

brown skin extract at 50 ppm effectively inhibited copper-induced oxidation of human LDL cholesterol compared to whole seed and green shell cover extracts. So, the consumption of almond nut can increase plasma polyphenol concentrations and total antioxidant capacity but reduces plasma lipid peroxidation (Torabian *et al.*, 2009). The cholesterol content of eggs is shown in Table 5. There were no significant differences for the egg yolk cholesterol content among different dietary treatments.

**Table 5.** Effects of dietary treatments on breast meat quality and egg yolk cholesterol content in Japanese quails

Treatments	CP (g/kg)	pH	MDA <sup>1</sup> (mg/kg)	MDA <sup>2</sup> (mg/kg)	MDA <sup>3</sup> (mg/kg)	Cholesterol (mg/g yolk)
Control	205.0	6.4	0.23	2.64 <sup>a</sup>	0.88 <sup>a</sup>	13.5
SAM 100 g/kg	211.3	6.3	0.20	0.70 <sup>b</sup>	0.50 <sup>b</sup>	14.1
SAM 200 g/kg	205.7	6.4	0.21	0.59 <sup>b</sup>	0.48 <sup>b</sup>	13.8
SAM 300 g/kg	215.7	6.3	0.19	0.45 <sup>b</sup>	0.47 <sup>b</sup>	13.7
SEM	3.51	0.04	0.024	0.092	0.036	0.31
<i>P-values</i>	NS	NS	NS	0.0001	0.0001	NS

<sup>1</sup>MDA concentration in fresh meat at 42 d of age; <sup>2</sup>MDA concentration in frozen meat at 42 d of age; <sup>3</sup>MDA concentration in frozen meat at 21 d of age.

<sup>a,b</sup> Means within a column with no common superscripts differ significantly ( $P < 0.05$ ).

### Conclusion

The finding presented here opened a new window towards using SAM in poultry diets. It seems that SAM could be used up to 300 g/kg in poultry diets without any adverse and side effect. However, for commercial applications of this by-product in future, some other data must be collected.

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