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MORE DIETARY BIOTIN IS NEEDED WHEN DIET OF QUAILS CONTAINS SOYBEAN OIL*

SOYA YAĞI İÇEREN BILDIRCIN RASYONLARINDA DAHA ÇOK BİYOTİNE İHTİYAÇ VARDIR

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ABSTRACT
The objective of this study was to evaluate the effects of dietary biotin supplementation with different fat sources (soybean oil or beef tallow) on live performance and plasma concentrations of some minerals and metabolites in Japanese quails. One hundred and twenty 10-day-old Japanese quails were assigned to 4 treatment groups. The experiment was designed in a 2 X 2 factorial arrangement using two types of fat source (5% soybean oil or 5% tallow) and two levels of biotin supplements (0 or 300 mcg/kg diet). Feed intake was not influenced by either fat source or biotin supplementation in the diet ($p > 0.05$). Final body weights and feed efficiency were greater when the diet of quails contained soybean oil compared with that of tallow ($p \leq 0.012$). Liver weights ($p = 0.017$) and abdominal fat accumulation ($p = 0.17$) were greater in quails fed tallow compared with of soybean oil. Plasma protein, triglyceride and cholesterol concentrations were greater ($p \leq 0.04$) when tallow was included in the diet. It was concluded that biotin can be supplemented at 300 mcg/kg diet to the quail diets containing 5% soybean oil.

Keywords: Biotin, soybean oil, tallow, quail.

ÖZ
Bu çalışmanın amacı Japon bıldırcın (*Coturnix coturnix Japanica*) rasyonlarına ilave edilen biyotinin farklı yağ kaynakları (soya yağı veya don yağı) ile birlikte kullanıldığında performansı ve kimi serum mineral ve metabolit konsantrasyonlarının nasıl etkilendiğini araştırmak idi. On günlük yaşta 120 adet Japon bıldırcını 4 gruba ayrılmıştır. Deneme, 2 X 2 faktöriyel düzende ve rasyonda 2 farklı yağ kaynağı (%5 soya yağı veya %5 don yağı) ve 2 farklı biyotin saplement düzeyi (0 ve 300 mcg/kg rasyon) olacak şekilde oluşturulmuştur. Yem tüketimi rasyondaki yağ türünden ya da biyotin saplement düzeyinden etkilenmemiştir ($p > 0.05$). Çalışma sonu canlı ağırlık ve yemden yararlanma oranı soya yağı içeren rasyon tüketen bıldırcınlarda don yağı içeren rasyon tüketen bıldırcınlara oranla daha fazla bulunmuştur ($p \leq 0.012$). Karaciğer ağırlığı ($p = 0.017$) ve karın etrafı yağ birikimi ($p = 0.17$) don yağı ile beslenen bıldırcınlarda daha yüksek bulunmuştur. Plazma protein, trigliserit ve kolesterol konsantrasyonları rasyona don yağı ilavesi ile birlikte artmıştır ($p \leq 0.04$). Bıldırcın rasyonlarına ilave edilen 300 mcg/kg rasyon biyotinin %5 soya yağı ile birlikte kullanılabileceği sonucuna varılmıştır.

Anahtar kelimeler: Biyotin, soya yağı, don yağı, bıldırcın.

INTRODUCTION
Biotin, formerly known as vitamin H or coenzyme R, is a B complex vitamin containing sulfur. Biotin is an essential co-factor for the enzymes carboxylases involved in gluconeogenesis and provision of intermediates into the citric acid cycle, fatty acid synthesis, leucine catabolism, and propionate catabolism (1). Biotin has also been found to be correlated with certain diseases such as diabetes mellitus, liver disorders, and immunological abnormalities (2-5). Since remarkable synthesis of biotin occurs through intestinal fermentation in farm animals, deficiency is rare particularly with corn-based rations (6). However, supplemental biotin may increase the biotin status influencing the performance of Japanese quails. Baez-Soldana *et al* (7) reported that biotin administration increased the activity of propionyl-CoA carboxylase, pyruvate carboxylase, and acetyl-CoA carboxylase in healthy individuals. Therefore, changes in the activities of metabolic enzymes may influence nutrient partitioning, type of tissue deposition, and thus live performance of farm animals. Fats and oils are included in poultry diets due mainly to their high-energy ingredi-

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ents and also other beneficial effects. Therefore, the objective of this study was to evaluate the effects of dietary biotin supplementation with different fat sources (soybean oil or beef tallow) on live performance, organ weights and plasma concentrations of Ca, P, total protein, cholesterol, triglyceride, and glucose in Japanese quails (*Coturnix coturnix Japanica*).

MATERIALS AND METHODS

One hundred and twenty 10-day-old healthy Japanese quails (*Coturnix coturnix Japanica*) were used in the study. The birds were randomly assigned, according to their initial body weights, to 4 treatment groups, 3 replicates of 10 birds each. The experiment was designed in a 2 X 2 factorial arrangement using two types of fat source (5% soybean oil or 5% tallow) and two levels of biotin supplements (0 or 300 mcg/kg diet). Vitamin H2 (Biotin) 2% white crystalline powder (Kartal Kimya, Istanbul-Turkey) was used as biotin source. Tallow (Kemsan, Kayseri-Turkey) and soybean oil (Maygroup, Istanbul-Turkey) were specifically produced as feed grade. Beef tallow was chosen as a source of mainly saturated fatty acids, whereas soybean oil served as source of long-chain unsaturated fatty acids, predominantly linoleic acid. The basal diet was formulated using NRC (8) guidelines (Table 1). The diets and fresh water were offered ad libitum. The birds were kept in cages (19 cm x 19 cm x 19 cm). Light was provided all the time (24 h) inside the henhouse. The length of the study was 30 days. At weekly intervals, feed intake and body weight were determined on group basis. Weight gain and feed efficiency of groups were then calculated. The experiment was in accordance with animal welfare and ethics, and was conducted under protocols approved by the Erciyes University School of Veterinary Medicine Ethical Committee (23.03.2006-005-006).

At the end of day 42, the experiment was terminated. For carcass evaluations, 10 birds randomly chosen from each treatment group were slaughtered. The carcasses were manually eviscerated and hot carcasses weights were obtained. Carcasses were chilled and weighed, and abdominal fat separated by hand and weighed. The weight (± 0.001) of liver, heart, spleen, emptied gizzard, and abdominal fat were calculated as percentages of BW. At the end of study, blood samples were collected by Vena brachialis puncture under the wing from 9 birds randomly chosen from each treatment. Blood samples were taken into tubes, centrifuged at 3000 rpm for 10 minutes to yield plasma, and stored at -20 °C for later analysis. Plasma samples were thawed at room temperature and were analyzed for Ca, P, total protein, cholesterol, triglyceride, and glucose concentrations using

Table I: Ingredients and chemical analyses of basal diets containing either soybean oil or tallow fed to quails

	Added dietary 5% of	
Ingredients, % of DM	soybean oil	tallow
Ground corn	47.40	48.02
Soybean meal, 46% CP	44.91	44.30
Soybean oil	5.00	-
Tallow	-	5.00
Limestone*	1.20	1.19
Dicalcium phosphate	0.92	0.92
Vitamin-Mineral premix**	0.32	0.32
Salt (NaCI), iodized	0.23	0.23
DL-Methionine	0.02	0.02
DM, %	88.79	88.45
Nutrient composition ***		
ME, MJ/kg	13.37	13.37
Crude protein, %	24.00	24.00
Ether extract, %	7.49	7.54
Calcium, %	0.8	0.8
Nonphytate phosphorus, %	0.3	0.3
Methionine, %	0.5	0.5
Methionine + cystine, %	0.76	0.75
Lysine, %	1.39	1.38
Biotin, mcg	197.75	196.17

* Limestone contains 0.55 % Mg.

** Premix (KAYTAS YEM Vitamin Mineral Formula CVM, Kayseri-Turkey) supplied (2,5 kg/ton diet) as 12.500.000 IU vitamin A, 3.000.000 IU vitamin D3, 20.000 mg vitamin E, 3.000 mg vitamin K3, 2.500 mg vitamin B1, 7.000 mg vitamin B2, 5.000 mg vitamin B6, 20 mg vitamin B12, 20.000 mg niacin, 15.000 mg Cal-D-Pan, 1.000 mg folic acid, 20 mg biotin, 50.000 mg vitamin C, 300.000 mg cholin chloride, 80.000 mg manganese, 70.000 mg iron, 50.000 mg zinc, 6.250 mg copper; 1.250 mg iodine, 200 mg cobalt, 150 mg selenium, 90.000 mg lasolocid sodium.

*** Except crude protein and ether extract, values were calculated from NRC (8) tables.

commercial kits (Chema) in a spectrophotometer (Shimadzu UV-1700). Chemical analysis of the diet was run using international procedures of AOAC (9). The data were analyzed by use of ANOVA with fat source and biotin as main effects using SAS (10).

RESULTS

Feed intake was not influenced by either fat source or biotin level in the diet ($p > 0.05$) (Table 2). Initial body weights were also similar among treatment groups ($p > 0.05$). Final body weights ($p = 0.012$) and feed efficiency ($p = 0.001$) were greater when the diet of quails contained soybean oil compared with that of tallow. There were no interactions for live performance parameters except feed efficiency ($p = 0.019$). Feed efficiency increased when soybean oil was included in the diet with greater biotin level; however, feed efficiency decreased when the diet included tallow with biotin supplementation. Liver weights (2.5% vs. 3.64% of hot carcass weight, $p = 0.017$) and abdominal fat accumulation (0.56% vs. 0.68% of cold carcass weight, $p = 0.17$) were greater in quails fed to tallow compared with that of soybean oil (data not shown). Diets containing soybean oil with no respect to biotin level did not change the plasma Ca ($p > 0.05$) but increased the plasma phosphorus concentrations ($p = 0.001$) (Table 3). Plasma protein concentrations increased with the inclusion of tallow ($p = 0.001$) and also with the biotin supplementation ($p = 0.01$) in the diet of quails. In addition, plasma protein concentrations were greater when tallow, at particular, was included in the diet with biotin supplementation (interaction, $p = 0.04$). Plasma cholesterol concentrations increased with the inclusion of tallow ($p = 0.002$) and also with the biotin supplementation ($p = 0.03$) in the diet of quails. In accord with plasma cholesterol concentrations, plasma triglyceride concentrations also

Table II: Supplementary effects of biotin and dietary fat sources (5%) on performance of Japanese quails*

Treatment					
Fat source	Biotin, mcg/kg	Feed intake, g	Initial body weight, g	Final body weight, g	Feed efficiency[a]
Soybean oil	0	590.80	46.31	193.84	0.249
Soybean oil	300	598.06	44.64	195.71	0.253
Tallow	0	597.80	44.39	186.16	0.237
Tallow	300	599.60	44.53	178.90	0.224
Pooled SEM		10.86	0.97	3.80	0.003
Main effects					
Soybean oil		594.43	45.48	194.77	0.251
Tallow		598.70	44.46	182.53	0.230
	0	594.30	45.35	190.00	0.243
	300	598.83	44.58	187.30	0.238
ANOVA					
Source		Probabilities			
Fat source		0.71	0.33	0.012	0.001
Biotin		0.69	0.46	0.50	0.106
Fat source x Biotin		0.81	0.38	0.27	0.019

* Values are means, n = 3

[a] gram of gain : gram of feed consumed

increased with the inclusion of tallow (p = 0.003) in the diet of quails with no respect to biotin supplementation. Biotin supplementation increased plasma glucose concentrations (p = 0.01) with no regards to the fat source in the diet of quails.

Table III. Supplementary effects of biotin and dietary fat sources (5%) on some plasma parameters of Japanese quails*

Treatment							
Fat source	Biotin, mcg/kg	Ca mg/dl	P mg/dl	Protein g/dl	Cholesterol mg/dl	Triglyceride mg/dl	Glucose mg/dl
Soybean oil	0	9.57	10.85	2.57	159.81	169.97	226.62
Soybean oil	300	10.50	11.67	2.89	200.10	210.51	259.22
Tallow	0	11.69	1.92	6.64	218.67	367.85	225.45
Tallow	300	13.76	2.52	9.99	258.93	561.48	248.61
Pooled SEM		1.77	1.61	0.68	18.79	90.56	10.60
Main effects							
Soybean oil		10.03	11.26	2.73	179.95	190.24	242.92
Tallow		12.72	2.22	8.31	238.80	464.67	237.03
	0	10.63	6.39	4.60	189.24	268.91	226.03
	300	12.13	7.10	6.44	229.52	386.00	253.92
ANOVA							
Source		Probabilities					
Fat source		0.12	0.001	0.001	0.002	0.003	0.56
Biotin		0.38	0.65	0.01	0.03	0.18	0.01
Fat source x Biotin		0.74	0.94	0.04	0.99	0.38	0.64

* Values are means, n = 9

body and therefore is an interest in terms of nutrient partitioning and other metabolic activities. High energy in the diet of broilers sometimes may cause liver enlargement (16) besides other known and unknown factors. However, although all quails were reared at the same conditions and consumed an isoenergetic diet free of mycotoxines (based on the feed manufacturer's declaration), liver weights were greater in quails fed tallow, indicating an enlargement of the liver with sometimes yellowish and friable but with no mortality or hemorrhage at all. Therefore, the results were due most probably to fat sources in the diet. Consistent with the result that abdominal fat is grater with tallow feeding, enlarged liver indicates a different metabolic path of the saturated fatty acids. Beynen and Katan (17) stated that dietary polyunsaturated fatty acids in the liver are preferentially oxidized compared with that of saturated fatty acids, probably causing saturated fatty acids to accumulate in the liver leading an enlargement (fatty liver).

DISCUSSION

Neither fat source nor biotin supplementation influenced feed intake of quails. Similar to results of the present study, several researchers reported no feed intake differences in broilers fed various amount (3-10%) and type (sunflower oil, tallow, fish oil, olive oil, or soybean oil) of fats (11-14). Soybean oil inclusion compared with that of tallow in the diet of quails at the present study yielded a better feed efficiency. Crespo and Esteve-Garcia (15) also reported a greater feed efficiency in broilers fed unsaturated fatty acid-enriched diets compared with that of saturated fatty acid-enriched diets. However, some researchers found no differences of feed efficiency in broilers fed a diet containing fat with various degree of saturation (11-12). At the present study, there was also an interaction between biotin supplementation and fat type for feed efficiency in a way that biotin supplementation yielded a better feed efficiency when soybean oil was included in the diet of quails.

The liver is the most important metabolic organ of the

Abdominal fat weights were greater, although not significantly, in quails fed tallow compared with that of soybean oil with no respect to biotin supplementation in the diet. Results obtained at the present study are in agreement with results reported by many researchers.

Feeding unsaturated fatty acid-enriched diets compared with that of saturated fatty acid-enriched diets (decreased degree of saturation) resulted in a lower abdominal fat accumulation (18-20) and a lower whole body fat accumulation in broilers (12, 21). In addition, Wongsuthavas *et al* (14) reported that increased dietary intake of polyunsaturated fatty acids (oil rich) at the expense of a more saturated fat sources reduced the number of fat cells in the breast meat of broiler chickens. The mechanism by which dietary polyunsaturated fatty acids decrease the abdominal fat mass may be due to their preferential oxidation compared with that of saturated fatty acids, yielding ATP thus shifting carbohydrates from oxidative through lipogenic pathway (17). In this case, however, the conversion of glucose into triglycerides is less efficient in terms of energy deposition than is the conversion of the fatty acids into triglycerides (22). Another mechanism by which dietary polyunsaturated fatty acids decrease the abdominal fat mass is explained through activities of enzymes involved in fatty acid synthesis. Sanz *et al* (11) found a greater β-oxidation enzyme activity (carnitine palmitoyltransferase I and L-3-hydroxyacyl-CoA dehydrogenase) and a lower fatty acid synthesis enzyme activity (fatty acid synthetase) resulting in a lower abdominal fat deposition in broiler chickens fed the sunflower oil-enriched diet compared with that of chickens fed tallow.

Soy bean oil-containing diet resulted in a decrease in plasma total protein concentrations, while an increase in plasma phosphorus concentrations. Biotin itself had no effects on plasma calcium or phosphorous concentrations. Contrary to the results of the present study, Watanabe et al. (23) reported a negative correlation between the biotin level (measured without any supplementation) and the serum albumin, triiodothyronine, phosphate, and calcium levels in the elderly people.

Plasma cholesterol and triglyceride concentrations decreased with soybean oil feeding, but cholesterol concentrations increased with biotin supplementation. Decreased plasma concentrations of triglyceride and cholesterol were also reported upon feeding polyunsaturated fatty acid-rich diets compared with that of saturated fatty acid-rich diets in broiler chickens (13). Similar results to the present study for biotin supplementation were also reported in elderly people in a way that biotin levels were correlated positively with the total cholesterol levels (23).

Unexpectedly, biotin supplementation increased the plasma glucose concentrations. Due to an important role of biotin in decreasing blood glucose concentrations, supplemental biotin was expected to decrease the plasma glucose concentration. A relationship between biotin status and glucose metabolism has been reported in a way that biotin deficiency has been associated with hyperglycemia and decreased utilization of glucose (24). Biotin treatment was shown to lower fasting blood glucose in diabetic patients (2). Biotin has been demonstrated to affect two critical enzymes of glucose metabolism: hepatic glucokinase (25) and hepatic phosphoenolpyruvate carboxykinase (26). Furthermore, administration of the vitamin in diabetic rats has been reported to increase hepatic pyruvate kinase and phosphofructokinase activity but not phosphohexose isomerase, suggesting a role of biotin on glycolysis in the liver (26,27). Romero-Navarro et al. (28) also showed that pancreatic islet glucokinase activity and expression (mRNA) are reduced by 50% in the biotin deficient rat. In the same work (28), insulin secretion in response to glucose was also impaired in pancreatic islets isolated from the deficient rat.

In general, biotin supplementation in diets of quails containing soybean oil provided a better live performance and lower abdominal fat accumulation. In addition, soybean oil with biotin supplementation decreased both plasma cholesterol and triglyceride concentrations. Therefore, results of the present study indicated that, based on the market demands, biotin can be supplemented at 300 mcg/kg diet to the quail diets containing %5 soybean oil.

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