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EFFECT OF DIETARY PALM KERNEL OIL SUPPLEMENTATION ON BIOTIN REQUIREMENT OF BROILERS

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Abstract

An experiment was conducted to study the effect of feeding supplemental palm kernel oil on the biotin requirement of broilers. A total of 480 day-old broilers were fed varying levels of palm kernel oil (0% and 2%) and biotin (40, 80, 120, 160, 200 and 240 µg/kg diet) in a 2 x 6 factorial arrangement of treatments. The results showed that 2% supplemental palm kernel oil affected the minimum biotin requirement and the appearance of biotin deficiency symptoms. Incidence of dermatitis, mortality (due to fatty liver and kidney syndrome) and leg deformities were lower in presence of the oil supplement. Lower amount of biotin (120 µg/kg diet) was required in the presence of supplemental palm kernel oil as compared with that needed (160 µg/kg diet) to prevent biotin deficiency symptoms in chicks fed diets without supplemental oil. Feed utilization data indicated that 120 µg biotin/kg diet was required to promote better feed intake and body weight gain whether or not supplemental palm kernel oil was fed.

সারমর্ম

ব্রয়লারের রেশনে বায়োটিন-এর প্রয়োজনীয়তা দেখার জন্য পাম কারনাল অয়েল সরবরাহ করে একটি পরীক্ষা চালানো হয়। এজন্য একদিন বয়সের ৪৮০টি ব্রয়লার বাচ্চার উপর ০% এবং ২% হারে পাম কারনাল ওয়েল ও ৪০, ৮০, ১২০, ১৬০, ২০০ এবং ২৪০ µg/kg হারে রেশনে বায়োটিন দেয়া হয় এবং এই পরীক্ষাটির জন্য ২ x ৬ ফ্যাকটরিয়াল এক্সপেরিমেন্ট ব্যবহার করা হয়। পরীক্ষায় দেখা যায় যে, ২% পাম কারনাল ওয়েল সরবরাহে সবচেয়ে কম বায়োটিন আসে এবং এক্ষেত্রে বায়োটিনের অভাবজনিত লক্ষণ দেখা যায়। এইসব ক্ষেত্রে ডারমাটাইটিস, মৃত্যুহার (ফ্যাটি লিভার এবং কিডনি সিনড্রম-এর কারণে) এবং পা বিকৃতি দেখা যায়। পাম ওয়েলের সাথে ১২০ µg/kg বায়োটিন সরবরাহ করতে হয়। খাদ্য উপযোগিতা তথ্য থেকে দেখা যায় যে, রেশনে পাম কারনাল ওয়েল দেয়া হউক বা না হউক ১২০ µg/kg বায়োটিন সরবরাহ করলে খাদ্য গ্রহণ এবং দৈহিক বৃদ্ধি ভাল হয়।

Key words : Palm kernel oil, Broiler chicken, Biotin.

Introduction

Involvement of biotin-dependent enzymes in the pathway of long-chain fatty acid synthesis and in the conversion of linoleic acid to arachidonic acid (Wakil and Bressler, 1962) is suggestive of a possible effect on biotin requirement for fat metabolism. Indeed, incidence of fatty liver and kidney syndrome (FLKS), a biotin deficiency symptom in chicken was demonstrated to be significantly affected by the type and content of fat in the diet (Blair *et al.,* 1975; Whitehead *et al.,* 1975). Roland and Edwards (1971) observed that in chicks, the dermatitis of biotin deficiency resembled due to the deficiency of essential fatty acids.

The objective of this study was to investigate the effect of dietary supplemental palm kernel oil (PKO) on the biotin requirement of broilers. The PKO was chosen in this study because its inclusion in poultry diets is becoming increasingly popular in Nigeria.

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Materials and Methods

Duplicate groups of day-old commercial broilers with 20 chicks per group were given a basal biotin-deficient diet (Diet 1; Table 1) supplemented with biotin to achieve graded levels of 40, 80, 120, 160, 200 and 240 µg biotin/kg diet. Broilers were housed in deep litter pens, 20 birds per pen (4.1 m^2), and they were supplied with feed and clean water *ad libitum*. Experimental birds were kept on these treatments for a period of 28 days. All birds that died were sent to the veterinary clinic, Faculty of Veterinary Medicine, University of Ibadan for post mortem examination.

Liveweights were measured at 1 and 28 days of age. Feed intake was recorded on a weekly basis. Weight gain and feed efficiency (gain/feed intake) were calculated. Weekly records of incidence of dermatitis (mild to very severe signs), FLKS mortality and incidence of leg deformities in terms of the percentage of birds within a treatment showing either (i) legs with crooked toes, or (ii) legs with bowed or twisted toes, or (iii) difficulty in standing or walking were kept.

At the 28th day, 2 ml of blood was collected in heparinized tubes from wing veins of each of four replicate samples of experimental chicks randomly selected from each treatment for the determination of glucose (Dubois *et al.*, 1956) and free fatty acids (Pearson, 1976) contents. These birds were slaughtered, then the livers and kidneys were excised, drained of fluid with blotting paper and weighed. These organs were freeze-dried and kept for subsequent estimation of total lipid (Folch *et al.*, 1957) and triglyceride (Fletcher, 1968) contents.

Diet 2 was formulated by including PKO in the biotin-deficient basal diet (Diet 1) at 2% level (Table 1) such that both diets had similar metabolizable energy value and protein content. Biotin-deficient diet with 2% oil inclusion, therefore, served as the second basal diet. Graded levels of biotin were added to the basal diet with 2% oil to provide experimental diets containing 40, 80, 120, 160, 200 and 240 µg biotin/kg diet.

These diets were fed to duplicate groups of 20 day-old chicks each for a period of 28 days. The liveweight, weight gain, feed intake, feed efficiency, and incidence of dermatitis, FLKS mortality and leg deformities were recorded.

Results obtained in this study were subjected to analysis of variance in accordance with the procedures of Steel and Torrie (1960). The differences between treatment means were tested using the multiple range test of Duncan (1955).

Results

Feed Utilization and Biotin-Related Features

At 28 days of age, birds given 40 and 80 µg biotin/kg diet with or without supplemental 2% PKO consumed significantly less feed, gained less weight and had lower body weight

Table 1. Composition of basal biotin-deficient diets

Ingredients	Diet 1 (%)	Diet 2 (%)
Yellow maize	54.0	49.5
Palm Kernel meal	18.0	18.0
Blood meal	10.0	10.5
Fish meal	2.5	2.5
Brewer's grain	12.2	12.2
Oyster shell	1.0	2.0
Bone meal	2.0	3.0
Vitamin/Mineral premix*	0.1	0.1
Salt (NaCl)	0.2	0.2
Palm Kernel oil	0.0	2.0
Total	100.0	100.0
Calculated composition		
Crude protein (%)	21.26	21.25
Metabolizable energy (Kacal/kg)	2741.92	2751.82
ME/CP	128.73	129.48
Fat (%)	3.50	5.50
Linoleic acid (%)	1.40	1.33
Biotin (µg/kg)	38.90	36.20
Crude fibre (%)	5.61	5.52

+Premix provides the following vitamins and minerals per kg of diet: Vitamin A, 8,000 i.u.; Vitamin D_3, 1500 i.u.; Vitamin E, 3 i.u.; Menadione sodium bisulphite (Vitamin K), 1.5 mg.; Vitamin B_2, 2.5 mg.; Calcium d-panthothenate, 3 mg.; Nicotinic acid, 8 mg.; Vitamin B_6, 0.3 mg.; Vitamin B_{12}, 0.008 mg.; Iron, 15 mg.; Manganese, 25 mg.; Copper, 2.5 mg.; Zinc, 10 mg.; Iodine, 0.3 mg.

($P<0.05$) than those given 120 to 240 µg biotin/kg diet (Table 2). Comparing the performance of birds on the same dietary biotin levels with or without oil supplementation indicated that the addition of 2% oil to the diet led to a marked reduction in feed intake, poorer body weight gain and lighter body weight at 28 days of age.

A 160 µg biotin/kg was quite satisfactory level to prevent FLKS mortality and leg deformities in the absence of oil, a lower level of 120 µg of the vitamin was required in the presence of 2% PKO to prevent FLKS mortality and leg deformities. Although dietary biotin level of 240 µg/kg prevented the occurrence of dermatitis in the birds whether or not their diets were supplemented with PKO, the levels of 120, 160 and 200 µg/kg kept the incidence of such lesion at a minimum level whether the diets were supplemented with or without 2% PKO.

The overall mean values during the 28-day experimental period clearly showed lower incidence of all the biotin-related features when diets were supplemented with 2% PKO. The overall mean values of dermatitis, FLKS mortality and leg deformities of birds fed diets supplemented with 2% PKO were 13.8, 1.7 and 1.7% respectively, being lower than the values of 20.0, 3.3 and 2.5% recorded for the corresponding unsupplemented diets.

Weights and Lipid Contents of Liver and Kidney

Weights and total lipid contents of liver and kidney were significantly ($P<0.05$) affected by the dietary biotin level when the diets were not supplemented with oil, yet no appreciable effects on these parameters were obtained when the diets were supplemented with 2% PKO (Table 3). In the affected group, weights and total lipid contents of livers and kidneys of chicks given 40 and 80 µg biotin/kg diet were appreciably higher than those of birds given 120 to 240 µg/kg diet. However, the supplementation of 2% oil to the diet had a great effect in decreasing the weight and total lipid contents of liver and kidney, compared with those of the unsupplemented diets.

Whether or not the diets were supplemented with 2% oil, birds given 40 and 80 µg biotin/kg gave significantly ($P<0.05$) higher liver and kidney triglyceride content and triglyceride percentages of total lipid than those given 120 to 240 µg/kg. The additionof 2% PKO to diets containing 40 and 80 µg biotin/kg significantly ($P<0.05$) decreased the total lipid concentrations in both organs.

Blood Glucose and Free Fatty Acids

Whether or not diets were supplemented with 2% oil, blood glucose concentration increased with increasing dietary biotin levels, whereas the reverse occurred with blood free fatty acids content (Table 4).

Discussion

Feed Utilization

Essentially of biotin for better feed utilization and growth in chicken has been demonstrated, and the reported values of its requirement for growth in broilers varied from 0.09 mg/kg to 0.17 mg/kg depending on the composition of the test diets (Wagstaff *et al.*, 1961; Ogunmodede, 1978; Whitehead and Bannister, 1980; NRC, 1994). Results of this study indicated that 120 µg/kg was required to promote better feed intake and growth whether or not PKO was included in the diet. The reduction in feed intake of birds fed 40 and 80 µg biotin/kg might be due to the reduction in their free movement as a result of the abnormal development of leg bones as well as the dermal lesions developed in the feet.

Supplementation of diets with fat increased transit time of ingesta in the gastro-intestinal tract with consequent reduction in feed intake (Maner *et al.*, 1962; Mateos *et al.*, 1982). Also poor performance of broiler chicks fed high fat rations were attributed to the failure of such rations to provide sufficient amino acids and perhaps other essential nutrients required in the higher energy ration (Leong *et al.*, 1955; Sunde, 1956). Indeed, Rand *et al.*, (1958); and Marion and Woodroof (1965) noted that body weight and feed efficiency were greater at higher levels of protein when diets were supplemented with fat. Since the two basal diets used in this study (Table 1) had similar protein contents, the 2% supplemental oil in the second basal diet (Diet 2) might have raised the protein requirement of birds for maximum body weight gain. Therefore, poorer weight gain in birds given 2% supplemental oil may be attributed to the lower dietary nutrient intake.

Table 2. Feed utilization and incidence of dermatitis, FLKS mortality and leg deformities in broilers fed diets supplemented with biotin and palm kernel oil at 28 days of age.

Biotin level (µg/kg)	Oil (%)	Body weight (g)	Body weight gain (g/bird/day)	Feed intake (g/bird/day)	Feed efficiency (g gain/g feed)	Dermatitis (%)	FLKS mortality (%)	Leg deformities (%)
40	0	252.3de*	7.58bc	26.07b	0.29	60.0a	10.0a	7.5a
80	0	266.5cd	8.09b	25.55b	0.32	45.0b	7.5b	5.0b
120	0	310.0a	9.64a	29.16a	0.33	10.0c	2.5d	2.5c
160	0	297.5ab	9.20a	28.79a	0.32	2.5d	0.0e	0.0d
200	0	297.5ab	9.20a	29.20a	0.32	2.5d	0.0e	0.0d
240	0	300.0ab	9.29a	28.61a	0.32	0.0d	0.0e	0.0d
Mean		287.3	8.83	27.90	0.32	20.0	3.3	2.5
40	2	229.0d	6.75c	22.50c	0.30	45.0b	7.5b	5.0b
80	2	237.0d	7.04c	21.29c	0.33	30.0b	2.5d	5.0b
120	2	286.0c	8.79ab	25.63b	0.34	2.5d	0.0e	0.0d
160	2	270.0c	8.21b	25.34b	0.32	2.5d	0.0e	0.0d
200	2	264.0c	8.00b	25.30b	0.32	2.5d	0.0e	0.0d
240	2	272.0c	8.29b	25.91b	0.32	0.0d	0.0e	0.0d
Mean		259.7	7.84	24.33	0.32	13.8	1.7	1.7
	±SEM	7.24	0.25	0.70	3.62E-03	6.05	1.04	0.71

* Means within a column for each variable with no common subscript differ significantly (P<0.05)
** SEM, Standard error of the mean.

Table 3. Weight and lipid contents of liver and kidney from broilers fed diets supplemented with biotin and palm kernel oil at 28 days of age.

Biotin level (μg/kg)	Oil (%)	Liver				Kidney			
		weight (g)	Total lipid (mg/g)	Triglyceride (mg/g)	Triglyceride (% Total Lipid)	Weight (g)	Total lipid (mg/g)	Triglyceride (mg/g)	Trigly-ceride (% Total Lipid)
40	0	9.56a*	299.1a	210.02a	70.3a	3.56	349.5a	247.9a	70.9a
80	0	9.11ab	254.1b	165.4b	65.1b	3.51	322.5a	225.1b	67.7a
120	0	8.29bc	169.9cd	79.9d	47.0d	3.09	189.7bc	60.0e	31.6c
160	0	7.90bc	160.9d	73.9d	45.9d	3.14	196.3bc	63.1e	32.1c
200	0	8.00bc	163.0cd	75.3d	46.1d	3.13	187.4bc	56.0e	29.9c
240	0	7.65c	159.5d	74.7d	46.8d	2.96	190.0bc	60.5e	30.8c
Mean		8.42	201.1	113.2	53.5	3.23	240.9	118.8	43.8
40	2	7.34cd	188.5c	100.9c	53.8bc	2.86	212.5b	143.3c	67.4a
80	2	6.50d	183.2cd	96.5c	52.7c	2.81	204.5bc	104.6d	51.4b
120	2	7.50cd	167.7cd	76.4d	45.5d	3.46	168.7c	49.9e	29.6c
160	2	7.34cd	165.8cd	75.8d	45.7d	3.25	186.6bc	61.4e	32.9c
200	2	7.48cd	166.0cd	77.5d	46.7d	3.02	191.5bc	54.7e	28.6c
240	2	7.50cd	166.8cd	78.6d	47.2d	3.05	189.4bc	56.8e	30.0c
Mean		7.28	173.0	84.3	48.6	3.08	192.2	78.4	40.0
	±SEM	0.228	12.09	12.04	2.29	0.068	16.29	19.32	4.76

* Means within a column for each variable with no common subscript differ significantly (P<0.05)
** SEM, Standard error of the mean.

Dermatitis

Foot dermatitis characteristic of biotin deficiency (Ogunmodede, 1978; Whitehead and Bannister, 1978) developed distinctly by the second week in the affected birds. The foot pads were swollen and contained haemorrhagic fissures. Cracked and swollen areas occurred on pads on the bottom of the feet. While Ogunmodede (1978) reported that 120 µg biotin/kg prevented dermal lesions in broiler chicks, Whitehead and Bannister (1978) reported that severity of foot lesions was only reduced by increasing the level of biotin supplementation but was not eliminated by 500 µg/kg. The present results indicated that whereas a minimum of 160 µg biotin/kg was required to effectively reduce the incidence rate in chicks fed diets without supplemental oil, at least 120 µg biotin/kg was needed to reduce the incidence when 2% PKO was added to the diets.

Information on the effect of dietary fat on the severity of dermal lesion characteristic of biotin deficiency seemed to suggest that the type and nature of free fatty acid predominating in the dietary fat might be an important factor. Maize and olive oils, reported to have aggravated the severity of dermal lesion in chicks (Roland and Edwards, 1971; Whitehead *et al.,* 1976), are unsaturated in nature, whereas coconut oil and hydrogenated cotton seed oil that were reported to reduce severity of lesions are saturated in nature (Godin and Spensley), 1971). Roland and Edwards (1971) suggested that the beneficial action of saturated fatty acids on the dermal lesions in biotin-deficient chicks might be by influencing bacterial synthesis of biotin in the gastro-intestinal tract. Whitehead and Bannister (1980) estimated the quantity of biotin contributed from microbial synthesis in broiler fed *ad libitum* at 7 to 8 weeks of age to be 5-10% of the amount required for growth. Dubos (1947) showed that long chain fatty acids exert a toxic effect on bacteria and their toxicity was directly related to the number of unsaturated bonds in the molecule. Mohrhauer *et al* (1967) also showed that myristic acid was effective in the inhibition of chain elongation systems in the microsomes. PKO is saturated in nature and it is rich in myristic acid (Godin and Spensley, 1971). Its inclusion in the diet of broiler may logically decrease the incidence and requirement for biotin in the prevention of dermatitis.

Fatty Liver and Kidney Syndrome

The FLKS, a metabolic disorder is characterized by morbidity followed by death in young chicks, is variable in its occurrence. Mortality can be as high as 20% in occasional outbreaks but, even when mortality is not apparent there is often a low background incidence (Bannister, 1976). In FLKS-affected chicken, Whitehead (1975) and Wight and Siller (1975) noted that the liver and kidney were enlarged because of marked involvement in fatty infiltration. Triglyceride was confirmed to be the main extra lipid in these organs (Johnson *et al.,* 1972; Whitehead, 1975). Also, elevated plasma free fatty acid and markedly reduced glucose level were observed in affected birds (Bannister *et al.,* 1975; Balnave *et al.,* 1977). Furthermore, Whitehead *et al.,* (1976) indicated that the primary abnormality in FLKS condition is the failure of hepatic gluconeogenesis via pyruvate carboxylase, a biotin-dependent enzyme, and this results in several hypoglycaemia which was believed to be the cause of death.

Significantly higher liver weight, lipid (total lipid and triglyceride) contents of liver and kidneys, and blood free fatty acid concentration and lower glucose content in blood of birds given 40 and 80 µg biotin/kg diet supplemented with and without PKO indicated hypoglycaemia condition and excessive lipid deposition in the organs, hence higher incidence of FLKS mortality recorded in these groups of birds. Incidence of FLKS can be reduced by increasing the level of dietary fat (Blair *et al.*, 1975; Whitehead *et al.*, 1975) and eliminated by biotin supplementation in the diet (Whitehead *et al.*, 1976). The results of this study showed that in addition to reducing the incidence of FLKS mortality, 2% supplemental PKO lowered the birds' biotin requirement for preventing its syndrome.

Table 4. Effect of supplemental biotin and palm kernel oil levels on the blood glucoseand free fatty acids contents of broilers at 28 days of age.

Biotin level (ug/kg)	Oil (%)	Glucose (mg/ml)	Free fatty acids (mg/ml)
40	0	0.61d*	0.90a
80	0	0.63cd	0.74b
120	0	0.69cd	0.53c
160	0	0.78bc	0.46c
200	0	0.89ab	0.51c
240	0	0.94a	0.46c
40	2	0.66cd	1.02a
80	2	0.75bc	0.72a
120	2	0.82ab	0.57c
160	2	0.95a	0.59c
200	2	0.99a	0.59c
240	2	1.01a	0.57c
±SEM**		0.040	0.048

Depression of lipid contents of liver and kidneys due to 2% supplemental oil was an indication of reduced lipid deposition in both organs. Hill *et al.*, (1958) observed a measurable depression of lipogenesis in rat liver due to the presence of 2% fat in the diet. The depression was later identified by Bortz *et al.* (1963) to be enzymatic in nature and thus indicated that the block was localized at the step involving the carboxylation of acetyl CoA. Whitehead *et al.* (1978) reported that liver pyruvate carboxylase activity was low in birds fed a diet causing a high incidence of FLKS but the addition of fat to this diet reduced the activity of another biotin-dependent enzyme, acetyl CoA carboxylase. This enzyme seemed to be able to sequester, biotin preferentially, thus a decrease in its activity allows more biotin to become available for pyruvate carboxylase. This explains why the oil inclusion in the ration reduced dietary biotin requirement of the chicken.

Leg Deformities

There is ample evidences supporting that biotin is involved in causing leg bone abnormalities in chicken (Cook *et al.*, 1984 a;b). Perosis, chondrodystrophy, crooked tibia and shortened or twisted tarsometatarsus were the deformities reported in biotin-deficient chicks. Leg deformities observed among the experimental broilers given 40 to 80 µg biotin/kg diet without supplemental oil and those given 40 to 80 µg biotin/kg diet supplemented with 2% oil included crooked toes bowed or twisted toes with majority of affected birds having difficulty in standing or walking.

Conclusion

It may be concluded that 2% supplemental PKO affected the minimum biotin requirement of broiler chicks. Lower amount of biotin (120 µg/kg) was needed in the presence of oil compared with that needed (160 µg/kg) in the absence of PKO to prevent incidence of dermatitis, FLKS mortality and leg deformities. However, feed utilization data indicated that whether or not supplemented oil was given, 120 µg biotin/kg was required to promote better feed intake and body weight gain.

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