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Evaluation of Dietary Calcium Level and Fat Source on Growth Performance and Mineral Utilization of Heat-distressed Broilers

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Abstract: Male broilers (commercial strain) were used to evaluate the effects of diets differing in fat source on performance of heat-distressed broilers. Dietary treatments included corn oil (CO), animal fat (AF), fish oil (FO) and a dry blended (animal and vegetable) fat product (DB) at either 0.9 or 1.5 % calcium. Diets were isocaloric with each containing an equal number of calories from fats. Birds were reared in floor brooder pens and fed experimental diets from Day 1 to 21 and then assigned the same dietary treatments in one of two environmentally controlled chambers. One chamber was maintained at 23.9 °C, whereas birds in the second chamber were exposed to 8 hours of 23.9 °C, 4 hours of 23.9 to 35 °C, 4 hours of 35 °C and 8 hours of 35 to 23.9 °C. At 42 days of age, plasma concentration of calcium and magnesium were higher ($p > 0.01$) in heat distressed (HD) birds than in their thermoneutral (TN) counterparts. Dietary calcium level, but not fat source, affected plasma calcium concentration. Temperature significantly ($p < 0.05$) affected the relative mineral retention (feed minus fecal mineral content) of magnesium while relative mineral retention of copper was affected by fat source. There was no effect of calcium level on performance but HD birds gained 31 % less weight than TN. Birds fed AF gained 10 % more than FO and 14 % more than DB. Data suggest that both fat source and environmental temperature influence mineral utilization and body weight gain.

Key words: Broilers, heat distress, dietary fats, fatty acids, mineral retention

Introduction

One consequence of heat distress is increased mineral excretion. Broilers raised at high cycling ambient temperatures (24 to 35 °C) had lower rates of phosphorus, potassium, sodium, magnesium, sulphur, manganese, copper, and zinc retention compared with birds housed at 24 °C (Belay and Teeter, 1996). This could lead to reduced bone strength and bone weight, which in turn, will have an impact on the bird's performance. It has also been demonstrated that mineral availability is affected by exposure to high temperatures (Smith *et al.*, 1995) while body weight gain is seriously compromised. Interaction among minerals and other nutrients are extensive and may well be important in the determination of biological availability. The retention of magnesium and calcium were shown to be reduced by fatty acid supplementation (Leeson and Atteh, 1995), while high levels of calcium have resulted in a reduction in the absorption of manganese, zinc and calcium (Shafey *et al.*, 1991). Fats have been reported to reduce mineral absorption with the formation of insoluble soaps when cations come in contact with free fatty acids released during digestion as the proposed mechanism (Whitehead *et al.*, 1971). Meat type poultry are prone to losses associated with very rapid growth. Skeletal abnormalities and sudden death are examples of maladies that result in losses from mortality, condemnation and down grading. Rotter *et al.* (1985) observed differences in tissue copper, calcium and zinc in birds on treatments differing in dietary fatty acids and that succumbed to sudden death syndrome. Heat distress periodically results in sudden death losses of market

Table 1: Composition of experimental diets

Ingredient	0.93 % Ca	1.5 % Ca
Corn	51.35	51.35
Soybean meal 49 %	36.5	36.5
Fat[a]	4.7	4.7
Dicalcium phosphate	2.0	2.0
Limestone	1.0	2.5
Vitamin-trace mineral mix[b]	1.0	1.0
Salt	0.4	0.4
DL-methionine	0.15	0.15
Coccidiostat	0.1	0.1
Sand[c]	2.8	1.3

[a]Fat sources were either corn oil, animal fat, fish oil or a dry blend of animal and vegetable fat. Amounts varied in order to make diets isocaloric.
[b]Supplied per kilogram of diet: copper, 8 mg; iodine, 0.4 mg; iron, 100 mg; selenium, 0.3 mg; zinc, 75 mg; vitamin A (retinyl acetate), 4540 IU; vitamin D_3, 1543 ICU; vitamin E, 15 IU; choline, 284 mg; niacin, 34 mg; d-pantothenic acid, 5.7 mg; menadione, 0.85 mg; vitamin B_{12}, 0.01 mg; biotin, 0.1 mg; folic acid, 0.5 mg; thiamine, 0.6 mg.
[c]Amount of sand varied at the expense of fat in order to make diets isocaloric.

Table 2: Effect of calcium level, fat source[1] and temperature[2] on body weight gain, feed consumption and feed efficiency

Fat source	Calcium level	Gain (g/day)		Feed (g/day)		Feed Efficiency	
		TN	HD	TN	HD	TN	HD
CO	0.93	48.5	31.8	117.9	113.7	0.41	0.29
	1.50	46.7	33.3	121.8	115.3	0.38	0.30
AF	0.93	50.0	34.2	122.7	112.6	0.41	0.31
	1.50	48.2	35.3	116.8	112.7	0.41	0.32
FO	0.93	46.4	29.0	123.1	115.8	0.38	0.26
	1.50	47.7	29.6	118.2	110.9	0.40	0.28
DB	0.93	42.9	30.3	123.3	109.4	0.35	0.27
	1.50	41.8	32.2	123.4	115.9	0.34	0.28
SEM		0.44		1.03		0.004	

ANOVA	df	Probabilities		
Temp	1	0.00004	0.0002	0.0001
Calcium	1	0.80	0.82	0.79
Fat	3	0.002	0.93	0.001
Temp x Calcium	1	0.22	0.53	0.52
Temp x Fat	3	0.05	0.82	0.11
Calcium x Fat	3	0.95	0.37	0.68
Temp x Cal x Fat	3	0.83	0.82	0.79
Main Effects				
Temperature				
TN		46.5[a]	120.9[a]	0.39[a]
HD		31.9[b]	113.3[b]	0.29[b]
Calcium				
0.93		39.3	117.3	0.34
1.50		39.1	116.9	0.34
Fat				
CO		40.0[ab]	117.1	0.35[ab]
AF		42.0[a]	116.2	0.37[a]
FO		38.2[bc]	117.0	0.33[b]
DB		36.8[c]	117.9	0.32[b]

[1]CO = corn oil; AF = animal fat; FO = fish oil; DB = dry blend (animal and vegetable fat); [2]TN = thermoneutral, 23.9 °C; HD = heat distress, 23.9 – 35 °C; [abc]Means in columns within main effect differ significantly ($P < 0.05$)

age broilers. It is possible that some of these losses are as a result of mineral inadequacies during heat stress.

The study reported here was conducted to evaluate the effects of diets differing in fat source and calcium level on production performance and mineral utilization in broilers reared under high temperature stress.

Materials and Methods

Animals and Diets: Two hundred and forty, 1-day old male broilers (commercial strain) were used in this experiment. Birds were randomly assigned to eight treatments with thirty birds per treatment and raised on floor pens in a brooder house for 21 days posthatching. The eight dietary treatments were achieved by using one of four different fat sources in the formulation of the experimental diets (Table 1): corn oil (CO), animal fat (AF), fish oil (FO), and a dry blended (animal and vegetable) fat product (DB) at either 0.9 or 1.5 % calcium. Diets were formulated to contain 3066 kcal/kg of metabolizable energy and 22.3 % crude protein and were isocaloric with 14.7 % of the calories from the individual fat source. The calorie:protein ratio was 137 for each diet while the Ca:P ratio was 1.82 for the low 0.93 % calcium diet and 2.9 for the 1.5 % calcium diet. Chicks were allowed ad libitum access to feed and water.

Growth Environment: On day 22 of the experiment, 15 chicks per treatment were randomly placed in individual 60 x 40 x 45 cm wire cages within each of two environmental chambers under controlled temperature and humidity. Each chamber was 14.6 x 7.3 m and contained four rows of single-level cages placed 91 cm off the floor. Cages in each chamber were fitted with feed and cup-type water dispensing equipment. During week 4 of the experiment, birds were adapted to chamber surroundings and peak chamber temperature in the cycling high temperature environment increased at the rate of 2 °C per day to a high of 35 °C. The temperature in one chamber was maintained at 23.9 °C constant temperature (thermoneutral, TN). For each 24 hour period in the

Table 3: Effect of calcium level, fat source[1] and temperature[2] on plasma mineral concentration

Fat source	Dietary calcium	Calcium (mg/dL)		Magnesium (mg/dL)		Copper (µg/dL)		Zinc (µg/dL)	
		TN	HD	TN	HD	TN	HD	TN	HD
CO	0.93	8.12	10.17	2.02	2.09	14.15	14.60	140.8	98.5
	1.50	10.25	11.25	2.00	2.15	14.95	13.15	118.9	82.8
AF	0.93	8.94	10.47	2.08	2.14	14.75	14.70	123.7	100.4
	1.50	10.23	10.83	2.15	2.16	15.25	14.65	113.7	307.7
FO	0.93	8.35	11.96	1.78	2.35	16.50	15.68	113.8	69.4
	1.50	9.92	11.30	2.07	2.08	13.80	12.10	106.2	64.4
DB	0.93	11.48	10.74	2.16	2.32	11.64	14.61	103.2	77.8
	1.50	10.08	11.54	2.03	2.13	15.20	18.15	107.7	79.3
SEM		0.28		0.04		0.89		20.5	

ANOVA	df	Probabilities			
Temp	1	0.0003	0.0002	0.87	0.82
Calcium	1	0.07	0.009	0.94	0.46
Fat	3	0.20	0.49	0.96	0.16
Temp x Cal	1	0.47	0.15	0.68	0.28
Temp x Fat	3	0.21	0.35	0.53	0.23
Cal x Fat	3	0.29	0.48	0.22	0.34
T x C x F	3	0.15	0.15	0.98	0.34
Main Effects					
Temperature					
TN		9.67[b]	2.04[b]	14.53	116.0
HD		11.04[a]	2.18[a]	14.70	112.1
Calcium					
0.93		9.94[b]	2.11	14.52	105.1
1.50		10.70[a]	2.09	14.66	122.6
Fat					
CO		9.94	2.06	14.12	110.8
AF		10.12	2.13	14.84	161.4
FO		10.21	2.04	14.41	91.9
DB		10.96	2.16	14.90	90.2

[1]CO = corn oil; AF = animal fat; FO = fish oil; DB = dry blend (animal and vegetable fat); [2]TN = thermoneutral, 23.9 °C; HD = heat distress, 23.9 – 35 °C; [ab]Means in columns within main effect differ significantly ($P < 0.05$)

second chamber, the environmental temperature cycled from a low of 23.9 °C to a high of 35 °C (heat distress HD). Birds were exposed to 8 hours of 23.9 °C, 4 hours of 23.9 to 35 °C, 4 hours of 35 °C, and 8 hours of 35 to 23.9 °C. The relative humidity in the HD environment peaked at 60% during the daily heat episodes. Birds were weighed weekly and feed consumption determined. At the end of the experimental period, body weight gain, total feed consumption and feed efficiency were calculated.

Plasma Analysis: On day 42, six birds from each treatment and growth environment were bled by brachial venipuncture using EDTA as an anticoagulant. Following blood collection, samples were centrifuged at 3000x g for 15 minutes to separate supernatant (plasma) from red blood cells. Plasma Ca, Mg, Cu and Zn were determined by using a flame atomic absorption spectrophotometer.

Relative Mineral Retention: Daily feed consumption was recorded on days 43, 44 and 45. On day 46, birds were fasted for 24 hours following which each bird was allowed to consume 25 % of its average daily feed intake and then fasted for another 12 hours. Total bird excreta was collected during the feeding and second fasting periods. Excreta samples were dried, ground and analyzed for mineral content. Feed samples were subjected to proximate analysis and mineral contents. Calcium, Mg, and Cu were determined by flame atomic absorption spectrophotometer. All samples were analyzed in duplicate. The difference in the mineral content of the consumed feed and the excreted feces was used to calculate relative mineral retention.

Tibia Mineral Analysis: Birds were killed on day 49 and the left tibia removed. All adhering tissue was removed and bones were dried, ashed and analyzed for ash minerals (AOAC, 1990). Bone Ca, Mg, Cu and Zn were determined by flame atomic absorption spectrophotometry.

Statistical Analysis: Data were subjected to analysis of

Table 4: Effect of calcium level, fat source[1] and temperature[2] on tibia mineral concentration

Fat source	Dietary calcium	Calcium (mg/g)		Magnesium (mg/g)		Copper (µg/g)		Zinc (µg/g)	
		TN	HD	TN	HD	TN	HD	TN	HD
CO	0.93	369	387	8.45	8.25	3.84	4.93	415	421
	1.50	365	370	8.06	7.98	4.21	6.21	445	413
AF	0.93	365	356	8.33	8.05	4.17	4.51	416	412
	1.50	368	371	8.35	8.23	5.03	5.14	414	441
FO	0.93	354	359	8.19	8.49	4.18	4.80	393	384
	1.50	360	364	8.41	7.91	4.74	4.28	391	408
DB	0.93	357	369	8.41	8.44	4.62	4.44	427	419
	1.50	365	373	8.10	8.17	3.96	4.17	437	431
SEM		2.62		0.07		0.1 9		5.61	

ANOVA	df	Probabilities			
Temp	1	0.08	0.25	0.06	0.86
Calcium	1	0.30	0.04	0.25	0.10
Fat	3	0.05	0.88	0.46	0.005
Temp x Cal	1	0.62	0.47	0.98	0.71
Temp x Fat	3	0.39	0.75	0.09	0.62
Cal x Fat	3	0.37	0.25	0.20	0.99
T x C x F	3	0.35	0.17	0.54	0.28
Main Effects					
Temperature					
TN		363	8.28	4.34	417
HD		363	8.17	4.81	417
Calcium					
0.93		363	8.32[a]	4.41	412
1.50		367	8.15[b]	4.72	422
Fat					
CO		372[a]	8.18	4.79	423[ab]
AF		365[ab]	8.24	4.71	420[ab]
FO		359[b]	8.22	4.47	395[b]
DB		366[ab]	8.28	4.3 0	428[a]

[1]CO = corn oil; AF = animal fat; FO = fish oil; DB = dry blend (animal and vegetable fat); [2]TN = thermoneutral, 23.9 °C; HD = heat distress, 23.9 – 35 °C; [ab]Means in columns within main effect differ significantly ($P < 0.05$)

variance using the General Liner Models procedures of SAS® software (SAS Institute, 1987) with the main effects being calcium level, fat source and environment. Least square treatment means were compared if a significant F statistic (5 % level of probability) was detected by analysis of variance (Steel and Torrie, 1960).

Results

Body Weight Gain, Feed Consumption and Feed Efficiency: The effects of calcium level and fat source in two growth environments on body weight gain, feed consumption and feed efficiency are shown in Table 2. Both temperature and fat source, but not calcium level, had an effect on body weight gain. Birds in the heat distress environment gained 31% less weight ($p < 0.05$) than their counterparts raised in the thermoneutral environment. Fat source also affected body weight gain with birds fed the diet containing animal fat gaining significantly more weight than those consuming diets containing fish oil or the dry blend of animal and vegetable fats. Feed consumption and feed efficiency were greater in birds reared in the thermoneutral environment, while the greatest feed efficiency was exhibited by birds consuming animal fat. Calcium level had no effect on the performance parameters measured.

Plasma Minerals: Table 3 shows a summary of the effects of dietary calcium level, fat source and environmental temperature on plasma mineral content. Plasma calcium and magnesium were significantly ($p < 0.05$) elevated in the heat distress environment while the higher level of dietary calcium increased plasma calcium level. There was no effect of fat source on plasma mineral content nor was there any two-way or three-way interactions.

Table 5: Effect of calcium level, fat source[1] and temperature[2] on relative mineral retention[3]

Fat source	Dietary calcium	Calcium		Magnesium		Copper	
		TN	HD	TN	HD	TN	HD
		relative retention %					
CO	0.93	44.4	29.9	32.5	27.4	30.8	31.9
	1.50	35.6	48.3	22.3	36.5	28.6	22.0
AF	0.93	47.6	41.6	29.0	30.5	44.1	28.3
	1.50	43.5	48.9	38.2	45.5	41.2	50.9
FO	0.93	46.1	51.5	32.7	38.8	19.3	22.3
	1.50	42.9	45.9	28.0	41.0	20.4	26.0
DB	0.93	39.8	40.5	24.2	31.7	21.0	20.6
	1.50	45.1	40.3	25.8	28.3	31.0	21.8
SEM		2.37		2.02		2.54	
ANOVA	df	Probabilities					
Temp	1	0.95		0.02		0.92	
Calcium	1	0.71		0.35		0.19	
Fat	3	0.31		0.06		0.0001	
Temp x Cal	1	0.20		0.18		0.95	
Temp x Fat	3	0.89		0.88		0.84	
Cal x Fat	3	0.75		0.17		0.21	
T x C x F	3	0.20		0.41		0.08	
Main Effects							
Temperature							
TN		43.1		29.0[b]		28.3	
HD		43.3		34.9[a]		27.9	
Calcium							
0.93		42.7		30.8		26.1	
1.50		43.8		33.2		30.2	
Fat							
CO		39.6		29.7		28.4[b]	
AF		45.3		35.8		41.1[a]	
FO		46.6		35.1		22.0[b]	
DB		41.4		27.5		21.1[b]	

[1]CO = corn oil; AF = animal fat; FO = fish oil; DB = dry blend (animal and vegetable fat); [2]TN = thermoneutral, 23.9 °C; HD = heat distress, 23.9 – 35 °C; [ab]Means in columns within main effect differ significantly ($P < 0.05$)
[3]Feed mineral content minus fecal mineral content

Tibia Minerals: Tibia bones were used to evaluate the temperature, dietary calcium level and fat source on bone deposition of minerals (Table 4). Growing temperature had no effect on tibia mineral content. However, the higher dietary calcium level had a significant negative impact on bone magnesium but did not elevate bone calcium. Birds consuming the fish oil diet had significantly ($p < 0.05$) less bone calcium and zinc than those consuming other diets containing fat from other sources.

Relative Mineral Retention: The effects of dietary calcium level, fat source and growing temperature are summarized in Table 5. Temperature had a significant ($p < 0.05$) effect on the relative retention of magnesium while fat source affected the relative retention of copper. Birds consuming diets containing animal fat retained a greater percentage of copper than those consuming diets containing fats from other sources. There was no effect of calcium level on relative mineral retention.

Discussion

Calcium is necessary for maintenance of bone integrity, however, decreased retention of calcium in chicks have been reported with increasing dietary fat content (Pepper *et al*., 1955). Other researchers (Biely and March, 1967) indicated that dietary fat levels as high as 12 % did not increase calcium requirement while still others (Lipstein and Bornstein, 1968) indicated that supplemental fat type had no effect on bone calcification. The study reported here evaluated the effects of calcium level, fat source and environmental temperature on growth performance and mineral utilization. High environmental temperature had significant detrimental impact on growth performance and feed utilization. This is consistent with studies reported previously (Smith, 1994; Smith and Teeter, 1993). The elevation of plasma calcium and magnesium concentration in the heat distressed growing environment may be as a result of birds losing significant amounts of water through the panting process thereby concentrating the blood mineral content. Increased dietary calcium level resulted in increased plasma but not bone calcium concentration. Tallow improved both body weight gain and feed efficiency while fish oil decreased tibia calcium and zinc concentration when compared with other sources of fat. There was no significant interaction between dietary calcium level and fat source for any of the parameters examined. While it is recognized that the use of fat in the diets of heat distressed poultry is desirable, the source of fat used should perhaps depend on economic considerations.

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