Comparison of Dietary 25-Hydroxycholecalciferol and Cholecalciferol in Broiler Chickens

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ABSTRACT We conducted a series of 10 feeding trials involving over 36,000 broilers. The effects of various dietary levels of cholecalciferol (vitamin D_3) or 25-hydroxycholecalciferol (25-OH-D_3) were compared using a "basal" dosage level of 69 μg/kg feed, as well as levels ranging from .5 to 1.5 times the basal level. For all 10 studies, average body weight increased by an average of .042 ± .03 kg ($P < .001$) and adjusted feed efficiency decreased (improved) by an average of .026 ± .0046 kg/kg ($P < .001$) in birds fed 25-OH-D_3 in comparison to those fed vitamin D_3 at the basal level. Changes in mortality were not detected. Evaluation of different dietary levels of 25-OH-D_3 revealed a significant dose-response relationship, with maximal effects on weight gain, feed efficiency, and breast meat yield being observed in the range of 50 to 70 μg/kg feed. Preliminary studies with different levels of vitamin D_3 suggested no additional benefits on weight gain or feed efficiency with higher dietary levels of vitamin D_3. Serum 25-OH-D_3 concentrations increased more rapidly in birds fed 25-OH-D_3 than in birds fed vitamin D_3. There were significant correlations with body weight, feed conversion, and serum 25-OH-D_3 concentrations, with no correlations observed between serum 1,25-$(OH)_2D_3$ concentrations and these variables.

(*Key words*: broiler performance, cholecalciferol, 25-hydroxycholecalciferol, serum concentrations, feeding studies)

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INTRODUCTION

The United States poultry broiler industry continues to be one of the most intensively managed industries in the world for the production of animal protein. Because chickens in many commercial broiler operations are grown in the presence of limited sunlight, cholecalciferol (vitamin D_3) must be provided in the diet in levels ranging from 2,200 to 3,300 IU/kg (55 to 83 μg/kg) of feed to prevent rickets and other bone problems and optimize growth and performance variables such as feed efficiency and weight gain (McNaughton, 1990; Dudley-Cash, 1994). In order for vitamin D_3 to carry out its physiological functions, it must undergo a two-step hydroxylation, first in the liver at the 25 position to produce 25-hydroxycholecalciferol (25-OH-D_3), the major circulating form of vitamin D_3, followed by a second hydroxylation in the kidneys to produce 1,25-dihydroxycholecalciferol (1,25-$(OH)_2$-D_3), the biologically active, hormonal form of vitamin D_3 (Norman, 1985; DeLuca, 1988). It is this form of vitamin D_3 that interacts with a specific receptor, found in a wide variety of tissues, to carry out the

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functions of vitamin D_3, which include calcium transport and cellular differentiation (Walters, 1992).

This greater understanding of the metabolism and mechanism of action of vitamin D_3 has led to an evaluation of several vitamin D_3 metabolites as sources of vitamin D_3 activity in broiler rations (Boris *et al.*, 1977; Cantor and Bacon, 1978). It is thought that many broilers are raised under stressful conditions, including bird density, heat stress, and diseases such as mycotoxicosis, enteritides, malabsorption syndrome, and certain immune disorders. These conditions may impair absorption or liver hydroxylation of vitamin D_3. This provides a rationale for the use of 25-OH-D_3 in broiler rations. The use of 25-OH-D_3 bypasses the requirement for liver hydroxylation but still avoids adverse side effects because of the tight regulation of the renal 1α-hydroxylase and the discrimination of the vitamin D_3 receptor against the 25-hydroxy metabolite in comparison to 1,25-$(OH)_2D_3$ (Hughes *et al.*, 1976; Walters, 1992).

Previous studies of the use of 25-OH-D_3 as a source of vitamin D_3 activity in broiler rations have suggested that feeding of this metabolite results in improved weight gain and feed efficiency in comparison to feeding the same amount (wt/wt) of vitamin D_3 (McNutt and Haussler, 1973; Cantor and Bacon, 1978). However, many of these studies were limited by the high cost and limited availability of 25-OH-D_3, which has prevented thorough evaluation of the use of this form of vitamin D_3 under commercial conditions. Amoco BioProducts Corporation has recently developed technology that has made it feasible to synthesize 25-OH-D_3 in kilogram quantities using a proprietary process. With the availability of large quantities of 25-OH-D_3 in a stable, formulated form, the present series of studies was undertaken to evaluate the efficacy of 25-OH-D_3 as a source of vitamin D_3 activity in poultry broiler rations under commercial conditions.

MATERIALS AND METHODS

Materials

25-Hydroxycholecalciferol was synthesized by Amoco BioProducts Corporation by photolysis of 25-hydroxyprovitamin D_3. The starting material for this process, cholesta-5,7,24-triene-3b-ol (cholestatrienol) was isolated using a proprietary yeast strain and the 25-hydroxyprovitamin D_3 was synthesized from the cholestatrienol also by a proprietary process. This process results in the production of kilogram quantities of 25-OH-D_3, which is stabilized through formulation into food grade, hydrogenated vegetable oil based beadlets containing butylated hydroxytoluene and citric acid as antioxidants.[6] The formulated beadlet (containing 50 mg of 25-OH-D_3/g of beadlet) was further diluted into a premix containing 138 mg of 25-OH-D_3/kg using commercially available rice hulls as an inert carrier. A similar premix containing 138 mg vitamin D_3/kg was obtained from Hoffmann-LaRoche.[7]

Feed Trials, Animals, Diets, and Treatments

We conducted a series of 10 feeding trials at two locations over a 4-yr period (Table 1) with 8 of the 10 studies completed during the past 2 yr. Five of the studies (Studies 1 and 6 to 9) were carried out at PARC Institute in Maryland and the other five studies (Studies 2 to 5 and 10) were done at Colorado Quality Research at Fort Collins, CO. To simulate commercial conditions, all of the studies were floor pen studies involving 5 to 10 pens per treatment and 50 to 90 birds per pen (bird density .7 ft^2 per bird, .065 m^2 per bird). Two common commercial broiler strains were used: Arbor Acres × Arbor Acres and Peterson × Arbor Acres. All birds were fed commercial starter, grower, and finisher broiler rations containing 23, 20, and 18% protein for periods of approximately 10, 28, and 9 d, respectively. Representative diet compositions from each location are shown in Table 2. Calcium and available phosphorus levels ranged from .8 to 1% and .35 to .46% in all diets, respectively (Table 2). All other nutrients were added at NRC (1984) recom-

[6]Particle Dynamics, St. Louis, MO 63144.
[7]Hoffmann-LaRoche, Nutley, NJ 07110.

TABLE 1. Broiler trials comparing dietary 25-hydroxycholecalciferol and cholesterol

Trial	Location[1]	Date completed	Duration	Breed[2]	Pens per treatment	Birds per pen
			(d)			
1	PARC	March 1990	46	P × AA	5	80
2	CQR	September 1991	49	AA × AA	10	50
3	CQR	April 1992	48	AA × AA	10	60
4	CQR	November 1992	50	AA × AA	8	70
5	CQR	September 1993	48	AA × AA	5	56
6	PARC	September 1993	47	P × AA	10	74
7	PARC	September 1993	46	P × AA	10	74
8	PARC	December 1993	47	P × AA	10	74
9	PARC	January 1994	47	P × AA	10	90
10	CQR	January 1994	52	P × AA	10	60

[1]PARC = PARC Institute; CQR = Colorado Quality Research.

[2]P × AA = Peterson × Arbor Acres; AA × AA = Arbor Acres × Arbor Acres.

mended levels and all grower and finisher diets were pelletized to simulate industry practice. In all but one study (Study 5), the diets contained an antibiotic and coccidiostat. In all studies, the chicks were also vaccinated for Marek's disease and were given Newcastle Disease-Infectious Bronchitis vaccine via water at 5 d. In the early studies, diets contained either vitamin D_3 or 25-OH-D_3 at a level of 69 μg/kg, a level of vitamin D_3 commonly used in the industry (McNaughton, 1990). In the later studies, (6 to 10), different levels of 25-OH-D_3 ranging from .5 to 1.5 times the basal level of 69 μg/kg were used in order to obtain additional dose-response information for the variables studied. In two of these studies (8 and 9), additional levels of vitamin D_3 ranging from 1.25 to 1.5 times the basal level were also studied.

Measurements

Average body weights as well as feed conversion efficiency were measured on a per pen basis. Feed efficiency was calculated in two ways: 1) on an adjusted basis by dividing the weight of the feed for each pen by the total weight of the birds in each pen plus the weight of the birds that had died during the study; and 2) on an unadjusted basis by simply dividing the weight of the feed per pen by the weight of the birds per pen. The former calculation is a method for comparing results from different studies with different mortalities, whereas the latter method is more common in the industry, as mortality tends to be more constant at a given facility. Mortality of the birds was monitored twice daily in all of the studies. All of the birds that were removed from the pens due to mortality or humane reasons were necropsied. In eight of the studies, a subset of the birds (4 to 12 per pen) were processed for carcass evaluation. Breast meat was expressed as percentage of live body weight and analyzed on a pen basis. In two of the studies (6 and 7), serum samples were collected from one male and one female from each pen for measurement of vitamin D metabolite concentrations. Serum 25-OH-D_3 concentrations were measured after acetonitrile extraction by radioimmunoassay using a commercially available kit from INCSTAR (Hollis *et al.*, 1993). Serum 1,25-$(OH)_2$-D_3 concentrations were measured by radioreceptor assay as previously described (Reinhardt *et al.*, 1984; Hollis, 1986). Intra- and interassay CV were less than 13% (Hollis, 1986).

Statistical Analysis

Statistical analysis was performed using the ANOVA that was appropriate for the design of the study. One-way ANOVA were used for comparisons of different vitamin D_3 and 25-OH-D_3 levels, but some studies also included additional experimental factors. Experimental errors for statisti-

TABLE 2. Representative starter broiler rations for 10 feeding trials at PARC Institute (PARC) and Colorado Quality Research (CQR)

Ingredients and analysis	PARC			CQR		
	Starter	Grower	Finisher	Starter	Grower	Finisher
	(%)					
Corn	56.2	65.2	71.9	54.9	65.1	72.2
Soybean meal	32.5	26.7	21.1	33.15	25	19.3
Fat	4.23	2.96	1.7	4.5	2.9	1.75
Meat meal	5	3	3.3	5	5	5
Limestone	.413	.61	.78	.67	.65	.55
Salt	.367	.32	.35	.35	.35	.35
Defluorinated phosphate	.908	.99	.72	.45	.2	.05
Methionine	.141	.063	.02	.21	.1	.07
Trace mineral premix[1]	.05	.05	.05	.25	.25	.25
Vitamin premix[1]	.05	.05	.05	.5	.5	.5
Calculated analysis						
ME, kcal/g	3.2	3.2	3.2	3.2	3.2	3.2
CP	23	20	18	23	20	18
Calcium	.85	.82	.8	1	.91	.81
Available P	.45	.4	.35	.46	.4	.37
TSAA	.93	.76	.67	.94	.73	.63
Lysine	1.2	1	.85	1.3	1.1	.91

[1]Vitamin and mineral premixes at each location were added to supply the following to each kilogram of finished feed: vitamin A, 7,000 IU/kg; vitamin E, 27 IU/kg; menadione, 3.5 mg; thiamine, 2.9 mg; riboflavin, 7 mg; pantothenic acid, 17 mg; biotin, 289 μg; folic acid, 1.13 mg; choline chloride, 600 mg; potassium, 7,022 mg; magnesium, 1,481 mg; sulfur, 1,560 mg; vitamin B_{12}, 16 μg; niacin, 52 mg; pyridoxine, 3.27 mg; manganese, 75 mg; iron, 153 mg; copper, 21 mg; zinc, 44 mg; selenium, 177 μg; and iodine, 383 μg.

cal tests were pooled across all treatments. For the results presentation, (Tables 3 to 6), only the comparison of the trial averages for the 69 μg/kg dietary level is shown, because this was the common level comparison across all 10 trials. The significance level was computed using a one-sided least significant difference test (Daniel, 1995). The sample size for each treatment mean is the number of pens for the treatments, which are shown in Table 1.

RESULTS

Performance Characteristics at a Normal Commercial Dietary Level

Tables 3 to 6 show a comparison of performance characteristics of broiler chickens fed either vitamin D_3 or 25-OH-D_3 at the basal level of 69 μg/kg feed. In 9 of the 10 studies, average body weight was greater ($P \leq .05$) in birds fed 25-OH-D_3 than in those fed vitamin D_3 (Table 3). The birds fed 25-OH-D_3 weighed an average of .04 ± .03 kg ($P < .01$) more than birds fed the same level of vitamin D_3. In 7 of the 10 studies, adjusted feed efficiency was lower ($P \leq .05$) in birds fed 25-OH-D_3 than in those fed vitamin D_3 (Table 4). Adjusted feed efficiency fell by an average of .026 ± .0046 ($P < .01$) kg feed/kg bird in the birds fed 25-OH-D_3. As expected, unadjusted feed efficiency was more variable and was lower ($P \leq .05$) in five of the studies in birds fed 25-OH-D_3 (Table 4). However, the average change of .039 ± .072 kg/kg was greater than that for adjusted feed efficiency. Mortality was variable in all of the studies and there were no treatment-related differences in mortality (Table 5). Pathologic examination of the dead birds from all trials revealed no treatment-related abnormalities. In three of the eight studies in which it was evaluated, percentage breast meat was greater ($P \leq .05$) in birds fed 25-OH-D_3 (Table 6). Percentage breast meat increased by an average of .53 ± .56% in all eight studies.

Effects at Different Dietary Levels

Figures 1 to 4 show the effects of various dietary levels of 25-OH-D_3 on the perfor-

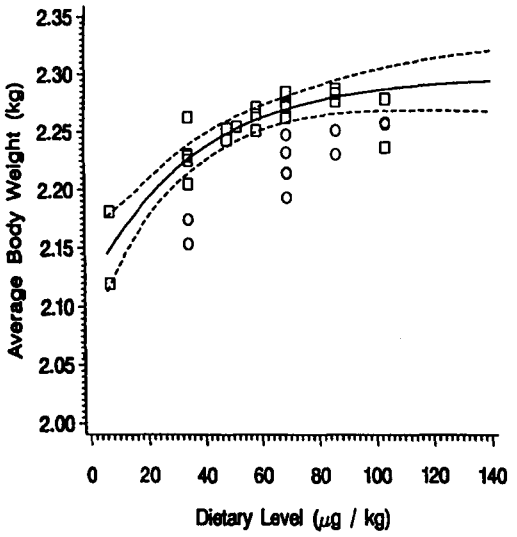

FIGURE 1. Effect of various dietary levels of cholecalciferol (vitamin D_3) or 25-hydroxycholecalciferol (25-OH-D_3) on average body weight. Data at various dietary levels of 25-OH-D_3 (□) from the five most recent studies (Studies 6 to 10) were fitted to the general equation: $y = a - be^{-c(x)}$ where y = average body weight; a = the theoretical maximum body weight (intercept); b = the change in body weight from the initial value; c = rate of change in body weight (slope); and x = the dietary 25-OH-D_3 level. The regression line shown is the mean regression line for all five studies and the dotted lines show the 95% confidence limits for the mean regression line. Data from vitamin D_3 treatments (o) are also shown on the same plot, but because of fewer data points, these were not fitted to the equation.

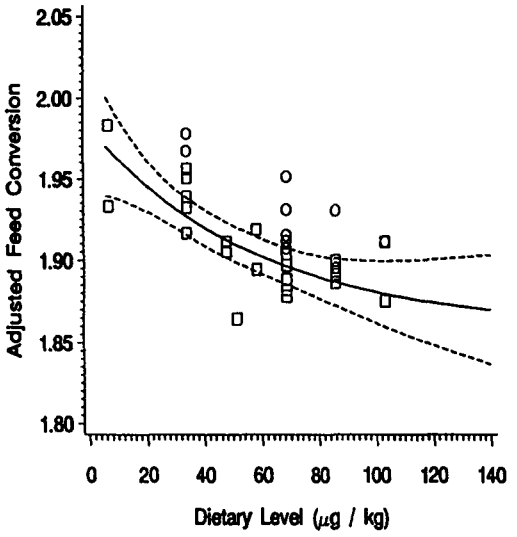

FIGURE 2. Effect of various dietary levels of cholecalciferol (vitamin D_3) or 25-hydroxycholecalciferol (25-OH-D_3) on adjusted feed efficiency. Data were fitted as described in Figure 1 except that the equation $y = a + be^{-c(x)}$ was used because of the fall in adjusted feed efficiency with increasing dietary level. Regression line with 95% confidence limits is drawn as in Figure 1.

mance characteristics evaluated above. These data were taken from the later studies (6 to 10) in which different dietary levels of vitamin D_3 and 25-OH-D_3 were fed. Average body weight increased and adjusted feed efficiency decreased with increasing dietary level of 25-OH-D_3 up to approximately 50 to 70 μg/kg feed (Figures 1 and 2). Unadjusted feed efficiency showed a similar relationship to dietary 25-OH-D_3 level as adjusted feed efficiency (not shown). As expected, there was no relationship between percentage mortality and dietary level of 25-OH-D_3 (Figure 3). Percentage breast meat also increased with increasing dietary level of 25-OH-D_3 (Figure 4).

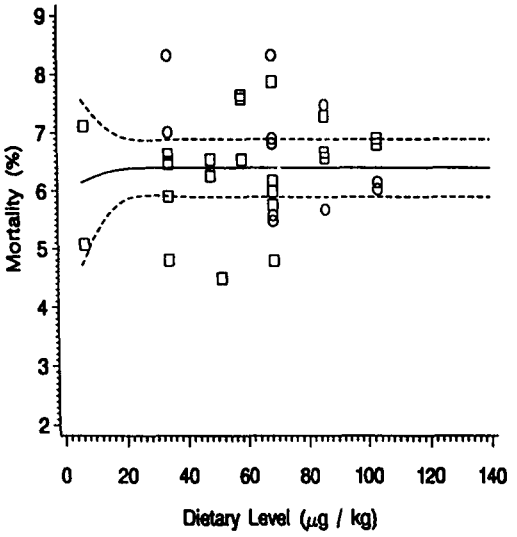

FIGURE 3. Effect of various dietary levels of cholecalciferol (vitamin D_3) or 25-hydroxycholecalciferol (25-OH-D_3) on percentage mortality. Data were taken from the five most recent studies and fitted as in Figure 1.

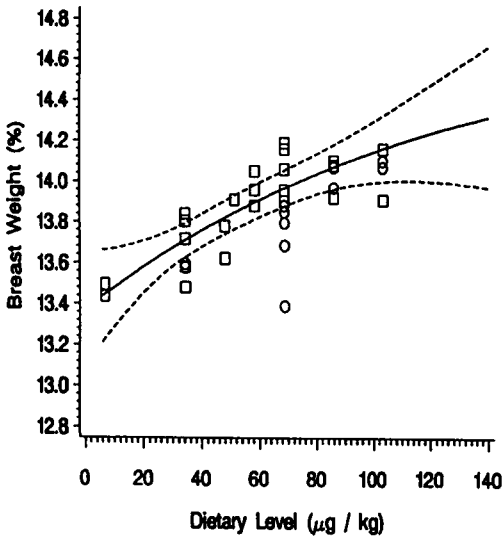

FIGURE 4. Effect of various dietary levels of cholecalciferol (vitamin D_3) or 25-hydroxycholecalciferol (25-OH-D_3) on percentage breast meat. Data were taken from the five most recent studies and fitted as in Figure 1.

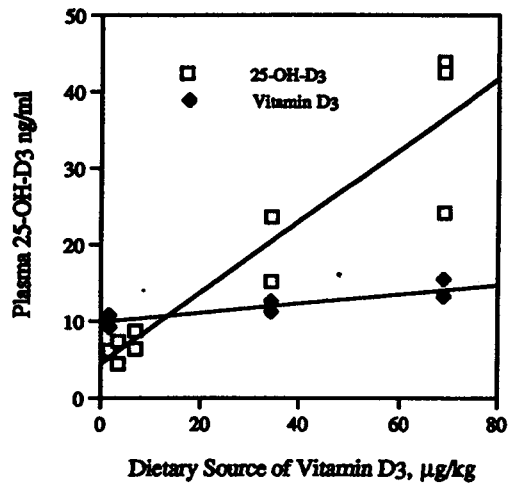

FIGURE 5. Effect of various dietary levels of cholecalciferol (vitamin D_3) (♦) or 25-hydroxycholecalciferol (25-OH-D_3) (□) on serum 25-OH-D_3 concentrations. Data were taken from Studies 6 and 7 and fitted to a linear regression model.

Vitamin D Metabolite Concentrations

In the two studies in which vitamin D_3 metabolite measurements were made, serum 25-OH-D_3 concentrations increased as expected with increasing dietary source of vitamin D_3 (Figure 5). The relationship for dietary 25-OH-D_3 was: serum 25-OH-D_3, ng/mL, = 4.4 + .465 × dietary 25-OH-D_3, µg/kg, ($r = .9$, $P < .01$), and that for dietary vitamin D_3 was: serum 25-OH-D_3, ng/mL, = 10 + .06 × dietary vitamin D_3, µg/kg, ($r = .9$, $P < .01$). These results would indicate that as dietary vitamin D_3 or 25-OH-D_3 increases, serum 25-OH-D_3 concentrations increase more rapidly in birds fed 25-OH-D_3 (slope = .465 ng/mL per µg/kg) than in those fed vitamin D_3 (slope = .06 ng/mL per µg/kg), so that at the "basal" level of 69 µg/kg serum 25-OH-D_3 concentrations averaged 35 ± 16 ng/mL and 14 ± 5 ng/mL ($P < .01$) in birds fed 25-OH-D_3 or vitamin D_3, respectively. Serum 1,25-$(OH)_2$-D_3 concentrations averaged 81 ± 20 pg/mL (n = 200 all treatments) and 84 ± 18 pg/mL (n = 120 all treatments) in birds fed 25-OH-D_3 or vitamin D_3, respectively, and did not change with dietary levels of either source of vitamin D_3 activity in the dietary range tested.

DISCUSSION

The present studies clearly document a consistent effect of 25-OH-D_3 to increase body weight and decrease feed efficiency (either adjusted or unadjusted) in broiler chickens grown under simulated commercial conditions. This is an extension of a previous study of caged broilers utilizing fewer birds per treatment and lower dietary levels of vitamin D_3 (Cantor and Bacon, 1978). Although the absolute magnitude of the changes in weight (.04 kg, 1.7%) and feed efficiency (changes of .026 adjusted or .039 unadjusted) are small, the potential benefits to the poultry broiler industry are considerable due to the large number of birds grown in the United States (7×10^9 birds per year) and the large amount of feed consumed (32×10^6 tons/yr, 1993). The dose-response relationships for these performance variables (Figures 1 and 2) lend further support to a cause and effect relationship between dietary 25-OH-D_3 and improvement in weight gain and feed efficiency. Preliminary studies with higher dietary levels of vitamin D_3 would suggest that the same benefits cannot be obtained with greater vitamin D_3 supplementation. The increase

TABLE 3. Average body weight (46 to 52 d) for broilers fed either 25-hydroxycholecalciferol (25-OH-D_3) or cholecalciferol (vitamin D_3) at a level of 69 μg/kg

Trial	Mean BW		Change	SD[1]	*P*
	25-OH-D_3	Vitamin D_3			
	(kg)				
1	2.10	2.07	.03	.028	.05
2	2.58	2.58	0	.024	NS
3	2.54	2.51	.03	.021	.01
4	2.52	2.48	.04	.026	.01
5	2.50	2.43	.08	.027	.01
6	2.06	2.02	.04	.052	.04
7	2.09	2.06	.03	.038	.04
8	2.05	2.03	.02	.027	.02
9	2.39	2.30	.09	.038	.01
10	2.74	2.67	.07	.037	.01
Weighted average[2]			.04	.033	

[1]Standard deviation represents the average SD of each treatment.

[2]Weighting was done taking into account the variance in each trial and the number of birds in each trial.

in breast meat with dietary 25-OH-D_3 is unique. Previous studies have shown that more breast meat is produced as the birds become larger (Walters *et al.*, 1963). However, in the present study there was a significant increase in percentage breast meat in two of the studies (3 and 4, Table 6) in spite of only a small increase in body weight in the birds fed 25-OH-D_3 (Table 3). Substitution of 25-OH-D_3 for vitamin D_3 as a source of dietary vitamin D_3 clearly had no adverse effects on survival (Table 5 and Figure 4) and no adverse effects were detected upon gross pathologic examination of the dead birds. In 6 of the 10 studies, percentage mortality was lower in birds fed 25-OH-D_3 than in those fed the same amount of vitamin D_3, but the changes were not significant.

The possible mechanism by which dietary 25-OH-D_3 may affect weight gain or feed efficiency of broilers cannot be determined from the present studies. If one accepts that the physiological effects of vitamin D_3 are mediated by 1,25-$(OH)_2$-D_3, then the present results are difficult to

TABLE 4. Adjusted (Adj) and unadjusted (Unadj) feed conversions for broilers fed either 25-hydroxycholecalciferol (25-OH-D_3) or cholecalciferol (vitamin D_3) at a level of 69 μg/kg feed

Trial	25-OH-D_3		Vitamin D_3		Change		SD[1]		*P*	
	Adj	Unadj	Adj	Unadj	Adj	Unadj	Adj	Unadj	Adj	Unadj
1	1.863	1.906	1.887	1.926	.024	.020	.0234	.0336	NS	NS
2	1.862	1.896	1.890	1.937	.028	.041	.0154	.0318	.01	.01
3	1.855	1.913	1.875	1.951	.020	.038	.0098	.0496	.01	.05
4	1.930	2.053	1.945	2.088	.015	.035	.0933	.1216	NS	NS
5	1.836	1.981	1.852	2.059	.016	.078	.0084	.0593	.01	.03
6	1.889	1.980	1.923	2.043	.034	.063	.045	.0995	.05	NS
7	1.888	1.907	1.925	1.943	.037	.036	.0405	.0470	.02	.05
8	1.816	1.824	1.841	1.848	.025	.024	.0333	.0341	.05	NS
9	1.876	1.938	1.939	2.005	.063	.067	.0346	.0463	.01	.01
10	1.981	2.112	1.989	2.084	.008	−.028	.052	.0918	NS	NS
Weighted average[2]					.026	.039	.0446	.0702		

[1]Standard deviation represents the average SD of each respective treatment.

[2]Weighting was done taking into account the variance in each trial and the number of birds in each trial.

TABLE 5. Percentage mortality for broilers fed either 25-hydroxycholecalciferol (25-OH-D_3) or cholecalciferol (vitamin D_3) at a level of 69 μg/kg feed

Trial	25-OH-D_3	Vitamin D_3	Change	SD[1]	*P*
1	3.2	3.2	.0	.93	NS
2	4.0	4.4	.4	.80	NS
3	5.9	7.7	1.8	.88	NS
4	10.2	11.9	1.7	.70	NS
5	12.9	16.8	3.9	.60	NS
6	10.5	13.1	2.6	.83	NS
7	4.9	7.0	2.1	.61	<.01
8	1.9	1.4	−.5	.83	NS
9	3.8	4.5	.7	.67	NS
10	10.2	7.8	−2.4	.61	NS
Weighted average[2]			1.0	.74	

[1]Standard deviation represents the average SD of each respective treatment.

[2]Weighting was done taking into account the variance in each trial and the number of birds in each trial.

explain because serum 1,25-$(OH)_2$-D_3 concentrations were similar to those reported by others and did not change in birds fed 25-OH-D_3 in comparison to those fed vitamin D_3 (Hughes *et al.*, 1977; Horst *et al.*, 1981). However, in the two studies analyzed (Studies 6 and 7), serum 25-OH-D_3 concentrations were higher in the birds fed 25-OH-D_3 and, as might be expected, there were positive correlations between serum 25-OH-D_3 concentrations and body weight ($r = .45$, $P < .01$) and breast meat ($r = .33$, $P < .01$) and an inverse correlation with adjusted feed efficiency ($r = .42$, $P < .01$), with no correlation being observed between serum 1,25-$(OH)_2D_3$ and these variables (not shown).

These results would suggest that 25-OH-D_3 may have some direct or indirect effect on growth and performance of the birds. One possibility is that increased serum 25-OH-D_3 may increase free 1,25-$(OH)_2D_3$ concentrations, because the latter metabolite binds much less tightly to the plasma vitamin D binding protein (DBP). This has been proposed as a mechanism for vitamin D toxicity (Vieth, 1990). However, in the present studies, total vitamin D metabolite concentrations were not high enough to saturate the DBP, and it is uncertain whether free 1,25-$(OH)_2D_3$ concentrations would be affected under these conditions. A final possibility is that 25-OH-D_3 may have some direct effect on growth and perfor-

TABLE 6. Percentage breast meat for broilers fed either 25-hydroxycholecalciferol (25-OH-D_3) or cholecalciferol (vitamin D_3) at a level of 69 μg/kg feed

Trial	25-OH-D_3	Vitamin D_3	Change	SD[1]	*P*
2	11.65	11.36	.29	.44	.08
3	13.12	12.62	.50	.51	.02
4	13.67	13.05	.62	.75	.05
6	14.02	13.81	.21	.70	.20
7	13.70	13.54	.16	.65	.26
8	14.12	13.81	.31	.52	.09
9	14.12	13.94	.18	.49	.20
10	14.29	13.42	.83	.45	.01
Weighted average[2]			.53	.56	

[1]Standard deviation represents the average SD of each treatment.

[2]Weighting was done taking into account the variance in each trial and the number of birds in each trial.

mance of the birds. However, because no specific intracellular receptor for 25-OH-D_3 has yet been identified, it is uncertain how 25-OH-D_3 could carry out such a function unless it were able to interact in some way with the receptor for 1,25-$(OH)_2D_3$. Whatever the mechanism, the present studies document the efficacy of 25-OH-D_3 in improving weight gain and feed efficiency of poultry broilers with no adverse effects on mortality.

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