ENVIRONMENT, WELL-BEING, AND BEHAVIOR

Evaluation of Models Using Corticosterone and Adrenocorticotropin to Induce Conditions Mimicking Physiological Stress in Commercial Broilers[1,2]

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ABSTRACT Three experiments (Exp) were conducted to delineate a suitable model for inducing conditions mimicking physiological stress with minimal bird handling. For Exp 1, Ross × Ross 308 male chicks were fed a control diet or a diet containing 5 mg of corticosterone (CS)/kg from d 1 to 7. For Exp 2, Ross × Ross 508 broilers received 1 of 4 dietary treatments: control; control + 4 IU/kg of BW of adrenocorticotropin (ACTH)/d i.m. from d 21 to 27; control + 8 IU/kg of BW of ACTH/d i.m. from d 21 to 27; or control + 15 mg of CS/kg of diet for 14 d from 21 to 35 d of age. In Exp 3, Ross × Ross 308 broilers were fed high or low nutrient density (ND) from 1 to 41 d of age, and 0 or 20 mg of CS/kg of diet from 18 to 21 d of age. Performance parameters (BW gain, feed intake, feed conversion, and mortality) were measured in all 3 experiments. In Exp 1, CS administration significantly decreased BW gain and decreased feed intake and mortality. In Exp 2, although ACTH injection resulted in significantly depressed performance responses relative to the control, CS administration yielded significantly stronger results. In Exp 3, ND and CS interacted ($P = 0.04$) to affect feed intake from d 0 to 34. Broilers fed diets containing high ND and CS had higher feed intake than broilers fed low ND and CS. From 0 to 21 and 0 to 42 d, CS decreased feed intake. Increased dietary ND improved BW gain and feed conversion in Exp 3. Also, CS decreased and increased BW gain and feed conversion, respectively, during all periods in Exp 3. Dietary addition of CS negatively impacted performance of broilers, and increasing dietary amino acid density did not ameliorate these effects.

Key words: broiler, stress, corticosterone, adrenocorticotropin, nutrient density

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INTRODUCTION

Prolonged stress in broilers results in the secretion of the stress hormone corticosterone (**CS**) (Holmes and Phillips, 1976). If CS levels remain elevated, decreases in meat production can be experienced as a result of CS-induced gluconeogenesis. The potential exists for poultry to encounter a myriad of stressors during live production, from environmental stress caused by exposure to temperature fluctuations, to behavioral stress due to disruptions in the social hierarchy. When a bird encounters a stressor, its first line of defense involves activation of the neurogenic system (Siegel, 1980). Epinephrine and norepinephrine are released into the bloodstream, causing a marked increase in blood pressure, muscle tone, nerve sensitivity, blood sugar, and respiration (Siegel, 1980). After this reaction, assuming the stressor goes away, the bird's previously elevated responses are normalized with minimal consequences. However, if the stress is chronic, such that the bird must acclimate to the stressor to maintain homeostasis, growth and the efficiency of nutrient utilization may be decreased. For example, chronic stress results in the activation of the hypothalamic pituitary adrenal system (Siegel, 1980). Activation of the hypothalamic pituitary adrenal system causes the hypothalamus to produce corticotrophin releasing factor, which in turn stimulates the pituitary gland to release adrenocorticotropin (**ACTH**; Holmes and Phillips, 1976). The presence of ACTH in the bloodstream causes the adrenals to secrete CS (Holmes and Phillips, 1976). If blood CS levels remain elevated, cardiovascular diseases, hypercholesterolemia, gastrointestinal lesions, alterations in immune system function, and changes in glucose and mineral metabolism may occur (Siegel, 1995). One of the most important effects of CS is its ability to alter metabolic processes (Siegel, 1995). The most detrimental effect of long-term stress on broiler

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production is the catabolism of structural protein through CS-induced gluconeogenesis (Puvadolpirod and Thaxton, 2000d). It is evident that glucose production occurs at the expense of protein degradation as increases in uric acid secretion have been reported in chickens treated with CS (Brown et al., 1958; Adams, 1968; Siegel and Van Kampen, 1984; Davison et al., 1985). The former metabolic alterations are well documented in birds treated with CS or ACTH and typically accompany significant decreases in BW as a result of physiological stress (Dulin, 1956; Garren et al., 1961; Freeman and Manning, 1975; Bartov et al., 1980; Thaxton et al., 1982; Siegel and Van Kampen, 1984; Davison et al., 1985; Klasing et al., 1987; Siegel et al., 1989; Puvadolpirod and Thaxton, 2000a,b,c,d; Post et al., 2003). This weight loss is often independent of high feed intake (Bartov et al., 1980; Siegel and Van Kampen, 1984; Puvadolpirod and Thaxton, 2000d).

Although much literature has been devoted to cataloging the physiological effects of stress in broilers, fewer data exist quantifying the effect of different nutritional regimes on the stress response (Siegel, 1980, 1995). One notable exception is nutritional research involving heat stress. However, nutritional research on broilers using models that induce controlled stress at the adrenal level (i.e., those utilizing CS or ACTH) are much more sparse.

The purpose of this research was to evaluate models to induce conditions which mimic physiological stress in broilers, different carriers for stress hormones, and the effects of a dietary amino acid (**AA**) regimen in the presence of conditions that mimic physiological stress.

Table 1. Composition of test diets for experiments (Exp) 1 and 2

Ingredient	Test prestarter diet[1] (Exp. 1)	Test grower diet[2] (Exp. 2)
	(%)	
Corn	56.38	65.07
Soybean meal	31.08	26.36
Poultry byproduct meal	5.00	
Poultry fat	3.79	3.27
Dicalcium phosphate	1.33	1.68
Filler oil[3]	1.00	1.00
Limestone	0.95	1.02
NaCl	0.46	0.51
Vitamin-mineral premix[4]	0.25	0.25
DL-Met	0.19	0.25
Sacox 60	0.05	
L-Lys·HCl	0.03	0.17
Choline·Cl	0.01	0.06
L-Thr		0.05
Calculated analysis		
CP, %	23.00	18.50
ME, kcal/kg	3,150.00	3,175.00
Lys, %	1.33	1.11
TSAA, %	0.99	0.87
Thr, %	0.89	0.76
Ca, %	0.94	0.84
Available P, %	0.47	0.42
Na, %	0.22	0.22

[1]Prestarter diet indicates diets fed to broiler chicks from 1 to 7 d during Exp 1 (i.e., during stress administration). From d 8 to 21 birds received a common diet that met or exceeded NRC (1994) nutrient recommendations. Corticosterone (CS) inclusion was 5 mg/kg of diet at the expense of filler oil.

[2]The test grower diet was fed to broilers from 21 to 35 d during Exp 2 (i.e., during stress administration). The CS was added (15 mg/kg of diet) at the expense of filler oil from 21 to 35 d. Adrenocorticotropin was injected i.m. (4 IU/kg of BW/7 d or 8 IU/kg of BW/7 d). Common diets were administered from 1 to 20 d and met or exceeded NRC (1994) nutrient recommendations.

[3]Filler oil represented food grade soybean oil.

[4]Broiler vitamin and mineral premix contained the following per kilogram of diet: vitamin A (vitamin A acetate), 7,718 IU; cholecalciferol, 2,200 IU; vitamin E (source unspecified), 10 IU; menadione, 0.9 mg; B_{12}, 11 μg; choline, 379 mg; riboflavin, 5.0 mg; niacin, 33 mg; D-biotin, 0.06 mg; pyroxidine, 0.9 mg; ethoxyquin, 28 mg; Mn, 55 mg; Zn, 50 mg; Fe, 28 mg; Cu, 7 mg; I, 1 mg; and Se, 0.2 mg.

MATERIALS AND METHODS

Three experiments (**Exp**) were conducted to determine an optimal model for conducting nutritional research on stressed broilers. Characteristics sought after in this model should include depressed BW, increased feed intake, and increased feed conversion.

Experiment 1

In Exp 1, 576 Ross × Ross 308 male broiler chicks were placed into 48 pens of a floor pen facility (12 birds/pen). Each pen contained 1 tube feeder, a nipple drinker line (4 nipples/pen), and built-up soft-wood shavings. Pen dimensions measured 0.92 × 1.22 m. Thus, stocking density for this experiment was approximately 0.9 m^2/bird. These chicks received 1 of 2 dietary treatments during the prestarter period (d 1 to 7). Treatment 1 consisted of a control diet containing no CS (Table 1). Treatment 2 consisted of the control diet plus 5 mg of CS/kg of diet (24 replications/treatment). The dietary CS treatment was made by dissolving CS into soybean oil. Two kilograms of ground corn was mixed with the soybean oil/CS solution and then added to the diet. Soybean oil was added to the diet at the expense of poultry fat at a level of 1% of the diet. Its energy contribution was given credit in the nutrient matrix. Dietary treatments were removed on d 7, and the chicks received a common starter diet from d 8 to 21. All diets met or exceeded nutrient specifications established by the National Research Council (1994). On d 21, chicks and feed were weighed by pen, and growth performance measurements for d 8 to 21 (i.e., BW gain, feed intake, feed conversion, corrected feed conversion, and mortality) were obtained. Corrected feed conversion utilized the weight of dead birds to adjust for feed consumed by birds that died.

Experiment 2

In Exp 2, 240 Ross × Ross 508 male broilers were placed into 20 pens of a floor pen facility (12 birds/pen). Stocking density and pen implements were identical to those listed for Exp 1. From d 1 to 20, chicks received a common starter diet. On d 21, chicks were weighed, starter feed was removed, and from d 21 to 35 broilers received 1

Table 2. Composition of test diets for experiment (Exp) 3

Ingredient	Starter		Grower		Finisher	
	H diet[1]	L diet[2]	H diet[3]	L diet[4]	H diet[5]	L diet[6]
	(%)					
Corn	55.53	58.76	65.67	71.89	65.67	71.89
Soybean meal	33.91	31.27	24.63	19.34	24.63	19.34
Poultry fat	4.36	3.79	4.19	3.16	4.19	3.16
Poultry byproduct meal	2.50	2.50	2.50	2.50	2.50	2.50
Dicalcium phosphate	1.60	1.62	1.18	1.22	1.18	1.22
Limestone	1.02	1.03	0.85	0.87	0.85	0.87
NaCl	0.49	0.49	0.49	0.49	0.49	0.49
Vitamin-mineral premix[7]	0.25	0.25	0.25	0.25	0.25	0.25
DL-Met	0.20	0.17	0.10	0.11	0.10	0.11
L-Lys•HCl	0.07	0.04	0.06	0.08	0.06	0.08
Sacox 60	0.05	0.05	0.05	0.05	0.05	0.05
Choline•Cl	0.02	0.03	0.02	0.04	0.02	0.04
L-Thr				0.02		0.02
Calculated analysis						
CP, %	22.50	21.50	19.00	17.00	19.00	17.00
ME, kcal/kg	3,100.00	3,100.00	3,200.00	3,200.00	3,200.00	3,200.00
Lys, %	1.35	1.25	1.08	0.94	1.08	0.94
TSAA, %	0.99	0.94	0.80	0.75	0.80	0.75
Thr, %	0.88	0.82	0.74	0.67	0.71	0.67
Ca, %	0.94	0.94	0.76	0.76	0.76	0.76
Available P, %	0.47	0.47	0.38	0.38	0.38	0.38
Na, %	0.22	0.22	0.22	0.22	0.22	0.22

[1]The H diet indicates high amino acid (AA) density diet fed to broilers from 1 to 17 d of age.

[2]The L diet indicates low AA density diet fed to broilers from 1 to 17 d of age.

[3]The H diet indicates high AA density diet fed to broilers from 18 to 21 d of age, which contained 20 mg of corticosterone (CS)/kg of diet.

[4]The L diet indicates low AA density diet fed to broilers from 18 to 21 d of age, which contained 20 mg of CS/kg of diet.

[5]The H diet indicates high AA density diet fed to broilers from 22 to 41 d of age.

[6]The L diet indicates low AA density diet fed to broilers from 22 to 41 d of age.

[7]Broiler vitamin and mineral premix contained the following per kilogram of diet: vitamin A (vitamin A acetate), 7,718 IU; cholecalciferol, 2,200 IU; vitamin E (source unspecified), 10 IU; menadione, 0.9 mg; B_{12}, 11µg; choline, 379 mg; riboflavin, 5.0 mg; niacin, 33 mg; D-biotin, 0.06 mg; pyroxidine, 0.9 mg; ethoxyquin, 28 mg; Mn, 55 mg; Zn, 50 mg; Fe, 28 mg; Cu, 7 mg; I, 1 mg; and Se, 0.2 mg.

of 4 treatments: 1) control diet (Table 1); 2) control diet plus 4 IU of ACTH/kg of BW i.m. for 7 d; 3) control diet plus 8 IU of ACTH/kg of BW i.m. for 7 d; and 4) control diet plus 15 mg of CS/kg diet for 14 d (5 replications/treatment). Preparation of ACTH injections for treatments 2 and 3 was accomplished by dissolving ACTH into gelatin. A stock ACTH solution was created by dissolving 5,000 IU of ACTH into 625 mL of sterile water containing a 2% gelatin solution. The ACTH was injected in the pectoralis major muscle i.m. The appropriate amount of solution to equal 4 or 8 IU/kg of BW was removed from the stock solution daily. Daily growth estimates were obtained from the National Research Council (1994) and 21 d BW measurements to create a daily standard for dosage calculation. Injections of ACTH were delivered i.m. to each bird daily at 0100 h. Addition of CS to the feed was accomplished by dissolving CS in soybean oil, using a method identical to that used in Exp 1. On d 35, broilers and feed were weighed and performance measurements (i.e., BW gain, feed intake, feed conversion, corrected feed conversion, and livability) for the d 21 to 35 period were obtained. Also, on d 35, 3 birds per pen were bled via vein puncture so that heterophil to lymphocyte ratios could be calculated. One milliliter of blood was collected from each chick into vacutainers containing a solution of EDTA. Differential leukocyte counts were performed using the method of Cook (1959).

Experiment 3

In Exp 3, 1,280 Ross × Ross 308 male broilers were placed into 32 pens of a floor pen facility (40 birds/pen). Each pen contained 1 tube feeder, nipple drinker lines (10 nipples/pen), built-up pine shavings, and measured 1.52 × 2.96 m. Thus the stocking density for this experiment was approximately 0.11 m^2/bird. This experiment utilized a factorial array of dietary nutrient density (**ND**) and CS. From d 1 to 42, chicks received diets containing either low ND (**L**) or high ND (**H**; Table 2). The difference between H and L diets is that AA density was higher for H diets than L diets. This was accomplished by formulating using a higher lysine value for H diets and increasing other AA relative to Lys using the ideal protein ratio. From d 18 to 21, chicks received 1 of 4 dietary treatments: 1) L without CS addition; 2) L plus 20 mg of CS/kg of diet; 3) H without CS addition; and 4) H plus 20 mg of CS/kg of diet (8 replications/treatment). From d 22 to 42, broilers continued to receive diets containing L or H ND. Addition of CS was accomplished

Table 3. Live performance of broiler chicks fed corticosterone (CS) in experiment 1[1]

	CS, mg/kg			
Parameter	0	5	SEM	Probability
BW gain (kg)				
1 to 7 d	0.098	0.092	0.0025	0.072
8 to 21 d	0.660[a]	0.609[b]	0.0145	0.018
Feed intake (g/bird per d)				
1 to 7 d	17.2	16.9	0.58	0.675
8 to 21 d	35.5[a]	31.7[b]	1.03	0.010
Feed conversion (kg/kg)				
1 to 7 d	1.23	1.34	0.079	0.142
8 to 21 d	1.38	1.37	0.027	0.818
Corrected feed conversion (kg/kg)				
1 to 7 d	1.23	1.34	0.059	0.158
8 to 21 d	1.38	1.36	0.027	0.835
Mortality (%)				
1 to 7 d	0.73[b]	4.86[a]	1.298	0.025
8 to 21 d	0.57[b]	3.06[a]	1.177	0.018

[a,b]Means within the same row followed by uncommon superscripts differ ($P \leq 0.05$).

[1]Corticosterone treatments were administered from 1 to 7 d.

by dissolving CS in ethanol and blending with 2 kg of ground corn as previously described by Gross et al. (1980). All diets met or exceeded nutrient specifications established by the National Research Council (1994). Growth performance measurements (i.e., BW gain, feed intake, feed conversion, corrected feed conversion, and mortality) were measured from d 1 to 21, d 1 to 34, and d 1 to 41.

Statistical Analysis

Experiments 1 and 2 were completely randomized designs using pen as the experimental unit. Experiment 3 utilized a factorial array of treatments in a completely randomized design. Pen was also the experimental unit for Exp 3. All data were analyzed using the GLM procedure of SAS (1998). Means differing significantly were separated using Tukey's Studentized range procedure of SAS (1998).

RESULTS AND DISCUSSION

Experiment 1

Chicks fed diets containing 5 mg of CS/kg of diet during the prestarter period displayed lower ($P < 0.05$) 8 to 21 d BW and feed intake than chicks fed the control diet (Table 3). Inclusion of CS increased ($P < 0.05$) 1 to 7 d and 8 to 21 d mortality. Inclusion of CS had no significant effects on feed conversion or corrected feed conversion.

The decrease in feed intake noted in this experiment is not in agreement with previous research in broilers, which demonstrated increased feed intake due to CS or ACTH treatments (Bartov et al., 1980; Siegel and Van Kampen, 1984; Puvadolpirod and Thaxton, 2000d). However, none of the previously mentioned studies administered stress treatments during the first 7 d of age, as was done herein. Research by Siegel (1962a,b) suggests that adrenal function is relatively low prior to 18 d of age. However, the decrease in BW gain and livability observed in the current research seems to indicate that chick adrenals are at least somewhat sensitive to CS administration at this age. In this experiment, CS was fed from d 1 to 7, but negative impacts on performance were only realized during the 8 to 21 d period. Perhaps this was caused by negative effects of feeding CS, which did not manifest themselves until a later age. It is difficult to draw meaningful conclusions from these results based on the scarcity of similar research.

Experiment 2

Injecting chickens with 8 IU of ACTH/kg of BW resulted in significantly reduced ($P < 0.05$) BW gain compared with broilers fed the control diet (Table 4). However, feeding broilers a diet containing 15 mg of CS/kg of diet resulted in birds with lower ($P < 0.05$) BW gain than in all other treatments. Broilers receiving either level of ACTH injection displayed no significant differences in feed conversion than that of controls. However, broilers receiving 15 mg of CS/kg of diet had higher ($P < 0.05$) feed conversion than birds receiving all other treatments. Feed intake and mortality did not differ among treatments. Broilers receiving injections of 8 IU of ACTH/kg of BW displayed a higher ($P < 0.05$) heterophil to lymphocyte ratio over chicks receiving injections of 4 IU of ACTH/kg of BW or control chicks. However, chicks receiving diets containing 15 mg of CS/kg of diet had higher ($P < 0.05$) heterophil to lymphocyte ratios than birds receiving all other treatments.

Although ACTH treatments significantly altered some parameters tested, broilers appeared to be significantly more sensitive to dietary CS. This is probably simply due to the fact that CS was administered for 14 d, and both ACTH treatments were only given for 7 d. Also, in this experiment, feed intake was not significantly decreased by CS, as noted in the previous experiment. One

Table 4. Live performance of broilers receiving dietary corticosterone (CS) or adrenocorticotropin (ACTH) injections for experiment 2[1]

Treatment	BW gain (kg)	Feed conversion (kg/kg)	Corrected feed conversion (kg/kg)	Feed intake (g/bird/d)	Mortality (%)	H:L ratio[2]
Control	1.00[a]	1.90[b]	1.87[b]	132.1	3.64	0.67[c]
Control + 4 IU of ACTH/kg of BW	0.91[ab]	2.03[b]	1.99[b]	127.2	3.48	0.91[c]
Control + 8 IU of ACTH/kg of BW	0.89[b]	1.96[b]	1.96[b]	123.5	0.00	1.31[b]
Control + 15 mg of CS/kg of diet	0.55[c]	3.42[a]	3.40[a]	130.3	1.67	1.65[a]
SEM	0.027	0.072	0.073	2.96	1.753	0.085
Probability	<0.01	<0.01	<0.01	0.22	0.44	<0.01

[a–c]Means within the same column without a common superscript differ ($P \leq 0.05$).

[1]Broilers receiving dietary CS were fed treatment feed from d 21 to 35. Broilers receiving either level of ACTH injection received treatments from d 21 to 27.

[2]The H:L ratio indicates levels of circulating heterophils to lymphocytes per 100 white blood cells.

plausible explanation is that the stress period was much longer, and the birds were beginning to acclimate to the stressor. However, feed intake was not measured on a daily basis. Thus, it is not known if feed intake was depressed early in the stress period, followed by a return to normal eating patterns due to acclimation to the stressor.

It has been well established that chickens experiencing stress display an increased heterophil to lymphocyte ratio (Siegel, 1995), and the results of this research are in agreement, as treatment with both ACTH and CS resulted in a significant increase in this response. However, 8 IU of ACTH, but not 4 IU of ACTH, increased the heterophil to lymphocyte ratio, whereas the addition of CS in feed further increased the heterophil to lymphocyte ratio over the treatment utilizing 8 IU of ACTH.

Experiment 3

Results for Exp 3 are presented in Table 5. No interactions between ND and CS were observed for BW gain throughout the experiment. However, the main effect of increasing dietary ND was higher ($P < 0.05$) BW gain from d 1 to 34 and d 1 to 41, although no significant differences were noted from d 1 to 21. The main effect for CS inclusion demonstrated decreased ($P < 0.05$) BW gain in broilers fed CS from d 1 to 21, d 1 to 34, and d 1 to 41. Dietary ND and CS interacted to affect feed intake from d 1 to 34 (Table 5) because broilers fed the diet containing H ND plus CS had higher ($P < 0.05$) feed intake than chicks fed the diet containing L ND plus CS (data not presented). However, no interactions were observed from d 1 to 21 or d 1 to 41. Dietary ND exerted

Table 5. Live performance of broilers fed diets differing in nutrient density (ND) and corticosterone (CS) for experiment 3[1]

Parameter	ND		CS		SEM	Source of variation		
	L[2]	H[3]	0	20		ND	CS	ND × CS
BW gain (kg)								
1 to 21 d	0.543	0.566	0.613[a]	0.496[b]	0.0085	0.072	<0.0001	0.667
1 to 34 d	1.333[b]	1.422[a]	1.499[a]	1.256[b]	0.0128	<0.0001	<0.0001	0.396
1 to 41 d	1.747[b]	1.852[a]	1.916[a]	1.683[b]	0.0158	<0.0001	<0.0001	0.761
Feed intake (g/bird per d)								
1 to 21 d	44.5	45.2	46.0[a]	43.8[b]	0.58	0.372	0.012	0.063
1 to 34 d	96.5	98.0	102.3[a]	92.1[b]	0.67	0.325	<0.0001	0.037
1 to 41 d	162.9	164.9	172.4[a]	155.3[b]	0.68	0.390	<0.0001	0.121
Feed conversion (kg/kg)								
1 to 21 d	1.74	1.70	1.58[b]	1.86[a]	0.020	0.188	<0.0001	0.245
1 to 34 d	1.76[a]	1.68[b]	1.68[b]	1.76[a]	0.013	<0.0001	<0.0001	0.261
1 to 41 d	1.94[a]	1.85[b]	1.88[b]	1.92[a]	0.012	<0.0001	0.013	0.226
Corrected feed conversion (kg/kg)								
1 to 21 d	1.73	1.70	1.57[b]	1.85[a]	0.020	0.207	<0.0001	0.317
1 to 34 d	1.76[a]	1.68[b]	1.68[b]	1.76[a]	0.124	<0.0001	<0.0001	0.315
1 to 41 d	1.94[a]	1.85[b]	1.88[b]	1.92[a]	0.012	<0.0001	0.014	0.246
Mortality (%)								
1 to 21 d	1.09	0.78	1.25	0.63	0.498	0.660	0.382	0.194
1 to 34 d	1.09	1.09	1.41	0.78	0.531	1.000	0.413	0.223
1 to 41 d	1.25	1.09	1.41	0.94	0.565	0.846	0.562	0.337

[a,b]Means within the same row followed by uncommon superscripts differ ($P \leq 0.05$).

[1]The ND treatments were administered from d 0 to 41. The CS treatments (0 or 20 mg/kg of diet) were administered from 18 to 21 d.

[2]The L diet indicates low AA density diet fed to broilers during CS administration (18 to 21 d). The L diets were also fed from 1 to 17 and 21 to 35 d (without CS) and contained TSAA levels of 0.94 and 0.75% and Lys of 1.25 and 0.94%, respectively.

[3]The H diet indicates high AA density diet fed to broilers during CS administration (18 to 21 d). The H diets were also fed from 1 to 17 and 22 to 35 d (without CS) and contained TSAA levels of 0.99 and 0.80% and Lys of 1.35 and 1.08%, respectively.

no main effect on feed intake from d 1 to 21 or d 1 to 41. However, inclusion of dietary CS depressed ($P < 0.05$) feed intake from d 1 to 21 and decreased ($P < 0.05$) feed intake from d 1 to 41. No interactions between ND and CS were observed for corrected or uncorrected feed conversion in any period. However, chicks fed diets containing H ND had lower ($P < 0.05$) feed conversion and feed conversion corrected for mortality weight than chicks fed diets containing L ND from d 1 to 34, and d 1 to 41. Inclusion of dietary CS increased ($P < 0.05$) feed conversion over that of broilers fed diets without CS in all periods measured. No interactions between ND and CS were observed for mortality in any period. Furthermore, no main effects on mortality were observed in any period for ND or CS inclusion.

Broilers treated with CS had much lower BW gain and higher FCR than birds fed control diets in this experiment compared with those in Exp 1. This could be due to the fact that these birds were given CS treatments from d 18 to 21. As previously discussed, it has been demonstrated that broiler adrenals are much more functional after 18 d of age (Siegel, 1962a,b). The results observed due to the main effects of ND were in agreement with previous research (Lemme et al., 2003; Eits et al., 2003; Corzo et al., 2004; Kidd et al., 2004, 2005a,b). Also, dietary CS significantly lowered feed intake from d 1 to 21, and d 1 to 42. This result was unexpected as previous research in chickens of this age (Bartov et al., 1980; Siegel and Van Kampen, 1984; Puvadolpirod and Thaxton, 2000d) noted significantly increased feed intake when CS or ACTH treatments were given. One possible explanation for this is that CS treatments were dissolved in ethanol for this experiment. A fairly large amount of ethanol (about 750 mL/treatment) had to be used to dissolve the CS completely. Thus, the feed had a fairly pungent odor. It is possible that the birds refused the feed due to the smell or taste of alcohol. However, previous research conducted by Gross et al. (1980) noted no significant depression in feed intake despite the use of the same methods to incorporate CS into the feed.

Based on this research, it would appear that feeding broilers CS dissolved in soybean oil may be a suitable model for inducing physiological stress-mimicking conditions in broilers as measured by decreased productive efficiency and an increased heterophil to lymphocyte ratio. Furthermore, as most nutritional research is done using pen as the experimental unit, this model requires the devotion of less time to treatment preparation and administration.

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