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Lipid Metabolism and Peroxidation in Broiler Chicks under Chronic Heat Stress*

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ABSTRACT: The effects of taurine supplementation on growth performance, serum and liver concentrations of lipid, fatty acid composition and lipid peroxidation in the livers of broilers under chronic heat exposure conditions were investigated. The chicks with a similar body weight were equally assigned to one of three controlled-environment chambers. The brolier chicks, which were kept at 34°C were fed either with a control diet or the control diet supplemented with 0.8% taurine, whereas broiler chicks kept at 22°C were fed a control diet. Both of the BW and BW gains of broilers maintained at a temperature of 34°C were significantly lower than those of the control group, which was maintained at a temperature of 22°C (p<0.05). However, taurine addition in the diet of birds submitted to heat stress significantly improved BW gain (p<0.05). The feed intake of chicks declined with increases in temperature. The relative liver and gall bladder weights of chicks fed the control diet and maintained at 34°C were significantly lower than those measured in the control birds (p<0.05). However, dietary taurine was found to compensate for these reductions in liver and gall bladder weights. Relative weights of abdominal fat did not differ significantly among the three groups. Serum triglyceride concentrations were significantly lower in the chicks fed the control diet and maintained at 34°C compare to those measured in the chicks fed the control diet at 22°C (p<0.05). Heat stress resulted in a significant reduction in total lipid and triglyceride levels, but also increased the levels of total cholesterol in the liver (p<0.05). However, dietary taurine supplementation under the heat stress condition resulted in the recovery, to control levels, of serum triglyceride concentrations, as well as the amounts of total lipids, triglycerides, and cholesterol in the liver. The livers of chicks fed on taurine diets at 34°C showed significantly higher proportions of C14:0, C16:1, C18:1, C18:2, and 20:3, and lower C18:0 and C20:4 proportions than those of chicks fed on control diets at the same temperature (p<0.05). The total levels of saturated fatty acids decreased, but monounsaturated fatty acids and unsaturated fatty acid levels increased in chicks fed the taurine diet, as compared to chicks fed the control diet at 34°C (p<0.05). Peroxidizability indices were significantly lower in the heat-exposed chicks fed the taurine diet than in the non-taurine heat-exposed groups (p<0.05). In conclusion, dietary taurine results in an increase in the growth performances of chicks under heat stress conditions via improvements in lipid absorption and metabolism, as well as an induced reduction in lipid peroxidation. (**Key Words:** Broiler, Heat Stress, Lipid Metaboilsm, Taurine)

INTRODUCTION

Heat stress exerts negative effects on the growth

performance of broilers (Cooper and Washburn, 1998; Mahmoud and Yaseen, 2005). Although heat-exposed broilers require more metabolic energy for the maintenance of homeothermy (Wolfenson et al., 1981), these negative effects can be associated primarily with reductions in the feed intake of broilers under heat stress (Hurwitz et al., 1980). Mckee et al. (1997) reported that heat-exposed birds reduced their feed intakes in order to reduce the thermogenic effects associated with nutrient absorption, assimilation, and utilization. Furthermore, the reduction in feed intake associated with heat stress also affects lipid metabolism in broilers, in that lipids, especially triglycerides, have the highest energy value. For example, previous studies have shown decreases in serum triglyceride concentrations in heat-stressed broilers (Moraes et al., 2003) and the amounts of total lipids in the livers of male

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chicks (Takahashi et al., 1983).

Taurine, also referred to as 2-aminoethanesulfonic acid, is found in tissues primarily as a free amino acid, and is one of a number of low-molecular-weight organic constituents present in abundance in mammals (Sturman and Hayes, 1980). Unlike methionine and cysteine, taurine is neither involved in protein synthesis nor an energy source, but rather influences membrane stabilization, bile salt formation, growth modulation, and calcium homeostatsis (Huxtable, 1992; Redmond et al., 1998), and plays an important antioxidant function (Li et al., 1993; Balkan et al., 2001, 2002; Kocak-Toker et al., 2005). The best-known function of this sulfur amino acid involves its conjugation with bile acid (Hayes, 1976).

Bile salts are produced and conjugated in the liver, and excreted in the small intestine. Belli et al. (1987) reported that patients suffering from cystic fibrosis and severe steatorrhea showed significant improvements in the absorption of triglycerides, total fatty acids, and linoleic acid, after they received taurine supplements. Numerous studies have been done to investigate the effect of taurine on lipid and cholesterol metabolism (Herrmann, 1959; Cantafora et al., 1986; Sugiyama et al., 1989; Petty et al., 1990; Gandhi et al., 1992; Yan et al., 1993; Park et al., 1998; Murakami et al. 1999; Yokogoshi et al., 1999) in a variety of species, including rats, guinea pigs, rabbits, and cats. Almost all of these experiments have been conducted using animals with hypercholesterolemia induced by the administration of a high-cholesterol diet.

Anderson et al. (1975) observed that the addition of dietary taurine improved the growth performance of chicks fed on a purified diet deficient in sulfur amino acids. In our previous study (Park and Choi, 1997), an increased growth rate was reported in heat-exposed broiler chicks fed on 0.5% and 1.0% taurine-supplemented corn-soybean basal diet. In this previous study, there was no data regarding any association between taurine and lipid metabolism in broiler chicks exposed to heat stress conditions.

Therefore, the primary objective of our study was to characterize the effects of taurine on growth performances, triglyceride absorption, lipid concentration, fatty acid composition and lipid peroxidizability indices in the livers of broiler chicks under chronic heat stress conditions.

MATERIALS AND METHODS

Animals and management

One hundred newly-hatched male broilers were obtained from a commercial hatchery. After weighing, the birds were wing-banded for identification, maintained all together in electrically-heated battery brooders, and raised on a commercial broiler starter diet for 21 days. The

ambient temperature was then decreased gradually beginning at 32°C at day 1, to a final temperature of 26°C at day 21. Water was available at all times, and continuous lighting was provided.

Experimental design

At day 22, 36 chicks of similar body weights (636±9.5 g) were selected from the flock, and allocated to three different environment-controlled chambers, each containing 12 birds. The temperature in the control chamber was reduced from 26°C to 22°C until day 42, whereas the temperature in the two heat chambers was maintained at 34°C until day 42. Relative humidity was maintained at 55±5% in all chambers. The three groups of chicks were provided with a commercial broiler starter diet consisting of 19% CP, at 3,000 kcal ME/kg. One group of the two heated-chamber groups were fed diet supplemented with 0.8% taurine (Dong-A Pharm., Co., Ltd., Korear). The chicks were provided with relevant diets and water *ad libitum*, and continuous light was provided throughout the experimental period.

Sample collection

At day 44, all of the birds were individually weighed and samples were taken after 24 h of fasting. Blood samples were obtained from the jugular vein, and the serum was separated. The chicks were then decapitated, and the livers, gall bladders, and abdominal fat were removed and weighed. The abdominal fat was segregated by the methods of Kim and Park (2002). The serum and liver were stored in a -20°C freezer for further analysis.

Serum and liver lipid analysis

Serum triglyceride concentrations were measured using analytical kits purchased from Asan Pharm. Co., Ltd. (Seoul, Korea). Extractions of total lipids from the liver were conducted as described by Folch et al. (1957). The contents of extracted lipids were estimated as described by Bligh and Dyer (1959). The cholesterol and triglyceride concentrations in the lipids extracted from the livers were then analyzed using an enzymatic kit (Asan Pharm. Co., Ltd. Seoul, Korea).

Fatty acid analysis

For fatty acid determination, the extracted lipids were transmethylated with BF $_3$ and methanolic NaOH, using an AOCS Official Method Ce 2-66. The fatty acid profiles were determined with a gas chromatograph (Hewlett-Packard Co., Wilmington, DE), equipped with a flame ionization detector (FID). One microliter of fatty acid solution was injected into a capillary column (30 m×0.25 mm×0.25 μ m, Pewlett-Packard Co.). Helium was used as

Table 1. The effects of taurine supplementation on growth and relative weights of liver, abdominal fat, and gall bladder in broilers exposed to excessive heat¹

Items	Control diet		Taurine diet
	22°C	34°C	34°C
BW (g/bird)	1,962±76 ^a	1,664±40 ^b	1,764±34 ^b
BW gain (g)	1,323±71 ^a	1,020±35°	1,125±24 ^b
Liver weight/BW (%)	2.21 ± 0.09^{a}	1.98 ± 0.09^{b}	2.20 ± 0.09^{a}
Abdominal fat weight/BW (%)	1.17±0.13	1.34 ± 0.08	1.23±0.08
Gall bladder weight/BW (%)	0.12 ± 0.01^{a}	0.09±0.00 ^b	0.12 ± 0.01^{a}

Values are means±SE.

^{a, b, c} Means within a row with no common superscript differ significantly (p<0.05).

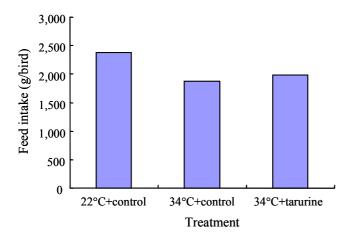


Figure 1. Effects of taurine supplementation on feed intake in broilers exposed to excess heat.

the conductor gas at a flow rate of 0.5 ml/min. The split relationship was 1/100. The operating conditions of the gas chromatograph were as follows: the initial temperature was maintained at 150°C for 2 minutes, with a programmed increase 175°C, at a rate of 3.5°C/min, at which point it was maintained for 10 minutes, followed by an increase at 4°C /min to 210°C and then an increase at 25°C/min to 250°C, with a final 5-minute hold period. The injector and detector temperatures were 250 and 270°C, respectively. The fatty acid peaks were identified via comparison with retention times of fatty acid methylester standards.

Peroxidizability indices (PI) were calculated by the methods of Andriamampandry et al. (1996), as follows: PI = (% dienoic 1)+(% trienoic 2)+(% tetraenoic 3)+(% pentaenoic 4)+(% hexaenoic 5).

Statistical analysis

All experiments were simultaneously conducted in order to eliminate variances as the result of the storage and handling of samples prepared at different times. The experimental data were analyzed with SAS (1999) statistical software. The general linear model was used to analyze variance, and Duncan's new multiple range test was applied to compare the differences between treatments. A significance level of 0.05 was used.

RESULTS AND DISCUSSION

As is shown in Table 1, the BW and BW gain in broilers exposed to high temperature was significantly less than that of the control group. These results were comparable to those reported by Howlider and Rose (1987). However, the BW gains of broilers fed dietary taurine at 34°C were significantly higher than those of broilers fed with control diets at the same temperature (p<0.05). This result suggests that dietary taurine alleviated the deleterious effects of heat stress on the growth of broilers. This result was also consistent with the findings reported by Park and Choi (1997). Bonnet et al. (1997) demonstrated that the reductions in the BW of broilers exposed to heat stress were associated with reduced feed intake. Our results also evidenced a decrease in feed intake by the broilers at 34°C (Figure 1).

Plavnik and Yahav (1998) reported that heat stress induced a reduction in the relative liver weights of chicks, as the result of a decrease in metabolic needs. Our data also showed that the relative liver weights of chicks fed on a control diet at 34°C were significantly lower than those of the control group (p<0.05) (Table 1). Furthermore, the relative gall bladder weights were significantly lower in chicks fed on the control diets at 34°C, than in the chicks fed on the control diets at 22°C (p<0.05). However, our data clearly showed that dietary taurine supplementation exerted a compensatory effect on these reductions in the heat-exposed chicks.

Table 1 also illustrates the effects of dietary taurine on the relative amounts of abdominal fat in the heat-exposed chicks. The relative amounts of abdominal fat did not differ significantly among the three groups, similar to the reports of Sonaiya (1998) and Plavnik and Yahav (1998). However, the effects of heat exposure on fat deposition have long been the subject of considerable controversy. Keshavarz and Fuller (1980), and Smith and Teeter (1987) reported a significant reduction of fat deposition, but Swain and Farrell (1975), Howlider and Rose (1987), and Geraert et al. (1996) reported an increase in fat contents under heat conditions. Further study is required in this resspect.

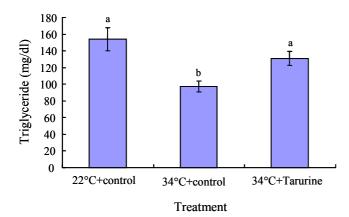


Figure 2. Effects of taurine supplementation on serum triglyceride concentrations in broilers exposed to excessive heat. Values are expressed as means±SE. ^{a, b} Means without bearing the same superscripts differ significantly (p<0.05).

Table 2. Effect of taurine supplementation on the hepatic lipid contents of broilers exposed to excessive heat¹

Items	Control diet		Taurine diet
	22°C	34°C	34°C
Total lipid, %	5.86±0.30 ^a	4.82 ± 0.20^{b}	5.93±0.08 ^a
Triglyceride, (mg/g wet tissue)	15.3±2.18 ^b	9.15±0.86°	19.8±1.16 ^a
Total cholesterol (mg/g wet tissue)	2.80 ± 0.07^{b}	3.00 ± 0.07^{a}	2.68 ± 0.04^{b}

¹ Values are means±SE.

The effects of dietary taurine on serum triglyceride concentrations in heat-exposed broilers are depicted in Figure 2. Serum triglyceride concentrations significantly lower in chicks fed on control diets at 34°C (p<0.05) than in chicks fed on control diets at 22. These findings are consistent with the results reported by Moraes et al. (2003), in which the triglyceride concentrations were shown to be significantly lower in chicks kept at 30°C than at 22°C. However, dietary taurine supplementation induced an elevation of the serum triglyceride concentration to control levels at 22°C, thereby implying that dietary taurine supplementation increased triglyceride absorption in the heat-exposed chicks. These results were in agreement with the report of Belli et al. (1987), in which patients suffering from severe steatorrhea manifested significant improvements in triglyceride absorption due to the taurine supplementation. Therefore, taurine may exert an important effect on triglyceride absorption in heat-exposed chicks.

The effects of taurine supplementation on total lipid, triglyceride, and cholesterol contents in the liver are shown in Table 2. The heat stress resulted in a significant decrease in the amounts of total lipids and triglycerides, but increased total cholesterol contents in the liver (p<0.05), consistent with what was reported in the studies conducted

Table 3. Effect of taurine supplementation on hepatic total lipid fatty acid compositions in broiler chicks exposed to excessive heat¹

Control diet Taurine diet						
Fatty acid (%)	Contro	Control diet				
	22°C	34°C	34°C			
Saturated (SFA)						
14:0	0.23 ± 0.02^{ab}	0.20 ± 0.02^{b}	0.26 ± 0.01^{a}			
16:0	18.6±0.30	19.4±0.52	19.1±0.26			
17:0	0.23 ± 0.01	0.24 ± 0.01	0.23 ± 0.01			
18:0	17.1 ± 1.31^{b}	20.0±1.10 a	15.2±0.37 ^b			
Σ SFA	36.1 ± 1.28^{b}	39.8 ± 1.36^{a}	34.8 ± 0.49^{b}			
Monounsaturated (MUFA)						
16:1	1.91 ± 0.22^{a}	1.44 ± 0.12^{b}	2.19 ± 0.09^{a}			
18:1	21.7 ± 1.34^{ab}	19.4±1.11 ^b	23.4 ± 0.48^{a}			
20:1	0.22 ± 0.01	0.22 ± 0.03	0.19 ± 0.01			
Σ MUFA	23.8 ± 1.53^{ab}	21.1±1.21 ^b	25.8 ± 0.56^{a}			
Polyunsaturated (PUFA)						
18:2	25.7 ± 0.96^{ab}	24.0 ± 1.29^{b}	27.3 ± 0.62^{a}			
20:3	0.61 ± 0.05^{ab}	0.56 ± 0.05^{a}	0.72 ± 0.07^{b}			
20:4	9.8 ± 40.98^{ab}	10.7 ± 0.96^{a}	8.04 ± 0.36^{b}			
$\sum \omega 6$	36.1±0.56	35.4±0.71	35.9 ± 0.44			
18:3	1.27 ± 0.14	1.24 ± 0.05	1.00 ± 0.15			
20:5	0.39 ± 0.04	0.41 ± 0.03	0.32 ± 0.09			
22:6	1.68 ± 0.22	1.65 ± 0.20	1.32 ± 0.09			
$\Sigma \omega 3$	3.34 ± 0.14^{a}	3.10 ± 0.09^{ab}	3.00 ± 0.06^{b}			
20:2	0.64 ± 0.05	0.60 ± 0.07	0.53 ± 0.02			
Σ PUFA	40.1±0.70	39.1±0.70	39.5±0.48			
Σ USFA	63.9 ± 1.28^{a}	60.2 ± 1.36^{b}	65.2 ± 0.49^{a}			
$\Sigma \omega 6/\omega 3$	10.9 ± 0.33^{b}	11.6 ± 0.41^{ab}	12.0±0.21a			
PI^2	85.8±2.71 ^a	87.1 ± 2.17^{a}	77.0 ± 1.66^{b}			

¹ Values are means±SE.

by Balnave (1972) and Takahashi et al. (1983). However, our data indicated that dietary taurine supplementation under heat stress conditions resulted in a reversion of these factors to control levels, thereby suggesting that dietary taurine increased lipogenesis in the livers of chicks under heat stress conditions.

The hepatic fatty acid compositions of the taurine-supplemented heat-exposed chicks are shown in Table 3. Chronic heat exposure clearly altered the hepatic fatty acid profiles. In the same heat-exposed chicks, taurine supplementation resulted in significant increases in the proportions of C14:0, C16:1, C18:1, C18:2 and C20:3 and, conversely, a reduction in the proportions of C18:0 and C20:4 in the liver (p<0.05). The total saturated fatty acid contents decreased, but the proportion of monounsaturated and unsaturated fatty acids increased in the heat-exposed chicks fed the taurine-rich diet (p<0.05). Peroxidizability indices were also shown to decrease in the heat-exposed chicks fed the taurine-rich diet (p<0.05), thereby suggesting the antioxidant functions of taurine.

^{a, b} Means within a row with no common superscript differ significantly (p<0.05).</p>

a, b Means within a row with no common superscript differ significantly (p<0.05).</p>

² PI; Peroxidizability index = (%dienoic×1)+(% trienoic×2)+(% tetraenoic ×3)+(% pentaenoic×4)+(% hexaenoic×5).

Essential fatty acids are not synthesized in chickens, but rather come from dietary sources (Fisher, 1984). Their presence in the body, therefore, depends on both their presence in the diet, and their rate of oxidation in the tissues. Belli et al. (1987) reported that taurine supplementation resulted in improvements in total fatty acid and linoleic acid contents in cystic fibrosis patients exhibiting ongoing fat malabsorption and essential fatty acid deficiency. Accordingly, in the present study, linoleic acid proportion was improved significantly in the heat-exposed chicks fed the taurine diet, indicating that dietary taurine increased the lipid absorption characteristics of chicks exposed to heat stress.

Antioxidant systems are important with regard to the scavenging of free radicals and their metabolic products, as well as in the maintenance of normal cellular physiology, via the restoration of various depleted antioxidants in stressed poultry (Halliwell and Gutteridge, 1989). Taurine is also known to play an antioxidant role, preventing lipid peroxidation (Li et al., 1993; Balkan et al., 2001, 2002; Kocak-Toker et al., 2005). Lipid peroxides induce a diminution of the production performance of chicks. Therefore, our data suggested that taurine might be applicable as a biological antioxidant for improving the production performances of chicks against peroxidative stress.

In conclusion, dietary taurine supplementation was found to result in increases in the growth performance of chicks under heat stress conditions, via improvements in lipid absorption and metabolism, as well as reductions in lipid peroxidation.

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