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Effect of ascorbic acid supplementation on plasma heat shock protein 70 and production parameters during heat stress in commercial broilers

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Abstract
The present work was conducted to investigate the effect of ascorbic acid on plasma heat shock protein 70 and certain production parameters during heat stress in commercial broilers. A total number of 96 birds were randomly divided into 4 groups and each group consist of 12 birds in two replicates. Work was done in two conditions, heat and comfort. Heat stressed groups were maintained at 37±5.0 °C ambience whereas Comfort groups were maintained at 26±1.0 °C. G1 was taken as control whereas G2, G3 and G4 were supplemented with 100 mg, 200 mg and 300 mg of ascorbic acid respectively. The overall mean concentration of plasma HSP70 showed non-significant difference between comfort and heat stressed broilers in all the groups. However, a significantly ($p<0.05$) higher concentration of HSP70 were observed in control group of comfort condition as compared to treatment groups. In both comfort and heat stressed condition, significantly higher ($P<0.05$) body weight gain, total feed intake (g), feed efficiency ratio (FER) and performance index (PI) was observed in G3 group of broilers. In the present investigation, supplementation of ascorbic acid significantly reduces plasma HSP70, which provide an indication that ascorbic acid may be useful in combating rigors of heat stress in chickens.

Keywords: Ascorbic acid, commercial broilers, heat stress, plasma HSP70

1. Introduction
Meteorological factors such as high ambient temperature and high relative humidity exert adverse effects on poultry production [1]. They also cause heat stress in poultry during the hot dry season [2]. Chickens also have other physiological mechanisms to improve thermal resistance [3]. When living organisms are exposed to thermal and non-thermal stressors, the synthesis of most proteins is retarded; however, a group of highly conserved proteins known as HSPs are rapidly synthesized. These proteins are essential for organisms living at the edge of their thermal range. It is well documented that one of the most important functions of HSPs is to protect organisms from the toxic effects of heating [4]. HSPs may play important roles in protein assembly and disassembly protein folding and unfolding protein translocation and the refolding of damaged proteins. Compared to other species of domestic animals, broiler chickens are more sensitive to high ambient temperatures. They have no sweat glands, a rapid metabolism and high body temperature. As environmental temperature rise food consumption, growth rate, feeding efficiency and survivability all decline [5]. The most significant increase in ascorbic acid demand take place during acute environmental stress such as excessive hot or cold weather and stress conditions increases the metabolic need for this vitamin or that decrease the innate capacity of biosynthesis. Under such conditions, supplementing the poultry diet with vitamin C may have a beneficial effect on performance [6]. The inhibitory action of ascorbic acid on adrenal steroidogenesis is mediated via modulation of steroid hydroylating enzymes in the adrenal gland [7]. Substantial attention has been paid to the role of nutritional additives to minimize the effects of heat stress. The aim of the study was to investigate the effect of ascorbic acid on plasma heat shock protein 70 and certain production parameters during heat stress in commercial broilers.

2. Materials and Methods
2.1 Location of the study
The present research was carried out in the Department of Veterinary Physiology and Biochemistry, College of Veterinary Science and Animal Husbandry, N.D.V.S.U., Jabalpur (M.P.). Jabalpur is located at latitude 23°10'N and longitude 79°56'E at an altitude of 411 meters in central India. It has a humid subtropical climate, typical of North-Central India.

2.2 Experimental design
A total number of ninety six (96) day old chicks of commercial broiler birds were procured from private hatcheries of Jabalpur. The experimental chicks (Cobb-400 broiler) were reared in the battery brooder house. Artificial heat was provided to chicks during early period (brooding period) of growth using thermostatically controlled electric brooders. The battery brooders were cleaned, washed and disinfected by blow lamping and complete house was fumigated using formaldehyde and potassium permanganate four days prior to start of the experiment. Diets were formulated as per NRC 1994 specifications. All mash system of feeding was practiced during the experiment. Broiler birds randomly divided into eight groups. Four groups of birds were maintained in summer conditions (May to June) maintained in heat stress (37±5.0 °C) ambience, whereas other four groups of birds was maintained at 26±1.0 °C (comfort temperature) using an air conditioner. G1 (Heat) group was kept as control whereas G2, G3, G4 groups was supplemented with 100 mg, 200 mg and 300 mg of ascorbic acid (AA) along with feed. Temperature and humidity of the experimental poultry unit was recorded using a digital thermo-hygrometer.

2.3 Collection of blood samples for plasma HSP70 analysis
Blood samples (2ml) collected from all the birds on 15th, 30th and 45th day of experiment from wing vein. The blood samples were collected with proper sepsis. The blood samples were collected in heparinized polypropylene tubes (20 IU heparin/ml of blood) were kept in the ice bucket and carried back to the laboratory immediately. In the laboratory, all the blood samples were centrifuged at 3000 rpm for 30 min and plasma was separated in the labelled vials of 2 ml capacity and stored at -20°C till analysis for plasma HSP70 analysis. Enzyme Linked Immuno Sorbent Assay (ELISA) was done for the estimation of plasma HSP70 as per the protocol prescribed by commercially available Kits procured from Cusabio, China.

2.4 Feed intake
Feed intake was recorded by weighing the offered feed and residual feed on weekly basis. Weekly feed consumption of broilers replicate was recorded till the end of week. The feed consumed was determined by subtracting the residual feed from total feed offered during each of the period.

2.5 Body weight
Body weight was recorded individually at an interval of three weeks basis (0, 3 and 6 weeks) by weighing all the birds in each treatment group using electronic weighing balance in the morning hours before feeding. The individual bird was kept on platform of the balance. The reading was recorded when it became stable.

2.6 Feed efficiency ratio
Feed efficiency ratio was calculated on the basis of body weight gain and feed intake in three interval basis of the experiment. FER was calculated using following formula.
FER = Body weight gain (g) / Feed consumption (g)

2.7 Performance index
Performance index was calculated as per the formula proposed by [8].
PI = Body weight gain (g) X FER

2.8 Mortality
Mortality was recorded on daily basis and the mortality rate was calculated for the period from 0 to 6 weeks.

2.9 Statistical analysis
The recorded data of present study were statistically analyzed using Completely Randomized Design [9]. Various conditions and treatment groups were compared by using Duncan Multiple Range test (DMRT).

3. Results and Discussion
The mean plasma HSP70 concentration of broilers is presented in Table 1. The overall mean concentration of HSP70 showed non-significant difference between comfort and heat stressed birds in all the groups. However, significantly ($p<0.05$) higher concentration of HSP70 (0.43±0.03 ng/ml) was observed in control group of comfort condition as compared to other treatment groups. Also, significantly ($p<0.01$) higher concentration of HSP70 (0.51±0.04 ng/ml) was observed in heat stressed control group as compared to other heat stressed treatment groups with varying concentrations of ascorbic acid supplementation.
On day 15, non-significant difference was observed in HSP70 concentration between comfort and heat stressed group. However, all the groups in comfort conditions showed non-significant difference in HSP70 concentrations. In heat stressed condition, significantly ($p<0.05$) higher concentration (0.41±0.02 ng/ml) of HSP70 was observed in G1 group as compared to other treatment groups but all treatment groups differ non-significantly.
On day 30, non-significant difference was observed in HSP70 concentration between comfort and heat stressed group. Further, the heat stressed birds in control group as well as control group of comfort condition showed significant difference ($p<0.01$) in HSP70 concentration as compared to treated groups. The maximum mean plasma level of HSP70 was observed 0.67±0.02 ng/ml in control group of comfort condition, whereas minimum concentration of 0.19±0.01 ng/ml was observed in G3 (200 mg AA) group. Further, the maximum mean plasma level of HSP70 was observed 0.82±0.07 ng/ml in heat stressed control group, whereas minimum concentration of 0.20±0.01 ng/ml was observed in G3 (200 mg AA) group. On day 45, interaction between different conditions and treatment was non-significant.

Table 1: Mean plasma HSP70 concentration (ng/ml) of broilers at different intervals.

Period	Condition	G1	G2	G3	G4
15th day	Comfort	0.29±0.02 (12)	0.23±0.01 (12)	0.20±0.02 (12)	0.23 ± 0.02 (12)
	Heat	0.41^{a}±0.02 (12)	0.21^{b}±0.01 (12)	0.19^{b}±0.01 (12)	0.20^{b}±0.02 (12)
30th day	Comfort	0.67^{A}±0.02 (12)	0.30^{B}±0.02 (12)	0.21^{B}±0.01 (12)	0.29^{B}±0.02 (12)
	Heat	0.82^{A}±0.02 (12)	0.24^{B}±0.02 (12)	0.20^{B}± 0.01 (12)	0.24^{B}±0.02 (12)
45th day	Comfort	0.32±0.02 (12)	0.26±0.02 (12)	0.21±0.01 (12)	0.25±0.02 (12)
	Heat	0.35±0.02 (12)	0.23±0.01 (12)	0.22±0.01 (12)	0.23±0.02 (12)
Overall Mean	Comfort	0.43^{a}±0.03 (36)	0.26^{b}±0.01 (36)	0.21^{b}±0.01 (36)	0.25^{b}±0.01 (36)
	Heat	0.51^{A}±0.04 (36)	0.22^{B}±0.01 (36)	0.20^{B}±0.01 (36)	0.22^{B}±0.02 (36)

Means bearing different superscripts within same row differ significantly (^{AB}; p<0.01, ^{ab}; p<0.05).
Comfort (26±1° C), Heat (37±5° C)
G1 (Control), G2 (100 mg AA), G3 (200 mg AA), G4 (300 mg AA)

Feed intake (g/bird/day) of broilers at weekly intervals is presented in Table 2 and Mean body weight (g) of broilers birds at different intervals are presented in Table 3. The production performance of broilers is presented in Table 4. Significant difference (p<0.05) was observed in body weight gain between comfort and heat stressed condition in G1 group, whereas, non-significant difference was observed in other treatment groups. In comfort condition G1 and G2 groups differed significantly (p<0.05) from G3 and G4. However, non-significant difference was observed between G1 and G2; G3 and G4 groups. The maximum body weight gain (2116.95±32.58 g) was attained in G3 group, supplemented with 200 mg AA and minimum body weight gain (1928.11±19.40 g) was attained in control group. In heat stressed condition, all the four groups differed significantly (p<0.05) from each other. The maximum body weight gain (2062.82±34.93 g) was attained in G3 group, supplemented with 200 mg AA and minimum body weight gain (1656.33±28.17 g) was attained in control group.

Non-significant difference in total feed intake (kg) was observed between comfort and heat stressed condition in all the groups during entire experimental duration. In comfort condition, the total feed intake differed significantly (p<0.05) in G1 and G2 groups as compared to G3. However, non-significant difference was observed between G1, G2 and G4; G3 and G4 groups. In heat stressed condition, the total feed intake differed significantly (p<0.05) in G1 and G2 groups as compared to G3 and G4. However, non-significant difference was observed between G1 and G2; G3 and G4 groups.

Non-significant difference in FER was observed between comfort and heat stressed condition in all the groups during entire experimental duration. In comfort condition, significant (p<0.05) difference was observed between all groups. The maximum FER (0.68±0.00) was found in G3 group, supplemented with 200 mg AA and minimum FER (0.64±0.00) was found in control. In heat stressed condition, G1 and G3 groups differed significantly (p<0.05) from G2 and G4, whereas, non-significant difference was observed between G2 and G4. The maximum FER (0.67±0.00) was found in G3 group, supplemented with 200 mg AA and minimum FER (0.60±0.01) was found in control group.

Performance index showed non-significant difference between comfort and heat stressed condition in all the groups during entire experimental duration. In comfort condition, significant (p<0.05) difference was observed between all groups. The maximum PI was observed in G3 group, supplemented with 200 mg AA and minimum PI was observed in control. In heat stressed condition, significant (p<0.05) difference was observed between all groups. The maximum PI was observed in G3 group, supplemented with 200 mg AA and minimum PI was observed in control. On perusal of data, it was observed that, highest mortality in broilers was found in heat stressed control group. However, in heat stressed condition, the highest mortality (16.67%) of broiler was found in G1 group (heat stressed control group without any supplementation), whereas no mortality was observed in G2, G3 and G4 groups.

Table 2: Feed intake (g/bird/day) of broilers at weekly intervals.

Week	Condition	G1	G2	G3	G4
1st	Comfort	18.41±00.54	18.40±00.49	19.17±00.58	18.93±00.29
	Heat	17.08±00.43	18.20±00.27	18.33±00.35	18.11±00.49
2nd	Comfort	32.80±00.65	32.63±00.75	33.93±00.78	33.39±00.98
	Heat	31.28±00.91	31.96±00.83	32.65±00.98	32.29±00.72
3rd	Comfort	42.94±01.31	42.61±01.03	43.76±00.89	42.91±01.06
	Heat	36.17±00.90	40.88±00.61	41.16±00.63	42.08±01.27
4th	Comfort	49.25±01.69	48.62±01.62	50.19±01.18	49.56±01.49
	Heat	44.35±01.25	47.76±01.66	48.68±01.45	48.64±01.10
5th	Comfort	69.54±01.22	72.62±01.31	74.58±01.34	73.16±01.30
	Heat	64.64±00.99	71.64±00.97	72.86±01.37	71.83±01.27
6th	Comfort	85.65±01.54	84.78±00.93	87.49±01.16	84.92±01.23
	Heat	80.53±00.86	83.92±01.02	86.94±00.90	84.26±00.86
Total FI (Kg)	Comfort	2.98^{b}±06.20	2.99^{b}±06.77	3.09^{a}±06.56	3.02^{a}±06.13
	Heat	2.74^{b}±05.13	2.94^{b}±05.82	3.00^{a}±05.63	2.97^{a}±05.11

Means bearing different superscripts within same row differ significantly (^{ab}; p<0.05)
Comfort (26±1 °C), Heat (37±5 °C)
G1 (Control), G2 (100 mg AA), G3 (200 mg AA), G4 (300 mg AA).

Table 3: Mean body weight (g) of broilers birds at different intervals.

Week	Condition	G1	G2	G3	G4
0	Comfort	62.96±01.80 (12)	61.75±01.11 (12)	64.85±01.47 (12)	63.51±01.20 (12)
	Heat	61.67±01.11 (12)	64.01±01.32 (12)	62.32±01.03 (12)	64.23±01.46 (12)
3	Comfort	774.20±08.68 (12)	797.40±14.80 (12)	862.50±19.70 (12)	817.10±12.60 (12)
	Heat	758.90±09.19 (12)	772.20±08.05 (12)	828.50±17.80 (12)	751.70±16.30 (12)
6	Comfort	1991.07^{b}±20.90 (12)	2040.00^{b}±14.40 (12)	2181.80^{a}±32.20 (12)	2115.20^{a}±27.30 (12)
	Heat	1718.00^{b}±32.40 (12)	1980.55^{b}±21.20 (12)	2082.20^{a}±34.80 (12)	2058.10^{ab}±30.70 (12)
Body weight gain (g)	Comfort	1928.11^{bp}±19.40 (36)	1978.23^{b}±14.56 (36)	2116.95^{a}±32.58 (36)	2051.71^{a}±27.19 (36)
	Heat	1656.33^{dq}±32.17 (36)	1916.54^{c}±20.52 (36)	2019.88^{a}±34.93 (36)	1993.91^{b}±31.55 (36)

Means bearing different superscripts within same row differ significantly (^{ab}; p<0.05)
Means bearing different superscripts within same column differ significantly (^{pq}; p<0.05)
Comfort (26±1 °C), Heat (37±5 °C)
G1 (Control), G2 (100 mg AA), G3 (200 mg AA), G4 (300 mg AA)

Table 4: Production performance of broiler birds

Parameters	Condition	G1	G2	G3	G4
Wt. Gain (g)	Comfort	1928.11^{bp}±19.40	1978.23^{b}±14.56	2116.95^{a}±32.58	2051.71^{a}±27.19
	Heat	1656.33^{dq}±28.17	1916.54^{c}±20.52	2019.88^{a}±34.93	1993.91^{b}±31.55
F I (g)	Comfort	2985.90^{b}±06.20	2996.60^{b}±06.77	3091.20^{a}±06.56	3028.70^{a}±06.13
	Heat	2740.50^{b}±05.13	2943.60^{b}±05.82	3006.40^{a}±05.63	2972.10^{a}±05.11
FER	Comfort	0.64^{d}±00.00	0.66^{c}±0.01	0.68^{a}±00.00	0.68^{b}±00.00
	Heat	0.60^{c}±00.01	0.65^{b}±0.01	0.67^{a}±00.00	0.67^{b}±0.01
PI	Comfort	1233.99^{d}±09.79	1305.63^{c}±12.42	1439.52^{a}±13.54	1395.16^{b}±12.17
	Heat	993.80^{d}±05.81	1245.75^{c}±10.86	1382.08^{a}±12.86	1335.91^{b}±12.56
Mortality (%)	Comfort	00.00	00.00	00.00	00.00
	Heat	16.67	00.00	00.00	00.00

Means bearing different superscripts within same row differ significantly (^{abc}; p<0.05).
Means bearing different superscripts within same column differ significantly (^{pq}; p<0.05).
Comfort (26±1 °C), Heat (37±5 °C)
G1 (Control), G2 (100 mg AA), G3 (200 mg AA), G4 (300 mg AA)

Based on the data recorded the overall mean plasma concentration of HSP70 showed non-significant difference between comfort and heat stressed birds in all the groups on varying concentrations of AA supplementation. [10] Reported hepatic HSP70 content during heat stress and found out that increase in hepatic HSP70 concentrations were significantly less in broilers, which do not match to present findings. [11] Reported that hepatic concentrations of HSP70 at in broiler birds exposed at 41ºC and 70 to 80 percent relative humidity had higher HSP70 concentrations than those birds subjected to more heat stress during experimental period. In present findings AA supplemented group of chickens had significant reduction in the concentrations of plasma HSP70. This confirms the role of ascorbic acid in reducing stress at cellular levels acting as potent anti-oxidant and a molecular chaperone. The increase in the plasma HSP70 concentrations in present investigation may be due to heat stress induced production of ROM, inducing secretion of HSP70 titers in plasma. In the present investigation, significantly (p<0.05) higher concentration of HSP70 was observed in control group of comfort condition as compared to other treatment groups. Also, significantly (p<0.01) higher concentration of HSP70 was observed in heat stressed control group as compared to other heat stressed treatment groups with varying concentrations of ascorbic acid supplementation. The probable reason for such increase in HSP concentration might be due to thermo tolerance, which was acquired by the birds when they are exposed to cyclic heat stress. Such thermo tolerance is associated with HSP70 induction and heat conditioning resulted in an increase in the expression of HSP70, when chickens were further challenged with stressful stimuli [12]. The chickens experience less stress when they were provided with AA during heat stress. Increase in HSP70 helps the cells to recover from previous stressors and provide them transitory degree of protection [13] which is also revealed in the present research investigation.

Significant difference (p<0.05) was observed in body weight gain (BWG) between broilers maintained in comfort and heat stressed condition in control group, whereas, non-significant difference was observed in other treatment groups supplemented with 100, 200 and 300 mg AA. Results of present study revealed that the birds received 200 mg AA mg/bird/day in diet had about 10% and 20% improvement in BWG during comfort and heat stressed condition as compared to control (un-supplemented) birds respectively. Various workers reported the effect of AA supplementation on BWG during heat stress in broilers. In a recent study by [14] broilers subjected to chronic heat stress had significantly reduced feed intake (16.4%), lower body weight (32.6%) and higher feed conversion ratio (25.6%) at 45 days of age in AA non-supplemented group of broilers. [15] Reported significant improvement in BW of birds provided with AA in the feed, which is similar to present findings. [16] Observed that AA supplementation increased BWG in broilers exposed to high ambient temperature. [17] Reported that BW and BWG of broilers fed diet supplemented with combination of different levels of AA and Zn, reared at 40 °C was significantly higher than those with non-supplemented diet, with the maximum weight being recorded at 200 mg AA and 70 mg Zn per kg diet, which is similar to our findings. In agreement to present reports, [18] reported that dietary addition of AA did not show any effect on BWG of broilers subjected to heat stressed condition. Dietary AA supplementation of either 150 or 300 mg/kg did not affect BW and BWG of the turkeys [19]. [20] Reported the effects of heat conditioning and dietary ascorbic acid supplementation on growth performance in heat-stressed broilers. Findings indicate that heat stress significantly decreased final body weight and weight gain (p<0.001) in broilers, which are in disagreement to present findings. The mechanism (s) of heat stress amelioration imparted by AA supplementation supported by two hypotheses: (1) AA decrease heat load by lowering heat production. (2) AA increases heat loss by influencing avenues of thermal exchange between the body and the surrounding environment [21] results from the present study support this hypothesis. Thus, supplemental AA resulted in increased feed consumption because of lowering effect of AA on heat load. Moreover, AA is involved in reducing free radical formation and thus providing welfare besides less secretion of corticosterone, a catabolic hormone.

In present investigation, the differences in feed intake among the AA supplemented groups at different conditions were non-significant. However, the feed intake within conditions (comfort and heat stressed) differed significantly ($p<0.05$). Significant effects of higher ambient temperature on feed intake have been reported by [22]. The low feed intake at higher ambient temperature would be because of hyperthermia in birds. Furthermore, higher the feed intake, higher will be the heat generated by metabolism of feed; therefore, in order to reduce heat increment, birds in AA un-supplemented group might have reduced the feed intake. Higher levels of epinephrine also decreases feed intake by reducing blood supply to gastro intestinal tract [23]. Apart from these, the gene controlling the feed intake and appetite like Grehlin might also be down regulated. [24] Reported that reduced feed intake at 34 °C were 50 percent as compared to at 24 °C during 2 to 4 weeks of age. The average daily feed intake (g) in 3 weeks old broiler chickens under normal temperature (23 °C) and heat stress condition (41 °C) were 67.8 and 64.8 g respectively. They also observed that the feed efficiency at 34 °C were 32 percent lower as compared to at 24 °C. [20] Reported that heat stress significantly decreased feed consumption ($p<0.01$) in broiler birds. Heat stress significantly increased feed conversion ratio ($p<0.05$) in broiler birds, which is in disagreement to our findings.

Non-significant difference in FER was observed between comfort and heat stressed condition in all the groups during entire experimental duration. In comfort and heat stressed condition, significant ($p<0.05$) difference was observed between all groups. The maximum FER was found in 200 mg AA supplemented birds. In present study the efficiency of feed utilization was significantly more due to AA supplementation therefore, feed intake remained lower and FCR was significantly improved in comparison to un-supplemented control in both comfort and heat stressed condition. Feed intake of broilers was not drastically affected with improvement of FER as a result of AA supplementation during hot-humid summer. Present findings are in agreement with earlier reports of [25]. AA could be implicated in these observations because it is associated with the conversion of body proteins and fats into energy for production and survival through increased corticosterone secretion [26]. [27] Reported that AA supplementation does not significantly increase feed intake among groups. However, from the 3rd week, birds received 40 mg AA/day had the best FCR and heaviest in comparison with those birds received 20 mg and 0 mg of AA/bird/day. [28] Also reported non-significant difference in feed efficiency among the control and AA supplemented birds (100 g/tone of feed) until the 3rd week of the experiment.

[20] Reported the effects of dietary ascorbic acid supplementation on growth performance in heat-stressed broilers. Heat stress significantly increased feed conversion ratio ($p<0.05$) in broiler birds. [29] Reported that mean FCR was significantly lower ($p\leq0.05$) in vitamin C supplemented group, comparison to untreated control group indicating efficacy of AA in improving gut functions, metabolism, nutrient assimilation and utilization. Higher growth associated with feed (and water) intake, lower heat load and low blood corticosterone in heat stressed AA supplemented birds might be attributed to better feed conversion.

Effect of AA on performance index of broilers showed non-significant difference between comfort and heat stressed condition in all the groups during entire experimental duration. In comfort and heat stressed condition, significant ($p<0.05$) difference was observed between all groups and with maximum PI in 200 mg AA supplemented group. The probable reason for increased PI in 200 mg AA supplemented group is due to higher BGW and better FER values.

The highest mortality (%) in broilers was found in heat stressed control group which was supplemented with no AA. [30] Indicated that keeping birds at constant temperature of 39°C for 2 hrs resulted in 50% mortality, whereas temperatures oscillating from 35 to 38°C for 6 hrs produce less mortality. In the present investigation in heat stressed condition, the highest mortality (16.67%) was found in control group of broilers (AA un-supplemented group). The absence of temperatures greater that 37°C prevented reduction in the mortality of birds. The sudden exposure to heat might be the cause of higher mortality as observed by earlier workers, while in the present study the birds were reared in cage system with supplementation of varying concentration of AA, wherein the birds were continuously exposed to heat stress and thus had adaptation to high temperature. [31] Also recorded the lower mortality rate for broilers exposed to 36ºC at 5 days of age than those in control group under heat stress conditions. Higher mortality percentage at high temperature might be due to hyperthermia, acid base imbalance and inability of birds to cope up with higher ambient temperature and humidity resulted from insufficient stress responses. Decreased mortality with dietary AA supplementation may be due to the fact that AA ameliorates stress inducing suppression of hormonal and cell mediated immunity and improves the response of chickens to cell-mediated immunity [32].

4. Conclusion

In conclusion it was found that supplementation of 200 mg of ascorbic acid has a significant effect to decrease plasma HSP70 concentration during heat stress. Also it was found that ascorbic acid (200 mg) supplementation during heat stress tend to improve production performances in heat stressed broilers. Thus supplementation of ascorbic acid is essential to overcome the deleterious effects of heat stress conditions on the oxidative status and performance of broilers.

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