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Acute heat stress in broiler chickens and its impact on serum biochemical and electrolyte parameters

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ABSTRACT

Biochemical and electrolyte parameters play an assistant role for the faster and realistic evaluation of multiple organ dysfunction. The present study was designed to investigate the changes in serum biochemical and electrolyte parameters of Arbor Acres (AA) broiler chickens during acute heat stress. AA broiler chickens (N=120) were randomly allocated to four groups: control group at 22 ± 1 °C and heat stress group at 2 h, 5h and 10h, respectively (HS2, HS5, and HS10) at 38±1°C. All groups of serum electrolyte and biochemical parameters, including serum concentrations of sodium, potassium, chloride, calcium, total protein, albumin, total bilirubin (TBIL), direct bilirubin (DBIL), blood urea nitrogen (BUN), creatinine, amylase, cholesterol, alanine transaminase (ALT), aspartate aminotransferase (AST), creatine kinase (CK), and alkaline phosphatase (ALP) were measured. These results indicated that heat stress have significant (p <0.05) effect on serum biochemical and electrolyte parameters of broiler chickens, which assist preliminary evaluation of the damage degree of corresponding organs.

Key words: Acute heat stress, Biochemical parameters, Broiler chickens, Electrolyte.

INTRODUCTION

The livestock industry, especially poultry industry has suffered a lot due to constantly rising ambient temperature in recent decades. Therefore, heat stress leads to huge losses, especially in hot regions of the world because of its negative effect on feed consumption, growth performance, organ weight, immunosuppression and mortality (Smith, 2003; Mujahid *et al*., 2005; Gupta *et al*., 2014; Habibian *et al*., 2014; Hosseinivashan *et al*., 2016; Wan *et al*., 2017). Some previously studies have been reported on heat stress in poultry, using specific biochemical and electrolyte parameters could help by realistic evaluation of management practices, nutritional status, organ function and health conditions (Amrutkar *et al*., 2016; Hosseinivashan *et al*., 2016; Wan *et al*., 2017), with main focused on the changes of serum total protein, albumin, aspartate aminotransferase (AST), creatine kinase (CK), alkaline phosphatase (ALP), potassium, sodium, chloride and glucose (Azad *et al*. 2010; Habibian *et al*., 2014; Hosseinivashan *et al*., 2016; Wan *et al*., 2017). Thus, the objective of the present study was to evaluate the effects of AHS for various periods of time of the day in Arbor Acres (AA) broiler serum to calculate the fluctuation in the biochemical and electrolyte parameters during the early four weeks after hatching. These parameters may prove to be helpful in interpreting related organ functional changes in the animal body.

MATERIALS AND METHODS

The animal experiments and procedures were performed in strict accordance with the guidelines of the regional Animal Ethics Committee and the protocol was approved by the Institutional Animal Care and Use Committee of Henan Agricultural University.

One-day old AA broiler chickens (n=120) were purchased from Zhengda Poultry Breeding Company, Kaifeng, China. The chicks were given 28 days to acclimatize to their new growing environment and allowed them free access to a commercial feed (Broiler Formula Feed 701, Wuwei Tieqishi Feed Co., LTD, China) and water *ad libitum* during heat stress exposure. All chicks were randomly allocated to four groups (n=30 per group and n=10 per cage) and were raised in single layer metal cages (size, 100cm×80cm×60cm). The control group was maintained at (22 ± 1) °C and (50 ± 10) % relative humidity and the experiment group, which consisted of the heat-treated 2, 5 and 10 hours group (HS2, HS5 and HS10, respectively) was maintained at (38 ± 1) °C and (50 ± 10) % relative humidity in experimental rooms by using electric heaters and air humidifier throughout the experiment. 2 chicks of equal weight (average weight, approximately 1.2 kilogram) and same physical conditions were selected from each group and slaughtered rapidly by cervical dislocation. Blood samples were collected in anticoagulant (EDTA) tubes (2 mL) and centrifuged at 3, 000 g for 5 min. Serum was separated and transferred to Eppendorf tubes (1.5 mL) stored at -20°C for performing subsequent biochemical and electrolyte parameters.

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Serum biochemical parameters (ALT (alanine transaminase), AST, ALP, CK, total protein, albumin, globulin, TBIL (total bilirubin), DBIL (direct bilirubin), BUN (blood urea nitrogen), creatinine, amylase and cholesterol) were measured by using a Idexx VetTest 8008 Chemistry Analyzer (IDEXX Biotechnology Company, USA) in the Animal Hospital of Henan Agricultural University, Zhengzhou, China, according to the manufacturer's instructions.

The concentrations of sodium (Na), chloride (Cl), calcium (Ca) and potassium (K) were measured by a KD100VET electrolyte analyzer (Shenzhen Kindle Medical Devices Co. Ltd., China)

All data were presented as means±SEM. The comparisons between groups were performed using one-way ANOVA followed by Duncan multiple range test (Duncan, 1955) and differences were considered statistically significant at the level of (p<0.05) by applying SPSS statistics software of Windows (version 17; SPSS, Chicago, Illinois).

RESULTS AND DISCUSSION

Heat stress can be studied generally by using indicators such as clinical behavior, blood biochemical parameters and histological observations, which can indicate tissue damage (Zhang *et al*., 2015). Especially specific biochemical and electrolyte parameters could help with a faster and realistic evaluation of organ function and tissue damage in poultry (Sasipriya *et al*., 2013; Ma, 2014). Amrutkar *et al*., (2016) reported that different tropical stress conditions have an obviously influence on biochemical parameters in various broiler strains. Similarly, the results of present study showed significant changes in Na, Cl, K, ALT, AST, ALP, CK, total protein, albumin, globulin, TBIL, DBIL, BUN, creatinine, amylase and cholesterol levels (except Ca) in AHS for various periods of time (exposing 2h, 5h and 10h) in broiler chickens compared to the control group (Table 1).

Changed levels of Na, Cl, K, BUN and creatinine are consistently correlated with kidney function and usually used to evaluate kidney damage. The primary function of the kidney is the regulation of acid-base balance to maintain electrolyte equilibrium (Lunn and Mcguirk, 1990; Koeppen, 2009; Shioji *et al*., 2016). In this study, Na level was significantly increased at HS2 (p<0.05) and K level was also significantly increased at HS5 (p<0.05) in broiler chickens compared to the control. The Cl level was significantly decreased at HS10 (p<0.05) and BUN level was significantly increased at HS5 ($p<0.05$) and HS10 ($p<0.01$), respectively. However, creatinine level was significantly increased at HS2 (p<0.01), HS5 (p<0.01) and HS10 (p<0.001) in broiler chickens compared to control. These showed that AHS significantly affect kidney related indicators. Similar findings were made by Belay and Teeter (1996) and Amrutkar *et al*., (2016) indicated that Na and K excretion levels increased, and Cl excretion decreased in urine and feces during heat stress. As Na and K are alkalogenic ions, its loss can lead to body fluid acidification (Sayed and Scott, 2007). Hence, kidney related parameters were changed during acute heat stress, which demonstrate that the function of kidney may be affected leading to acid-base disorder.

When the integrity of the hepatocellular membrane, muscles and cardiac muscle fibers are compromised, there is extrusion of the marker enzymes into the blood (Hsueh *et al*., 2011; Oche *et al*., 2014). In the present study, ALT levels were significantly increased at HS5 (p<0.05) and HS10 (p<0.01) and CK levels at HS5 (p<0.001) in broiler chickens compared to the control group. AST levels were significantly decreased at HS2 (p<0.05) and at HS5 (p<0.05) but were elevated at HS10. Meanwhile, ALP levels were also significantly decreased at HS10 (p<0.05) in broiler chickens compared to the control. It is observed that, the release of these enzymes into the serum is as a result of tissue injury or changes in the permeability of liver, heart or muscle membranes. Therefore, the level may increase with acute heat damage to liver, heart or muscle cells. Interference with the normal liver functions affects the rate of conjugation and excretion of bilirubin. Thus, a high level of bilirubin is used as indices for liver function and bile excretion status (Oche *et al*., 2014). The present study showed a significant increase at HS2 (p<0.05) in TBIL and DBIL while TBIL levels were significantly decreased at HS5 (p<0.001) and HS10 (p<0.01) in broiler chickens compared to the control. These demonstrated that AHS significantly affect hepatobiliary indicators, which was almost similar with the findings of Ye *et al*., (2015) reported that Kirin chicken liver injury induced by heat stress caused an extremely significant increasing in the activity of liver damage enzymes AST and ALT. In addition, Sahin (2012) also reported that heat stress inhibited the levels of protein and albumin and alleviated the effect of heat stress by supplement vitamin E in diet. In the present study, total protein and albumin levels were significantly decreased at HS2 (p<0.01)and globulin levels were also significantly decreased at HS2 (p<0.05) in broiler chickens compared to the control and the reduced serum total protein, albumin and globulin concentrations returned to control levels at HS5 (p>0.05, p<0.05 and p<0.01), respectively. Therefore, liver and gallbladder related biochemical parameters were also affacted, which implied that the function of the liver and gallbladder were damaged under AHS.

Reference to a large number of documents found that the latest research of serum amylase and cholesterol are rare in the study of biochemical indexes of broiler chickens. Wilding and Dawson (1967) reported that serum amylase mainly comes from the pancreas and intestine. In addition, it is generally accepted that hyperamylasemia only results if

Table 1: Effects of AHS on biochemical and electrolyte parameters of the broiler chickens

Parameters[1,2]	Control group (n=6)	Heat stress group[3]		
		HS2 (n=6)	HS5 (n=6)	HS10 (n=6)
Sodium (mmol/L)	145.42±0.33	150.57±0.13*	146.06±0.37	143.66±0.64
Potassium(mmol/L)	15.16±0.78	12.40±0.25	17.376±2.03*	12.89±0.67
Chloride (mmol/L)	107.10±0.97	111.33±1.24	107.86±1.25	103.16±0.92*
Calcium (mmol/L)	0.59±0.01	0.39±0.04	0.44±0.01	0.58±0.01
BUN (mmol/L)	0.86±0.03	0.91±0.01	1.046±0.04*	1.26±0.05**
Creatinine (μmol/L)	59.60±1.78	82.60±1.50**	93.00±1.30**	129.80±4.42***
ALT (U/L)	4.00±0.71	4.33±0.33	5.25±0.50*	5.40±0.68**
AST (U/L)	247.60±5.31	214.00±6.00*	185.75±36.23*	284.60±26.66
ALP (U/L)	861.20±188.83	818.909±93.01	897.75±163.92	497.40±109.00*
CK (U/L)	530.40±227.56	1364.67±761.36	2854.25±174.07***	715.80±214.73
Total protein (g/L)	32.60±1.40	22.67±0.33**	36.00±2.74	34.80±0.86
Albumin (g/L)	17.20±0.80	13.00±0.00**	15.25±0.48*	16.80±0.49
Globulin (g/L)	15.40±0.75	9.67±0.33*	20.750±3.119**	18.00±0.84
TBIL, (mmol/L)	9.86±0.453	15.83±3.13*	4.750±0.296***	4.98±1.23**
DBIL (mmol/L)	1.28±0.26	2.87±0.92*	1.77±0.07	1.64±0.40
Amylase (U/L)	375.20±24.15	291.33±9.39*	442.50±133.85	287.00±23.61
Cholesterol(mmol/L)	3.04±0.14	1.99±0.04***	3.18±0.19	3.18±0.13

[1]Abbreviations: ALT: alanine transaminase; AST: aspartate aminotransferase; ALP: alkaline phosphatase; CK: creatine kinase; TBIL: total bilirubin; DBIL: direct bilirubin; BUN: blood urea nitrogen; U/L=unit of enzyme activity/L.

[2]*asterisks* indicate values that are significantly different between the control and heat stress groups (*p<0.05; **p<0.01; ***p<0.001); Values are means±SEM (Standard Error of Mean).

[3]HS2, HS5 and HS10: heat stress at 2 hours, 5 hours and 10 hours.

the excretory channels of the organ are obstructed. Winslet *et al*. (1992) indicated that the amylase diagnostic value of pancreatitis was found in 96.1% of all mild cases and 87.4% of severe cases. In this study, amylase levels were significantly decreased at HS2 (p<0.05) and not significantly increased at HS5 in broiler chickens compared to the control group. It can be infer that pancreas function may suffer from slight influence. In addition, cholesterol levels were significantly decreased at HS2 (p<0.001) in broiler chickens compared to the control and the elevated serum cholesterol concentrations returned to control levels at HS5 and HS10. In clinic, the diagnosis of this change does not have much meaning, although cholesterol index has a fluctuation during heat stress. All in all, these results have demonstrated that the serum biochemical and electrolyte parameters were markedly changed and can be used as a sero-marker on diagnosis corresponding to organ dysfunction under AHS in broiler chickens.

CONCLUSION

Biochemical and electrolyte parameters play an assistant role for the faster and realistic evaluation of multiple organ dysfunctions. The parameters in this study changed under AHS and have a significant diagnostic aid of various organs (such as liver, kidney, heart and pancreas) and can help in interpreting related organ dysfunction of animal body.

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