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Biochemical studies in experimentally *Salmonella* Gallinarum infected broiler chicken intoxicated with alpha-cypermethrin

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Abstract

Biochemical studies were conducted on two hundred and thirty, day old broiler chicken experimentally infected with Salmonella Gallinarum and exposed to alpha-cypermethrin. These chicks at the age of 7 days were divided randomly into three groups viz. group A, B and C containing 70, 80 and 80 chicks, respectively. All the birds of group B and C were given alpha-cypermethrin @ 63.94 mg/kg bwt (1/5th of MTD) and @ 31.97 mg /kg bwt (1/10th of MTD), respectively orally through drops throughout the experiment. The group A was kept as control i.e. no alpha-cypermethrin was given. At the age of 14 days, the birds of groups B and C were divided into two subgroups (group B into B1 and B2, group C into C1 and C2) of 40 birds each. Similarly, birds of group A were divided into two subgroups A1 and A2 of 30 and 40 birds, respectively. Thereafter, each bird of subgroup A_2 , B_2 and C_2 were injected intraperitoneally 107 CFU Salmonella Gallinarum in 1 ml normal saline solution. Blood samples were collected from six birds of each subgroup directly from heart subsequently on 0, 7th, 14th, 21st and 28th day post infection in sterile tubes for serum separation for biochemical parameters analysis. Serum samples were stored in aliquots at -20°C until its use for different biochemical parameters. Biochemical studies revealed significantly higher mean serum AST, ALT, LDH, ALP activities, creatinine concentration and significantly lower total protein and albumin concentration in all the infected groups as compared to control group.

Keywords: Alpha-cypermethrin, chicken, biochemical

Introduction

Alpha-cypermethrin is a highly active pyrethroid insecticide consisting essentially of two of the four cis-isomers comprising cypermethrin. Alphamethrin and alkoxylate are synonyms used for alpha-cypermethrin. The main effects of pyrethroids are achieved through action on sodium and chloride channels (Narahashi et al., 2007; Boumba et al., 2017)^[31, 8]. In veterinary medicine it is applied topically, as a spray or pour on to cattle and sheep (150 mg per animal) and as a spray to poultry (8 to 10 mg per bird) for controlling ectoparasites such as ticks, fleas, lice and blow flies (Narayani, 2012)^[32]. Instances of poisoning due to alpha-cypermethrin are on increase in recent years due to their indiscriminate use. Repeated exposure of poultry to chemical insecticides causes health consequences to poultry culminating in great economic loss, while also posing a potential threat to public health due to the presence of insecticide residues in chicken (Mondal, 2014) ^[29]. The extensive use of synthetic pyrethroids in agriculture where both plants and domestic animals are affected necessitates the detailed study on the concentrations of their residues in foodstuffs. Indiscriminate use of insecticides has led to a widespread concern over the potential adverse effects of these chemicals on animal and human health (Al-Saleh, 1994)^[1] as these chemicals interfere with the defense mechanisms of the hosts which in turn makes host primarily poultry susceptible to the potentially highly invading pathogens like Salmonella spp. (Cabassi, 2007)^[10] which is responsible for deadliest disease in poultry i.e. fowl typhoid. Keeping in view the above facts biochemical studies were undertaken in experimentally Salmonella Gallinarum infected broiler chicken exposed to alpha-cypermethrin.

Materials and methods

Ethical approval

The study was conducted after the approval of the Institutional Animal Ethics Committee.

Study design

For the present study two hundred and thirty, day old broiler chicks were procured. These chicks at the age of 7 days were divided randomly into three groups viz. group A, B and C containing 70, 80 and 80 chicks, respectively. All the birds of group B and C were given alpha-cypermethrin @ 63.94 mg/kg bwt (1/5th of MTD) and @ 31.97 mg /kg bwt (1/10th of MTD), respectively orally through drops throughout the experiment. The group A was kept as control i.e. no alphacypermethrin was given. At the age of 14 days, the birds of groups B and C were divided into two subgroups (group B into B_1 and B_2 , group C into C_1 and C_2) of 40 birds each. Similarly, birds of group A were divided into two subgroups A₁ and A₂ of 30 and 40 birds, respectively. Thereafter, each bird of subgroup A₂, B₂ and C₂ were injected intraperitoneally 107 CFU Salmonella Gallinarum in 1 ml normal saline solution.

Collection of sample

Blood samples were collected from six birds of each subgroup directly from heart subsequently on 0, 7th, 14th, 21st and 28th day post infection in sterile tubes for serum separation for biochemical parameters analysis. Serum samples were stored in aliquots at -20°C until its use for different biochemical parameters.

Serum biochemical examination

Serum samples were analyzed for different biochemical parameters by using semi-automatic biochemistry analyzer (Erba Mannheim Chem-5 Plus, Transasia) and different kits procured from ERBA diagnostics Mannheim GmbH (Transasia Bio-Medicals Ltd.). The levels of following serum biochemical parameters were determined: Total protein (TP), albumin, creatinine concentrations and enzyme activities of alanine transaminase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH),

Statistical analysis

The data for various parameters were subjected to statistical

analysis by using Duncan Multiple Range Test as modified by Krammer (1957)^[24] at 5 per cent level of significance using SPSS 16.0 version software. Individual means were compared for statistical significance using least significance difference.

Results and discussion

Total protein

Mean total serum protein (TSP) concentrations of different groups at various intervals are illustrated in Table -1. Mean total serum protein concentrations in all infected groups (A₂, B_2 , and C_2) were lower as compared to control group (A₁) throughout the experiment. Though this decrease was significant (P \leq 0.05) in group A₂ from 14DPI to 21DPI, in group C_2 from 14DPI onward and in group B_2 from 7DPI onwards as compared to control group (A_1) . The total serum protein concentration in the birds of group B_2 as compared to group A₂ was found significantly (P ≤ 0.05) decreased from 21DPI onwards. More or less similar observation have been reported by other workers in Salmonella Gallinarum infection (Shah et al., 2007; Saha et al., 2012; Shah et al., 2013; Kumari et al., 2015; Biazus et al., 2017; Singh, 2017) [39, 37, 41, ^{26, 5, 42]}. In contrast to our findings, some workers have found increase in the concentration of total serum protein in Salmonella Gallinarum infection (Deshmukh et al., 2007)^[12]. In contrary to our findings, no significant alteration in serum protein was reported by Nehra (2013) [34] in Salmonella Gallinarum infection. The mean total serum protein concentration in group B_1 and C_1 was significantly ($P \le 0.05$) lower from 28DPAT onwards as compared to control group (A₁). Garg *et al.* (2004) ^[14] observed decrease in total serum protein in broiler chicks intoxicated with synthetic pyrethroid, organophosphate and chlorinated pesticides. Grewal et al., 2010 ^[16] concluded toxicity of cypermethrin in rats by administering cypermethrin @ 14.5mg/kg b.wt.once daily for 30 days caused significant decrease in level of serum proteins. Narayani, 2010^[32] found that subacute exposure of alphamethrin in broiler chicks leads to significant decrease in total serum protein concentration.

Mean ± S.E total serum protein concentration (g/dl) of broiler chicks						
Crouns	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
Groups	0/7	7/14	14/21	21/28	28/35	
A ₁	4.00 ^a ±.31	4.12 ^b ±.27	4.28 ^b ±.24	4.26°±.15	4.34°±.23	
A ₂	4.08 ^a ±.23	$3.59^{ab} \pm .22$	2.85 ^a ±.23	3.22 ^b ±.18	$3.82^{bc} \pm .38$	
B 1	3.76 ^a ±.27	$3.62^{ab} \pm .19$	3.58 ^b ±.20	$2.88^{ab} \pm .18$	2.47 ^a ±.21	
B ₂	3.66 ^a ±.36	$3.24^{a} \pm .31$	2.45 ^a ±.19	$2.46^{a}\pm.17$	2.52 ^a ±.33	
C1	3.86 ^a ±.27	$3.72^{ab} \pm .28$	3.65 ^b ±.33	3.33 ^b ±.19	3.29 ^{ab} ±.13	
C ₂	$3.84^{a} \pm .28$	3.41 ^{ab} ±.29	2.65 a±.22	3.02 ^{ab} ±.25	2.97 ^{ab} ±.36	

Table 1: Mean ± S.E total serum protein concentration (g/dl) of broiler chicks in different experimental groups at different intervals

Means \pm S.E-Different superscripts in the same column differ significantly (P \leq 0.05)

Albumin

Mean serum albumin concentrations of different groups at various intervals are illustrated in Table -2. Mean serum albumin concentrations in all infected groups (A₂, B₂ and C₂) were lower as compared to control group (A₁) throughout the experiment though this decrease was significant ($P \le 0.05$) in group A₂ from 14DPI to 21DPI and groups B₂ and C₂ from 14DPI onwards as compared to control group (A₁). Almost similar observation have been reported by other workers in *Salmonella* Gallinarum infection (Shah *et al.*, 2007; Saha *et al.*, 2012; Shah *et al.*, 2013; Kumari *et al.*, 2015; Biazus *et al.*

2017; Singh, 2017) ^[39, 37, 41, 26, 5, 42]. However, some workers have found increase in the concentration of serum albumin in *Salmonella* Gallinarum infection (Deshmukh *et al.*, 2007) ^[12]. In contrary to our findings, no significant alteration in serum albumin was reported by Nehra (2013) ^[34] in *Salmonella* Gallinarum infection. The mean serum albumin concentration in groups B₁ was significantly (P \leq 0.05) lower from 28DPAT onwards as compared to control group (A₁). More or less similar observations have been reported by other workers in alpha-cypermethrin toxicity (Narayani, 2010) ^[32]. According to Blood *et al.*, 1994 ^[7] liver is the prime site for albumin synthesis. In the present study, inappetance, damage to liver and kidneys as evidenced by gross and histopatholocal studies were observed leading to decrease in total albumin concentration.

Table 2: Mean ± S.E. serum albumin concentration (g/dl) of broiler chicks in different experimental groups at different intervals

			Mean ± S.E serum albumin concentration (g/dl) of broiler chicks						
Days post in	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)								
Groups 0/7	7/14	14/21	21/28	28/35					
A ₁ $2.54^{a}\pm0.10$	2.78 ^a ±0.28	3.18 ^b ±0.31	3.10 ^b ±0.29	2.69°±0.29					
A ₂ $2.74^{a}\pm0.22$	2.39 ^a ±0.32	1.79 ^a ±0.30	$1.98^{a}\pm0.28$	2.30 ^{abc} ±0.56					
B ₁ $2.85^{a}\pm0.35$	2.62 ^a ±0.10	2.66 ^b ±0.21	2.07 ^a ±0.19	1.69 ^a ±0.25					
B ₂ $2.48^{a}\pm0.21$	1.99 ^a ±0.27	1.64 ^a ±0.20	1.79 ^a ±0.13	1.59 ^a ±0.32					
C ₁ $2.66^{a} \pm 0.24$	2.73 ^a ±0.32	3.01 ^b ±0.28	2.45 ^{ab} ±0.17	2.43 ^{bc} ±0.17					
C ₂ $2.73^{a}\pm0.22$	2.09 ^a ±0.23	1.71 ^a ±0.22	1.93ª±0.25	1.83 ^{ab} ±0.17					

Means \pm S.E.-Different superscripts in the same column differ significantly (P \leq 0.05)

Mean values of serum albumin-globulin ratio were lower in the birds of all infected groups (A₂, B₂, and C₂) though this decrease was not significant ($P \le 0.05$) as compared to group A₁,B₁,C₁ throughout the experiment. Kokosharov (2006) ^[23] also observed a decrease in albumin globulin ratio in experimental acute fowl typhoid infection in birds.

Aspartate transaminase (AST) and alanine transaminase (ALT) activities

Mean serum aspartate transaminase (AST) and alanine transaminase (ALT) activities of different groups at various intervals are illustrated in Table -3 and 4, respectively. Mean serum aspartate transaminase (AST), and alanine transaminase (ALT) activities were found to be significantly increased in all the *Salmonella* Gallinarum infected groups as

compared to control group. The increased serum ALT and AST activity has also been reported in many other studies (Shah *et al.*, 2007; Garcia *et al.*, 2010; Shah *et al.*, 2013; Barde, 2014; Kumari *et al.*, 2015; Biazus *et al.*, 2017) [^{39, 13, 41, 3, 26, 5]} due to experimental *S*. Gallinarum infection in broilers chicks. The increase in serum AST is indicative of cellular injury to cardiac muscle and hepatocytes where as elevated serum ALT is mostly due to hepatic injuries and to some extent due to other visceral organs such as heart, skeletal muscles, kidneys, pancreas, spleen and lungs (Campbell and Coles, 1986) ^[11] which have been noticed in the present study too as evidenced by pathological finding. ALT a highly liver specific enzyme (Lukaszewicz-Hussain and Moniuszko-Jakoniuk, 2005) ^[27] is one of the most reliable indicators of hepatotoxic damage (Ozer *et al.*, 2008) ^[35].

Table 3: Mean ± S.E. serum aspartate transaminase activity (IU/L) of broiler chicks in different experimental groups

Mean ± S.E. serum aspartate transaminase activity (IU/L) of broiler chicks						
Crowna	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
Groups	0/7	7/14	14/21	21/28	28/35	
A ₁	188.09 ^a ±9.49	196.39 ^a ±10.61	203.40 ^a ±10.50	199.00 ^a ±9.77	210.80 ^a ±12.79	
A ₂	189.35 ^a ±4.24	241.44 ^{bc} ±10.77	280.92°±9.29	234.02 ^b ±8.78	218.26 ^a ±9.25	
B 1	191.52 ^a ±7.28	219.15 ^{ab} ±6.90	248.74 ^b ±8.68	275.15°±8.25	283.97 ^b ±9.32	
B ₂	193.71 ^a ±7.93	265.83°±9.73	347.82 ^d ±10.75	310.04 ^d ±11.06	292.00 ^b ±9.95	
C1	191.65 ^a ±7.15	200.81 ^a ±9.15	200.74 ^a ±9.56	221.82 ^{ab} ±11.94	225.64 ^a ±8.79	
C ₂	191.69 ^a ±6.12	245.13 ^{bc} ±10.61	281.06°±6.16	239.04 ^b ±9.47	230.63 ^a ±11.69	

Means \pm S.E-Different superscripts in the same column differ significantly (P \leq 0.05)

Table 4: Mean \pm S.E. serum alanine transaminase activity (IU/L) of broiler chicks in different experimental groups

Mean ± S.E. serum alanine transaminase activity (IU/L) of broiler chicks						
Caracter	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
Groups	0/7	7/14	14/21	21/28	28/35	
A1	9.23 ^a ±.50	9.72 ^a ±.55	8.93 ^a ±.54	9.13 ^a ±.60	9.55 ^a ±.43	
A ₂	10.35 ^{ab} ±.59	20.26 ^b ±.72	31.28°±2.40	25.37°±1.97	14.48 ^a ±1.75	
B 1	11.70 ^b ±.63	18.05 ^b ±1.92	22.55 ^b ±1.89	29.51°±1.59	31.97°±1.75	
B ₂	11.16 ^b ±.68	24.71°±1.48	42.70 ^d ±2.84	41.89 ^d ±2.29	41.10 ^d ±2.21	
C1	10.22 ^{ab} ±.78	12.89 ^a ±1.37	$14.14^{a}\pm1.44$	18.18 ^b ±1.23	21.54 ^b ±1.79	
C2	11.04 ^{ab} ±.73	20.65 ^b ±1.16	32.92°±1.97	29.07°±1.87	24.50 ^b ±1.89	

Means \pm S.E-Different superscripts in the same column differ significantly (P \leq 0.05)

Present studies also revealed significantly increased mean serum aspartate transaminase, alanine transaminase activities in alpha-cypermethrin intoxicated groups as compared to control group. These results are in consonance with the findings from earlier studies (Manna *et al.*, 2006; Hocine, 2016; Ghorzi *et al.*, 2017) ^[28, 29, 15]. The increase in serum AST is indicative of degenerative changes/alteration in permeability of cell membrane in hepatocytes and cardiac muscles, resulting in release of the transaminases in the blood stream (Shah and Gupta, 1997) ^[40]. Elevation in ALT activity

indicates liver damage (Brar *et al.*, 2000) ^[9], which have been noticed in the present study to as evidenced by pathological finding.

Lactate dehydrogenase (LDH) activities

Mean serum lactate dehydrogenase (LDH) activities of different groups at various intervals are illustrated in Table -5. The present study revealed that the mean values of lactate dehydrogenase (LDH) activities were significantly higher in the birds of all infected groups (A_2 , B_2 , and C_2) as compared

to the birds of control group (A₁) throughout the experiment. Kumari *et al.* (2015) ^[26] and Singh (2017) ^[42] reported that lactate dehydrogenase activities were significantly higher in *Salmonella* Galiinarum infected groups. It was noticed that the mean values of LDH activities showed significant (P \leq 0.05) increase in the birds of group B₁from 14DPAT onwards as compared to the birds of group A₁ and C₁. Grewal *et al.*

(2010) ^[16] observed a significant increase in serum LDH in toxicity of cypermethrin in rats. Increase in serum LDH level indicates hepatocellular necrosis, myocardial damage, renal necrosis, pancreatic necrosis and hemolysis (Benjamin, 1985) ^[4] which have been recorded in the present study as evidenced by pathological findings.

Table 5: Mean ± S.E. serum lactate dehydrogenase (IU/L) of broiler chicks in different experimental groups

Mean ± S.E. serum lactate dehydrogenase (IU/L) of broiler chicks						
C	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
Groups	0/7	7/14	14/21	21/28	28/35	
A ₁	351.86 ^a ±3.98	$346.92^{a} \pm 5.48$	360.32 ^a ±5.59	350.59 ^a ±6.13	342.41 ^a ±7.16	
A_2	350.32 ^a ±6.07	$361.89^{a} \pm 5.86$	496.30°±8.64	443.49°±9.67	385.39 ^b ±10.68	
B 1	355.44 ^a ±5.59	405.31° ±10.15	416.58 ^b ±8.24	467.00°±8.95	491.46 ^d ±10.31	
\mathbf{B}_2	353.64 ^a ±5.58	$435.23^{d} \pm 7.41$	$566.52^{d} \pm 2.94$	525.51 ^d ±12.60	519.26 ^d ±10.00	
C1	356.53 ^a ±5.58	385.46 ^b ±10.05	386.55 ^a ±2.10	391.55 ^b ±13.79	419.46°±18.22	
C ₂	356.79 ^a ±4.67	427.38 ^d ±9.01	506.73°±9.14	445.53°±7.00	421.47°±6.75	

Means \pm S.E-Different superscripts in the same column differ significantly (P \leq 0.05)

Alkaline phosphatase (ALP) activities

Mean serum alkaline phosphatase (ALP) activities of different groups at various intervals are illustrated in Table -6. The mean values of alkaline phosphatase (ALP) activities were higher in the birds of all infected groups $(A_2, B_2, and C_2)$ as compared to the birds of control group (A₁) throughout the experiment though this increase was significant ($P \le 0.05$) in the birds of group A₂ from 7DPI to 21DPI and in the birds of groups B₂ and C₂ from 7DPI onwards as compared to the birds of control group (A_1) . There was also an increase in the mean values of ALP activity in the birds of group (B₂) as compared to the birds of group A₂ and C₂ throughout the experiment though this increase was significant (P \leq 0.05) from 7DPI onwards. Similarly, the mean values of ALP activity was found significantly ($P \le 0.05$) increased in the birds of group (C_2) as compared to the birds of group A_2 on 21DPI. According to Barde (2014)^[3], experimentally infected quail with 10⁶ Salmonella enterica serovar Gallinarum revealed increase in serum ALP. Singh (2017) [42] also reported elevated level of serum ALP in Salmonella Gallinarum infected broiler chickens. In contrast to our findings, some workers have reported decreased serum ALP activity due to S. Gallinarum infection (Kokosharov et al., 1997; Kumari, 2012)

 $^{[22, 25]}$. Mean values of ALP activity showed significant (P \leq 0.05) increased in the birds of group B_1 from 21DPAT onwards and in the birds of group C_1 on 35DPAT as compared to the birds of group A_1 . It was also noticed that there was significant increase ($P \le 0.05$) in the mean values of ALP activity in the birds of group B_1 from 14DPAT onwards as compared to the birds of group C_1 . More or less similar observations have been reported by other workers in birds and rats (Manna et al., 2006; Narayani, 2010; Hocine et al., 2016) ^[28, 32, 19]. Alkaline phosphatase catalyzes several reactions in the body and is involved in the active transport of phosphate, synthesis of proteins, and DNA turnover in the nucleus (Nehra, 2013)^[34]. It is also involved in hydrolysis of esters of phosphoric acids and phosphate fixation in the tissue and bones during development (Biogin et al., 1981)^[6]. According to Kaplan and Righetti (1970)^[21], any cellular damage in the body of animal releases phosphatases into the systemic circulation and thereby increases their concentration in the blood. So, increased activity of alkaline phosphatase in serum in both alpha-cypermethrin treated and infected groups might be due to lesions observed in liver and intestine (Msoffe et al., 2006; Hendawi et al., 2016) [30, 18].

Table 6: Mean ± S.E. serum alkaline phosphatase activity (IU/L) of broiler chicks in different experimental groups

Mean ± S.E. serum alkaline phosphatase activity (IU/L) of broiler chicks						
Crowne	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
Groups	0/7	7/14	14/21	21/28	28/35	
A ₁	340.28 ^a ±14.02	377.07 ^a ±13.92	380.24 ^a ±11.27	400.17 ^a ±8.61	415.56 ^a ±11.98	
A ₂	341.48 ^a ±12.33	426.47 ^b ±12.69	580.26° ±12.33	460.43 ^b ±11.73	440.66 ^{ab} ±10.83	
B 1	343.38 ^a ±9.28	391.45 ^a ±12.36	500.41 ^b ±15.34	545.56 ^d ±11.60	580.45° ±15.59	
B ₂	344.40 ^a ±8.33	475.35 ^c ±11.86	650.40 ^d ±13.74	630.27 ^e ±16.41	590.46° ±11.99	
C1	342.48 ^a ±7.42	365.63 ^a ±6.39	390.41 ^a ±7.04	420.41 ^a ±9.16	$460.55^{b} \pm 9.98$	
C2	342.55 ^a ±5.83	428.40 ^b ±6.93	581.57° ±11.55	500.54° ±13.22	$470.45^{b} \pm 7.14$	

Means \pm S.E-Different superscripts in the same column differ significantly (P \leq 0.05)

Serum creatinine

Mean serum creatinine concentrations of different groups at various intervals are illustrated in Table -7. Mean values of serum creatinine was higher in the birds of all infected groups (A₂, B₂, and C₂) as compared to the birds of control group (A₁) throughout the experiment though this increase was significant in the birds of group B₂from 14DPI onward and in the birds of groups C₂ only on 14DPI as compared to the birds of control group (A₁). There was no significant change in

other groups as compared to control group. This increase might be due to renal damage (Benjamin, 1985; Kaneko *et al.*, 2009) ^[4, 20]. Creatinine, a by-product of muscle metabolism (Quintanilla, 1982) ^[36] is excreted in the urine by the kidneys and thus renal insufficiency or obstruction results in an elevated creatinine concentration (Ambali *et al.*, 2007) ^[2]. Nehra (2013) ^[34] observed elevated level of serum creatinine in *Salmonella* Gallinarum infection in broiler chicken. Increased serum creatinine has also been observed in guinea pigs due to *Salmonella Dublin* infection (Gupta *et al.*, 1999) ^[17] and in Hubbard breed broilers due to *Salmonella*

typhimurium infection (Salim et al., 2011)^[38].

Table 7: Mean \pm S.E. serum creatinine concentration (mg/dl) of broiler chicks in different experimental groups

Mean ± S.E. serum creatinine concentration (IU/L) of broiler chicks						
Groups	Days post infection (DPI)/Days post alpha-cypermethrin treatment (DPAT)					
	0/7	7/14	14/21	21/28	28/35	
A ₁	0.62 ^a ±0.01	$0.60^{a}\pm0.02$	$0.61^{a} \pm 0.02$	$0.62^{a} \pm 0.02$	$0.61^{a} \pm 0.02$	
A ₂	0.61 ^a ±0.01	$0.62^{a}\pm0.01$	$0.65^{ab} \pm 0.01$	$0.63^{a} \pm 0.01$	$0.62^{a} \pm 0.01$	
B ₁	0.62 ^a ±0.01	0.61 ^a ±0.01	$0.63^{ab} \pm 0.01$	$0.64^{a} \pm 0.01$	0.65 ^{ab} ±0.01	
B ₂	0.63 ^a ±0.01	$0.62^{a}\pm0.01$	0.71° ±0.02	$0.70^{b} \pm 0.01$	$0.67^{b} \pm 0.01$	
C1	0.62 ^a ±0.01	0.61 ^a ±0.01	$0.62^{a} \pm 0.01$	$0.62^{a} \pm 0.01$	$0.63^{a} \pm 0.01$	
C2	0.62 ^a ±0.01	$0.62^{a}\pm0.01$	$0.67^{bc} \pm 0.02$	$0.66^{a} \pm 0.01$	$0.62^{a} \pm 0.01$	
Means \pm S.E-Different superscripts in the same column differ significantly (P ≤ 0.05)						

Conclusion

Based upon present study, it is concluded that there was significant decrease in serum total protein, albumin concentrations whereas significant increase in serum ALT, AST, LDH, alkaline phosphatase activities and serum creatinine concentrations, in all the infected groups as compared to control group A₂. The biochemical changes were more severe in B₂ (infection+1/5thMTD alpha-cypermethrin) and C₂ (infection+1/10thMTD alpha-cypermethrin) groups in comparision to infection alone (A₂ group).

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Competing Interests

The authors declare that they have no competing interests.

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