

BIOCHEMICAL ALTERATIONS IN BROILER CHICKENS EXPERIMENTALLY INFECTED WITH *SALMONELLA GALLINARUM* FOLLOWING ADMINISTRATION OF TULSI (*OCIMUM SANCTUM*) LEAF POWDER

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ABSTRACT

Experimental *Salmonella Gallinarum* infection in broiler chickens resulted into significant decrease in total serum proteins, albumin and alkaline phosphatase activity. However, alanine transaminase (ALT), aspartate transaminase (AST), and lactate dehydrogenase (LDH) activities increased significantly in infected chickens. Administration of Tulsi (*Ocimum sanctum*) dried leaf powder @ 5g/kg in feed also resulted in only milder reduction in total serum proteins, albumin and alkaline phosphatase activity and increase in the activities of ALT, AST and LDH but these changes were of lesser degree as compared to non-supplemented birds. Tulsi dried leaf powder had protective effects against fowl typhoid.

Key words: *Salmonella Gallinarum*, Tulsi (*Ocimum sanctum*), broiler chickens

Fowl typhoid is acute septicemic disease caused by *Salmonella Gallinarum* and affects all ages of poultry. The disease is of great economic concern due to high morbidity and mortality, low growth rate, low fertility and hatchability and drop in egg production. Control of fowl typhoid is difficult due to endemicity of the disease, both horizontal and vertical transmission, presence of carrier stage, facultative intracellular nature and multiple drug resistance of the organism. These days various herbal plants have been used to manage chicken diseases (Dold and Cocks, 2001). Tulsi (*Ocimum sanctum*), a commonly used medicinal plant in India has been reported to possess various biological activities like anti-bacterial, immuno-modulatory, anti-inflammatory and hepato-protective properties (Gupta *et al.*, 2002). Perusal of literature reveals very limited information in respect of biochemical profile in broiler chickens following feeding of Tulsi (*Ocimum sanctum*) dried leaf powder (DLP). The present paper places on record the effects of Tulsi on biochemical changes during experimental fowl typhoid.

MATERIALS AND METHODS

One hundred sixty, day-old broiler chicks were procured from a commercial hatchery and reared under strict hygienic conditions. These chicks were randomly divided into 2 groups each comprising 80 chicks. Chicks in group I were given normal autoclaved feed while group II chicks were supplemented with dried Tulsi (*Ocimum sanctum*) leaf powder @ 5g/kg in feed. All the chicks were examined bacteriologically for detecting *Salmonella* carrier stage and were found negative. On day seven, the chicks in each group were subdivided into 2 groups i.e. group I into A, B and group II into C, D each having 35, 45, 35 and 45 chicks, respectively. Chicks in group B and D, on day seven were infected with one ID₅₀ dose (8.2x10⁸ bacteria/ml) of 18 h old broth culture of *S. Gallinarum* by s/c route. The chicks in groups A and C were kept as uninfected controls. The blood from three randomly selected chicks from each group was drawn by intracardiac puncture and collected in sterile glass tubes without adding any anti-coagulant on 0, 3, 5, 7, 10, 14, 21, 28 and 35 days post infection (DPI). Subsequently, serum was separated and collected in sterile vials and stored at -20°C. The sera were subjected to the

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estimation of total serum proteins (TSP), albumin, globulin, alanine transaminase (ALT), aspartate transaminase (AST), lactate dehydrogenase (LDH), and alkaline phosphatase (AkPase) by auto chemical analyzer (RA-50) using single step reagent kit procured from M/S Trans Asia Bio Medical Ltd. The globulin concentration was estimated on subtraction the values of albumin from total serum protein.

RESULTS AND DISCUSSION

Total serum protein: The mean TSP in groups A, B, C and D varied from 3.04 ± 0.09 to 3.99 ± 0.10 , 1.84 ± 0.18 to 3.70 ± 0.15 , 3.08 ± 0.06 to 4.11 ± 0.13 and 2.60 ± 0.06 to 4.02 ± 0.15 g/dl, respectively (Fig 1). The TSP concentrations were higher in group C than group A but the values were not significantly ($P < 0.05$) different. It may be due to hepatoprotective property of TLP (Chattopadhyay *et al.* 1992). After the infection, TSP values declined gradually in both

the infected groups and were the lowest (1.84 ± 0.18 and 2.60 ± 0.06 g/dl) on 5 DPI in groups B and D, respectively. Mean TSP values in group D were higher than group B and were significant ($P < 0.05$) on 5 and 7 DPI. The lowering of TSP in *S. Gallinarum* infection was in accordance with Ganovska (1981). It may be due to degenerative and inflammatory changes in liver caused by the *S. Gallinarum* infection. Blood *et al.* (1994) reported that hypoproteinemia may be caused due to liver damage leading to failure of synthesis of plasma proteins or renal injury leading to protein loss through urine. Degenerative and necrotic changes observed in liver and kidneys on gross and histopathological examination justify the decrease of TSP.

Albumin: Albumin concentration varied in sera of chickens from 2.25 ± 0.13 to 2.98 ± 0.18 , 0.90 ± 0.05 to 2.85 ± 0.18 , 2.18 ± 0.16 to 3.03 ± 0.11 and 1.08 ± 0.16 to 2.90 ± 0.17 in groups A, B, C and D, respectively (Fig 2). After *S. Gallinarum* infection, a significant ($P < 0.05$)

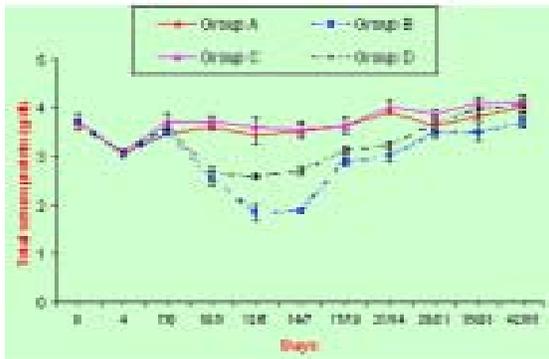


Fig 1. Total serum protein (g/dl) values in broiler chickens of different experimental groups.

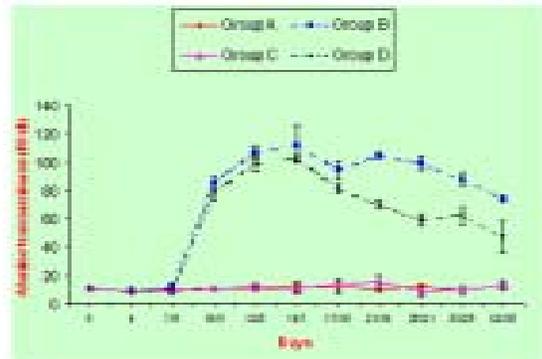


Fig 3. Mean values of alanine transaminase (IU/dl) in different experimental groups at different intervals.

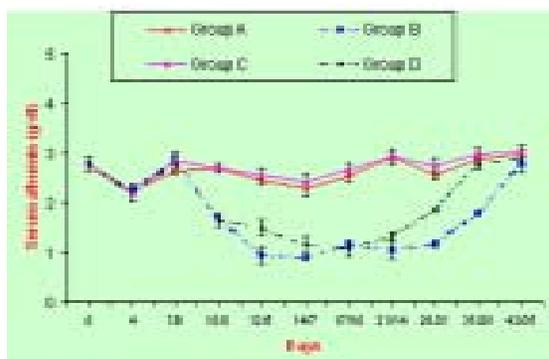


Fig 2. Mean concentration of serum albumin (g/dl) in chickens from different experimental groups at different intervals.

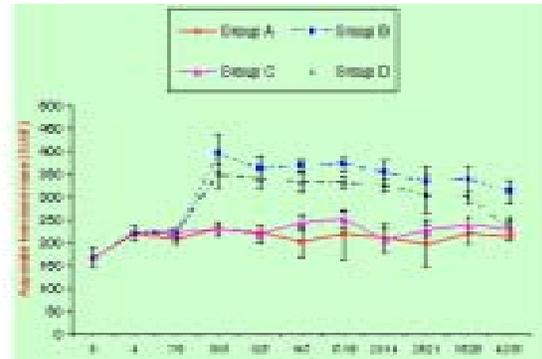


Fig 4. Mean values of aspartate transaminase (IU/dl) in different experimental groups at different intervals.

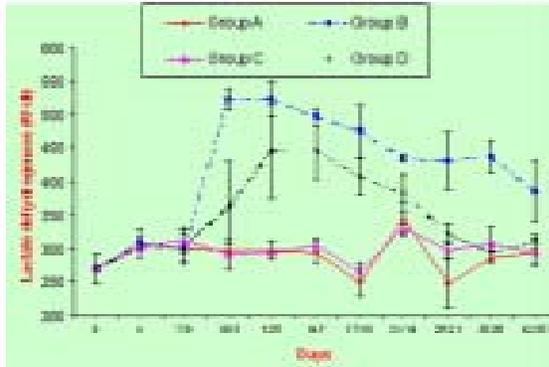


Fig 5. Mean values of lactate dehydrogenase (IU/dl) in different experimental groups at different intervals.

decrease in albumin was observed in both infected groups (B and D). The minimum albumin concentration (0.90 ± 0.05 and 1.08 ± 0.16 g/dl) was observed in groups B and D on 7 and 10 DPI, respectively.

The albumin concentrations were higher in group D as compared to group B but a significant increase was observed only on 28 DPI. The significant decrease in albumin concentration during experimental fowl typhoid may be due to degenerating and necrotic changes in liver and kidneys caused by *S. Gallinarum*.

Globulin: Mean values of globulin in groups A, B, C and D ranged from 0.78 ± 0.5 to 1.07 ± 0.13 , 0.78 ± 0.05 to 2.18 ± 0.06 , 0.86 ± 0.08 to 1.11 ± 0.12 and 0.79 ± 0.08 to 2.09 ± 0.09 g/dl, respectively. A significantly higher globulin concentration was observed in group B from 10 to 21 DPI and from 7 to 21 DPI in group D, when compared with uninfected groups. There appears to be no published report on increased globulin concentration however, Gao *et al.* (1987) reported low albumin globulin ratio in chicks during *S. Pullorum* outbreak.

Serum Alanine transaminase: The mean (ALT) values in sera of broiler chickens of group A ranged from 9.56 ± 1.21 to 12.89 ± 3.08 IU/dl while the corresponding values for groups B, C and D and were 9.56 ± 1.21 to 112.18 ± 13.30 , 8.66 ± 1.38 to 15.27 ± 4.92 and 8.66 ± 1.38 to 102.62 ± 2.30 IU/dl, respectively (Fig 3). In group B chicks, ALT activity showed significant increase from 11.67 ± 2.75 to 85.03 ± 4.98 between 0 and 3 DPI, reaching peak value (112.18 ± 13.30 IU/dl) on 7 DPI and remained so till the end of experiment. In group D,

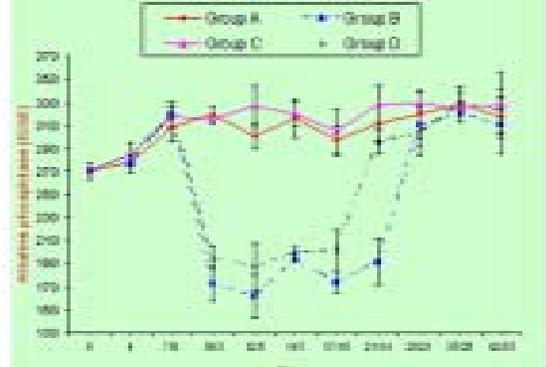


Fig 6. Mean values of alkaline phosphatase (IU/dl) in different experimental groups at different intervals.

significant rise was also observed in ALT values with maximum of 102.62 ± 2.30 IU/dl, on 7 DPI and decreased thereafter reaching 46.99 ± 11.62 IU/dl on 35 DPI. The increase in ALT activity was less in group D as compared to group B.

Aspartate transaminase: The average values of serum (AST) in groups A, B, C and D varied from 166.87 ± 21.36 to 230.37 ± 10.25 IU/dl, 166.87 ± 21.36 to 395.89 ± 39.32 , 166.87 ± 21.36 to 251.62 ± 20.25 and 166.87 ± 21.36 to 347.07 ± 25.88 IU/dl, respectively (Fig 4). A gradual increase in AST values in group B was observed following infection with maximum activity (395.89 ± 39.32 IU/dl) on 3 DPI. Thereafter, it decreased and a value of 311.98 ± 26.47 IU/dl was observed on 35 DPI. AST values were significantly ($P < 0.05$) higher in group B from 3 to 35 DPI as compared to uninfected groups (A and C). In group D also ALT values increased significantly from 3 to 14 DPI, thereafter, it decreased gradually reaching 233.91 ± 12.42 IU/dl on 35 DPI. AST values in group D, remained lower than that of group B at all intervals but decrease was significant ($P < 0.05$) on 35 DPI only. The increased serum ALT and AST activity has also been reported by Kokosharov and Goranov (1997) during experimental *S. Gallinarum* infection. Increased activities of these enzymes can be attributed to degeneration and necrosis in liver, heart and skeletal muscles. The increased activity of lesser degree in Tulsi supplemented chicks suggests lesser degenerative and necrotic changes in these organs, which was evident during histopathological studies (Mamta, 2004). Hepatoprotective and cardioprotective properties of Tulsi (*Ocimum sanctum*) have been reported

by Chattopadhyay *et al.* (1992) and Sharma *et al.* (2001).

Lactate dehydrogenase: The average (LDH) values in sera of chickens of groups A, B, C and D ranged from 247.42±36.62 to 343.66±25.08, 269.11±21.30 to 522.82±26.81, 264.47±13.85 to 328.93±6.23 and 269.11±21.30 to 448.10±45.48 IU/dl, respectively (Fig 5). In group B, a significant increase in LDH concentration was observed from 301.90±26.06 to 522.82±26.81 IU/dl between 0 and 5 DPI thereafter it decreased gradually and reached 384.84±44.92 IU/dl on 35 DPI. In group D, maximum LDH value (448.10±45.48 IU/dl) was observed on 7 DPI, which declined thereafter to normal levels. The overall values in group D remained lower than group B and were significant on 21 and 28 DPI.

Alkaline phosphates: The mean values of AkPase in groups A, B, C and D ranged from 276.08±7.58 to 328.42±15.50, 161.41±18.76 to 320.67±8.11, 270.08±7.58 to 329.68±9.58 and 186.92±20.35 to 328.66±9.87 IU/dl, respectively (Fig 6). Mean AkPase activity in group B decreased significantly ($P<0.05$) from 318.89±11.89 to 161.41±18.76 IU/dl between 0 and 5 DPI. These values remained lower till 14 DPI and then increased gradually and were comparable to control values on 35 DPI. In group D also, a similar decrease in enzymatic activity was noticed. The lowest value 186.92±20.35 IU/dl was recorded on 5 DPI. The values increased gradually and on 35 DPI, it was 322.83±18.96 IU/dl, which were comparable to control group. The decrease in AkPase activity has been reported by Itoh *et al.* (1996) during experimental *S. Typhimurium* and by Kokosharov and Goranov (1997) in *S. Gallinarum* infection. Reduction in severity of biochemical alterations induced by *S. Gallinarum* in group D indicates the protective effects of Tulsi DLP against injury caused by inoculated organisms.

It may be concluded that Tulsi administration

@ 5g/kg in feed resulted into lesser degree of bio-chemical alterations observed during experimental *S. Gallinarum* infection, suggesting protective role of Tulsi DLP against the *S. Gallinarum* infection in broiler chickens.

REFERENCES

- Blood, D.C., Radostits, O.M., Gay, C.C., Arundel, C.H., Ikede, B.O., McKenzie, R.A., Trembley, R.R.M. and Henderson, J.A. (1994). *Veterinary Medicine* (8th edn.), The English Language Book Society and Bailliere Tindall, Eastbourne.
- Chattopadhyay, R.R., Sarkar, S.K., Ganguly, S., Medda, C. and Basu, T.K. (1992). Hepatoprotective activity of *Ocimum sanctum* leaf extract against paracetamol induced hepatic damage in rats. *Indian J. Pharmacol.* **24**: 163-165.
- Dold, A.P. and Cocks, M.L. (2001). Traditional veterinary medicine in the Alice district the Eastern Cape Province, South Africa. *South African J. Sci.* **97**: 375-379.
- Ganovska, M. (1981). Effect of dietary proteins on blood proteins of fowls infected with *S. gallinarum*. *Vet. Med. Nauki.* **18**: 64-72.
- Gao, Q.Y., Cheng, W., An, L.Y., Di, B.Q., She, R.P., Liu, R.P., Ma, Y.X., Huang, W.Q. and Ji, R.Z. (1987). Investigation of pullorosis in growing chicks. *Chinese J. Vet. Sci. Tech.* **4**: 15-18.
- Gupta, S.K., Prakash, J. and Srivastava, S. (2002). Validation of traditional claim of Tulsi, *Ocimum sanctum* Linn. as a medicinal plant. *Indian J. Expt. Biol.* **40**: 765-773.
- Itoh, N., Kikuchi, N. and Hiramune, T. (1996). Biochemical changes in fowl serum during infection with *Salmonella typhimurium*. *J. Vet. Med. Sci.* **58**: 1021-1023.
- Kokosharov, T. and Goranov, T. (1997). Enzyme activities and lipid levels in serum of poultry with experimental acute *Salmonella gallinarum* infection. *Veterinarski Archiv.* **67**: 53-58.
- Mamta (2004) Studies on effects of Tulsi (*Ocimum sanctum*) leaf powder on pathology and pathogenesis of experimental fowl typhoid in broiler chicken. M.V.Sc thesis, CCS Haryana Agricultural University, Hisar, Haryana.
- Sharma, M., Kishore, K., Gupta, S.K., Joshi, S. and Arya, D.S. (2001). Cardioprotective potential of *Ocimum sanctum* in isoproterenol induced myocardial infarction in rats. *Mol. Cell Biochem.* **225**: 75-83.