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Haematological and Biochemical Changes in Chicks Fed with *Aspergillus terreus* Infested Feed

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ABSTRACT

Influence of *Aspergillus terreus* infested feed on the growth and metabolism of chick was investigated. The relative weight gain of different organs and feed intake gradually decreased with the increased amount of intake of infested feed. Serum Glutamate Pyruvate Transaminase (SGPT) and Serum Glutamate Oxaloacetate Transaminase (SGOT) activities and bilirubins increased with increase of intake of *A. terreus* infested feed, indirectly with territrem B intake. However, proteins, iron, urea, calcium and albumins with increased exposure to *A. terreus* infested feed. Different biochemical components of blood also decreased with increased *A. terreus* infested feed intake by the chicks.

Key words: *Aspergillus terreus* infested feed, territrem B, SGPT, SGOT, bilirubins, chicks

INTRODUCTION

Natural contamination of foods and feeds with mycotoxins is widespread and thus the mycotoxins reach human beings through a food chain. The ingested mycotoxins affect the health of man varying from acute to chronic. Fung and Clark (2004) elegantly discussed various aspects of health hazards of man caused by mycotoxins. The inhalation of dust containing mycotoxins can also cause a variety of toxic effects humans (Bunger *et al*., 2004). Mycotoxins also cause severe damage to the vital organs. Mycotoxins such as aflatoxins (hepatotoxic, carcinogenic), zearalenone (estrogenic and teratogenic), ochratoxin A and citrinin (nephrotoxic), DON (feed refusal and vomiting), penicillic acid and citroviridin (cardiotoxic), T2 toxin (mouth lessions and loss of appetite), fumonisins (neurological disorders and liver damage), penitrems and territrems (tremorgenic) and terreic acid (diabetogenic) are reported to cause a wide range of health hazards (Ratcliff, 2002). Some mycotoxins are also reported to cause genetic disorders in animals and plants (Fink-Gremmels and Malekinejad, 2007). Galvano *et al*. (2001) reported that fumonisins cause DNA damage in human fibroblasts. Morgavi and Riley (2007) have excellently reviewed the role of fusarial toxins in animal health.

Similarly, a large number of workers including (Chowdhury *et al*., 2005; Akande *et al*., 2006; Abbes *et al*., 2006; Sharma *et al*., 2008; Che *et al*., 2011; Iheshiulor *et al*., 2011) have investigated biological effects of different mycotoxins in chicks. Reddy *et al*. (1997) have reviewed the incidence and biological effects of tremorgenic mycotoxins on animals. Rafiyuddin *et al*. (2006) have reported the toxicity of tremorgenic mycotoxins in chicks. However, only limited information is

available with regard to toxicity of territrem B, produced by *A. terreus* on humans and animals (Jiang *et al*., 2005). Therefore, in the present investigations an effort was made to investigate the effect of territrem B on growth and development of chicks.

MATERIALS AND METHODS

Six week old chicks with their pre recorded initial average weight were selected and randomly assigned to five groups. They were fed with *Aspergillus terreus* infested feed for 6 weeks. The infested nature of feed was tested for the presence of *A. terreus* and territrem B. At the end of 2, 4 and 6th weeks of incubation, a set of chicks were sacrificed and analyzed for selected parameters. Blood was collected for haematological and biochemical analysis. Standard methods were adopted for analyzing blood serum amylase (Gomori, 1957), serum proteins (Kingsley, 1942), serum bilirubin (Jendrassik and Grafs, 1965), serum calcium (Clark and Collip, 1925), serum iron (Otto, 1958), serum urea (Wybenga *et al*., 1971), SGPT and SGOT (Reitman and Frankel, 1957), blood iron and haemoglobin (Wong, 1928) and blood sugars (Folin and Wu, 1920). The RBC and WBC counts were made with the help of haemocytometer. Biochemical changes such as glycogen (Klicpera *et al*., 1952), tissue proteins (Lowry *et al*., 1951) and total cholesterol in serum and tissue (Zlatkis *et al*., 1953) were also determined and body weight of chicks was also recorded.

Statistical analysis: The experiments were conducted thrice each with three replicates. The results obtained were subjected to statistical analysis using SPSS package (12.0 version) at $p = 0.05$ level of significance. The p-value less than 0.05 ($p<0.05$) were taken as significant.

RESULTS AND DISCUSSION

The results pertaining to feed intake/wastage are presented in Fig. 1 and other results are presented in Table 1, 2 and 3. A critical perusal of Fig. 2 reveals that weight of heart and liver decreased due to intake of *A. terreus* infested feed, which however, showed slow recovery with the progress of age. Weight of the brain increased after 2nd week of feed intake. On the other hand, weight of kidney increased marginally with the age of the chick ($p<0.05$). Increased organ weight observed in this study has also been recorded by other investigators (Chowdhury *et al*., 2005). Reports are also available on increased gizzard and bursa weights in broiler chicks fed with Fusarial mycotoxins like DON and T_2 toxins (Huff *et al*., 1986; Kubena *et al*., 1989). Aflatoxins were also reported to cause increase in liver, spleen and kidney weight of broiler chicks (Huff *et al*., 1986, 1988). Norred and Voss (1994) and Henry *et al*. (2000) have recorded more intense symptoms

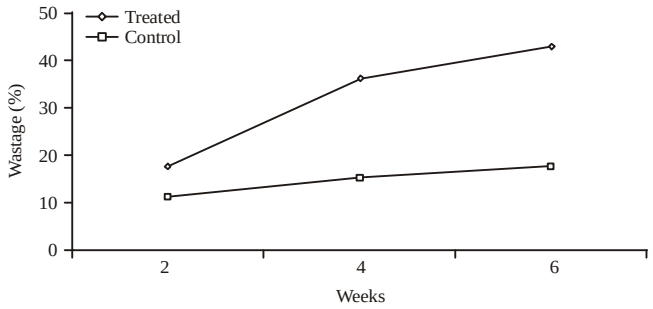


Fig. 1: Wastage and intake of infested feed

Table 1: Effect of *A. terreus* infested feed on components of blood serum of chicks

		Treated (week)		
Components of blood	Control	2nd	4th	6th
Serum amylase (U mL^{-1})	45.60	48.30	52.20	57.50
Serum proteins (mg mL^{-1})	1.15	1.14	1.10	1.06
Serum iron (µg mL^{-1})	10.20	9.34	9.15	8.16
Serum urea (mg %)	44.00	41.30	39.40	36.50
Serum calcium (mg mL^{-1})	25.70	23.50	21.10	20.60
Serum bilirubins (mg %)	0.83	0.89	0.91	0.95
Serum albumins (O.D)	0.13	0.11	0.08	0.06
SGPT (U mL^{-1})	24.40	26.60	27.10	28.90
SGOT (U mL^{-1})	58.50	60.10	62.10	63.30

Analysis of Variance (ANOVA)

Sources of variation	Sum of squares	df	Mean square	F	Significant	Inference
Between groups	16857.117	8	2107.140	372.784	0.000	
Within groups	152.616	27	5.652			S
Total	17009.733	35				

S: Significant at ($p<0.05$)

Table 2: Effect of *A. terreus* infested feed on haematological and biochemical composition of blood

		Treated (week)		
Blood component	Control	2nd	4th	6th
Blood iron (mg/100 mL)	26.40	24.50	23.40	22.40
Blood haemoglobin (g/100 mL)	12.40	12.00	11.50	10.30
Blood sugars (mg mL^{-1})	28.10	25.50	24.10	23.50
RBC/mm^3 (1×10^5)	15.30	15.00	14.50	13.10
WBC/mm^3 (1×10^5)	6.30	6.14	5.60	5.45
Neutrophils (%)	55.00	54.00	46.00	37.00
Lymphocytes (%)	30.00	28.00	25.00	21.00
Monocytes (%)	04.00	02.00	02.00	02.00
Basophils (%)	00.00	00.00	00.00	01.00

Analysis of Variance (ANOVA)

Sources of variation	Sum of squares	df	Mean square	F	Significant	Inference
Between groups	7240.668	8	905.083	85.158	0.000	
Within groups	286.962	27	10.628			S
Total	7527.630	35				

S: Significant at ($p<0.05$)

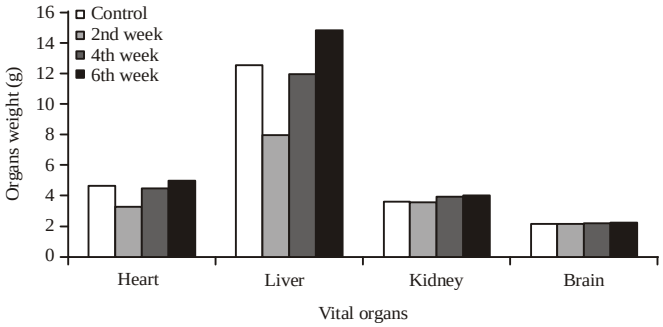
Table 3: Effect of *A. terreus* infested feed on biochemical composition of different organs of chicks

			Treated (week)		
Constituents (mg mL^{-1})	Organs	Control	2nd	4th	6th
Glycogen	Liver	0.21	0.18	0.18	0.15
	Kidney	0.32	0.31	0.29	0.27
	Brain	0.25	0.24	0.22	0.21
	Heart	0.19	0.17	0.16	0.14
Proteins	Liver	285.10	280.10	275.00	263.60
	Kidney	376.30	361.10	358.50	333.40
	Brain	278.10	250.40	200.00	185.50
	Heart	320.00	300.40	285.30	246.60
Cholesterol	Liver	0.22	0.24	0.29	0.33

Analysis of Variance (ANOVA)

Sources of variation	Sum of squares	df	Mean square	F	Significant	Inference
Between groups	0.043	3	0.014	30.758	0.000	
Within groups	0.006	12	0.000			S
Total	0.048	15				
Between groups	33933.095	3	11311.032	13.968	0.000	
Within groups	9717.145	12	809.762			S
Total	43650.240	15				
Between groups	0.219	3	0.073	118.216	0.000	
Within groups	0.007	12	0.001			S
Total	0.226	15				

S: Significant at ($p<0.05$)



Analysis of Variance (ANOVA)

Sources of variation	Sum of squares	df	Mean square	F	Sig.	Inference
Between groups	2275437.724	4	568859.431	23.197	0.000	
Within groups	367844.370	15	24522.958			S
Total	2643282.095	19				

S: Significant at ($p<0.05$)

Fig. 2: Influence of *A. terreus* infested feed on weight of different vital organs of chicks

such as diarrhea, decreased feed consumption, decreased body weight and increased relative weights of liver in broilers. Girish *et al*. (2008) also reported the decreased weight of turkey poultry fed with fusarial mycotoxins.

Changes in the components of blood serum are presented in Table 1. Significant changes in the composition of blood serum were recorded due to intake of *A. terreus* infested feed. However, the degree of changes varied with the amount of feed intake. An increase in the amylase activity was recorded with the progress of growth of chick and feed intake. On the other hand, serum proteins decreased under the influence of *A. terreus* infested feed. Oguz *et al*. (2002), Allameh *et al*. (2005) and Safameher (2008) have also reported decreased serum protein in chicks fed with aflatoxin contaminated corn. A positive correlation could be observed with the intake of *A. terreus* infested feed and protein content of serum. Iron, urea, calcium and albumin levels decreased with the increase of intake of infested feed. A significant increase in serum bilirubins, SGPT and SGOT was recorded with the increased feed intake ($p<0.05$).

The effect of *A. terreus* infested feed on biochemical and haematology composition of blood was assessed and the results are presented in Table 2. The results clearly reveal a significant decrease in blood iron, haemoglobin and sugar. The decrease was progressive with the increase of feed intake. Reddy *et al*. (1997) have also recorded decreased haemoglobin, albumins, proteins and sugar of the blood of chicks under the influence of gliotoxin. The RBC, WBC, neutrophil and lymphocytes decreased with the increased exposure of *A. terreus* infested feed ($p<0.05$). Monocytes were affected only marginally. However, basophils number increased by the end of 6 weeks observation. The present observations are in agreement with those of Singh *et al*. (1992), who also recorded decrease in WBC counts of chicks receiving aflatoxin B_1. Verma *et al*. (1991) reported significant changes in the blood of rat under the influence of aflatoxins. In 1995, a number of histopathological changes in chick receiving trichothecene mycotoxins were reported. Reddy *et al*. (1997) have also recorded an increase in Serum Glutamate Pyruvate Transaminase (SGPT) and Serum Glutamate Oxaloacetate Transaminase (SGOT) of blood of chick under the influence of gliotoxin produced by *Trichoderma viride*.

Biochemical changes in different vital organs of chick under the influence of *A. terreus* infested feed were analyzed and the results are presented in Table 3. The glycogen content of liver decreased with the increased concentration and exposure time of territrem B, glycogen of kidney, brain and heart decreased marginally under the influence of territrem B. Protein content of liver, kidney brain and heart also decreased with the increased intake of *A. terreus* infested feed ($p<0.05$). On the other hand, cholesterol content in liver increased with the intake of *A. terreus* infested feed as recorded by Jassar and Singh (1993) have also recorded significant biochemical changes in the blood of broiler chicks treated with aflatoxins.

CONCLUSION

Feeding of chicks with *Aspergillus terreus* infested feed has significant influence on the vital organs in particular and overall growth of chick in general. Feed efficiency decreased with the age of chicks. Significant changes in the composition of blood serum were noticed. Serum bilirubins, SGPT and SGOT increased with the progress of growth of chick till the end of observation period. Similarly iron, haemoglobin and sugar contents in the blood decreased. Similarly, RBC, WBC, neutrophils and lymphocyte count also decreased with the increased intake of infested feed. However, basophils number increased. Significant decrease in glycogen, protein contents of the liver, kidney and heart decreased. On the other hand, cholesterol contents of liver increased. These observations confirms the toxicity of territrem B produced by *A. terreus* in the infested feed.

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REFERENCES

Abbes, S., Z. Ouanes, J.B. Salah-Abbes, Z. Houas, R. Oueslati, H. Bacha and O. Othman, 2006. The protective effect of hydrated sodium calcium aluminosilicate against haematological, biochemical and pathological changes induced by zearalenone in mice. Toxicon, 47: 567-574.

Akande, K.E., M.M. Abubakar, T.A. Adegbola and S.E. Bogoro, 2006. Nutritional and health implications of mycotoxins in animal feeds: A review. Pak. J. Nutr., 5: 398-403.

Allameh, A., A.R. Safamehr, S.A. Mirhadi, M. Shivazad and M. Razzaghi-Abyaneh, 2005. Evaluation of biochemical and production parameters of broiler chicks fed ammonia treated aflatoxin contaminated maize grains. Anim. Feed. Sci. Technol., 122: 289-301.

Bunger, J., G. Westphal, A. Monnich, B. Hinnendahl, E. Hallier and M. Muller, 2004. Cytotoxicity of occupationally and environmentally relevant mycotoxins. Toxicology, 202: 199-211.

Che, Z., Y. Liu, H. Wang, H. Zhu, Y. Hou and B. Ding, 2011. The protective effects of different mycotoxin adsorbents against blood and liver pathological changes induced by mold-contaminated feed in broilers. Asian Aust. J. Anim. Sci., 24: 250-257.

Chowdhury, S.R., T.K. Smith, H.J. Boermans and B. Woodward, 2005. Effects of feed-borne *Fusarium* mycotoxins on hematology and immunology of laying hens. Poult. Sci., 84: 1841-1850.

Clark, E.P. and J.B. Collip, 1925. A study of the Tisdall method for the determination of blood serum calcium with a suggested modification. J. Biol. Chem., 63: 461-464.

Fink-Gremmels, J. and H. Malekinejad, 2007. Clinical effects and biochemical mechanisms associated with exposure to the mycoestrogen zearalenone. Anim. Feed Sci. Technol., 137: 326-341.

Folin, O. and H. Wu, 1920. Determination of blood sugar by Folin and Wu method. J. Biol. Chem., 41: 367-374.

Fung, F. and F.R. Clark, 2004. Health effects of mycotoxins: A toxicological overview. J. Toxicol. Clin. Toxicol., 42: 217-234.

Galvano, F., A. Piva, A. Ritieni and G. Galvano, 2001. Dietary strategies to counteract the effects of mycotoxins: A review. J. Food Prot., 64: 120-131.

Girish, C.K., T.K. Smith, H.J. Boermans and N.A. Karrow, 2008. Effects of feeding blends of grains naturally contaminated with *Fusarium* mycotoxins on performance, hematology, metabolism and immunocompetence of turkeys. Poult. Sci., 87: 421-432.

Gomori, G., 1957. Assay of serum amylase with small amounts of serum. Am. J. Clin. Pathol., 2: 714-716.

Henry, M.H., R.D. Wyatt and O.J. Fletchert, 2000. The toxicity of purified fumonisin B_1 in broiler chicks. Poult. Sci., 79: 1378-1384.

Huff, W.E., L.F. Kubena, R.B. Harvey, D.E. Corrier and H.H. Mollenhauer, 1986. Progression of aflatoxicosis in broiler chickens. Poult. Sci., 65: 1891-1899.

Huff, W.E., R.B. Harvey, L.F. Kubena and G.E. Rottinghaus, 1988. Toxic synergism between aflatoxin and T-2 toxin in broiler chickens. Poult. Sci., 67: 1418-1423.

Iheshiulor, O.O.M., B.O. Esonu, O.K. Chuwuka, A.A. Omede, I.C. Okoli and I.P. Ogbuewu, 2011. Effects of mycotoxins in animal nutrition: A review. Asian J. Anim. Sci., 5: 19-33.

Jassar, B.S. and B. Singh, 1993. Biochemical changes in experimental aflatoxicosis in broiler chicken. Indian J. Anim. Sci., 63: 847-848.

Jendrassik, L. and P. Grafs, 1965. Recommendation of Uniform Bilirubin Standard: Methods of Clinical Chemistry. Vol. V, Academic Press, New York, USA., Pages: 55.

Jiang, X., L. Ao, C. Zhou, L. Yang and Q. Zhang *et al*., 2005. Design, synthesis and biological evaluation of new territrem B analogues. Chem. Biodivers., 2: 557-567.

Kingsley, G.R., 1942. The direct biuret method for the determination of serum proteins as applied to photoelectric and visual colorimetry. J. Lab. Clin. Med., 27: 840-845.

Klicpera, H., Z. Drahota and R. Zak, 1952. Notes on the Determination of Muscle Glycogcn. Vol. 3, Academic Press, New York, USA., Pages: 121.

Kubena, L.F., W.E. Huff, R.B. Harvey, T.D. Phillips and G.E. Rottinghaus, 1989. Individual and combined toxicity of deoxynivalenol and T-2 toxin in broiler chicks. Poult. Sci., 68: 622-626.

Lowry, O.H., N.J. Rosebrough, A.L. Farr and R.J. Randall, 1951. Protein measurement with the Folin phenol reagent. J. Biol. Chem., 193: 265-275.

Morgavi, D.P. and R.T. Riley, 2007. An historical overview of field disease outbreaks known or suspected to be caused by consumption of feeds contaminated with *Fusarium* toxins. Anim. Feed Sci. Technol., 137: 201-212.

Norred, W.P. and K.A. Voss, 1994. Toxicity and role of fumonisins in animal diseases and human esophageal cancer. J. Food Protect., 57: 522-527.

Oguz, H., F. Kurtoglu, V. Kurtoglu and Y.O. Birdane, 2002. Evaluation of biochemical characters of broiler chickens during dietary aflatoxin (50 and 100 ppb) and clinoptilolite exposure. Res. Vet. Sci., 73: 101-103.

Otto, S., 1958. American Association of Clinical Chemistry. Vol. II, Academic Press, New York, USA., pp: 69-78.

Rafiyuddin, M., N.J. Rao, S. Girisham and S.M. Reddy, 2006. Toxicology of tremorgenic mycotoxins on chicks. Natl. Acad. Sci. Lett., 29: 311-315.

Ratcliff, J., 2002. The Role of Mycotoxins in Food and Feed Safety. Animal Feed Manufacturers Association, South Africa.

Reddy, S.M., D.R. Kumari and V.K. Reddy, 1997. Toxicological studies on gliotoxin produced by *Trichoderma viride*. Indian J. Poult. Sci., 32: 169-172.

Reitman, S. and S. Frankel, 1957. A coloric method for determination of SGOT and SGPT. Am. J. Clin. Pathol., 28: 56-63.

Safameher, A., 2008. Effects of clinoptilolite on performance, biochemical parameters and hepatic lesions in broiler chickens during aflatoxosis. J. Anim. Vet. Adv., 7: 381-388.

Sharma, D., R.K. Asrani, D.R. Ledoux, N. Jindal, G.E. Rottinghaus and V.K. Gupta, 2008. Individual and combined effects of fumonisin B_1 and moniliformin on clinicopathological and cell-mediated immune response in Japanese quail. Poult. Sci., 87: 1039-1051.

Singh, A., K.C. Satija and S.K. Mahipal, 1992. Haematological and biochemical studies on broiler chicks fed aflatoxin B_1 and after its withdrawal. Indian J. Poult. Sci., 27: 153-156.

Verma, R.J., P.J. Raval and H.C. Dube, 1991. Effect of aflatoxin on liver and blood cells of rats. Indian J. Microbiol., 31: 87-89.

Wong, S.Y., 1928. Colorimetric determination of iron and hemoglobin in blood. J. Biol. Chem., 77: 409-412.

Wybenga, D.R., J. Di Giorgio and V.J. Pileggi, 1971. Manual and automated methods for urea nitrogen measurement in whole serum. Clin. Chem., 17: 891-895.

Zlatkis, A., B. Zak and A.J. Boyle, 1953. A new method for the direct determination of serum cholesterol. J. Lab. Clin. Med., 41: 486-492.