Bull. Environ. Contam. Toxicol. (1992) 48:549–553

Environmental Contamination and Toxicology

Hematological and Biochemical Changes in Rats Administered with *Aspergillus niger* Infested Diet

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Aspergilli and Penicillia are the common inhabitants of air, water and soil. In 1971, a total of 392 Aspergillus strains were evaluated for toxicity tests and toxicity was suspected in A. niger strains with other species of Aspergillus (Semeniuk et al. 1971). A. niger, resulting in black mold storage disease, produces cyclic pentapeptides called malformins (Takeuchi et al. 1967). Besides malformin A and B, malformin C was isolated from mold damaged grains (Anderegg et al. 1976 and Kobbe et al. 1977) which are antibiotic to bacteria (Suda and Curtis 1966). Natural occurrence of malformin was first reported in A. niger infected onion bulbs (Curtis et al. 1974). The fungus was also associated with 9 Yr old buffalo which aborted to 7 mon of gestation (Pal 1988). The present communication delineates the toxicity of A. niger to albino rats concerning hematological and biochemical alterations.

MATERIALS AND METHODS

Feeding trial experiments were performed with 100 d old female albino rats procured from the animal house of Defence Research and Development Establishment, Gwalior. Rats, randomized into five groups of six each, were housed in singlet inside galvanized iron cages and received their diet ad libitum.

Diet was prepared by placing 200 g of pearlmillet grains in 500 ml Erlenmeyer flask and autoclaved at 120°C for 40 min by adding distilled water to it. The substrate was allowed to cool down, inoculated with selected fungal species and the inoculated flasks were incubated at 26±2°C for 12 d. At the end of incubation period the stuf was allowed to dry in an oven at 40±2°C for three consecutive days. Finally the diet was prepared by mixing the infested diet with N diet in a 1:1 ratio. Autoclaved grains free from storage fungi were served as control diet. Experimental and control diets were further added with vitamins, minerals, vegetable oil and casein to make the diet perfect.

Animals were weighed twice, before and after the duration of the experiment. At the end of each duration (i.e. 8, 18, 28, 38 and

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48 d) blood was collected in heparinized tubes and serum was prepared by centrifuging the blood.

Estimations of glucose (Folin and Wu 1920) and hemoglobin were made in blood: protein, cholesterol and urea were estimated in serum by Biuret, Libermann-Burchard and Urease methods respectively (Henry et al. 1974, Henry 1968, Fawcett and Scott 1920).

RESULTS AND DISCUSSION

The present studies, performed at the different intervals, revealed rapid fall in hemoglobin and RBC in 8 d fed rats but subsequently the fall became gradual (statistically significant at 0.01 and 0.05 level) in rats fed upto 48 d (Table 1).

Table 1. Hematological alteration in rat

Experimental duration in d	RBC Count		WBC Count		Hemoglobin (g/100ml)	
	I	C	I	C	I	C
8	2.84 ±0.418	5.54 ±0.330	6783 ±1489	13850 ±410	9.6 ±0.879	14.23 ±0.027
18	4.09 ±0.307	5.52 ±0.330	15766 ±896	11268 ±410	16.8 ±0.472	14.45 ±0.027
28	5.78 ±0.306	5.50 ±0.330	12700 ±1065	10400 ±410	14.5 ±0.774	14.45 ±0.027
38	6.40 ±0.340	5.50 ±0.330	35033 ±4655	10401 ±410	11.0 ±0.679	14.45 ±0.027
48	4.90 ±0.117	5.51 ±0.330	36266 ±4888	10401 ±410	8.0 ±1.229	14.42 ±0.027

I Animals fed upon infested diet.
C Animals fed upon control diet.
± Standard error.

The percentage of blood glucose level was higher in rats receiving control diet (>75 mg/100ml) while a gradual decrease with statistical significance at 0.05 and 0.01 levels was observed in the rats receiving infested diet upto 48 d (Table 2).

Table 2. Blood glucose level in rat (mg/100ml)

Experimental duration in d	Experimental	Control
8	69.40 ± 5.172	75.00 ± 3.920
18	49.60 ± 6.792	75.00 ± 3.920
28	50.00 ± 6.584	75.00 ± 3.920
38	69.30 ± 5.821	74.00 ± 3.920
48	27.76 ± 9.932	75.00 ± 3.90

Reduction in RBC count and hemoglobin resulted in anemic condition of rats while gradual fall in blood glucose of animals rendered them to be inactive. Deficiency in the erythrocyte level might be due to malfunctioning of hematopoietic tissues.

Total protein level in serum varied considerably (Table 3) indicating loss with statistical significance at 0.05 level.

Table 3. Level of total protein in rat serum (g/100ml)

Experimental duration in d	Experimental	Control
8	7.00 ± 0.688	8.20 ± 0.49
18	7.06 ± 0.635	8.20 ± 0.49
28	6.60 ± 0.972	8.30 ± 0.49
38	6.60 ± 1.013	8.40 ± 0.49
48	6.46 ± 0.951	8.20 ± 0.49

Malformed kidney and elevated levels of urea in serum suggest the altered function of kidney. The urea level increased with the increase of experimental duration (Table 4).

Table 4. Urea level in rat serum (g/100ml)

Experimental duration in d	Experimental	Control
8	10.3 ± 1.030	5.0 ± 0.119
18	11.3 ± 1.215	5.0 ± 0.119
28	20.0 ± 2.919	4.9 ± 0.119
38	26.0 ± 3.894	5.5 ± 0.119
48	30.0 ± 4.633	5.5 ± 0.119

Ingestion of infested diet initially increased the cholesterol level in 8 d fed rats but later on the level declined in comparison to the controlled set of rats (Table 5).

Table 5. Level of cholesterol in rat serum (mg/100ml)

Experimental duration in d	Experimental	Control
8	180.0 ± 6.050	150 ± 3.242
18	156.6 ± 3.003	150 ± 3.242
28	151.6 ± 2.076	150 ± 3.242
38	145.6 ± 2.198	150 ± 3.242
48	139.6 ± 2.382	151 ± 3.242

Loss in RBC and hemoglobin percentage is designated as an impairment of hematopoietic system while increase in WBC count is suggestive of infection or disease procurement. Toxicologically trichothecenes possess dermal toxicity, hemorrhagic lesions, depression of immune responses and destruction of hematopoietic organs (Ueno 1983). The role of trichothecenes in hematological disorders was investigated in mice, rats and cats (Sato et al. 1978). Fall in RBC count and hemoglobin, and elevation of WBC count was observed in rats provided with oral citrinin and toxic diet (Bilgrami et al. 1981). Significant decrease in total protein level of different durations might be due to catabolism in protein metabolism and malfunctioning of liver (Harper et al. 1977). Concentration of serum protein was reported reduced significantly during aflatoxicosis (Tung et al. 1975) and mycotoxicosis induced by T-2 toxin (Kravchenko et al. 1986). The variations in cholesterol level implicated stimulation and inhibition of physical changes. Similarly blocked incorporation of acetate into hepatic cholesterol in rats was found due to aflatoxin B1 (Kato et al. 1969). The increased urea level in the present investigation suggests an altered membrane permeability due to mold toxicosis in the same way as the increase in the serum glycoproteins and electrolytes are due to the penicillic acid toxicosis (Pandiyan et al. 1987).

Acknowledgments. The authors thank the Head, School of Studies in Botany, Jiwaji University, Gwalior and Dr. P.K. Ramachandran, Director, Defence Research and Development Establishment, Gwalior for providing research facilities.

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Received April 15, 1991; accepted December 10, 1991.