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PROTECTIVE EFFECTS OF PIPERINE ON CYPERMETHRIN-INDUCED HAEMATOLOGICAL TOXICITY IN RATS

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Abstract: Cypermethrin is a synthetic pyrethroid insecticide. It is known for its wide toxic manifestations. The present experiment pertains to the protective role of piperine against haematological toxicity induced by cypermethrin during 28 days exposure. The rats were divided into five groups of six each; the first group served as control and second group was used as vehicle control. While, groups III, IV and V were orally treated with piperine (50 mg/kg body weight), cypermethrin (25 mg/kg body weight) and cypermethrin plus piperine, respectively for 28 days. Exposure of rats to cypermethrin caused significant changes of some haematological parameters like red blood cells, haemoglobin, haematocrit and white blood cells in treated rats compared to controls. Co-administration of piperine with cypermethrin restored some of the parameters cited above to near-normal values. Therefore, our investigation revealed that piperine given partial protection against cypermethrin-induced toxicity.

Keywords: Cypermethrin, Piperine, Haemaological toxicity, Rats.

Introduction

Cypermethrin (CYP) is a member of the family of synthetic pyrethroids, belongs to type II class pyrethroids and is widely used in agricultural and other domestic applications. Accidental exposure with pyrethroids in human and animals result from advertent use. In mammals, CYP can accumulate in body fat, skin, liver, kidneys, adrenal glands, ovaries, lung, blood, and heart. Several studies showed that CYP had immunotoxicity effect through alteration in the haematological parameters in human and animals. Institoris et al. (1999) reported that oral treatment with 55.4 mg/kg b.wt for 28 days of CYP decreased DTH reaction in rats. This dose of the insecticide also decreased the mean cell volume of erythrocytes and white blood cell count in the peripheral blood. Recently, we have reported that CYP given orally at a dose rate of 25mg/kg b.wt over a period of 4 weeks caused significant depression in leukocyte count, lymphocyte count, serum total protein, serum

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albumin, serum globulin, antibody titer against sheep red blood cells, and cell-mediated immunity in rats (Sankar et al., 2010).

Researchers are looking forward in search of protective agent in order to combat against CYP-induced haematological toxicity. Piperine, a main component of *Piper longum* L. and *Piper nigrum* L., is a plant alkaloid, which is known to exhibit a variety of pharmacological activities like antipyretic, antiinflammatory, immunoprotective, hepatoprotective and bioenhancer (Selvendiran et al., 2004). Therefore, the present study has been undertaken to evaluate the protective effect of piperine on cypermethrin-induced haemato-toxicity in rats.

Materials and Methods

Cypermethrin (CYP; 96%) was a kind gift from Gharda Chemicals, Mumbai. Piperine was purchased from M/s Sigma Chemicals, USA. All other chemicals were of analytical and molecular grade from different companies. Animals were maintained under standard management conditions and handled as per the Institute Animal Ethics Guidelines. Rats were given standard rat feed and water ad libitum throughout the experiment. All the animals were quarantined for a period of at least 7 days before beginning of the experiment. Rats were divided into five groups containing six animals each. Group I (control), was given normal saline, while Groups II was given once equivalent amount of ground nut oil (1%: Vehicle control). Group III was administered cypermethrin (25 mg/kg, orally) daily for 28 days. Group IV was administered piperine (50 mg/kg, orally) daily for 28 days. Group V was administered piperine (50 mg/kg, orally) and then cypermethrin (25 mg/kg, orally) daily for 28 days. Rats were sacrificed at the end of the exposure period. Blood was collected from heart in a dry, clean and sterilized test tube for different haematological parameters.

Total leukocytes, lymphocytes, monocytes, and granulocytes counts were done as per the method described by Jain (1986). Total protein and albumin levels (g/dL) in serum were estimated by Biuret method using Span diagnostic kit (Surat, India). Serum globulin (g/dL) was determined by subtracting albumin level from serum total protein level.

Results and Discussion

The mean values of various haematological parameters have been presented in Table.1 and 2. The haemoglobin level showed significant decrease in cypermethrin treated group. Reduction in haemoglobin content can be related to the decreased size of red blood cells or to the impaired biosynthesis of heme in bone marrow (Shakoori et al., 1990). Cypermethrin plus piperine group did not show significant amelioration as compared to cypermethrin treated group. The PCV value showed a significant decrease in cypermethrin treated group as

compared to control. Cypermethrin plus piperine group did not show significant amelioration as compared to cypermethrin treated group. Lymphocytopenia was observed in group treated with cypermethrin. The reduction in the blood parameters (PCV, RBC and Hb) may be attributed to a hyperactivity of bone marrow (Tung et al., 1975) leading to production of red blood cells with impaired integrity which were easily destroyed in the circulation. PCV is obviously decreased by the decreased cellular count in blood after pesticide treatment. Similar effects have also been found in sheep and rabbits (Yousef et al., 1999). Group exposed to cypermethrin alone exhibited significant reduction in serum protein, albumin and globulin concentration compared to control group (Table.3). These results are in agreement with those of Yousef et al. (1999) in rabbits and Baligar and Kaliwal (2001) in rats. Cypermethrin plus piperine and cypermehrin plus curcumin treated groups showed significant increase in serum protein and albumin as compared to cypermethrin. Pathak and Khandelwal (2007) demonstrated the cytoprotective and immunomodulatory activity of piperine on murine spleniocytes.

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Table 1: Effect on hematological parameters following 28-day exposure to cypermethrin, and cypermethrin plus piperine in rats

Groups	Hb(g/dl)	PCV (%)	RBC ($x10^6$/µl)	MCV (fl)	MCH (pg)	MCHC (g/dl)
Control	15.67±0.17	36.50±0.43	7.27±0.03	50.18±0.51	21.89±0.36	42.92±0.59
Ground nut oil	14.67±0.17	34.00±0.26	7.07±0.03	48.18±0.51	20.73±0.31	43.12±0.60
Piperine	13.50±0.18	32.67±0.49	7.02±0.06	45.87±1.12	19.20±0.30	40.51±1.50
Cypermethrin	14.00±0.29a	31.50±0.34a	6.85±0.02a	46.85±1.07	20.39±0.39	38.26±5.03
Cypermethrin +Piperine	14.08±0.15	33.00±0.52	6.78±0.08	48.66±0.93	20.73±0.33	42.69±0.84

Values are mean ± SE. Significant differences are indicated by a compare to control ($p<0.001$).

Table 2: Effect on TLC and DLC following 28-day exposure to cypermethrin and cypermethrin plus piperine in rats

Groups	TLC 10^3/µl	Lymphocyte (%)	Neutrophil (%)	Monocyte (%)	Eosinophil (%)
Control	9.34±0.14	78.17±0.65	16.83±1.14	4.67±0.80	0.50±0.34
Ground nut oil	8.77±0.03	77.17±0.48	18.50±0.85	4.17±0.54	0.50±0.22
Piperine	8.28±0.03	76.67±0.80	17.83±1.01	4.33±0.80	0.33±0.21
Cypermethrin	7.87±0.08a	69.83±0.60a	24.67±1.50a	3.83±0.60	0.50±0.34
Cypermethrin+ Piperine	8.27±0.07b	74.67±0.42c	19.17±0.83d	4.83±0.70	0.50±0.34

Values are mean ± SE. Significant differences are indicated by a compare to control ($p<0.001$), b compare to cypermethrin ($p<0.05$), c compare to cypermethrin ($p<0.001$), d compare to cypermethrin ($p<0.05$).

Table 3: Effect on biochemical parameters following 28-day exposure to cypermethrin and cypermethrin plus piperine in rats

Groups	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	A:G ratio
Control	6.08±0.06	3.78±0.05	2.30±0.04	1.600.04
Ground nut oil	6.05±0.03	3.85±0.04	2.20±0.04	1.72±0.04
Piperine	6.15±0.04	3.87±0.08	2.28±0.07	1.65±0.07
Cypermethrin	5.33±0.04	3.38±0.03a	1.95±0.04a	1.65±0.04
Cypermethrin+ Piperine	6.08±0.04b	3.95±0.02b	2.10±0.03	1.850.03

Values are mean ± SE. Significant differences are indicated by a compare to control ($p<0.001$), b compare to cypermethrin ($p<0.001$).