TOXIC EFFECTS OF DELTAMETHRIN AND FLUORIDE ON ANTIOXIDANT PARAMETERS IN RATS

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SUMMARY: Toxic effects of the pyrethroid pesticide deltamethrin and water-borne fluoride, alone and in combination, on free radical mediated parameters are reported in rats. Twenty-four healthy adult Wistar rats of both sexes were divided into 4 groups with 6 rats in each group. Group I receiving no treatment served as the control. Group II and group III were orally administered deltamethrin (1/100 of LD_{50}) and 20-ppm fluoride in their drinking water, respectively, for 28 days. An additional group IV was co-administered deltamethrin and fluoride at the same dosages as groups II and III. Enhanced oxidative stress was observed as shown by significantly increased lipid peroxidation and alterations in antioxidant parameters, especially in the fluoride-deltamethrin co-exposed group IV.

Keywords**:** Antioxidant parameters; Deltamethrin in rats; Fluoride intoxication; Oxidative stress.

INTRODUCTION

Deltamethrin, (S)-α-cyano-3-phenoxybenzyl-(1R,cis)-2,2-dimethyl-3-(2,2-dibromovinyl)-cyclopropanecarboxylate (see Figure), a type II pyrethroid, is a highly neurotoxic pesticide for vertebrates as well as insects.[1] Pyrethroids are reported to generate free radicals through hydrolytic ester cleavage and oxidative pathways by the CYP-450 enzymes.[2-4] Induction of oxidative stress has been reported with pyrethroids such as cypermethrin, deltamethrin, and fenvalerate.[5-7]

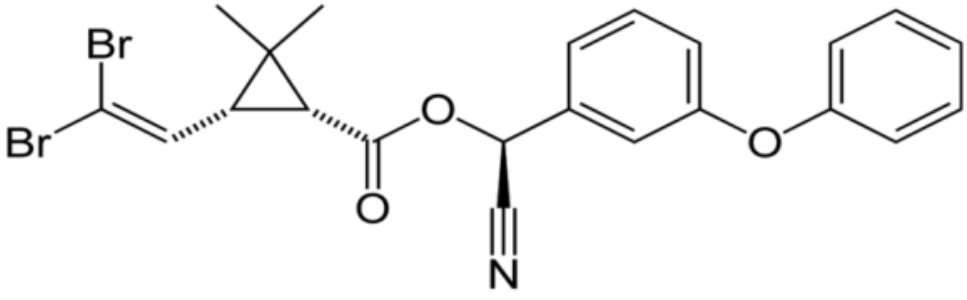

Figure. Structure of deltamethrin.

In many parts of the world, toxic effects of fluoride (F) causing dental and skeletal fluorosis are a major environmental problem.[8] In laboratory studies, excessive ingestion of F has been found to induce free radical injury and oxidative damage in various tissues of mice.[9-13]

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It is now well recognized that humans and animals are exposed concurrently to more than one chemical in the environment from various sources. However, relatively few studies have reported assessment of the degree of hazard posed by simultaneous exposure to toxic chemicals, especially, at lower doses.[14] The present research was aimed to investigate the interactive effect of deltamethrin and F on the antioxidant status in rats following sub-acute oral exposure.

MATERIALS AND METHODS

Adult Wistar rats of either sex weighing 150–200 g were purchased from the Indian Institute of Integrative Medicine (Council of Scientific & Industrial Research) Laboratory, Jammu, and maintained under standard experimental conditions with *ad libitum* feed and water. The experimental design with the animals was approved by the University Animal Ethical Committee. The rats were randomly allocated to four groups of six rats each. Group I was untreated and served as control. Group II received deltamethrin (Butox) by gavage @ 1.28 mg/kg bw/day (1/100 of LD_{50}) between 9:30 and 10:30 AM. Group III was administered F, as NaF, in drinking water @ 44.5 mg/L/day, providing 20 ppm F ion. Group IV received both deltamethrin and F at the same dosages as groups II and III, respectively. The duration of exposure of all the toxicants was 28 days. In order to minimize the possible instability, both toxicants were prepared freshly in water. All the rats were weighed weekly to make necessary corrections in the deltamethrin dosage as per body weight.

After 28 days of daily treatment with the above toxicants, the rats were anaesthetised with diethyl ether. Blood samples were collected from retro-orbital fossa using capillary tubes in aliquots containing heparin @10 IU/mL of blood. Prior to centrifugation, 200 µL whole blood was used for the estimation of blood glutathione (GSH).[15] Then 1 per cent of the haemolysate was used for the estimation of superoxide dismutase (SOD), catalase (CAT), glutathione S-transferase (GST), and glutathione peroxidase (GPx).[16-19] About a third of the haemolysate was used for estimation of lipid peroxidation (LPO), i.e., by malondialdehyde (MDA) levels.[20]

Statistical analysis: The results were subjected to analysis of variance (ANOVA) in completely randomized design (CRD) with statistical significance being tested using the Duncan Multiple Range Test.[21]

RESULTS

Changes in MDA content: As seen in the Table, a significant ($p<0.05$) increase in the level of MDA was observed in the red blood cells (RBCs, erythrocytes) of all three toxicant-exposed groups with higher lipid peroxidation in the combined group IV differing significantly ($p<0.05$) from all the other groups.

Changes in SOD and CAT activity: Though a significant ($p<0.05$) decrease in SOD activity was observed in all the toxicant-exposed groups, the results were at par with each other. In the present study, a significant ($p<0.05$) decrease of CAT activity was also observed in all toxicant-exposed groups, with each group differing significantly ($p<0.05$) from one another (Table).

Changes in GSH level, GPX and GST activity: A significant ($p<0.05$) decrease in the levels of blood glutathione was observed in the rats co-exposed to deltamethrin and F, whereas no significant change was observed in GSH levels of rats treated with these toxicants alone. Compared to the control group, the activity of GST was significantly ($p<0.05$) increased in all exposed groups. However, GST of the deltamethrin group was at par with both the F and the combined group, with the latter differing significantly ($p<0.05$) from one another. Moreover, a significant ($p<0.05$) decrease of GPx activity was found in the erythrocytes (RBCs) of all exposed groups (Table).

Table. Effect of repeated oral administration of deltamethrin, fluoride, and their combination on oxidative stress parameters in the erythrocytes of rats. (Values given are mean ± SEM, n=6)*

	Groups			
	Control	Deltamethrin	Fluoride	Deltamethrin+Fluoride
LPO (nmole MDA formed/mL RBCs)	2.84±0.62^{a}	4.10±0.31^{b}	4.15±0.14^{b}	5.27±0.20^{c}
SOD (U/mg protein)	33.97±5.93^{a}	13.12±2.21^{b}	11.33±3.61^{b}	8.50±1.49^{b}
CAT (µmole H_2O_2 decomposed/min/mg protein)	82.54±4.07^{a}	47.57±1.47^{b}	32.71±1.27^{c}	56.70±3.27^{d}
GSH (nmol/ mL RBCs)	31.38±3.28^{a}	33.19±1.93^{a}	31.08±2.86^{a}	22.64±1.41^{b}
GST (µmole GSH-CDNB/min/mg plasma protein)	0.010±0.001^{a}	0.015±0.001^{bc}	0.013±0.001^{b}	0.017±0.001^{c}
GPx (U/mg protein)	10.69±0.16^{a}	9.68±0.25^{b}	9.31±0.33^{bc}	8.96±0.12^{c}

*Means with at least one common superscript do not differ significantly ($p<0.05$).

DISCUSSION

Increased lipid peroxidation and decreased activity of SOD have also been reported from exposure of rats to various pyrethroids and to F.[22-25] Malondialdehyde (MDA) is an important reactive metabolite and an indicator of LPO (lipid peroxidation).[26] LPO represents one of the most frequent reactions caused by free radical attack on biological structures[27] as reflected in elevated MDA levels resulting from disturbance of the oxidant/antioxidant balance in the biological system,[28] referred to as oxidative stress.[29]

SOD is the first and major line of defense against the action of $\bullet O_2^-$ and other reactive oxygen species (ROS). Superoxide radicals are produced in mitochondria and endoplasmic reticulum as a consequence of auto-oxidation of electron transport chain components. SOD disproportionates superoxide into hydrogen peroxide and oxygen.[30] Decreased SOD activity in the present study is suggestive of free radical generation resulting in the depletion of this enzyme owing to its excessive utilization. The decrease in the activity of CAT has also been reported in rats treated with F [31] and deltamethrin.[32] Catalase is a heme-containing enzyme

that catalyzes the disproportionation of hydrogen peroxide into water and oxygen. This enzyme is important in the removal of hydrogen peroxide generated by SOD. Stress conditions in which free radical generation occurs result in the depletion in CAT activity.[33]

Similar decreases in glutathione (GSH) levels have been reported in broiler chicks treated with deltamethrin,[34] rats exposed to F,[35] and rats co-exposed to NaF and katron.[25] Decrease in GPx activity has been found in rats exposed to deltamethrin, cypermethrin,[36,37] and F.[38] Increase in GST activity has also been reported in rats treated with permethrin, cypermethrin, or fluoride.[4,23,24] GSH is a major endogenous antioxidant that participates in detoxification reactions and counter-balances free radical mediated damage by eliminating the compounds responsible for LPO or by increasing the efficiency of NADPH that protects detoxifying enzymes.[39] GSTs catalyze the conjugation of GSH via reaction of the sulphydryl group with electrophilic centers in a wide variety of substrates. In addition GST binds with varying affinities to a variety of hydrophobic compounds such as polycyclic aromatic hydrocarbons and other xenobiotics such as pyrethroids.[40] The induction of GST in this study could be a defensive mechanism to counter-balance the oxidative insult by utilizing endogenous antioxidant GSH. This could be the possible reason for the depletion of GSH, especially in the co-treated group with its exposure to a greater toxic insult thereby producing more free radicals. As the activity of GPx is dependent upon the level of GSH, depleted GSH, concomitant with increased utilization of GPx to detoxify the toxicant induced free radicals and H_2O_2 production, could result in the significant decrease of GPx in toxicant-exposed groups especially the co-exposed group.

Lower activities of SOD, CAT, and GSH-Px as well as the GSH levels suggest that F and deltamethrin toxicity might induce the accumulation of free radicals, consumption of the antioxidants, and production of oxidative stress marked by LPO. This oxidative injury is more severe with the concomitant exposure to these chemicals. Thus, the results of this study indicate that F and deltamethrin aggravate the toxicity of each another.

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