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Inhalation Exposure of Formulated Fenvalerate (20% EC): Toxicologic Alterations in Kidney of Rats

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Synthetic pyrethroid insecticides are logically gaining ground in agriculture, veterinary and in house pest control programmes (Leahey 1985) due to severe knock down effects on insects (Casida et al. 1983) and environment compatible over organochlorine insecticides with less toxicity to mammals (Aldridge 1990). Toxicity of pyrethroids was studied in different animals and it was found that these insecticides have neurotoxic (Crafton et al. 1995) and genotoxic effects (Amer et al. 1993). He et al. (1989) reviewed 573 cases of acute pyrethroid poisoning caused by deltamethrin followed by fenvalerate. Importantly, all of the pyrethroid exposures were attributed to formulated products, specified as emulsifiable concentrates, therefore undoubtedly containing organic solvent and surfactant. Thus, the health effects reported are not due to pyrethroids alone, but also due to the formulated solvent, surfactant and other uncharacterized co-exposures (Yang et al. 2002). Xylene may also be present in the concentrates (IARC 1989). Fenvalerate is also formulated in combination with oxydemeton–methyl (Royal society of chemistry 1986).

Fenvalerate [(RS)-a-cyano-3-phenoxy benzyl (RS)-2-(4-chlorophenyl)] isovalerate, a photo stable pyrethroid insecticide has been used worldwide for control of wide range of pest insects of cotton and vegetables (Shiba et al. 1990). India is an agricultural country and the farmers use array of pesticides for controlling agricultural pests and disease causing vectors. Fenvalerate is one among the several pesticides used in large quantity for plant protection purpose. Pesticides may reach the water bodies through different modes like, run-off from agricultural fields along with rainwater, accidental spills, atmospheric transport and also by direct application (Mushingeri and David 2005). Reports in respect of toxicity of synthetic pyrethroids including formulated fenvalerate in experimental animals is very scarce. Earlier studies from this laboratory reported the pulmonary toxicity and steroidogenic alterations in fenvalerate inhalation exposed rats (Mani et al. 2001, 2002). However reports on nephrotoxicity of formulated fenvalerate in individuals exposed through inhalation is not available in literature. The purpose of present work was to investigate the effects of formulated fenvalerate (20% EC) in kidney of rats.

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Materials and Methods

Formulated commercial fenvalerate (20% E.C) available in the market by the name MOTIFEN was obtained from M/s Moti Lal Pesticide Pvt. Ltd. Masani, Mathura, India. All the other chemicals used were of analytical grade. Healthy adult male Wistar rats weighing approximately 150 ± 20 g bred at Industrial Toxicology Research Center, Gheru Campus, Lucknow were used in the present experiment. Rats were fed ad libitum with pellet diet (Amrut Laboratory animal feed M/s Maharastra chakan oil mills Ltd.) and water. Rats were maintained at 22 ± 2°C ambient room temperature, 50–60% relative humidity with 12-h light and dark cycle. Different group of rats consisting of 6 rats per group were exposed to 1/15th, 1/10th and 1/5th LC_{50} of formulated fenvalerate 4 h daily, 5 days a week for 90 days using Flow Past Dynamic Nose Only Inhalation

Table 1 Inhalation exposure doses and chamber concentration of formulated fenvalerate

Groups	Exposure doses of fenvalerate[a]	Chamber conc. of fenvalerate[a]
Control	Compressed air	0
fenvalerate 20% EC	1/5th LC_{50}	6,500 ± 135 mg/m^3
fenvalerate 20% EC	1/10th LC_{50}	3,500 ± 70 mg/m^3
fenvalerate 20% EC	1/15th LC_{50}	2,200 ± 50 mg/m^3

[a] Formulated product containing 20% of fenvalerate

Exposure Chamber (Intox Products, USA) following the methodology reported by us earlier (Mani et al. 2001). Briefly, rats were restrained in the animal holding tubes, which were air tight, having pointed nose with round aperture through which nose portion of the animals are exposed. Exposure chamber has also got connection on the top for outlet of exhaled air with the help of vacuum pump. Under the inhalation chamber, there is provision to fit nebuliser (used for generate aerosol) as per need and the quantity of aerosol to be used for inhalation chamber. Air samples were collected from inhalation chamber at 30 min interval and the concentration of fenvalerate in each sample was analyzed by using Hewlett Packard (USA) Model 5890A Gas chromatograph. The amount of fenvalerate (chamber concentration) found in the samples for different dosages are shown in Table 1. The same number of age and sex matched rats were exposed to pure compressed air under similar experimental conditions and served as control. Body weight was measured weekly and clinical observations were made daily. After the termination of the exposure the animals were anaesthetized by sodium thiopentone injection, blood collected from retro orbital plexus and sacrificed by cervical dislocation. Sera samples were separated and urea was estimated with kit obtained from Qualigen Glaxo, Bombay, whereas serum creatinine was estimated with a kit from M/s Ark Diognostics Pvt. Ltd, Mumbai, India. Both kidneys were removed, blotted dry, weighed and fixed in neutral formal saline for histological evaluations. The tissues were processed and 5 µm paraffin sections were stained with hematoxyline and eosin (Lillie and Fuller 1976). Data were statistically analyzed by Student's *t* test. P values less than 0.05 were considered to be statistically significant (Fischer 1950).

Results and Discussion

Following inhalation to formulated fenvalerate, the exposed rats showed perinasal and perioral wetness, nasal and oral irritation, mild dyspnoea, lacrimation and discomfort in exposure chamber, but they recovered quickly from such clinical symptoms after the termination of the exposure. There was no change in the food and water intake after exposure. Clinical symptoms remained the same throughout the exposure period. Five out of the 06 animals in the 1/5th LC_{50} exposed group exhibited acute signs of pyrethroid toxicity viz., tremors, convulsions, salivation and hyper excitability. The results in Table 2 indicate significant increase ($p < 0.05$) in absolute and relative weight of kidneys of rats exposed to formulated fenvalerate (1/5th LC_{50}) by nose only inhalation for 3 months duration as compared to controls. However, only slight increase (but not significant) was observed in the weight of kidneys of rats exposed to 1/10th and 1/15th LC_{50} of fenvalerate for 3 months. These observations are in agreement with the findings of Parker et al. (1984, 1986), who also noticed an increase in the weight of kidneys in rats fed with fenvalerate. One of the parameters used to determine toxic effects of a compound in animals is organ weight change. Organ weight changes in relation to control group indicate toxic effect in animals (Anderson et al. 1999). The biochemical assay of serum urea and creatinine showed significant increase ($p < 0.05$) in group of rats exposed to 1/5th LC_{50} fenvalerate for 3 months. The values of blood urea and creatinine in serum slightly elevated after 3 months exposure with 1/10th and 1/15th LC_{50} but they were not significant (Table 3).

Comparative analysis of kidney lesions between 1/15th and 1/10th LC_{50} indicated lesser degree of damage during low dosage exposure than their respective higher concentration of exposures. During 3 months post exposure period of 1/15th and 1/10th LC_{50} concentrations showed hyper-

Table 2 Effect of formulated fenvalerate inhalation (4 h/day, 5 day a week) for 3 months on absolute and relative kidney (g/100gm body weight) weight of rats

Exposure Condition	Duration	Absolute kidney weight (g)		Relative kidney weight	
		Control	Experimental	Control	Experimental
1/15th LC_{50}	3 months	1.52 ± 0.09	1.58 ± 0.05	0.74 ± 0.04	0.73 ± 0.07
1/10th LC_{50}	3 months	1.48 ± 0.01	1.52 ± 0.09	0.76 ± 0.06	0.78 ± 0.05
1/5th LC_{50}	3 months	1.55 ± 0.05	1.77 ± 0.06*	0.66 ± 0.02	0.92 ± 0.05*

Data represent mean ± SE of 6 rats in each group

*$p < 0.05$

Table 3 Effect of formulated fenvalerate inhalation (4 h/day, 5 day a week) for 3 months on serum urea and creatinine

Exposure Condition	Duration	Serum urea (mg/dl)		Serum creatinine (mg/dl)	
		Control	Experimental	Control	Experimental
1/15th LC_{50}	3 months	40.98 ± 1.85	45.66 ± 1.68	1.22 ± 0.09	1.45 ± 0.59
1/10th LC_{50}	3 months	41.52 ± 2.07	48.05 ± 2.41	1.25 ± 0.76	1.89 ± 0.08
1/5th LC_{50}	3 months	43.23 ± 1.18	73.13 ± 3.61*	1.34 ± 0.14	2.72 ± 0.46*

Data represent mean ± SE of 6 rats in each group

*$p < 0.05$

trophy in glomeruli. Desquamated epithelial cells and the presence of tubular casts were also evident. Interstitial spaces showed focal presence of mononuclear cells and some necrotic cells Fig. 1(2,3). These lesions became more evident with more and more glomerular atrophy, tubular changes, presence of more tubular casts and having marked interstitial changes during 3 months with 1/5th LC_{50} post exposure period Fig. 1(4).

Kidney dysfunction (as judged by evaluating serum urea and creatinine) is apparent in group of rats exposed to 1/5th LC_{50} fenvalerate (20% EC) for 3 months. Serum urea and creatinine are not able to get filtered from kidney of rats exposed in above dose level hence their concentrations are elevated in serum. Parker et al. (1984) observed significantly higher values of serum cholesterol, glucose and BUN in male and female DC-fenvalerate fed rats. Administration of fenvalerate to mice produced a dose dependent increase in the specific activity of γ-Glutamyl transpeptidase in kidney homogenate (Balbaa and Bassiouny 1993).

The histopathological changes of kidney of rats exposed to formulated fenvalerate by inhalation for 3 months showed focal collections of mononuclear cells and necrotic debris. In this regard it is interesting to note that Majumdar et al. (1994), also noticed large size of glomeruli and glomerular and tubular necrosis in kidney of broiler chicks treated with sub acute dermal doses of fenvalerate for 28 days. Same authors reported accumulation of mononuclear cells and proliferation of connective tissue of kidney in broiler chicks following dermal application of fenvalerate once daily for 31 days (Majumdar et al. 1997). Parker et al. (1984) found an increased incidence of critical tubular basophilia and focal to multi focal accumulations of atrophic or dilated tubules in the cortex and hypercellularity, thickened mesangium and/or glomerular loops in 6 months feeding study of fenvalerate in dogs. In contrast, we observed that exposures of rats to formulated fenvalerate by inhalation resulted in presence of a few atrophied glomeruli, hypertrophied tubular epithelial cells along with the presence of casts in the tubular lumens of kidneys. Mohammad and Adam (1990) reported fenvalerate to produce some renal changes in Nubian goats poisoned with high doses of Sumicidin (fenvalerate) 112.5 to 1,350-mg/kg-body weight. Mandal et al. (1992) reported that single oral administration of fenvalerate 5 mg/kg produced necrosis of kidney in goats.

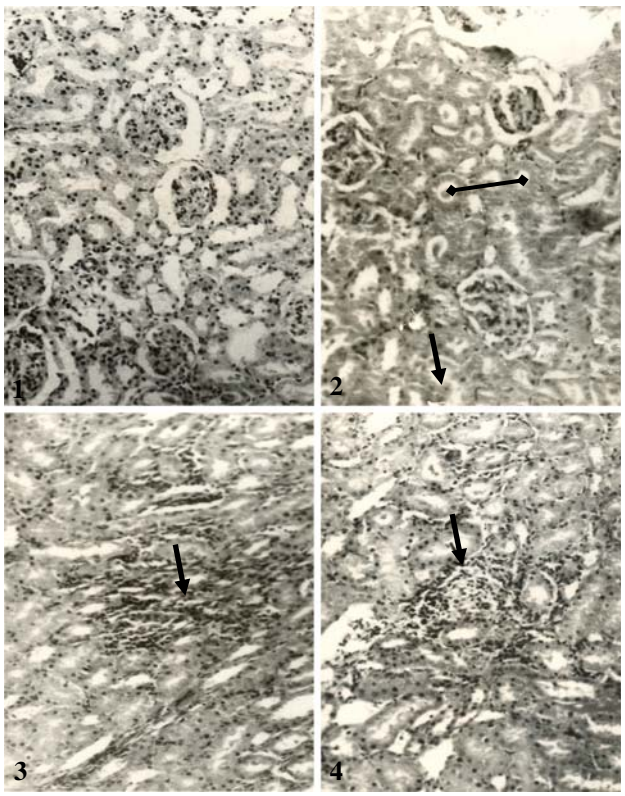

Fig. 1 **1.** Section of control rat kidney showing normal renal histo architecture. **2.** Section of rat kidney, three months post exposure to 1/15th LC50 showing the presence of hypertrophied tubular lining cells (→) and the marked presence of casts (↔) in the tubules. **3.** Section of rat kidney, 3 months post exposure to 1/10th LC50 of fenvalerate showing focal area of mononuclear cellular accumulation along with cellular debris (→) in the centro lobular area. **4.** Section of rat kidney, three months post exposure to 1/5th LC50 of fenvalerate showing localized presence of mono-nuclear cells in interstitial area and of necrotic cellular debris (→). (H & E ×400)

Abu EI-Zahab et al. (1993) observed congestion of blood vessels, hemorrhage, necrosis and inflammatory leucocytes in kidneys of rats inhaling pyrethroids. Abdeen et al. (1994) reported that treating mice with fenvalerate

induced renal damage of the epithelial lining of the renal tubule, ruptured distal tubules and enlargement of the glomeruli with hydropic degeneration. Abou-Zaid and EI-Balshy (1995) reported that inhalation of Ezalo (a synthetic pyrethroid) caused acute tubular necrosis and glomerulonephritis in kidneys of newborn mice. Sub chronic feeding of decarboxy fenvalerate was found to induce glomerulonephritis in kidneys of rats (Parker et al. 1984). Further, Sakr and Hanafy (2002) reported leukocyte infiltration and atrophied glomeruli in kidney of toads intoxicated with fenvalerate. Atrophy of renal glomeruli and hypertrophy of bowman capsules occurred in mice after poisoning with fenvalerate (Tos Luty et al. 2001).

Though the precise nature of emulsifying agent/solvents used in the formulated fenvalerate are not known. However, some studies in literature do indicate moderate toxicity associated with different formulated preparations possibly due to matrix/solvent/base used therein (Squibani et al. 1987; Oakes and Pollak 2000; Pereira et al. 2000; Yang et al. 2002). Toxicity manifestations in rats exposed to formulated fenvalerate (20% E.C) by inhalation route in the present study are being reported for the first time. Some of these might be due to solvent/emulsifier ingredients used in the formulated fenvalerate. The study suggests that the formulated fenvalerate is not toxic at lower doses. However, the biochemical and histopathological evidences at the highest dose warrant adequate training and use of protective gear by the sprayers using this formulation routinely.

References

Aldridge WN (1990) An assessment of the toxicological properties of pyrethroids and their neuro toxicity. Crit Rev Toxicol 21:89–104

Amer SM, Ibrahim AA, EI-Sherberry KM (1993) Induction of chromosomal aberrations and sister chromatid exchange in vivo and in vitro by the insecticide cypermethrin. J Appl Toxicol 13:341–345

Anderson H, Larsen S, Splid H, Christenson ND (1999) Multivariate statistical analysis of organ weights in toxicity studies. Toxicol 136:67–77

Abu-EI-Zahab HS, Amr MM, Awadallah R, Abdel Aal WE, Khyat EI (1993) Physiological and histopathological studies on long term inhalation of adult albino rats to pyrethorids. J Egypt Ger Soc Zool 11:311–334

Abdeen AM, Ameer M, EI-Habibi M. Sayed EI, Kamel M (1994) Histopathological and histochemical studies on the effect of fenvalerate insecticide on some organs of albino mice. J Union Arab Biol 2:129–166

Abou-Zaid FA, Balshy DI (1995) Toxic effect of Ezalo. A synthetic pyrethroid on liver, kidney and lung of newly born mice. J Union Arab Biol 4:235–264

Balbaa M, Bassiouny K (1993) Enhancenent of γ-Glutamyltranspeptidase by pyrethroids. J.Med Res Inst 14:9–21

Casida JE, Gammon DW, Glockman AH, Lawrence LJ (1983) Mechanisms of relative action of pyrethroid insecticides. Ann Rev Pharmacol Toxicol 23:413–438

Crafton KM, Kehn LS, Gilbert ME (1995) Vehicle and route dependent effects of a pyrethroid insecticide deltamethrin, on motor function in the rat. Neurotoxicol Teratol 17:489–495

Fischer RA (1950) Statistical methods for research workers, 11th edn. Oliver and Boid, London

He F, Wang S, Liu L, Chen S, Zhang Z, Sun J (1989) Clinical manifestations and diagnosis of acute pyrethroid poisoning. Arch Toxicol 63:54–58

IARC (1989) IARC Monographs on the evaluation of carcinogenic risks to Humans, vol 47 Some organic solvents, resin monomers and related compounds, pigments and occupational exposures in paint manufacture and painting, Lyon, pp 125–56

Leahey JP (1985) The pyrethroid insecticides. Taylor & Francis, London

Lillie RD, Fuller HM (1976) Histopathologic technique and practical histochemistry. McGrew Hill Book Co, New York

Majumdar S, Chakraborty AK, Mandal TK, Bhattacharya A, Basak DK (1994) Sub acute toxicity of fenvalerate in broiler chicks: concentration, cytotoxicity and biochemical profiles. Indian J Exp Biol 32:752–756

Majumdar S, Chakraborty AK, Bhattacharya A, Mandal TK, Basak DK (1997) Effect of short-term dermal toxicity of fenvalerate on residue, cell architecture and biochemical profiles in broiler chicks. Indian J Exp Biol 35:162–167

Mandal TK, Bhattacharya A, Chakraborty AK, Basak DK (1992) Disposition kinetics, cytotoxicity and residues of fenvalerate in tissues following oral administration to goats. Pestic Sci 35:201–207

Mani U, Islam F, Prasad AK, Kumar P, Maji BK, Dutta KK (2001) Pulmonary toxicity of a formulated preparation of fenvalerate in rats sub chronically exposed by nose only inhalation for 90 days. Biomed Environ Sci 14:333–340

Mani U, Islam F, Prasad AK, Kumar P, Kumar VS, Maji BK, Dutta KK (2002) Steroidogenic alterations in testes and sera of rats exposed to formulated fenvalerate by inhalation. Human Exp Toxicol 21:593–597

Mohammad OSA, Adam SEL (1990) Toxicity of Sumicidin (fenvalerate) to Nubian Goats. J Comp Pathol 102:1–6

Mushingeri SB, David M (2005) Fenvalerate induced changes in the AchE and associated AchE activity in different tissues of fish *Cirrhinus mrigala* (Hamilton) under lethal and sub-lethal exposure period. Environ Toxicol Pharmacol 20:65–72

Oakes DJ, Pollak JK (2000) The in vitro evaluation of the toxicities of three related herbicide formulations containing ester derivatives of 2,4,5-T and 2,4-D using sub-mitochondrial particles. Toxicology 151:1–9

Parker CM, Piccirillo VJ, Kurtz SL, Garner FM, Garner FM, Gardiner TH, Van Gelder GA (1984) Six month feeding study of fenvelerate in dogs. Fund Appl Toxicol 4:577–586

Parker CM, Wimberly HC, Lam AS, Gardiner TH, Van Gelder GA (1986) Sub chronic feeding study of decarboxy fenvalerate in rats. J Toxicol Environ Hlth 18:77–90

Pereira T, Cerejeira MJ, Santo JE (2000) Use of micro bio tests to compare the toxicity of water samples fortified with active ingredients and formulated pesticides. Environ Toxicol 15:401–405

Sakr SA, Hanafy SN (2002) Histopathological alterations in the liver and kidney of toads (*Bufo regularis*) intoxicated with a pyrethroid insecticide. J Biol Sci 2:208–211

Shiba K, Kaneko H, Kakuta N, Yositaka A, Miyamoto J (1990) Placental transfer of Esfenvelerate & fenvelerate in pregnant rats. J Pestic Sci 15:169–174

Royal Society of Chemistry (1986) European Directory of Agrochemical Products, vol 3. Insecticides, Acaricides, Nematicides, Cambridge, pp 4–10

Squibani AB, Marano F, Ronot X, Adolphe M, Dao SP (1987) Effects of Deltamethrin and its commercial formulation DECIS on different cell types in vitro: cytotoxicity, cellular binding, and intracellular localization. Pestic Biochem Physiol 28:103–113

Tos Luty S, Haratym Maj A, Latuszynska J, Przebirowska DB, Rodak MK (2001) Oral toxicity of deltamethrin and fenvalerate in swiss mice. Ann Agri Environ Med 8:245–254

Yang PY, Lin J-L, Hall AH, Tsaw TSY, Chern MS (2002) Acute ingestion poisoning with insecticide formulations containing the pyrethroid permethrin, Xylene and surfactant: A review of 48 cases. Clin Toxicol 40:107–113