Histopathological Changes In White Matter Of Cerebellum In Pyrethroid Exposed Albino Rats

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ABSTRACT

Introduction: The liquid vaporizers are very commonly used as residential insecticides in developing countries. Neurotoxic effects of pyrethroids have been reported earlier but studies regarding their effects on white matter of cerebellum are scanty in spite of its direct exposure. So the present study was planned to assess and compare the effects of long term prallethrin (a pyrethroid) exposure on white matter of cerebellum.

Method: Twenty albino rats were divided into two groups of control and experimental. Rats in experimental group were exposed to 3.2% w/v prallethrin vapors 12 hours daily for 180 days. Control animals were kept under identical conditions without exposure to said repellent. The animals were sacrificed after 180 days. Their Cerebellum removed and processed. Sections cut and stained with Haematoxylin and Eosin.

Result: The loosening of the white core along with cellular microcystic change with interstitial oedema was present in Experimental Group. White matter showed large empty looking cells with darkly stained nucleus and increased cytoplasmic vacuolation compared to control

Conclusion: : This study showed that long term pyrethroids exposure resulted in loss of white matter of cerebellum in rats. At present , the significance and manifestation of such a loss of white matter is not known . Further behavioural study is required to shed some light on this issue.

Keywords: pyrethroids, liquid vaporizers,rats,cerebellum,white core, histology

Introduction

Pyrethroids are synthetic compounds derived from natural pyrethrin (Elliot et al, 1976).These newer compounds emerged as potent cidal agents for vectors and pests. Pyrethroid insecticides are used worldwide and considered to be relatively safer than organochlorines and organophosphates and has successfully acquired an international market of insecticides. Pyrethroids are widely used insecticides in agriculture, household and for industrial purpose in India and other countries to get protection from mosquitoes, cockroaches and other insects (Das et al, 2003).

Crofton and Reiter (1988) studied the relative potencies for acute effects of pyrethroids on motor function in rats. They found that all pyrethroids regardless of structure and class produced dose dependent decrease in motor activity.

The major types of residential insecticidal products include aerosols, mosquito coils and vaporizing mats among which the liquid vaporizers have outnumbered others in popularity (Krieger, 2003).Their toxic effects have been observed in non target organs causing muscle pain, joint pain, ataxia, chronic fatigue, headache and difficulty in concentration

Pyrethroid induced neurotoxicity and their deleterious effects in humans and experimental animals caused a concern on their chronic use. Prolonged use of mosquito repellent has also been reported to cause convulsions exhibiting CNS toxicity in exposed infants beside other harmful manifestation such as corneal damage, shortness of breath, asthma and damage to liver (Briassoulis, 2001; Liu et al, 1988 and Sudakin et al, 2003). It has been shown that new born babies and pregnant and lactating mothers are more prone to toxic insults when exposed to pyrethroids as they stay for longer duration inside the houses and exposed to mosquito repellents in which Pyrethroid such as allethrin is the active ingredient (Sharma, 2001).Allethrin and prallethrin are the chief constituents of various mosquito repellent insecticides in India (Ramesh and Vijayalakshmi, 2001; Liu et al, 2003).The above reports provide evidence of the deleterious effect of pyrethroid based mosquito repellents exposure in humans These compounds are being extensively used and the product information leaflet enclosed by the manufacturers are too ambiguous to ensure the safety profiles on prolonged usage. Thus it has aroused an urgent need to reaffirm its toxicity.

Most of the previous reports are based on studies on immature mammals who received drug through different routes except respiratory. The latter being conventional route through which millions of people are exposed for several decades. Therefore the present study was carried on adult male albino rats exposed to 3.2% w/v of prallethrin vapors for a period of 180 days to assess sections of cerebellum for any changes.

The cerebellum is a central part of the major circuitry that links sensory to motor areas of the brain, and is required for the coordination of fine movement. In health, it provides corrections during movement, which are the basis for precision and accuracy, and it is critically involved in motor learning and reflex modification.Cerebellar output is mainly to those structures of the brain that control movement. Many workers studied the effect of pyrethroids on cerebellum and found depleting changes. White core of cerebellum was selected as most of the healthy population is exposed to Pyrethroid in liquid vaporizers for long period and very little attention is focused on its effect on white core.

Material and Method

The present study was carried out on adult Charles foster rats weighing between 100-150gms. The animals were provided with standard pellet laboratory diet (Lipton India Limited) and water ad-libitum. They were housed under identical diurnal conditions and temperature. The animals were weighed, marked and divided into two groups:

Group 1-Experimental
Group 2-Control
The experimental animals were kept in unit plastic cages (36cm x 22cm x 14cm) with many holes. They were exposed to liquid mosquito repellent inside a closed room (180cm x 240cm) according to the method of Sinha et al (2004). The animals were exposed to 3.2% w/v prallethrin vapors for 12 hrs daily for a period of 180 days. The control animals were kept under identical conditions without exposure to 3.2% w/v prallethrin vapors. The body weight was measured weekly and the water consumption was assessed daily.
Rats in the experimental group were exposed to mosquito repellant vapors every day for 12 hours for 180 days. On day zero that is before exposure, the weight and physical activities of both control and experimental rats were noted down.
At the end of exposure (after 180 days) the animals (experimental and control) were sacrificed and sections were taken from cerebellum to detect the neuro-histological changes, if any.
The animal was first made unconscious by exposure to diethyether vapors, and the heart was exposed through the thoracic approach. The needle of the blood transfusion set was introduced into the left ventricle (apex) and a niche was made in the right atrium. 10% Formalin in distilled water was then infused till the body tissues showed signs of fixation.
The brain was approached through the dorsal aspect of skull and spine by the method of Heffnera et al (1980).The skin of the head was cut from the base of the head to the mid eye area. After reflecting the skin the skull was cut open at the midline fissure using a pair of scissors. The scissors were lifted up while cutting to avoid any damage to the cortex. Dorsal part of the skull was removed using curved forceps. The brain was removed by releasing it gently from the bottom and sides of the brain calvarium. A tissue section of 5mm size was then taken from cerebellum and prepared for neuro-histological examination. The tissue sections were stained by H & E.

Results

In between the two cerebellar hemispheres, the compact mass of white matter extends into the folia as a core of white matter(Fig1a).White matter is located under the grey matter of the cortex and consists of nerve fibres which are myelinated running to and from the cortex(Fig 1a).The loosening of the core of white matter was observed in the experimental group as compared to control. Odema along with cellular microcystic change was also seen in experimental group (fig1b).The nucleus was darkly stained with swollen and empty looking cells suggestive of degeneration.

Discussion

The finding of this study showed that long term pyrethroid inhalation results in loss of white matter of cerebellum of albino rats. This study is suggestive of the sensitivity of white matter to the damaging effects of pyrethroids as indicated by oedema and cytoplasmic vacuolation in the experimental group.

The process leading to the depletion of white matter is not clear , however it is a possibility that the interaction of pyrethroid with some receptor in white core triggered apoptotic process leading to loss of white matter. Long term pyrethroid exposure has deleterious effects on cell development particularly in purkinje cell layer in cerebellum of albino rats characterized by reduced number of purkinje cell number.

Conclusion

The findings of our study establish that pyrethroids given by inhalational route do cause neurotoxicity on chronic exposure as shown by degenerative changes in the histological sections of cerebellum. The findings are paralleled by white matter changes in cerebellum in the present study eventually leading to cellular odema in white matter. This study showed that long term pyrethroid exposure resulted in loss of white matter of cerebellum in rat model system. At present the significance and manifestation of this loss is yet to be determined . It appears that for the manifestation of behavioral abnormality and to see if these histological changes are reversible, a further study with prolonged period of duration is required, as in today's scenario the exposure to pyrethroid containing mosquito repellent is continued and chronic .

Conflict of Interest: None declared.

References

1. Elliot M: Properties and application of pyrethroid. *Environment health Perspect*.1976,14: 3-13.
2. Das NG, Baruah I, Talukdar PK, Das SC: Evaluation botanical repellents against mosquitoes. *J. Vector Borne Dis*.2003, 40(1–2): 49–53.
3. Crofton KM, Reiter LW: The effects of type 1 and II pyrethroids on motor activity and the acoustic startle response in the rat. *Fundamental and Applied Toxicology*,1988,10(4): 624-634.
4. Krieger RI, T.M. Dinoff, X. Zhang: Octachlorodipropyl ether (S-2) mosquito coils are inadequately studied for residential use in Asia and Illegal in the United States. Environ. *Health Perspect.*2003,111: 1439–1442.
5. Briassoulis G, Narlioglou M, Hatzis T: Toxic encephalopathy associated with use of DEET insect repellents: a case analysis of its toxicity in children. *Human Exp. Toxicol*.2001,20: 8–14.

6. Liu WK, Sun SE: Ultrastructural changes of tracheal epithelium and alveolar macrophages of rats exposed to mosquito coil smoke. *Toxicol. Lett.*1988,41:145–157.
7. Sudakin D, Trevathan WR : DEET, a review and update of safety and risk in the general population. *J. Toxicol., Clin. Toxicol*.2003, 41: 831–839.
8. Sharma V P:Health hazards of mosquito repellents and safe alternatives. *Curr. Sci*. 2001,80: 341–343.
9. Ramesh A, Vijayalakshmi A: Monitoring of allethrin, deltamethrin,esbiothrin,prallethrin and transfluthrin in air during the use of household mosquito repellents. *J. Environ.Monit.*2001,3:191–193.
10. Liu W, Zhang J, Hashim JH, Jalaludin J, Hashim Z, Goldstein BD: Mosquito coil emissions and health implications. *Environ.HealthPerspect*.2003,111(12):1454–1460.
11. Sinha C, Agarwal AK, Islam F, Seth K, Chatuvedi RK, Shukla S, Seth PK: Mosquito repellent (pyrethroid- based) induced dysfunction of bloodbrain barrier permeability in developing brain. *Int. J. Dev. Neurosci*.2004, 22 (1):31–37.
12. Heffnera TG, Hartman JA, Seiden LS: A rapid method for the regional dissection of the rat brain. *Pharmacology Biochemistry and Behavior*.1980,13(3):453-456.

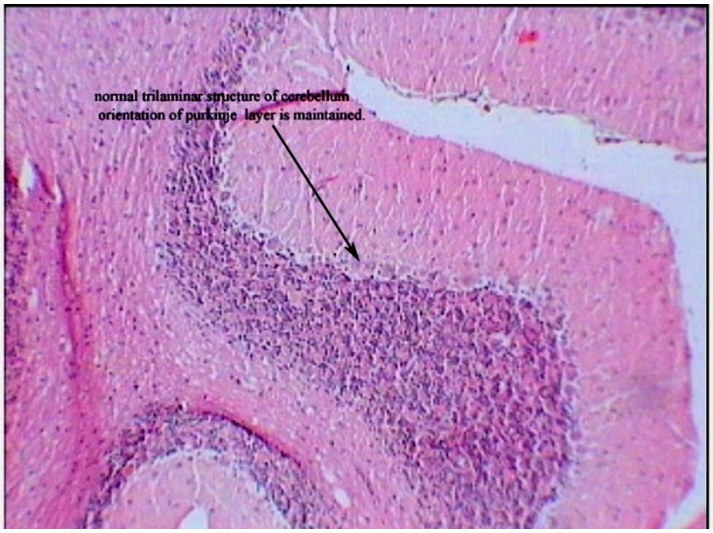

Fig 1a: control showing white core, granular layer and molecular layer.

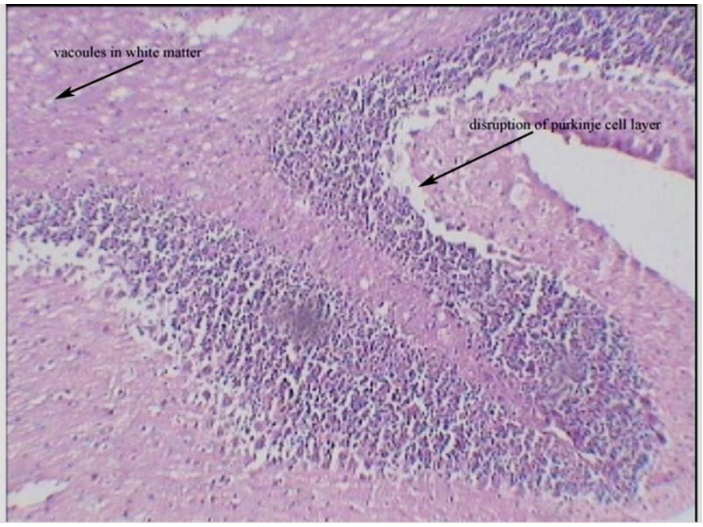

Fig 1b: experimental showing vacuolation in white core and disruption of purkinje cell layer.