

Sub-Chronic Exposure to Mosquito Coil Smoke in Mice: Effect on Motor Coordination

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Abstract: Mosquito coil is a common insect repellent used in many homes with the prevalence of mosquitoes. It burns and emits smoke that repels and immobilizes mosquito. However, the smoke emitted from the coil has deleterious effects. The aim of this study was to investigate the effect of sub-chronic exposure to mosquito coil smoke on motor coordination in mice. Twenty-eight adult mice weighing 20 – 38g were used for the study and divided into four groups of seven mice each. Group one was exposed to environmental air; group two, three and four exposed to Goldeer mosquito coil smoke (containing 0.03% transfluthrin) for one, two and three hours respectively for six weeks. Mice were passed through the Beam walking assay after one, three and six weeks of exposure. The mice were anaesthetized, sacrificed and brain sample harvested and homogenized. The homogenate was centrifuged, and the supernatant used for biochemical assays of malondialdehyde (MDA), superoxide dismutase activity (SOD) and catalase activity (CAT). Data were analysed using One Way Anova with Tukey post hoc tests. Results were expressed as Mean \pm SEM and P values of ($P < 0.05$) regarded as statistically significant. The result showed that prolong mosquito coil smoke inhalation could impair motor coordination and therefore its usage must be discouraged among the local population.

Keywords: Mice, mosquito coil smoke, motor coordination, beam walk.

I. INTRODUCTION

Malaria is a life-threatening disease caused by parasites that are transmitted to people through the bites of infected female Anopheles mosquitoes. According to recent WHO estimate, there were 212 million cases of malaria in 2015 and 429, 000 deaths [1]. The endemic nature of malaria infestation in Nigeria and the economic implication of being infected have left many families with no choice other than to look for any way possible to avoid it [2]. The mosquito coil is the commonest and cheapest methods used by many families to drive away mosquitoes from sleeping rooms [3]. The main active ingredients in mosquito coils are pyrethroids [4], which are effective against many genera of mosquito, including *Aedes*, *Anopheles* and *Mansonia* [5]. Other ingredients could also include Dibutylhydroxyl toluene (BHT), Piperonyl butoxide (PBO), aromatic and aliphatic hydrocarbons [7]. Finding by [7] showed that burning one mosquito coil would release the same amount of particulate matters as burning 75-137 cigarettes and emission of formaldehyde as high as that released from burning 51 cigarettes. Actually, there has been a growing concern among the public regarding the routine and prolonged use of mosquito coils [8]. It poses a serious public health hazards and ecological challenges because of the effects associated with their use, especially innocuous and chronic inhalation of the fumes [9, 2]. It has been shown by [10] that locomotion is affected by pyrethroids. Pyrethroids causes loss of coordination [11] and affect other behaviours that are dependent upon the coordinated movement of animals [12].

At low to intermediate levels of exposure, pyrethroids impair motor function in insects. This effect manifests as alterations in flying or walking patterns. At intermediate dose levels, motor activity is disorganized, eventually forcing non-flying species to become immobilized and flying species to fall down [13]. Looking at the health implications of prolonged exposure to mosquito coil smoke and its increased usage among the greater population of local communities in Nigeria. It is therefore necessary to look at its negative effect on the body system which includes the nervous system and motor coordination.

II. MATERIALS AND METHOD

A. EXPERIMENT AND ANIMAL GROUPING: Twenty eight adult mice weighing 20 – 38 grammes were obtained from the National Institute of Trypanosomiasis and Oncocerciasis Research (NITOR), Kaduna, Nigeria and allowed to acclimatize for two weeks at the Department of Human Physiology, Ahmadu Bello University, Zaria, Kaduna State, Nigeria. They were fed mainly with grower mash and water ad libitum. Goldeer mosquito coil containing 0.03 transfluthrin and manufactured in Kano, Nigeria was obtained from a retail shop in Zaria, Nigeria. The mice were divided into four groups of seven mice each. Group 1 was exposed to environmental air only while mice in groups 2, 3 and 4 were exposed to mosquito coil smoke 1, 2 and 3 hours respectively for 6 weeks in a partially ventilated inhalation chamber (2.00 x 0.98 x 1.55m). The mice were passed through beam walk assay after 1, 3, and 6 weeks of exposure. After which the brains of the animals were harvested. The whole brain was homogenized, centrifuged and the supernatant used for oxidative stress biomarkers. Data obtained were analyzed by SPSS version 17 using One Way ANOVA followed by Tukey multiple comparison tests and expressed as Mean \pm SEM. Values of $p < 0.05$ were taken to imply statistical significance.

All experimental protocols were in accordance with the Ahmadu Bello University research policy, ethics and regulations governing the care and use of experimental animals.

B. MOUSE BEAM WALKING ASSAY: The method of [14] was used in which mice were trained to walk from a start platform along a ruler (80cm long, 3cm wide) elevated 30cm above the bench to a goal box (enclosed hamster house). Training was performed for each mouse until the mice trained were aware that there was a goal box that could be reached. Once the mice had been trained on the ruler, they were moved to the beam test. The beam was made of wood, 8mm in diameter, 60cm long and elevated 30cm above the bench by metal support. The mice were placed on the beam at one end and allowed to walk to the goal box. Mice that fell were returned to the position they fell from, with maximum time of 60 sec allowed on the beam. The measurement taken was time on beam and the number of foot slips (one or both hind limbs slipped from the beam).

C. EVALUATION OF BRAIN LIPOPEROXIDATION: The level of thiobarbituric acid reactive substance, malondialdehyde (MDA) as an index of lipid peroxidation was evaluated on the brain sample using the method of [15] as modified by [16]. The principle of the method was based on spectrophotometric measurement of the colour developed during reaction of thiobarbituric acid (TBA) with MDA.

D. EVALUATION OF BRAIN ANTIOXIDANT ACTIVITY: Superoxide dismutase and Catalase activity were assayed using superoxide dismutase assay kits from NorthWest Life Science Specialties, Vancouver, WA 98662, NWLSS™ NWK-SODO2 based on the method of [17] and Catalase assay kits from Northwest Life Science Specialties CAT01, following the manufacturer’s manual. Ultrospec Plus Spectrophotometer Model number 4054 at wavelength of 540nm was used to measure absorbance.

III. RESULT

Table 1 showed the Beam walk latency across the groups at week 1, week 3 and week 6 of mosquito coil smoke-inhalation and the control group. There was statistical increase in Beam walk latency at week 1 for 2 hours exposure (11.6 ± 1.36) and 3 hours exposure (11.8 ± 1.70) as compared to control (8.0 ± 1.10) and at week 3 for 1 hour (23.6 ± 7.46) and 3 hours (28.8 ± 2.77) as compared to control (14.8 ± 1.93).

Table 2 showed the effect of mosquito coil smoke inhalation on foot slip in mice. There was statistical significance foot slips in mice exposed for 2 hours daily for 3 weeks (1.60 ± 0.25) compared to the control (0.40 ± 0.14) and those exposed for 6 weeks at 1 hour (1.60 ± 0.4); 2 hours (1.80 ± 0.37) and 3 hours (1.60 ± 0.40) as compared to control (0.20 ± 0.10).

Figure 1 shows the MDA concentration of the mosquito coil smoke-inhaled and control groups. There was increase in MDA concentration across the groups but not statistically significant.

Figure 2 shows the activity of SOD in mosquito coil smoke-inhaled and control groups. Result obtained showed increased in SOD activities in all the groups compared to the control but not significant statistically. Figure 3 shows CAT activity in mosquito coil smoke-inhaled and control groups. Result obtained does not show statistical significant increase or decrease when compared to the control group but showed statistical significant decrease ($P < 0.01$) in the 2 hours inhaled group as compared to 1 hour group. Result also showed a significant increase ($P < 0.05$) in the 3 hours group as compare to the 2 hour group.

TABLE 1: EFFECT OF MOSQUITO COIL SMOKE INHALATION ON BEAM-WALK LATENCY IN MICE

Group	Week 1	Week 3	Week 6
Control	8.0 ± 1.10	14.8 ± 1.93	8.0 ± 1.00
1 hour	6.0 ± 0.89	23.6 ± 7.46^a	8.4 ± 0.51
2 hours	11.6 ± 1.36^a	13.8 ± 3.00	8.8 ± 1.43
3 hours	11.8 ± 1.70^a	28.8 ± 2.77^a	7.6 ± 0.93

Results are presented as Mean \pm SEM; n = 5; a = statistical significant difference ($P < 0.05$).

TABLE 2: EFFECT OF MOSQUITO COIL SMOKE INHALATION ON BEAM-WALK FOOT SLIP IN MICE

Group	Week 1	Week 3	Week 6
Control	0.60 ± 0.24	0.40 ± 0.14	0.20 ± 0.10
1 hour	0.20 ± 0.20	0.80 ± 0.24	1.60 ± 0.40 ^a
2 hours	1.20 ± 0.13	1.60 ± 0.25 ^a	1.80 ± 0.37 ^a
3 hours	1.60 ± 0.40	1.40 ± 0.30	1.60 ± 0.40 ^a

Results are presented as Mean ± SEM; n = 5; a = statistical significant difference (P < 0.05).

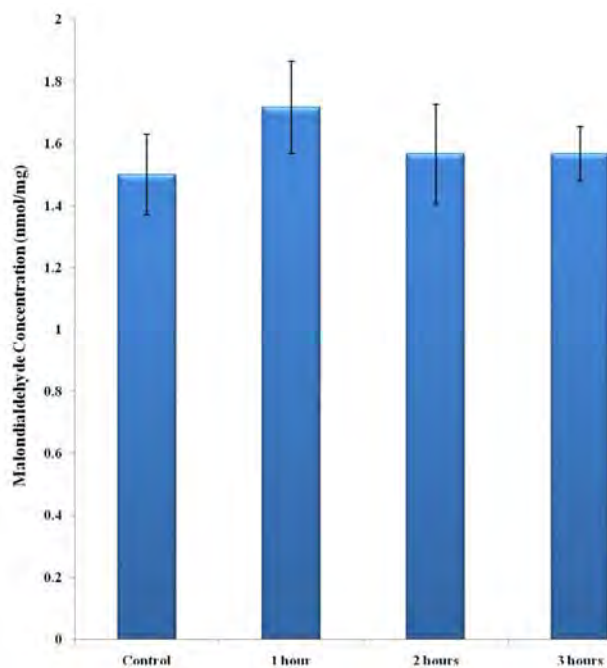


Figure 1: MDA concentration of mosquito coil smoke-inhaled and control groups (P < 0.05).

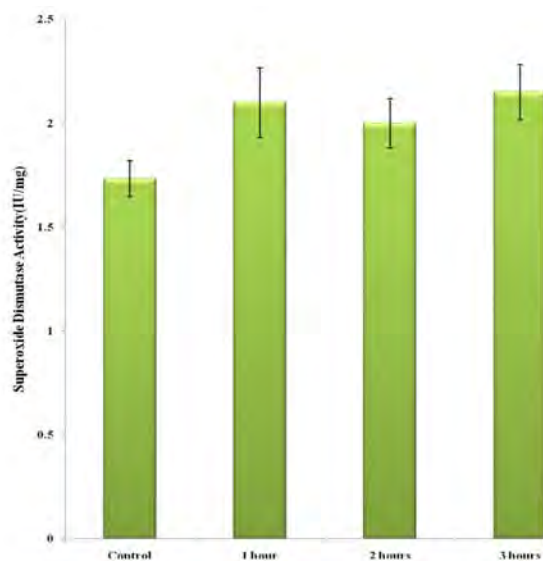


Figure 2: Activity of SOD in mosquito coil smoke-inhaled and control groups ($P < 0.05$)

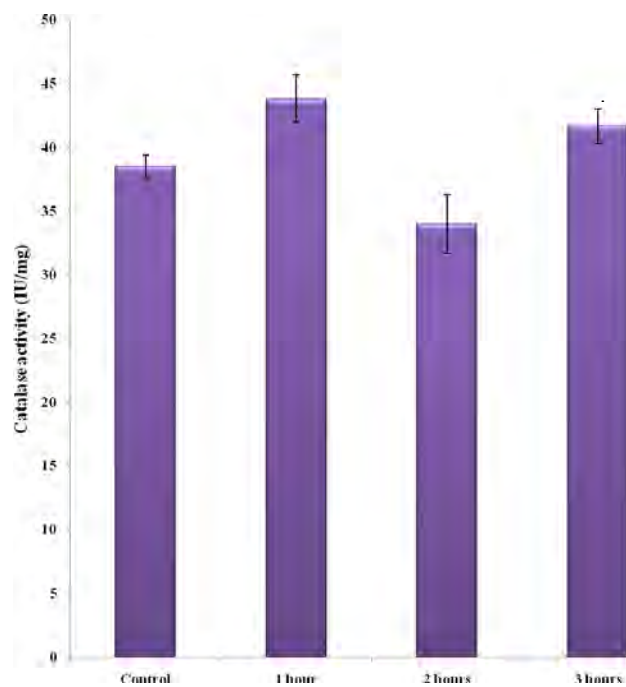


Figure 3: Catalase activity in mosquito coil smoke-inhaled and control group ($P < 0.05$).

IV. DISCUSSION

The result showed significant increase in foot slip, the most important measure of motor in coordination, in the group that got exposed to mosquito coil smoke for six weeks. This showed that mosquito coil smoke exposure for a longer duration affect neuronal activities in the brain which could impair motor function. [18] had stated that neuronal activity within basal ganglia and cerebellar loops with motor areas of the cerebral cortex is highly correlated with parameters of movement. This finding is also consistent with the findings of [12] who stated that motor functions including motor coordination is a behavioural domain impaired by all pyrethroids compound regardless of species and types. [19] also found that abnormal motor functions, decreased grip strength and reflex changes were observed in pyrethroid such as cypermethrin and permethrin [20]. Some components of mosquito coil combustion such as benzo(a)pyrene induced significant alteration of locomotors activity that correlated with relative brain concentration [20, 21]. It has been reported by [22] that the animals exposed to pyrethroid compounds (transfluthrin) showed signs of tiredness and mental confusion.

The increase in malondialdehyde (MDA) concentration in the mosquito coil smoke inhaled groups (though statistically insignificant) showed that oxidative stress could have been partly involved in the molecular mechanism of motor function deficits observed in the present study. This is because the brain, due to its biochemical and physiological properties is sensitive to free radicals which destroy its function and structures [23, 24]. MDA is the end – product of lipid peroxidation, which is a process where reactive oxygen species degrade polyunsaturated fatty acids [25]. Actually, [26] has shown that pyrethroid had the propensity to cause significant oxidative damage in rat brain. Mosquito coil smoke has been shown to elicit reactive oxygen species and interferes with the antioxidant defence system [27, 28]. The antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) showed a trend of increase activity except with CAT which decreased significantly ($P < 0.01$) in the 2 hours exposed group. This agrees with the finding of (29) who reported that increased oxidative stress caused increase in the activity of antioxidant enzymes such as SOD and CAT. The SOD provides a defence against oxidative damage of the tissue by inactivating superoxide radicals [30] and thus its role in central nervous system (CNS) becomes more important. The decreased in CAT activity in the 2 hours exposed group agreed with the finding of [31] that mosquito repellent exposure could lead to decrease in CAT which exhibits a higher affinity for H_2O_2 by degrading it to H_2O and O_2 , limiting the damage of H_2O_2 before it could diffuse to other parts of the cell.

V. CONCLUSION

From the findings, we conclude that prolong inhalation of mosquito coil smoke could lead to motor coordination deficit. Its prolong usage in human could also be detrimental. Therefore, there should be a concerted effort to raise public awareness on the health hazard of mosquito coil indiscriminate usage.

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