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Exposure to cigarette smoke altered the cytoarchitecture and anti-oxidant activity of the frontal cortex in Wistar rats

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ABSTRACT

This study was designed to determine the effects of cigarette smoke on the body weights of Wistar rats, anti-oxidant activity and the histology of the frontal cortex. Sixteen Wistar rats with mean weight of 131.40 ± 4.43 g were used. They were grouped into four. Group A had a stick of cigarette (Pall Mall brand ®) daily, group B had two sticks and group C had three sticks daily, while Group D served as the control group. They were exposed at 1800 hrs each day for thirty five days, and their weights were monitored. After sacrifice by cervical dislocation, the tissues were processed for histological and biochemical studies. The results showed minimal weight change in the exposed groups compared to the marked increase in the control group. The activity of glutathione peroxidase (GPx) enzyme increased while there was decrease in malondialdehyde (MDA) activity. The histology also showed decreased staining of the nucleic acids as well as increased vacuolations in the exposed groups. The authors suggested these findings could alter brain functions particularly those related to the frontal cortex.

Keywords: Cigarette smoke, frontal cortex, GPx, MDA, nucleic acids.

INTRODUCTION

Cigarettes are products consumed through inhalation of smoke and manufactured from finely reconstituted tobacco, often combined with other additives. Tobacco is consumed in every part of the world, most especially in the developing countries (Aghaji, 2008), with a world population of about 2.4 billion people consuming tobacco, either in form of snuff, chewing or smoking, and an annual five million death is attributed to tobacco smoking, the second leading cause of mortality among adults. The nicotine content is the highest and most toxic compound of aqueous extract of tobacco leaves and of neurotoxic importance (Penton and Lester, 2009). Exposure to nicotine alters the normal functions of the brain and the whole nervous system (Wickstrom, 2007) with active and passive smoking shown to have similar biological effect (Doll et al., 2005). Environmental

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tobacco smoke contains smoke release sed from the burning cigarette which is the part that emerges directly into the environment and the mainstream smoke is the part inhaled by the smoker (Raw et al., 1998). Therefore, passive smokers inhale the side-stream smoke which contains many toxic constituent like the mainstream smoke (Silver et al., 2003).

The frontal lobe of mammals is involved in voluntary motor functions, motivation, foresight planning memory, mood, emotions, social judgment and aggression (Fix, 2000). Therefore, because of this important fact, a structural change that may occur within the frontal lobe is vital to health state (Adeniyi et al., 2010).

Studies have linked cigarette smoking to deficits in frontal lobe functions such as attention and memory in adults. It thins the cerebral cortex in an area suggested to be linked to addiction, meaning long-term smokers could become more prone to addiction the longer they continue to smoke. The greater a teen's addiction to nicotine, the less active his prefrontal cortex, suggesting that smoking can affect brain function (Davis, 1997).

MATERIALS AND METHODS

Experimental animals

Sixteen male Wistar rats with mean weight 131.40 ± 4.43 g, were randomly divided into three treated groups A, B, C, and a control group D. The rats were allowed to acclimatise for two weeks, during which they were fed with animal feeds and water. They were housed in different cages at the Animal House of the Faculty of Basic Medical Sciences, University of Ilorin.

Each rat in group A was exposed to a stick of cigarette (Pall Mall brand ®), each rat in group B was exposed to 2 sticks, while each rat in group C was exposed to 3 sticks of cigarette daily. The control group D was not exposed to cigarette at all. Exposure was done at 1800 hrs daily, and lasted for average of 9 min, 22 min and 34 min for groups A, B and C respectively. Total duration of exposure was 35 days.

Smoking chambers

A smoking chamber was constructed using a plastic container with a lid. A hole of about 1 cm in diameter was made on the lid. The cigarette stick was suspended in the plastic container via a thread. Three of such plastic containers were constructed, and a rat from each group was exposed one at a time. The lids of the containers were opened periodically to avoid choking during the administration.

Specimen collection and processing for histological and biochemical studies

The rats were sacrificed 38 hours after the last exposure to cigarette smoke by cervical dislocation. The cranium was gently opened to expose the brain and the frontal lobe was identified and excised. The frontal lobe was placed in 0.25 M solution under a frozen condition. It was homogenized, centrifuged at 5000 rpm for 5 min. The supernatant was collected using pipette, to analyse the activities of malondialdehyde (MDA) and glutathione peroxidase (GPX). The tissue for histological studies was fixed in formal saline and processed using the Feulgen reaction staining techniques for nucleic acids.

RESULTS

Compared to the control, the rate of weight gain progressively reduced, according to the dose of cigarette, and animals exposed to the highest concentration of cigarette smoke gained no weight, but rather lost weight at the end of the 35 days treatment (Table 1).

The activity of GPx increased significantly in the exposed groups, being highest in the highest dose group C, and lowest in the lowest dose group A (Figure 1). As compared with the control animals,

activity of MDA reduced significantly in the exposed groups as shown in Figure 2.

The histology of the frontal cortex, using the Feulgen DNA staining technique showed a progressive reduction in the staining intensity when compared with the control, such that animals exposed to the highest dose of cigarette smoke had the least positivity for Nissl granules. Presence of vacuolar spaces also progressively increased in the treated groups, and were most numerous in the highest dose group C.

Table 1: Weights of the animals.

Weights	A: 1 stick	B: 2 sticks	C: 3 sticks	D: Control
Final Weight (g)	140.78±5.273	132.80±1.390	142.68±1.970	131.25±8.168
Initial Weight (g)	143.00±5.458	131.90±1.816	144.28±3.140	106.43±6.412
Weight Diff (g)	-2.22	0.9	-1.6	24.82*

*Statistically significant difference (p <0.05)

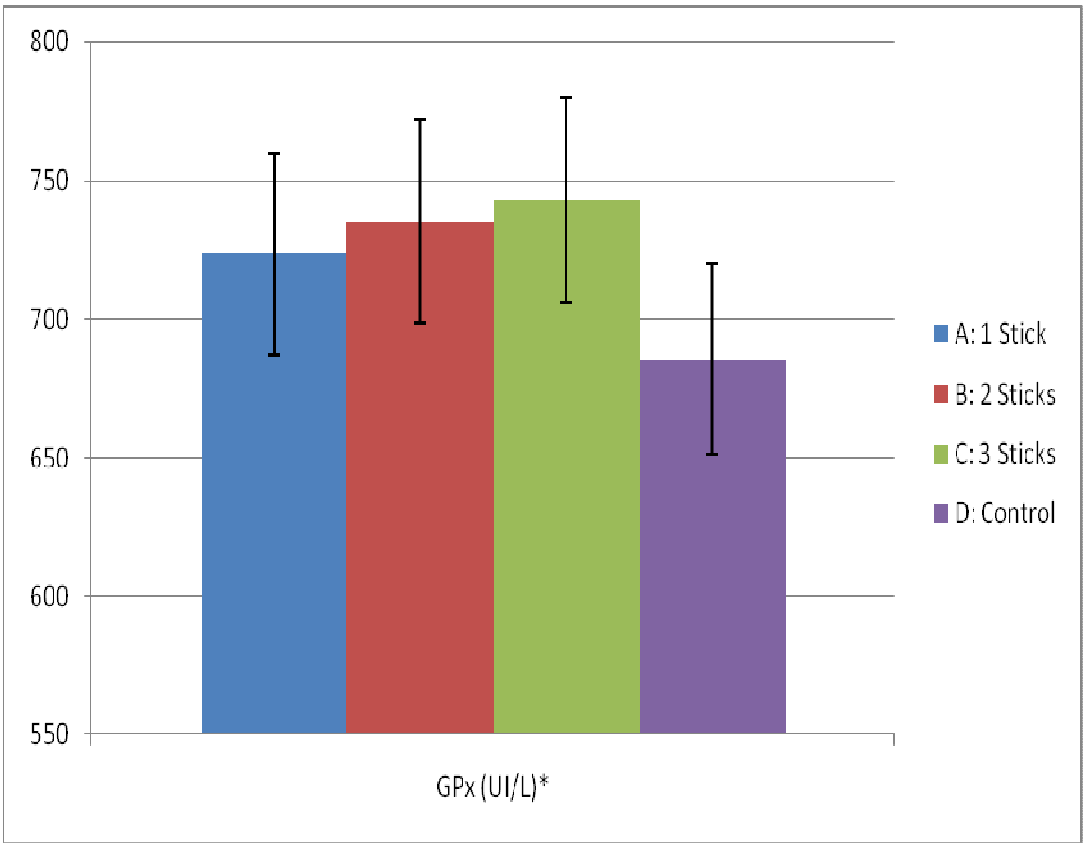


Figure 1: Activity of glutathione peroxidase (GPx) showing significant increase *(p <0.05) compared with control.

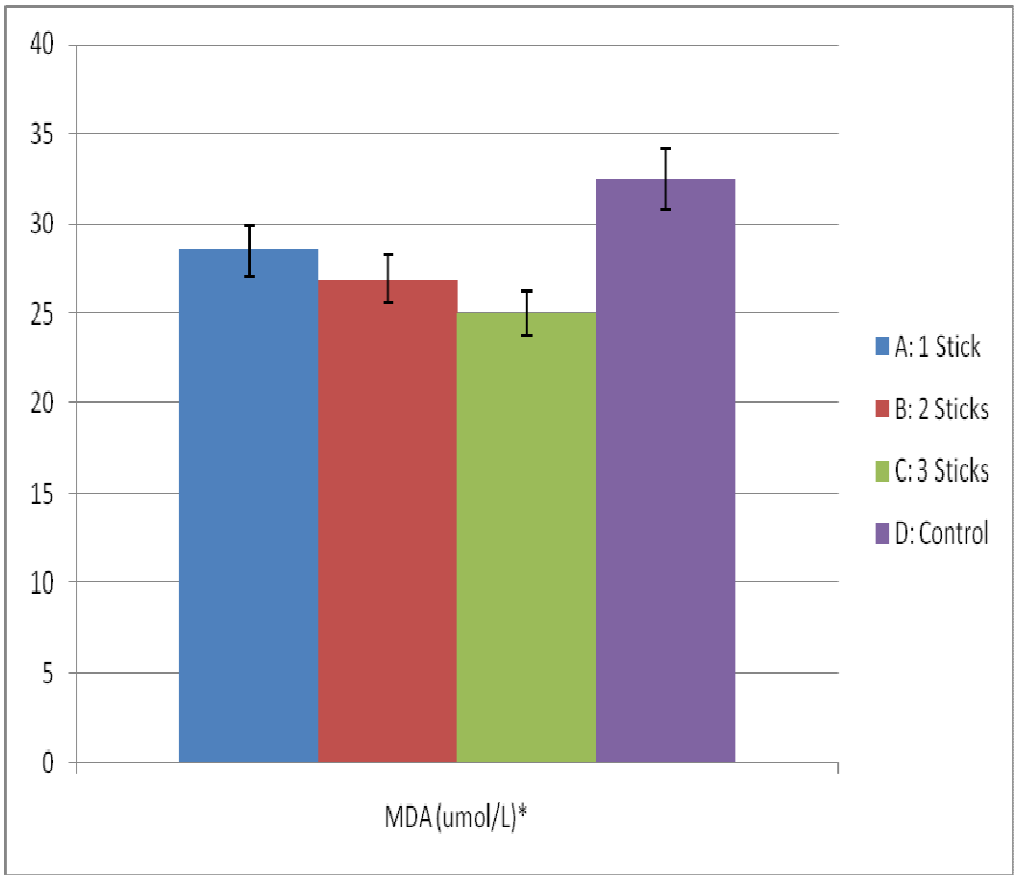
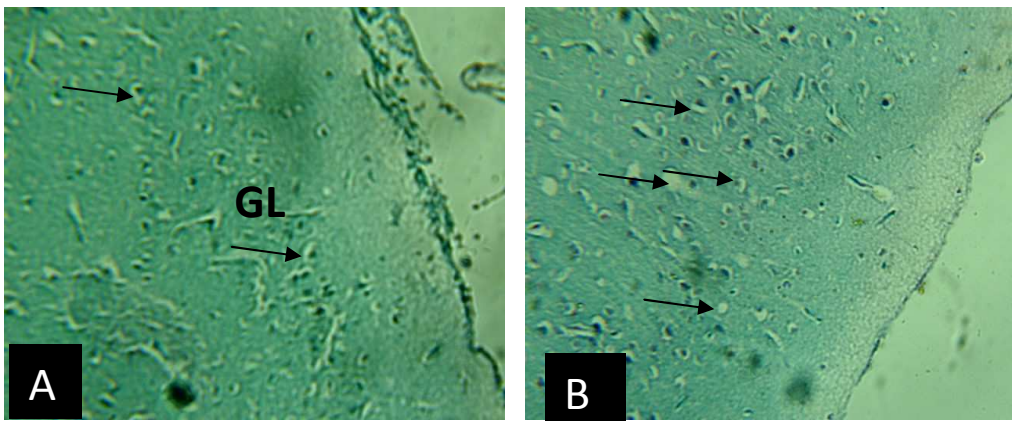


Figure 2: Activity of malondialdehyde (MDA) showing significant decrease *(p <0.05) compared with control.



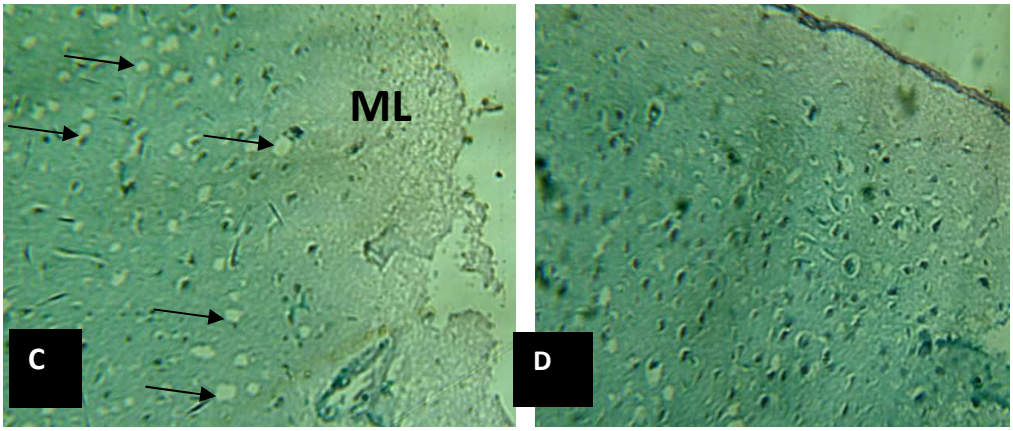


Figure 3: Photomicrographs of the frontal cortex of Wistar rats showing gradual reduction in staining intensity for nucleic acids, and increasing numbers of vacuolar spaces (arrows in A, B, C) in animals exposed to a stick of cigarette (A) and those exposed to 3 sticks (C), compared to the control (D). Feulgen DNA x100.

DISCUSSION

The administration of tobacco to the animals resulted in weight differences which were statistically significant. There was marked increase in weight in the control group while the treated group A had minimal increase in weight and group C even lost some weight. These weight differences may be due to decrease metabolic activities, reduction in appetite for food or reduction in fat deposition in the treated group, as earlier investigated (Chen et al., 2005). Cigarette smoking increases a person's metabolic rate in part by forcing the heart to beat faster. Although nicotinic receptors have been demonstrated in hypothalamic appetite-regulating areas and nicotine administration alters food intake and body weight in both animals and humans, the mechanisms underlying the effects of smoking on appetite circuits remain unclear (Chen et al., 2005).

Smokers weigh less because they consume less and they gain weight upon stopping smoking as a consequence of consuming more. In addition to affecting feeding behavior, however, cigarette smoking has a variety of physiological effects that may play a more important role than amount of calories consumed, in accounting for lower body weights among smokers and weight gain after cessation (Boross, 1991). Numerous cross-sectional studies have indicated that body mass index (BMI) is lower in cigarette smokers than in nonsmokers (Williamson et al., 1991; Flegal et al., 1995), and that leanness correlated directly with duration, but not intensity of smoking, with longer duration associated with lower BMI (Albanes et al., 1987; Ward et al., 2001).

The glutathione peroxidase enzyme activity was increased in a dose dependent manner of the treated group compared to the control group. This finding was similar to another study which showed that after 21 days of smoke exposure, the activities of glutathione peroxidase, glutathione reductase, and glucose-6-phosphate dehydrogenase were increased over control values (York et al., 1976). In another study, the activities of two principal antioxidant enzymes SOD and glutathione peroxidase as well as plasma levels of MDA were significantly increased compared to non smokers (Ozguner, 2005). Meanwhile, a significant decrease in the levels

of glutathione and glutathione peroxidase activity was found in lens, but not in blood of smokers when compared with nonsmokers (Sulochana et al., 2002). A study conducted by Bahri and colleagues showed a significant increase in GPx activities of smoking mothers and their infants, suggesting that they may have been exposed to more oxidant stress (Ermis et al., 2004).

The malondialdehyde enzyme showed a dose-dependent reduction in enzyme activity compared to the control group in this study. Systemic and local MDA levels are increased by smoking in addition to the impact of periodontitis according to Tonguc et al. (2011). However, another study showed that MDA levels negatively correlated with paroxynase 1 and glutathione peroxidase activities, whereas they positively correlated with glutathione reductase activities (Solak et al., 2005), which agrees with the finding in this study.

The histology showed a progressive reduction in the staining intensity for nucleic acids and increase in the vacuolations in the treated group compared to the control group, which might be a pointer to some forms of structural damage to neuronal components in the frontal cortex as a result of long term exposure to cigarette smoke.

Conclusion

The current study showed that consumption of cigarette smoking results in dose-dependent increase in oxidative stress in the frontal cortex, which might be responsible for some of the histological changes; and consequently, some forms of neurological dysfunction.

REFERENCES

Adeniyi PA, Ghazal KO, Enaibe BU, Jimoh OR, David JA, Adefolaju GA, Oyewopo AO, Caxton-Martins EA. 2010. The Cytoarchitectural alterations in the neocortex of Wistar rats: Effects of aqueous tobacco (*Nicotiana tabacum*) leaves extract exposure. *Afr. J. Biotechnol.*, **9**(44): 7539-7543.

Aghaji M. 2008. Cigarette smoking and quitting among young adults in Enugu, Nigeria. *Nig. Med. J*., **49**: 27-30.

Albanes D, Jones DY, Micozzi MS, Mattson ME. 1987. Associations between smoking and body weight in the U.S. population: Analysis of NHANES II. *Am. J. Public Health*, **77**(4): 439-444.

Boross M, Pénzes L, Izsák J, Rajczy K, Beregi E. 1991. Effect of smoking on different biological parameters in aging mice. *Z. Gerontol*., **2**: 76-80.

Chen H, Vlahos R, Bozinovski S, Jones J, Anderson GP, Morris MJ. 2005. Effect of short-term cigarette smoke exposure on body weight, appetite and brain neuropeptide Y in mice. *Neuropsychopharmacology*, **4**: 713-719.

Davis RM. 1997. Passive smoking-History repeats itself. *BMJ*, **15**(7114): 961-962.

Doll R, Peto R, Borehan J, Sutherland T. 2005. Mortality from cancer in relation to smoking; 50 Years Observations on male British doctors. *Br. J. Cancer*, **92**(3): 426-429.

Ermis B, Ors R, Yildirim A, Tastekin A, Kardas F, Akcay F. 2004. Influence of smoking on maternal and neonatal serum malondialdehyde, superoxide dismutase, and glutathione peroxidase levels. *Ann. Clin. Lab. Sci*., **34**(4): 405-409.

Fix JD. 2000. Cerebral Cortex. In *High Yield Neuroanatomy* (2nd edn). Lippincott Williams & Wilkins: Philadelphia; 121-128.

Flegal KM, Troiano RP, Pamuk ER, Kuczmarski RJ, Campbell SM. 1995. The influence of smoking cessation on the prevalence of overweight in the United States. *N. Engl. J. Med.*, **333**(18): 1165-1170.

Wickstrom R. 2007. Effects of nicotine during pregnancy: Human and Experimental Evidence**.** *Curr. Neuropharmacol*., **5**(3): 213–222.

Ozguner F. 2005. Active smoking causes oxidative stress and decreases blood melatonin levels. *Toxicol. Ind. Health*, **21**(10): 21-26.

Penton R and Lester R. 2009. Cellular event in nicotine addiction. *Semin. Cell Dev. Biol.*, **20**(4): 418-431.

Raw M, McNeill A, West R. 1998. Smoking cessation guidelines for health professionals. *Thorax*, **53**(5): 1-19.

Silver RA, Boudreaux ED, Woodruff PG, Clark S. 2003. Cigarette smoking among asthmatic adults presenting to 64 emergency Departments. *Chest*, **123**(5): 1472-1479.

Solak ZA, Kabaroğlu C, Cok G, Parıldar Z, Bayındır U, Ozmen P, Bayındır O. 2005. Effect of different levels of cigarette smoking on lipid peroxidation, glutathione enzymes and paraoxonase 1 activity in healthy people. *Clin. Exp. Med*., **5**(3): 99-105.

Sulochana KN, Punitham R, Ramakrishnan S. 2002. Effect of cigarette smoking on cataract: antioxidant enzymes and constituent minerals in the lens and blood of humans. *Indian J. Pharmacol.*, **34**: 428-431.

Tonguç MÖ, Öztürk O, Sütçü R, Ceyhan BM, Kılınç G, Sönmez Y, Yetkin Ay Z, Sahin U, Baltacıoğlu E, Kırzıoğlu FY. 2011. The impact of smoking status on antioxidant enzyme activity and malondialdehyde levels in chronic periodontitis. *J. Periodontol.*, **82**(9): 1320-1328.

Ward KD, Klesges RC, Vander Weg MW. 2001. Cessation of smoking and body weight. In *International Textbook of Obesity*, Björntop P (ed). Wiley & Sons Ltd: Chichester; 323-336.

Williamson DF, Madans J, Anda RF, Kleinman JC, Giovino GA, Byers T. 1991. Smoking cessation and severity of weight gain in a national cohort. *N. Engl. J. Med.*, **324**: 739-745.

York GK, Peirce TH, Schwartz LW, Cross CE. 1976. Stimulation by cigarette smoke of glutathione peroxidase system enzyme activities in rat lung. *Arch. Environ. Health*, **31**(6): 286-290.