Greener Journal of Medical Sciences

ISSN: 2276-7797

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Research Article

Evaluation of Haemorheological Parameters in Cigarette Smokers in Western Nigeria

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ABSTRACT

Tobacco use is widely spread throughout the world, from countries with low income economics to the most influential industrialized nations. The effect of smoking on human health are serious and in many cases, deadly. This study aimed at estimating the haemorheological parameters and examining the effects of this social behavior on them with a view to educate the smokers and the society. This study was carried out among the regular smokers that smoke not less than two sticks of cigarettes per day. Ninety (90) subjects were studied, comprising of sixty (60) smokers subjects, willingly volunteered to participate in the study and thirty (30) nonsmoker subjects that served as control for the study. The haemorheological parameters determined were packed cell volume (PCV), Erythrocyte sedimentation rate (ESR), Fibrinogen concentration (FIBC), Whole blood viscosity (WBV) and Plasma viscosity (PV). It was observed that there were statistically significant difference in WBC, ESR, FIB, and PCV ($p < 0.05$, $p < 0.01$, $p < 0.01$ and $p < 0.05$). Though experimental differences were noticed in PLT, WBV and PCV when the results of smokers were compared with the non smokers, the values were not statistically significant ($p > 0.05$). In the correlation results, WBC correlate negatively with PLT and PV ($r = -0.142$, $r = -0.042$), PLT correlate negatively with ESR and PCV ($r = -0.124$, $r = -0.172$), and ESR correlate negatively with WBV ($r = -0.065$). The correlation between FIBC and WBV, PV, ($r = -0.304$, $r = -0.077$), WBV and PV ($r = -0.333$) were negative and all these were not statistically significant ($p > 0.05$). The negative correlation that exist between the PV and PCV ($r = -0.846$) was statistically significant (< 0.01). The positive correlation noticed and statistically significant were between WBC and ESR ($r = 0.341$) (< 0.05), also between PLT and FIBC ($r = 0.477$) (< 0.01).

Conclusion: Smoking exerts negative influence on the haemorheological parameters and these are the contributing factors that lead to cardiovascular and atherosclerosis that usually occur in cigarette smokers and cause of their death. Also smokers are more prone to infection more than nonsmokers because of their low level of immunity.

Key Words: haemorheological, Fibrinogen, Platelets, White blood cells, Whole blood viscosity, Plasma viscosity.

INTRODUCTION

Tobacco smoking is one of the most potent and prevalent addictive, influencing behavior of human beings for over four centuries. Smoking is now increasing rapidly throughout the developing world and is one of the biggest threats to current and future health (Edward, 2004). Tobacco continues to be the second major cause of death in the world. By 2030, if current trends continue, smoking will kill over 9million people annually (World health report, 2002).

Smoking is an important preventable cause of mortality worldwide. The prevalence of pulmonary and cardiovascular disease, cataracts and some cancers is higher in smokers. Tobacco smoke contains some deadly carcinogenic chemicals formed from natural components of the tobacco plants and leads to the uptake of many hazardous compounds such as heavy metals and N-nitrosocompound (Wesley *et al.*, 2010). Apart from the natural constituents of tobacco, many substances are added to cigarette by manufacturers to enhance the flavor or to make smoking more pleasant. Some of the compounds found in tobacco smoke include ammonia, tar and carbon monoxide. Exactly what effects these substance have on the cigarettes smoker health is unknown, but there is no evidence that lowering the tar content of cigarette lower the health risk (U.S.FDA, 2011).

More than 3040 chemicals have been isolated from processed tobacco (Roberts, 1988); most are leaf constituents, but some arise from growing conditions such as the soil and atmosphere in an area, while others

originate from the use of agricultural chemicals from casings, humectants and flavorings added to the leaves and from curing methods. When tobacco is burn in the course of smoking, many pyrolysis and other reaction products are formed (WHO, 1999).

Tobacco smoking affects humoral and cellular immunity in humans, but the magnitude of the changes vary widely among studies. In humans, cigarette smoking has marked effect on alveolar macrophages morphology and physiology; it decrease serum immunoglobulins (IgA, IgG, IgM) but increases IgE, and has a range of effects on B and T lymphocytes (Spori *et al.*, 1994).

Base on the data collected from 1995 to 1999, the CDC estimated that adult male smokers lost an average of 13.2years of life and female smokers lost 14.5years of life because of smoking (CDC, 2002). Smoking in any form (Cigarette, Cigars, Pipe) is a strong independent risk factor for atherosclerosis, acute myocardial infarction, stable angina, stroke and sudden death. It markedly increases the risk of other cancers, particularly those of the oral cavity larynx, oesophagus, kidney, bladder, uterine cervix and pancreas. Cigarette smoking is the principal cause of chronic bronchitis and emphysema. Smoking during pregnancy increases baby low birth weight and results in other parental complications that are particularly hazardous to woman. The world health epidemiologist predicted that unless there is a dramatic change in present trends, tobacco smoking will be killing more than 9 million people yearly by the late 2020 (World health report, 2002). There are research publications on the effect of smoking on lung cancer risk (Doll and Hill, 1950; Wynder and Graham, 1950), relatively few people were aware that smoking caused serious illness.

MATERIAS AND METHODS

Subject Selection

This work was carried out among the regular tobacco smokers who smoke nothing less than two sticks of cigarette per day and samples were analyzed at the Department of Haematology, Obafemi Awolowo University Teaching Hospital Complex, Ile – Ife. Blood samples were collected from ninety (n=90) individual who gave consent of participation, Sixty (n=60) regular smokers and thirty (n=30) non smokers for haemorheological investigation. The venous blood was collected and used for all haemorheological investigations.

Specimen collection and Laboratory analysis

The blood sample was collected under aseptic conditions. Nine milliliters (9ml) of blood was collected from cubital vein by venepuncture into 0.5ml of 3.8% sodium citrate in a plastic tube and commercially prepared Ethylene Diamine Tetra acetic acid (EDTA) plastic tube. The blood collected into sodium citrate plastic tube was centrifuged immediately at 2500g for 15minutes and the plasma separated and stored into stopper tubes and used within 4 hours of collection for fibrinogen concentration (FIBC). The EDTA blood sample was used for PCV, WBV, PV, PLT and WBC. Stasis was avoided during blood collection to prevent activation of clotting factors. The Erythrocyte Sedimentation Rate (ESR) was estimated by Westengreen method, while the Packed Cell Volume was observed by standard method. Relative whole blood viscosity and plasma viscosity were observed by Ingram method (Ingrm, 1961). A simple technique and standardized method for fibrinogen concentration was done (Reid and Ugwu, 1987). Values obtained from each group were compared.

Results

During the period of this study, ninety (90) subjects were recruited, sixty (60) smokers and thirty (30) non smokers that served as controls, all within Ife Township in Ife East Local Government area of Osun State. All of them gave consent of participation. Table 1 shows the haemorheological profiles in smokers and non smokers.

Table 1: Haemorheological profiles in smokers and non- smokers (Mean±SEM)

Parameters	Smokers	Non Smoker	P-Value
FIBC(g/L)	5.70±0.10	3.13±0.01	<0.01
WBV(mPa.s)	5.44±0.09	5.28±0.14	>0.05
PV(mPa.s)	1.74±0.02	1.52±0.04	<0.01

Smokers n = 60; Control n = 30

All values in Mean $\pm$ SEM,

Key FIBC: Fibrinogen concentration, WBV: Whole blood viscosity, PV: Plasma Viscosity.

Statistically significant differences were found in FIBC ($p < 0.01$) and PV ($p < 0.01$). The differences seen in the WBV was not statistically significant. Some haematological parameters in both smokers and nonsmokers were shown in Table 2.

Table 2: Some haematological parameters of smokers and non- smokers

Parameters	Smokers	Non Smoker	P-Value
WBC ($\times 10^9/L$)	5.59 $\pm$ 0.56	3.91 $\pm$ 0.02	<0.05
PLT($\times 10^9/L$)	182.09 $\pm$ 9.54	197.66 $\pm$ 14.69	>0.05
ESR(mm/hr)	36.50 $\pm$ 5.16	11.33 $\pm$ 1.42	<0.01
PCV(%)	42.90 $\pm$ 0.50	41.73 $\pm$ 0.92	>0.05

Smokers n = 60; Control n = 30

All values in Mean $\pm$ SEM,

Key WBC: White Blood cell Count, PLT: Platelet, ESR: Erythrocyte Sedimentation Rate, PCV: Packed Cell Volume.

Statistically significant difference exist between the WBC and ESR of smokers and non smokers and the p-values were (< 0.05 and < 0.01). Table 3 shows the Pearson of correlation of all the parameters in smokers. WBC correlate negatively with PLT and PV ($r = -0.142$, $r = -0.042$), PLT correlate negatively with ESR and PCV ($r = -0.124$, $r = -0.172$), and ESR correlate negatively with WBV ($r = -0.065$). The correlation between FIBC and WBV, PV, ($r = -0.304$, $r = -0.077$), WBV and PV ($r = -0.333$) were negative and all these were not statistically significant ($p > 0.05$). The negative correlation that exist between the PV and PCV ($r = -0.846$) was statistically significant (< 0.01). The positive correlation noticed and statistically significant were between WBC and ESR ($r = 0.341$) (< 0.05), also between PLT and FIBC ($r = 0.477$) (< 0.01).

Table 3: Pearson Correlation of parameters of smokers.

	WBC	PLATELET	ESR	FIB	WBV	PV	PCV
WBC($\times 10^9/L$)	1	-.142	.341*	.060	.057	-.042	.153
PLT($\times 10^9/L$)	-.142	1	-.124	.477**	.009	-.038	-.172
ESR(mm/hr)	.341*	-.124	1	.064	-.065	.064	.091
FIB(mPa.s)	.060	.477**	.064	1	-.304	-.077	.013
WBV(mPa.s)	.057	.009	-.065	-.304	1	-.333	.123
PV(mPa.s)	-.042	.038	.064	-.077	-.333	1	-.846**
PCV(%)	.153	-.172	.091	.013	.123	-.846**	1

*Correlation is significant at the 0.05level

** Correlation is significant at the 0.01level

DISCUSSION

The effects of smoking on human health are serious and in many cases deadly. There are approximately about 4,000 chemicals in cigarette, hundreds of which are toxic. The ingredients in cigarettes affect everything from the internal functioning of organ to the efficiency of the body immune system.

Blood viscosity is an important determinant of rate of blood flow and the greater the viscosity, the less the flow in a vessel, if all other factors are constant. Furthermore, the viscosity of normal blood is about three times as great as that of water. What makes blood so viscous is mainly the large number of suspended red cells in the blood, each of which exerts frictional tray against adjacent cells and against the wall of the blood vessel (Guyton and Hall, 2006). This was reflected in this work, the whole blood viscosity (WBV) and Packed Cell Volume (PCV) were not statistically significant but experimental differences were noticed in the results. The whole blood viscosity of smokers

was slightly higher than that of non-smokers and this was in agreement with the previous work done (Haustein *et al.*, 2004). Smoking is associated with an increase in haematocrit (PCV) or red blood cell mass, this was also noticed in this work and the increased may be attributable to increased level of carbon monoxide (CO) and carboxyhaemoglobin. Haematocrit decreases with smoking cessation, but increased blood viscosity and deformability of red cells may persist (Haustein *et al.*, 2004). The report of Burtis and Ashwood also showed that differences observed may be due to the increased level of carboxyhaemoglobin in smokers, thereby creating continuous state of hypoxia (Burtis and Ashwood, 2001). Any condition that causes the quantity of oxygen transport to the tissue to decrease ordinarily increases the rate of red cell production by stimulating erythropoietin secretion (Guyton and Hall, 2006). The increased number of red cells compensates for impaired ability of red cells to transport oxygen. The blood erythrocyte count is therefore increase in smokers (Burtis and Ashwood, 2001).

The white blood cells are associated with the effective functioning of the immune system of the body and when the number or the functions of these white blood cells are affected, the immune system is compromised. Leucocytosis (increase level of leucocytes) is an indication of infection depending on which line of the leucocytes is affected. Leucocytes are essential element of inflammation process (Frohlich *et al.*, 2003), an independent predictor of coronary heart disease in smokers (Brown *et al.*, 2001). The result of this work showed statistically significant increase in white blood cells of smokers compared to non- smokers, and this was in line with the previous work done (Burtis and Ashwood, 2001). The increased level of white blood cells observed in the smokers might be due to infection, since cigarette smoking has marked effect on alveolar macrophages morphology and physiology; it decreases serum immunoglobulin (IgA, IgG, IgM, but increase IgE) and has a range of effects on B and T lymphocytes (Spori *et al.*, 1994). The previous work done also revealed that cigarette smoking impaired human lymphocytes function by inhibit transglutamine, and also expatiated that smoking induces a CD8-lymphocyte infiltrate in pulmonary arteries, which was associated with impairment of the vessel's structure and function (Berntorp *et al.*, 1989). An acute or chronic inflammatory response induced by particulates of cigarette smoke may also be another possible mechanism responsible for increased leucocytes count in smokers (Frohlich *et al.*, 2003). In this present study, increased leucocytes count among smokers is of a magnitude similar to the above mentioned which might forecast the possible high risk for developing fatal disease in the smokers. Research indicates that inflammation contribute to atherogenesis, because high leucocyte counts and high level of CRP and fibrinogen are all powerful predictor of future cardiovascular events (Libby *et al.*, 2002). Positive correlation exists between the white blood cells of smokers and the erythrocyte sedimentation rate. Both leucocytosis and increased erythrocyte sedimentation rate imply inflammation reaction or an infection within the vascular system of smokers.

Erythrocyte sedimentation rate (ESR) though is a non specific test but is a pointer to the presence of an inflammation response to disease in the body. Tobacco use leads most commonly to diseases affecting the heart and lungs. Other organs affected include cervix uteri, urinary bladder, kidney, etc and causes respiratory diseases such as pneumonia, influenza, Bronchitis, emphysema, chronic airway obstruction, etc (Sugeon General's Report (USDHHS, 2004). The result of this work confirm the previous work done, a statistically significant difference was noticed in the ESR of smokers compared to non- smokers. This indicates that there is an inflation reaction taking place in the system of smokers and this alters the albumin –globulin ratio. Inflammations enhance the increase formation of globulin at the expense of albumin which eventually enhances rouleaux formation. When the rouleaux formation of red blood cells increased, the rate of settling down of red blood cells are faster and this leads to increased ESR. Report has it that erythrocyte sedimentation rate also carries strong prognostic information in men with known or suspected coronary heart disease and, since an increasing erythrocyte sedimentation rate was associated with a particularly steep gradient in the percentages of men dying from coronary heart disease without prior myocardial infarction, it is hypothesized that a high erythrocyte sedimentation rate may be an indicator of aggressive, malignant forms of coronary heart disease, conceivably by being a marker of activated humoral immune mechanisms in widespread atheromatous tissues.(Erikssen *et al.*, 2000).

Plasma fibrinogen is an independent predictor of cardiovascular disease including coronary heart disease (CHD) (Maresca *et al.*, 1999). The results of this work revealed a statistically significant high values in the fibrinogen concentration of smokers compare to non- smokers and this confirm the previous work on smokers (Awodu and Famodu, 2007; Awodu *et al.*, 2005). The higher level value recorded in the plasma fibrinogen concentration might be due to the response of fibrinogen to the vascular inflammation which usually accompany cigarette smoking. Studies had indicated that circulating levels of fibrinogen increase in smokers and decrease with smoking cessation (Hunter *et al.*, 2001; Tuut and Hense, 2001). Also research suggests that elevated fibrinogen values are an independent risk factor for coronary heart disease (Paramo *et al.*, 2004) and deep-vein thrombosis (Vayá *et al.*, 2002). The effect of fibrinogen on CVD is partly attributable to smoking and seems to be mediated through alterations in rates of synthesis by the liver. To date, several prospective studies have convincingly documented an association between fibrinogen and coronary heart disease (Ernst and Resch, 1993).

The plasma viscosity directly determines blood flows in the micro circulation level (Lowe, 1986) and plasma hyperviscosity results in a deterioration of micro-circulatory blood flow, which theoretically limits tissue perfusion, particularly in poststenotic areas with low shear forces. The result of this study was in line with the previous work done (Awodu *et al.*, 2005) where high level of plasma viscosity was recorded in smokers compare to non-smokers. Plasma viscosity, as well as fibrinogen and white blood cell count, was positively associated with the incidence of coronary heart disease (CHD) events in a population based study of middle aged men. Plasma viscosity and fibrinogen were also associated with incidence of CHD and stroke in a study of older men and women (Lowe *et al.*, 1997).

Platelet is one of the cellular components of blood and perform a number of important functions; maintaining the integrity of vascular endothelium, performing the primary haemostatic plug following vessel injury, activation of blood coagulation system, producing mediators involved in vessel wall repair and regulation of vascular toxicity, as well as inflammatory reaction and producing growth factors (Stiene-Martin *et al.*, 1998). Literature reports on the effect of smoking on platelet count seem to be controversial. Brumit and Baker found no correlation between platelet count and smoking in healthy volunteers (Brumit and Baker, 2000). Also Dotevall *et al.* noted no changes in platelet count in female smokers and non-smokers (Dotevall *et al.*, 1992), and Suwansaksri *et al.* observed no alteration in platelet in smokers and non-smokers (Suwansaksri *et al.*, 2004). According to Blann *et al.*, smoking two cigarettes a day by chronic smokers of both sexes does not affect the platelet count (Blann *et al.*, 1998). The result of this work showed a reduction in the platelet number of smokers compare to non-smokers, and this was not in line with the previous work done. The number of platelet counted was still within the normal range, researchers related the number of newly formed reticulated platelets, rather than absolute count, to incidence of thrombotic events (Rinder *et al.*, 1998).

CONCLUSION

Smoking exerts negative influence on the haemorheological parameters and these are the contributing factors that lead to cardiovascular and atherosclerosis that usually occur in cigarette smokers and cause of their death. Also, this work confirms that smokers are more prone to infection than non-smokers, because of their low level of immunity.

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