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Evaluation of effect of smoking and hypertension on serum lipid profile and oxidative stress

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ABSTRACT

Objective: To determine the effect of smoking, hypertension individually on lipid profile and lipid peroxidation and the cumulative influence of smoking and hypertension on oxidative stress and lipid profile. **Methods:** Serum total cholesterol, high density lipoprotein (HDL), low density lipoprotein (LDL), very low density lipoprotein (VLDL), triglycerides and malondialdehyde (MDA) were estimated in sixty cases including twenty smokers, twenty hypertensives, and twenty smokers with hypertension and compared with those in twenty age and sex matched healthy controls. **Results:** Statistically significant increase in MDA, total cholesterol, LDL, VLDL and triglycerides and decrease in HDL in cases were observed in smokers, hypertensives and smokers with hypertension when compared to healthy controls. Smokers had significantly elevated levels of lipid profile and MDA except for HDL when compared to hypertensive group. Statistically significant increase in the levels of study parameters of smoking and hypertensive group was noticed when compared to group with hypertensives ($P < 0.05$) and there was a statistically significant decrease in HDL levels in smoking and hypertensive group when compared to healthy controls. All the biochemical study parameters had larger effects ($0.80 < d < 1.20$) for the smoking and hypertensive group in comparison with control group. **Conclusions:** Cigarette smoking, together with hypertension, has larger effect on lipid profile than in patients with cigarette smoking or hypertension alone and induces alteration in serum lipid levels and oxidative stress in the direction of increased risk for coronary artery disease.

1. Introduction

Cigarette smoking is acknowledged as one of the leading causes of preventable morbidity and mortality, and is one of the largest preventable causes of ill health in the world. Cigarette smoking is said to be responsible for 17%–30% of all deaths from cardiovascular illness. The effects of cigarette smoking are dose-related and life style modification measures involving quitting smoking are probably the single most important steps to decrease the chance of coronary artery disease and a heart attack[1].

Dyslipidemia, a strong predictor of cardiovascular disease, which causes endothelial damage and the loss of physiological vasomotor activity that results from endothelial damage may become manifested as blood

pressure increases. Therefore, factors like dyslipidemia that cause endothelial dysfunction may lead to hypertension[2]. Plasma lipoprotein abnormalities are said to be the underlying major risk factors and may even be essential for the common occurrence of atherosclerotic vascular diseases[3]. Free radical mediated lipid peroxidation has been associated with the pathogenesis of many diseases and clinical conditions. Oxidative pathway appears to be one important mechanism for modifying low density lipoprotein (LDL), because a wide variety of structurally unrelated antioxidants inhibit atherosclerosis. Lipid peroxidation is one of the pathological processes impairing the function of arterial endothelial cells. Oxidative damage to unsaturated lipids is a well established, general mechanism for oxidant mediated cellular injury. Oxidative stress appears to be a probable clinically relevant factor in cigarette smoke-related atherogenesis and increased lipid peroxidation is also a risk factor for myocardial infarction[4].

Hypertension is a risk factor for the development of atherosclerosis. There is increasing evidence that atherosclerosis should be viewed fundamentally as an inflammatory disease. Atherogenic stimuli such as

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hyperlipidemia appear to activate the inflammatory response by causing expression of mononuclear leukocyte recruiting mechanisms. Hypertension not only is a well-established cardiovascular risk factor but also increases the risk of atherosclerosis. Both of hypertension and dyslipidemia are independent risk factors for the development of atherosclerosis[5]. Though individually these two factors are known to have adverse effects on lipid profile and lipid peroxidation, there are limited studies to substantiate their cumulative influence on oxidative stress and lipid profile. Hence, the present study was aimed to find the collective effects of cigarette smoking and hypertension on lipid profile and lipid peroxidation.

2. Materials and methods

Eighty subjects were included in this study, out of which twenty healthy subjects were considered as group I who were nonsmokers and nonhypertensives, aged between 35 to 55 years as controls and whereas cases were age and sex matched sixty subjects who were enrolled from the Outpatient Department of Medicine. These sixty subjects were divided into 3 groups. The criteria for chronic smokers were taken as those who smoked more than 10 cigarettes per day for more than 10 years (10 pack years)[6]. The criteria for hypertension were taken as those who were having average blood pressure of 140/90 for a period of more than 10 years[6]. Twenty chronic smokers were included in group II. Hypertensives were included in group III but are not smokers. Twenty chronic smokers with high blood pressure were included in group IV. Patients with renal disorders, diabetes mellitus, liver disorders and other chronic infections and those who were on antioxidants or vasoactive medications were excluded from the study. Informed consent was taken from each patient and the study was approved by Institutional Ethical Committee.

A volume of 5 mL of fasting venous blood sample was

collected with aseptic precautions. Blood samples were allowed for clotting. Serum was then separated and analyzed for lipid profile and malondialdehyde (MDA) levels. Serum total cholesterol (TC) was estimated by the method of Zak. high density lipoprotein cholesterol (HDL-C) was estimated by the precipitation method. Serum triglyceride (TG) was estimated colorimetrically using the Hantzsch reaction[7]. Very low density lipoprotein cholesterol (VLDL-C) was calculated by using the formula of Rifai and Warnick[8]. Measurement of by-product of lipid peroxidation, MDA was done by thiobarbituric acid method[9].

The statistical software SPSS 11.0 was used for the analysis of the data and Microsoft Word and Excel have been used to generate tables. The analysis of variance (ANOVA) has been used to find the significance of mean of lipid profile and MDA between group I, group II, group III and group IV. $P < 0.05$ was considered as statistically significant.

3. Results

Statistically significant increase was found in serum TC, TG, VLDL and LDL-C of sixty cases together and each group of cases and serum parameters were compared with those in control group ($P < 0.001$ and $P < 0.05$) except for MDA levels between group III and group I. Statistically significant decrease in HDL in each group of cases was observed when compared to controls. When the levels of all the parameters in this study were compared between smokers (group II) and hypertensives (group III), statistically significant higher elevated levels of lipid profile were found in chronic smokers than hypertensive individuals ($P < 0.05$) except for HDL. Statistically significant greater increase in the levels of study parameters of group IV subjects with risk factors of smoking and hypertension was noticed when compared to group III with hypertensives ($P < 0.05$) except for HDL (Table 1). All the lipid parameters and MDA had larger effects ($0.80 < d < 1.20$) for group IV in comparison with control group.

Table 1

Mean pattern and comparison of lipid profile and MDA in cases and controls (mean±SD).

Parameters	Group I	Group II	Group III	Group IV	F value
TC	167.05±12.43	225.20±18.11 ^{a***}	215.50±8.87 ^{a***}	244.50±19.59 ^{abc***}	91.910
TG	123.35±16.48	218.00±15.42 ^{a***}	203.00±8.64 ^{ab***}	221.50±14.24 ^{ac***}	216.007
HDL	45.85±3.86	34.95±1.39 ^{a***}	37.00±1.52 ^{ab***}	35.00±2.29 ^{ac***}	87.907
LDL	100.35±17.20	147.35±17.50 ^{a***}	125.40±7.14 ^{ab***}	142.40±11.65 ^{ac***}	44.530
VLDL	24.17±3.20	42.90±2.49 ^{a***}	40.60±1.73 ^{ab***}	44.30±2.85 ^{ac***}	253.215
MDA	2.37±0.03	2.46±0.06 ^{a***}	2.38±0.03 ^{b***}	2.49±0.03 ^{ac***}	49.749

^a: $P < 0.05$ as compared with group I; ^b: $P < 0.05$ as compared with group II; ^c: $P < 0.05$ as compared with group III; ^{***}: $P < 0.001$ as compared with group I.

4. Discussion

Atherosclerosis is the underlying process involved in coronary artery disease, peripheral vascular disease and stroke[10]. Smoking causes a huge and increasing number of untimely deaths in India[11]. There are currently 240 million tobacco users aged 15 years and above (195 million male users and 45 million female users) in India. The prevalence of tobacco use is higher in rural population than that in urban areas[12].

The biological mechanisms linking smoking and atherogenesis are complex and not fully understood. In addition to inflammation, potential mechanisms by which smoking increases the risk of cardiovascular diseases include systemic haemostatic and coagulatory disturbances, lipid abnormalities, increase in oxidative stress, and vascular endothelial dysfunction[13]. Cigarette smoking leads to increase in the concentration of serum TC, LDL-C, TG, VLDL-C, and fall in the levels of anti-atherogenic HDL-C[14]. It is presumed that nicotine stimulates sympathetic adrenal system leading to increased secretion of catecholamines resulting in increased lipolysis and

increased concentration of plasma free fattyacids, which further results in increased synthesis of hepatic TG, along with VLDL-C in the blood stream^[15]. Cigarette smoking is associated with higher androstenedione levels, a higher total androgen to total estrogen ratio, and lower progesterone levels^[16].

High levels of LDL-C, VLDL-C, and TG are strongly associated with the development of coronary artery disease, while a low level of HDL-C remains a significant independent predictor of coronary artery disease^[17].

Hypertension is one of the major risk factors for coronary artery disease and stroke. Hypertension is an independent risk factor for atherosclerosis. It is well known that hypertension is associated with abnormal changes in lipid profile (dyslipidemia) which is a cause for atherosclerosis^[18]. Cigarette smoking and hypertension individually potentiates lipid abnormalities but the combined effect has to be studied.

Tobacco smoke contains large numbers of free radicals that are capable of initiating or promoting oxidative injury. Cigarette smokers have higher lipid peroxidation products in their blood when compared to nonsmokers and smoking increases the concentration of serum MDA levels^[14]. It is suggested that oxidative injury may play a major role in mediating the health risks associated with cigarette smoking. Oxidation of LDL might be an important mechanism whereby cigarette smoking can accelerate atherogenesis. LDL from smokers was more susceptible to peroxidative modification when compared to that from non-smokers^[14]. There is evidence to suggest that oxidatively modified LDL may contribute to the pathogenesis of atherosclerosis^[19].

Both cigarette smoking and hypertension are proven independent risk factors for atherosclerosis; both are associated with abnormalities in lipid profile and also show high values of serum MDA indicating increased rate of lipid peroxidation which suggests involvement of high rate of oxidative stress. Though theoretically we can assume that there is a cumulative effect of both cigarette smoking and hypertension on these cardiovascular risk factors. From this study, it is well established that in both cigarette smoking and hypertension there is dyslipidemia and increased oxidative stress. The constellation of these altered lipoproteins along with lipid peroxides suggests that smokers with hypertension are at a high risk for the development of coronary heart disease.

There is a cumulative effect of cigarette smoking and hypertension on lipid profile and lipid peroxidation, indicated by increased levels of serum MDA and dyslipidemia indicated by higher values of TC, TG, LDL, VLDL, and lower levels of cardio protective HDL. This observation suggests that there is a more risk of progression of atherosclerosis in individuals who have both the risk factors *i.e.* smoking and hypertension. Further large population studies are required to substantiate our findings.

Conflict of interest statement

We declare that we have no conflict of interest.

References

- [1] Yathish TR, Manjula CG, Deshpande SR, Gayathree L. A study on the association of coronary artery disease and smoking by a questionnaire method. *J Clin Diagn Res* 2011; **5**(2): 264–268.
- [2] Halperin RO, Howard D, Sesso HD, Jing Ma, Buring JE, Stampfer MJ, et al. Dyslipidemia and the risk of incident hypertension in men. *Hypertension* 2006; **47**: 45–50.
- [3] Steinberg D. The LDL modification hypothesis of atherogenesis: an update. *J Lipid Res* 2009; **50**(Suppl): S376–S381.
- [4] Venkatesan A, Hemalatha A, Bobby Z, Selvaraj N, Sathiyapriya V. Effect of smoking on lipid profile and lipid peroxidation in normal subjects. *Indian J Physiol Pharmacol* 2006; **50**(3): 273–278.
- [5] Janjigian YY, McDonnell K, Kris MG, Shen R, Sima CS, Rizvi NA, et al. Pack years of cigarette smoking as a predictor of survival in 2013 patients with stage IIIb/IV non-small cell lung cancer. *J Clin Oncol* 2008; **26**.
- [6] European Society of Hypertension–European Society. Cardiology guidelines for the management of arterial hypertension. *J Hypertens* 2003; **21**: 1011–1053.
- [7] Gowenlock AH, McMurray JR, McLauchlan DM. *Varley's practical clinical biochemistry*. 4th ed. New Delhi: CBS Publishers; 1988, p. 312–316.
- [8] Rifai N, Warnick GR. Lipids, lipoproteins, apolipoproteins and other cardiovascular risk factors. In: Burtis CA, Ashwood ER, Bruns DE. (eds.) *Tietz textbook of clinical chemistry and molecular diagnostics*. 4th ed. Missouri: Saunders; 2006, p. 903–981.
- [9] Buege JA, Aust SD. Microsomal lipid peroxidation. *Methods Enzymol* 1978; **52**: 302–310.
- [10] Romero JR, Pikula A. Carotid artery disease: current concepts on endothelial dysfunction and matrix remodeling. *Curr Drug Ther* 2009; **4**: 202–213.
- [11] Jha P, Jacob B, Gajalakshmi V, Gupta PC, Dhingra N, Kumar R, et al. A nationally representative case-control study of smoking and death in India. *N Engl J Med* 2008; **358**: 1137–1147.
- [12] Reddy KS, Gupta PC. *Report on tobacco control in India*. New Delhi: Ministry of Health and Family Welfare, Government of India; Centers for Disease Control and Prevention, USA; World Health Organization; 2004.
- [13] Yanbaeva DG, Dentener MA, Creutzberg EC, Wesseling G, Wouters EFM. Systemic effects of smoking. *Chest* 2007; **131**: 1557–1566.
- [14] Pasupathi P, Rao YY, Farook J, Saravanan G, Bakthavathsalam G. Effect of smoking on lipids and oxidative stress biomarkers in patients with acute myocardial infarction. *Res J Med Med Sci* 2009; **4**: 151–159.
- [15] Benowitz HL. Pharmacologic aspects of cigarette smoking and nicotine addiction. *N Engl J Med* 1998; **319**: 318–320.
- [16] Cochran CJ, Gallicchio L, Miller SR, Zacur H, Flaws JA. Cigarette smoking, androgen levels, and hot flushes in midlife women. *Obstet Gynecol*. 2008; **112**(5): 1037–1044.
- [17] Waqar A. Effect of tobacco smoking on the lipid profile of teenage male population in Lahore City. *Int J Med Med Sci* 2010; **2**(6): 172–177.
- [18] Halperin RO, Sesso HD, Jing Ma, Buring JE, Stampfer MJ, Gaziano JM. Dyslipidemia and the risk of incident hypertension in men. *Hypertension* 2006; **47**: 45–50.
- [19] Barbosa KB, Volp ACP, Hermsdorff HHM, Navarro-Blasco I, Zulet MA, Martínez JA, et al. Relationship of oxidized low density lipoprotein with lipid profile and oxidative stress markers in healthy young adults: a translational study. *Lipids Health Dis* 2011; **10**: 61.