

Platelet Aggregability And C - Reactive Protein In Male Smokers

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Abstract: Background: Cigarette smoking is major risk factor for many illnesses including cardiovascular diseases. Smoking is known to have adverse effects on endothelial function. This leads to thrombotic episodes by enhancing platelet aggregation. Raised C-reactive protein can be a predictor for future risk of athero-thrombotic events. In this study pro-thrombotic activity was studied in smokers in form of platelet aggregability and serum C-reactive protein. Method: Platelet aggregability was measured by method given by O'Brien and C-reactive protein was measured by rapid latex agglutination test in male smokers consuming minimum 5 cigarettes per day for more than 3 years. Result: Platelet aggregability was found significantly increased in smokers. C-reactive protein levels were also found raised in smokers. Conclusion: Increased platelet aggregability in smokers could be due to endothelial injury and increased sympathetic activity. Increased C-reactive protein could be due to chronic inflammation in lungs and damage to endothelium.

Key words: Platelet aggregability and C-reactive protein.

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Introduction: Cigarette smoking, a major risk behaviour adversely affecting public health, has reached epidemic proportions. WHO estimates that there are about 100 million smokers in the world representing about one third of the global population aged 15 years and over. The vast majority of the smokers are in developing countries.¹The history of smoking dates back to as early as 5000–3000 BC when the agricultural product began to be cultivated in South America; consumption later evolved into burning the plant substance either by accident or with intent of exploring other means of consumption.

Smoking is generally five times higher among men than women, however the gender gap declines with younger age.²Smoking is not only associated with lung cancer but also linked to cardiovascular diseases, chronic respiratory diseases and stroke. The risk of developing cardiovascular diseases (CVD) increases with length and intensity of exposure to cigarette smoke. Overall, smokers have a 70% greater risk of mortality from CVD than non-smokers.³

Pro-thrombotic effects of smoking are well documented which include increased circulating levels of fibrinogen, endothelial dysfunction, platelet activation and platelet aggregation. Thus the adverse prognostic effects of smoking may relate not only to increased risk of developing atherosclerosis but also to increased risk of occlusive thrombosis and myocardial infarction.⁴

Recently, acute phase reactants such as C-reactive protein (CRP), serum amyloid A, IL-6, and IL-1 have been correlated with the development of athero-thrombotic events. C-reactive protein, in particular, is proving to be a strong, independent prognostic indicator for the development of future athero-thrombotic events. Platelet aggregability and CRP concentrations are known to be affected by smoking.⁵ So present study was planned to see the effects of smoking on platelet aggregability and C-reactive protein.

Material and Method: The study was a cross-sectional type of study and was conducted in 100 young males from the staff members of medical college and hospital in the age group of 40-50 Yrs. The study was carried out between August 2012 to December 2012 after obtaining permission from the institutional ethical committee.

The study groups consisted of 50 healthy males who were smoking minimum 5 cigarettes per day for more than 3 years. The control group consisted of 50 age matched non-smoker males.

Inclusion criteria for selection of subjects of study group were:- Males of 40 to 50 years of age, history of smoking minimum 5 cigarettes/day for more than 3 years & having normal body mass index (BMI). Exclusion criteria for the subjects were:- subjects

undergoing regular exercise, subjects on medications such as statins, glitazones, fibrates, niacin, clopidogrel, aspirin, iron/vitamins supplements, subjects having history of any major disease such as ischemic heart disease, kidney or liver disease, hypertension, diabetes mellitus, dyslipidemia, malignancy, venous thrombosis, systemic or pulmonary embolism, congenital hemorrhagic disease, thrombocytopenia etc.

Written consent was taken from all the subjects after explaining the nature of the study to them. Detailed medical history was taken and thorough clinical examination was carried out to rule out presence of any disease.

Fasting blood samples were collected from both the groups from antecubital vein under all aseptic precautions. In order to avoid the effect of diurnal variations on platelet aggregability and C-reactive protein, the time of collection of blood was kept constant between 9 am to 11 am. Platelet aggregability was measured by method described by O'Brien et al as it is practicable and reliable.⁶ Qualitative estimation of C - reactive protein was done by rapid latex agglutination test.⁷ In this test, if sample showed agglutination, it was considered as a positive test indicating that C-reactive protein concentration equal or greater than 6mg/L.

Results were compared by using paired 'Z' test. SPSS software version II was used for statistical analysis.

Result: Table 1 showed demographic profile of subjects involved in the present study. Our study population is anthropometrically matched.

Table 1: Demographic distribution of subjects

Parameters	Control group	Study group
	Males	Males
Age(Yrs) Mean ± SD	44.9±3.43	46.5±3.55
Height (cms) Mean ± SD	166.45±7.82	167.83±8.26
Weight (Kgs) Mean ± SD	67.21±8.95	68.71±8.26

Table 2: comparison of platelet aggregability in males in control and study group.

Parameters	Control Group (N=50)	Study Group (N=50)	p Value
Platelet Aggregability (Adsorbance) Mean ± SD	0.023± 0.15	0.087± 0.15	<0.0001*

* Statistically Significant

Table 2 shows comparison of platelet aggregability in males in control and study group. There is statistically significant increase in platelet aggregability in males of study group (p value<0.0001).

Table 3: Percentage (%) of subjects having increased C-reactive protein levels

Parameter	Control group (n=50)	Study group (n=50)
Increased CRP levels (> 6 mg/L)	8%	78%

Table 3 Shows that increased CRP levels are seen in higher Percentage (%) of subjects in study group as compared to control group.

Discussion: Cigarette smoking contributes to several diseases including cardiovascular diseases by being both prothrombotic and atherogenic. It predisposes the individual to several clinical atherosclerotic events like peripheral vascular diseases; acute coronary syndromes and stroke.⁸ The association of smoking with cardiovascular disease is nonlinear. Smoking as little as 1-4 cigarettes per day confers almost threefold higher risk of dying from coronary heart disease as compared to non-smokers.⁹

The present study was undertaken to see the effects of cigarette smoking on platelet aggregation and C-reactive protein in healthy male adults. We have found statistically

significant increase in platelet aggregability of smokers as compared to non-smokers.

The effect of cigarette smoke on blood coagulation and on different body systems have been an object of investigation. Findings similar to ours were observed by Bliden et al. They found that there was increase in platelet aggregation in smokers.¹⁰ Saba and Mason et al reported enhancement of ADP induced platelet aggregation by addition of nicotine to human platelet-rich plasma.¹¹ Bordia et al has also shown that platelet aggregation was markedly increased after smoking and when treated with garlic oil showed inhibition of platelet aggregation.¹²

But few studies have found no correlation between platelet aggregation and cigarette smoking.^{13,14,15,16,17}

Platelet aggregation can be stimulated in vitro by a number of agonists that affect platelet receptors, including ADP, epinephrine, collagen and thrombin. The cellular events leading to platelet aggregation are mediated by the binding of fibrinogen to the glycoprotein (GP) IIb/IIIa receptor of platelets as a final common pathway.¹⁸ Cigarette smoke contains over 4,000 known harmful components including nicotine, tar, ammonia, carbon monoxide, free radicals and other gaseous products which exert a negative effect on the platelet function and augment platelet aggregability. Normally platelets are in quiescent state but after endothelial injury due to cigarette smoke they might be getting activated. After activation, morphologically platelets undergo conformational change and adhere to each other by surface integrins GP IIb/IIIa forming larger aggregates. Activated platelets are known to facilitate the coagulation cascade and the formation of fibrin through release of coagulation factors including thrombin formation on the platelet surface.¹⁸ There is also an increase in sympathetic activity due to nicotine of cigarette smoke. It is known to be a sympathomimetic chemical that promotes the release of catecholamines which increase platelet aggregation.¹⁹

We have also found increased CRP levels in higher percentage of subjects in study group as compared to control group. Many other studies have also shown similar findings. Ohsawa et al found an elevation in CRP levels in smokers than non-smokers which was attributed to detrimental effects of tobacco smoke on tissues.²⁰ Mahrukh S et al also found raised CRP as well as complements like C3, C4 in smokers. They have attributed it to activation of monocytes and complement recruitment, resulting in the secretion of inflammatory cytokines which could further lead to release of CRP from liver in the blood.²¹ Aral et al have also noted similar observations.²² National Health and Nutrition Examination Survey (NHANESIII) study has confirmed that cigarette smoking contributed significantly to low-grade systemic inflammation and led to reduced lung function. It was associated with elevated CRP, fibrinogen and blood leukocytes, contributing to higher levels of systemic inflammation in susceptible individuals such as smokers.²³ Sin et al found high levels of CRP in smokers having chronic obstructive pulmonary disease.²⁴ A study by Pinto-Plata et al also supported the same fact²⁵. Loughlin et al studied the association between cigarette smoking and CRP in adolescent girls and boys and found a positive relationship. They also showed a linear association of CRP with the number of cigarettes smoked per day.²⁶

Few studies have shown conflicting results showing no correlation of CRP levels with smoking such as a study done by Haket al.²⁷

Inflammatory changes in the bronchial epithelium due to exposure to cigarette smoke could have led to increased C-reactive protein levels among cigarette smokers. Also, the toxins present in cigarette smoke might also contribute to increased C-reactive protein levels. C-reactive protein is an acute phase reactant released from liver which has been shown to tilt the balance of endothelial health towards a proatherogenic and thrombotic state.¹⁹

Conclusion: Increased platelet aggregability in smokers could be due to endothelial injury and

increased sympathetic activity. While increased C-reactive protein levels in smokers could be due to chronic inflammation in lungs and damage to the endothelium. It appears that for a given scenario, increased platelet aggregability and increased C-reactive protein levels are the risk factors for thrombotic episodes in future. It is known that majority of the adverse health effects of smoking can be reversible. So there is an urgent need to sensitize smokers about its health hazards so that disease burden due to smoking on society in general could be brought down.

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