Original research article

Study of TLC, DLC and SpO_2 of hemoglobin changes in healthy smokers & non-smokers

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Abstract

Background: Tobacco cigarette smoking is one of the major leading causes of death throughout the world. Smoking has both acute and chronic effect on hematological parameters. **Aim:** To evaluate the changes associated with the extent of adverse effects of tobacco smoking in total and differential leukocyte count and oxygen saturation of hemoglobin in healthy smokers and non-smokers. **Materials and Methods**: This cross-sectional study was carried out in the Department of Physiology, Patna Medical College, Patna, Bihar, India from May 2019 to December 2019. Total of 200 clinically healthy volunteers of Bihar, in the age group of 20–60 years participated in the present study. Individuals with a history of smoking cigarettes/bidis daily for at least 6 months were considered as smokers. Another 100 non-smokers of the same age group were included separately in this study as a control group. TLC, DLC and other parameters were analyzed using standard methods. **Results**: A total of 200 subjects (100 non-smokers and 100 smokers cases), in which baseline demographic parameters (age and BMI) are compared between smokers and non-smokers. No significant difference between the baseline demographic parameters between the smokers and non-smokers ensures optimum comparison avoiding bias.The difference between TLC, lymphocyte count, monocyte count, granulocyte count, and oxygen saturation of hemoglobin among smokers and non-smoker subjects. The mean values of TLC (p <0.001), lymphocyte count(p<0.001),monocyte count(p=0.01),and granulocyte count (p=0.01) were significantly higher in smokers as compared to non-smokers, while the mean values of SpO_2(p = 0.02) were significantly lower in smokers as compared to non-smokers. **Conclusion**: The study has shown that altered values of TLC and DLC and oxygen saturation of hemoglobin in smokers should be considered during diagnosis, interpretation of result, and treatment of patients. A high TLC and DLCs exhibited in this research may be responsible for chronic inflammation and subsequent high risk of CVD in smokers. Therefore, quitting smoking should be encouraged for better health.

Keywords: SpO2,Oxygen Saturation of Hemoglobin, Total and Differential Leukocyte Count, Smokers

Introduction

Smoking is the most important public health problem. Many studies conducted have proved its deleterious effects on many organ systems mainly respiratory, reticuloendothelial system and cardiovascular systems.[1] According to data reported from the World Health Organization, there are about 2.4 billion people worldwide that have consumed tobacco in the forms of smoking, chewing, snuffing or dipping. WHO also estimates that tobacco-related deaths will amount to 8.3 million in2030 and one billion deaths during the 21st century.[2]

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It has been estimated that an average of 7 minutes of life is lost for each cigarette smoked, roughly the time taken to smoke it.[3] A person who begins smoking at the age of 15 years has an average of 8 years of reduced longevity, and one starting after 25 years of age faces an average 4-year reduction. Coronary heart disease, cancer, and various respiratory diseases account for the majority of excess mortality related to cigarette smoking.[4] Smokers average a 16 fold increased the risk of acquiring lung cancer; a 12 fold increased the risk of acquiring COPD and a two-fold increased risk of having a myocardial infarction as compared to non-smokers. Since early 1950, several studies have shown a direct relation between smoking, hematological parameters, peripheral vascular disease, and stroke.[5] The link between smoking and pulmonary diseases was first recognized in the 1870's but it was not until 1964

that the US Surgeon General's report warned of a potential relationship between smoking and emphysema.[6] Heavy smoking is the commonest cause of ischemic heart disease and death in 30-40 years of the age group who are likely to be free from other myocardial risk factors. Alterations in the hematological parameters may be responsible for the high risk of occlusive vascular disease in chronic smokers.[7] Chronic smoking seems to cause an upward shift of hemoglobin dissociation curve, which may decrease the utility ofhemoglobin levels in the detection of anemia in smokers, suggesting that hemoglobin cut-off values should be adjusted for smokers to compensate for masking the effect of smoking on detection of anemia.[8] It is evident that respiratory system is primarily meant for oxygenation. Once oxygen reaches the alveoli (mostly transported by hemoglobin), a small fraction of it is remains molten. The amount of dissolved O_2in the bloodstream 0.003ml/100ml,with 1gm of haemoglobin carrying around 1.34 ml of O_2. The amount of oxygen in the bloodstream which is transported by the hemoglobin is known as oxygen saturation(SpO_2).Smoking is a major cause of both morbidity and mortality, with a prevalence rate of 20–40% in females and 30–40% in males in developed countries while 2–10% in females and 40–60% in males in developing countries. Thus, cigarette smoking damages lungs and affects other organ systems in different ways.[9]The present study thus investigates the effect of tobacco smoking on total and differential leukocyte count and oxygen saturation of hemoglobin for better diagnosis,interpretation of results,and treatment.

Materials and Methods

This cross-sectional study was carried out in the Department of Physiology, Patna Medical College, Patna, Bihar, India from May 2019 to December 2019, after taking the approval of the protocol review committee and institutional ethics committee. After taking informed consent detailed history was taken from the participant.

Methodology

A total of 200 clinically healthy volunteers of Bihar, in the age group of 20–60 years participated in the present study. Individuals with a history of smoking cigarettes/bidis daily for at least 6 months were considered as smokers. Ex-smokers or past smokers were excluded from the study. Smokers are defined as someone who, at the time of the study, smokes any tobacco product either daily or occasionally, while a non-smoker is someone who, at the time of the study, does not smoke at all. Moreover, an ex-smoker is someone who was formerly a daily or occasional smoker but currently does not smoke at all.

Unhealthy adults with any history of acute or chronic illness, bleeding and bleeding disorders, drug addiction, and if they had donated blood within the previous 6 months were not included in the study. Pregnant women were also excluded from the study.

Anthropometric parameters which include height, weight, and body mass index (BMI) was taken. Information of the smoking habits was obtained by a questionnaire.

Estimation of TLC, DLC and oxygen saturation of hemoglobin: After taking antiseptic precautions, blood samples were taken from the antecubital vein and collected into 3-5 ml ethylenediaminetetraacetic acid (EDTA) vacutainers.The EDTA blood samples were processed using automated hematology cell counter for total leukocyte count (TLC) (in thousands) and DLC (in percentage). Oxygen saturation of hemoglobin was done using fingertip pulse oximeter.

Statistical analysis

The data were analyzed using statistical software, SPSS (ver. 20.0) (IBM Inc, Armonk, New York, USA). Descriptive statistics and bivariate and regression analysis were carried out to find association and correlation and considered significant at $p< 0.05$. The internal consistency, i.e., Cronbach's alpha value was 0.87 that was suggestive of high reliability.

Results

Table 1 shows that in a total of 200 subjects (100 non-smokers and 100 smokers cases), in which baseline demographic parameters (age and BMI) are compared between smokers and non-smokers. No significant difference between the baseline demographic parameters between the smokers and non-smokers ensures optimum comparison avoiding bias.

Table 1: Comparison of baseline demographic parameters of smokers and non-smokers subjects

Smoking status	N=200	Range	Minimum	Maximum	Mean	Standard deviation
Non-smoker						
Age	100	33	18	61	32.67	9.457
BMI	100	21.37	17.04	37.08	23.1897	3.16224

Smoker						
Age	100	30	20	57	33.24	7.276
BMI	100	12.78	19.11	30.87	25.1528	2.85245
p-value	>0.05					

Test applied: student t-test, BMI: Body mass index

Table 2 shows the difference between TLC, lymphocyte count, monocyte count, granulocyte count, and oxygen saturation of hemoglobin among smokers and non-smoker subjects. The mean values of TLC (p <0.001), lymphocyte count(p<0.001),monocyte count(p=0.01),and granulocyte count (p=0.01) were significantly higher in smokers as compared to non-smokers, while the mean values of SpO_2(p = 0.02) were significantly lower in smokers as compared to non-smokers.

Table 2: Comparison of TLC, DLC, and oxygen saturation among smokers and non-smoker subjects

Parameter	**Non-smokers (N=100)**	**Smokers (N=100)**	***P*-value**
TLC	6.8787	7.3577	<0.001
DLC			
Lymphocyte count	0.3619	0.3723	<0.001
Monocyte count	0.061	0.0537	0.01
Granulocyte count	0.5878	0.50	0.01
SpO2	0.9767	0.9830	0.02

Test applied: student t-test ,
TLC: Total leukocyte count, DLC: Differential leukocyte count

Discussion

The results of our study showed a significant increase in the total WBC, lymphocyte count, monocyte count, and the granulocyte count in smokers as compared to non-smokers. We have also exhibited that oxygen saturation of hemoglobin was found to be lower in smokers than innon-smokers.Pedersen *et al*. in the Copenhagen general population study found that smoking causes increased blood leukocytes, neutrophils, lymphocytes, and monocytes.[10]Asif *et al*. in their study also found that regular smokers exhibited significantly greater WBCs count compared to non-smokers (p = 0.027).[11]They also found that the WBC count among male smokers was higher which also suggests that they may have greater risk of developing both atherosclerosis and CVDs than female smokers and non-smokers.[11]Airway epithelium acts as a physical barrier obstructing the entry of inhaled noxious particles into the submucosa. Leukocytosis has emerged as a potential marker of tissue damaged caused by cigarette smoke. Moreover, a rise inits count may account for an increased incidence of CVD through a plethora of postulated pathogenic mechanisms that mediate inflammation, block microvasculature at various junctures, and induce hypercoagulability. Gitte and Taklikaralso found in their study a sharp increase in total leukocyte count values of smokers with respect to the non-smokers.[12]Anithaand Manjunath also confirm this empirical positive association between smoking and total leukocyte count.[13]Our study also aimed at DLCs due to a probable association between cigarette smoking with TLC. Evidence suggests a strong possibility of this association, however, its effect on the DLC is still a matter of debate. In our study, it was also demonstrated that there was a statistically significant increase in all leukocyte subtypes. Zei-Shung*et al*. in their study also found significantly higher TLCs along with its subtypes in smokers.[14]One of the possible mechanistic hypotheses of this increased TLC is the extracted glycoprotein from the tobaccoleaf, which stimulates lymphocyte proliferation and differentiation by intermingling with a specific membrane component, commonly seen in antigenic response.[15]As for lymphocyte count, Shenwaiand Aundhakar reveal that the lymphocyte count increases significantly from 32.4% in non-smokers to 38.3% in smokers, while neutrophil count showed a slight fall in smokers than non-smokers, however, the difference for neutrophil count is statistically non-significant. Furthermore, no significant change was observed in eosinophil,

basophil, and monocyte counts.[16]It is quite evident that lymphocytosis is attributed to both chronic tissue damage and inflammation produced bytoxic substances found in tobacco smoke. It has also been suggested that smoke causes stimulation of respiratory bronchial tract inflammatory markers, thus inducing their increase in the blood .Moreover nicotine induces an increase in blood lymphocyte counts too.[10]Cigarette smoking encompasses a myriad of effects on the immune response of lymphocyte cells. Some of the noteworthy examples include immunoglobulin production, T4/T8 lymphocyte ratio change, enhanced NK activity, and low mutagen induced lymphocyte transformation.[11]In his research, Silverman *et al*. found that that smokers exhibit marked elevation in leukocytes especially Tlymphocytes.[17]We are aware that saturation of arterial blood to oxygen is essential for all individuals. Ozdal*et al*. reported that non- smoker individuals had significantly higher oxygen saturation of haemoglobin than smoker individuals($p<0.05$)which was similar as found in our study. The two main ingredients of cigarette smoke that potentially reduces oxygen supply to all tissues of the body are nicotine and carbon monoxide by combining themselves to transport proteins such as hemoglobin and myoglobin.[9]

The strength of our study was that the authentic subject selection was done on the basis of inclusion and exclusion criteria. Meticulously statistical analysis was done and p value was obtained to prove statistically significance.Earlier detection of respiratory damage in asymptomatic smokers will prevent future complications.Reduction in smoking may prove useful in subjects undergoing treatment and cansurely serve pivotal and an empirical cornerstone in people who are resistant to quitting. Limitations involve the limited sample size; the research should be carried out with larger sample sizes.Future direction in this kind of research is necessary to determine whether smoking cessation is advantageous and if yes to what extent smoking needs to be reduced for health benefits tooccur.

Conclusion

Smoking is nonetheless one of the major preventable risk factors for CVD mortality and morbidity. Cigarette smoking enhances inflammatory responses which areexhibited in our study by increasing levels of WBCs count and its subtypes. This study has shown that the total and DLC were altered in smokers and thus should be considered during diagnosis, interpretation of result, and treatment of patients. Tobacco smoking has a negative impact on oxygen saturation of hemoglobin. Reduction in smoking can improve the changes which are sensitive to change in smoking intake. We advise regular monitoring of the above-mentioned hematological parameters in smokers to detect early changes and avoid future catastrophic outcomes.

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