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Physiology

Effect of Cigarette Smoking on Selected Coagulation Parameters in Apparently Healthy Male Smokers

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Abstract	Original Research Article

***Background*:** Smoking is a significant global health concern associated with increased mortality and morbidity, including cancer development. Tobacco smoke contains carcinogens that can disrupt cellular processes and immune function, potentially affecting coagulation parameters. ***Objective*:** This study aimed to investigate the impact of smoking on selected coagulation parameters in apparently healthy male smokers compared to non-smokers. ***Method*:** A cross-sectional analytical study was conducted at the Department of Physiology, Sir Salimullah Medical College (SSMC) in Dhaka, Bangladesh, from July 1, 2018, to June 30, 2019. Seventy male participants aged 20 to 40 years were enrolled, comprising 35 healthy non-smokers (Group A) and 35 male smokers (Group B). Smokers (Group B) were further categorized into two groups based on pack-years of smoking (group B1 and group B2). Coagulation parameters including prothrombin time (PT), activated partial thromboplastin time (APTT), bleeding time (BT), and clotting time (CT) were assessed. ***Results*:** In this study, age and BMI were almost similar between groups A (healthy non-smokers) and B (smokers), with mean ($\pm$ SD) ages of 33.17 ± 2.91 and 34.11 ± 3.18 years, and mean ($\pm$ SD) BMI values of 24.36 ± 2.28 and 24.68 ± 2.33 kg/m2, respectively. There was no statistically significant disparity between the two groups, indicating successful age and BMI matching. Smokers (group B) exhibited significantly lower mean ($\pm$ SD) prothrombin time (PT) values (11.56 ± 0.87 seconds) compared to non-smokers (13.07 ± 0.77 seconds), with a statistically significant difference ($p < 0.001$). Similarly, activated partial thromboplastin time (APTT) was significantly decreased in smokers (28.42 ± 2.00 seconds) compared to non-smokers (31.50 ± 1.00 seconds) ($p < 0.001$). Bleeding time (BT) and clotting time (CT) were also significantly reduced in smokers (2.40 ± 0.24 minutes and 5.35 ± 0.29 minutes, respectively) compared to non-smokers (3.48 ± 0.36 minutes and 6.37 ± 0.36 minutes, respectively), with p-values < 0.001 for both comparisons. Further subgroup analysis revealed a significant decrease in PT and APTT values in both group B1 and B2 compared to A, with B2 displaying significantly lower values than B1. Negative correlations were observed between pack-year of smoking duration with PT, APTT, BT, and CT, indicating a decrease in these parameters with longer smoking duration ($p < 0.001$). ***Conclusion*:** Smoking adversely affects coagulation parameters, leading to decreased PT, APTT, BT, and CT values, especially among long-term smokers as compared to non-smokers. This trend may be attributed to the enduring inflammatory response generated by tobacco smoke, leading to the generation of inflammatory mediators that may contribute to the heightened blood clotting linked to smoking cigarettes. These findings emphasize the importance of smoking cessation in promoting cardiovascular well being.
Keywords: Smoking, Coagulation parameters, Prothrombin time (PT), Activated partial thromboplastin time (APTT), Bleeding time (BT), Clotting time (CT).

INTRODUCTION

Smoking is the inhalation of the smoke of burned tobacco that may occur occasionally or habitually as a consequence of physical addiction to nicotine [1]. Cigarette smoking is one of the major leading causes of death throughout the world. In addition, it is a major

contributor to cardiovascular disease and cancer, which account for millions of preventable deaths worldwide each year [2]. The 50 recognized carcinogens included in tobacco products increase the risk of cancer by influencing the immune or endocrine systems by causing mutation in a way that interferes with the control of the cell cycle. About 20% of all cancers are caused by smoking [3].

In our nation, the prevalence of current smoking is 18%. Male cigarette smoking rates are reportedly far higher than those of female smokers at the moment. Worldwide, the prevalence of smoking is still rising, with low- and middle-income nations having the highest incidence (about 80%). Smokers have a lower life expectancy than non-smokers. Additionally, it is predicted that there will be about one billion tobacco-related fatalities worldwide by the 21st century [4, 5].

The respiratory and cardiovascular systems are the two main organ systems that are adversely impacted by smoking [6]. Cigarette smoke has several detrimental effects on human health as it causes alterations in the main hemostatic systems by affecting the functions of endothelial cells, platelets and coagulation factors [7]. Cigarette smoking plays a significant role in prothrombotic alterations of coagulation system leading to hypercoagulability [8]. Several studies have suggested that smoking adversely affects some of the parameters of coagulation profile such as prothrombin time (PT), activated partial thromboplastin time (APTT), bleeding time (BT), clotting time (CT) etc. [2, 9-12].

Prothrombin time (PT) and activated partial thromboplastin time (APTT), bleeding time (BT) and clotting time (CT) are the most commonly performed laboratory tests in patients with suspected abnormal coagulation. Prothrombin time evaluates the extrinsic pathway and APTT indicates efficacy of both the intrinsic and common coagulation pathway [13]. Earlier studies have revealed significantly shortened PT and APTT with increased duration of smoking in smokers compared to non smokers [10, 11, 14, 15]. This findings was not in accordance with some authors who found prolonged PT and APTT in smokers with increasing duration of smoking as well as number of sticks smoked per day [2, 13]. On the other hand some investigators found no significant changes in APTT and PT among smokers compared to nonsmokers [12, 16, 17]. Another study reported a shortened PT and APTT in smokers indicating greater clotting activities in smokers [11]. Other study reported that PT and APTT shortening in chronic smokers increases the risk of thrombosis [10]. Moreover, another study reported an inverse correlation between the duration of smoking and coagulation markers (BT, WBCT, PT, APTT) in chronic smokers [9]. Shortened APTT indicates Hypercoagulability which is associated with venous thromboembolism [18].

From the aforementioned researches, it has been revealed that long term smoking results in alteration of some of the coagulation parameters in smokers predisposing them to various life-threatening complications. However, the effect of smoking intensity on the degree of impairment of coagulation cascade still remains unclear. So, it is important to identify the effect of smoking on coagulation parameters among smokers. But there is less published data available regarding this topic in our country. So the present study has been designed to ascertain the effect of smoking on some coagulation parameters in apparently healthy male smokers.

OBJECTIVES

General Objective
The purpose of this study is to investigate the effect of cigarette smoking on selected coagulation parameters in apparently healthy male smokers.

Specific Objectives
- To estimate prothrombin time (PT), activated partial thromboplastin time (APTT), bleeding time (BT) and clotting time (CT) in apparently healthy male cigarette smokers and non-smokers.
- To compare coagulation parameters of male cigarette smokers with those of non-smokers.
- To correlate the coagulation parameters with pack year of smoking duration.

METHOD

Study Type: Cross sectional analytical study.

Study Place and Period: Department of Physiology, Sir Salimullah Medical College (SSMC), Dhaka. The study was conducted from 1st July 2018 to 30th June 2019.

Ethical Issue: The ethical permission was taken from the Institutional Ethics Committee (IEC) of Sir Salimullah Medical College (SSMC).

Study Population: Study population included hospital staff members of Sir Salimullah Medical College and Mitford hospital and Bangabandhu Sheikh Mujib Medical University, Dhaka and also from personal contact.

Selection Criteria:
Inclusion Criteria: Comparison group were apparently healthy non-smoker male subjects. Inclusion criteria for study group were apparently healthy male regular cigarette smokers taking at least 10 sticks per day for 10 years or $\geq$ 5 pack-years of smoking history (1 pack-year = 20 cigarettes per day for 1 year).

Exclusion Criteria:
For Both Groups: Subjects with history of preexisting hypertension, diabetes mellitus, heart disease, liver disease, renal disease, thyroid disorder, malignancy, infection or debilitating illness, drug addiction or alcoholism and any coagulopathy were excluded from the study.

Study Procedure:
A total number of seventy male participants, aged 20 to 40 years, were included in this study, selected through consecutive purposive sampling. Among them, 35 apparently healthy non-smoker male subjects age ranged from 20 to 40 years were considered as comparison group (Group A). Another 35 age and BMI matched apparently healthy smoker male subjects were considered as study group (Group B). Again, smokers were subdivided into two groups according to the pack-years of smoking history; Group B1 (smokers with 5-15 pack-years of smoking history) and Group B2 (smokers with > 15 pack-years of smoking history).

After proper counselling the aim, objectives, risk and procedure of the study were explained in details to the subjects. They were encouraged for voluntary participation and were allowed to withdraw themselves from the study even after participation whenever they like. Smokers were interviewed about duration of smoking and average number of cigarette sticks smoked per day to determine their smoking exposure by estimation of pack-year. Prior to blood collection, participants underwent detailed medical assessments and physical examinations which were recorded in a data information sheet.

Blood was collected from each participants for estimation of biochemical and hematological tests. First all the blood samples were analyzed for glucose, creatinine, and ALT levels to rule out underlying health issues. Then Coagulation profiles were evaluated by estimation of prothrombin time (PT), activated partial thromboplastin time (APTT), bleeding time (BT), and clotting time (CT).

Statistical Analysis:
Data were presented as mean ± SD (standard deviation). Statistical analysis was done by using Statistical Package of Social Science (SPSS) windows version-22. ANOVA test was performed for comparison among the groups and then Bonferroni test was done to compare between the groups. Unpaired 't' test and Pearson's Correlation test were done to compare the data as applicable. p value ≤ 0.05 was considered as level of significance.

RESULTS
In both groups, subjects had almost similar mean (± SD) ages of 33.17 ± 2.91 and 34.11 ± 3.18 years for group A and group B respectively, with no statistically significant difference observed. Additionally, the mean (± SD) BMI values were 24.36 ± 2.28 and 24.68 ± 2.33 kg/m2 for group A and group B respectively, with no statistically significant difference, indicating successful age and BMI matching across all study subjects.

Table-1: Age and BMI in both groups (N=70)

Variable	Group A (n=35)	Group B (n=35)	p-value
Age (years)	33.17 ± 2.91 (25.00 – 37.00)	34.11 ± 3.18 (24.00 - 39.00)	0.200
BMI (kg/m2)	24.36 ± 2.28 (20.02 - 29.05)	24.68 ± 2.33 (19.00 - 29.06)	0.564

In this study, the mean (± SD) prothrombin time (PT) value was 13.07 ± 0.77 and 11.56 ± 0.87 seconds in group A and group B respectively, with a statistically significant (p<0.001) lower PT value observed in group B compared to group A. Similarly, for activated partial thromboplastin time (APTT), the mean (± SD) values were 31.50 ± 1.00 and 28.42 ± 2.00 seconds in group A and group B respectively, with a significant (p<0.001) decrease in APTT observed in group B. Additionally, the mean (± SD) bleeding time was 3.48 ± 0.36 and 2.40 ± 0.24 minutes in group A and group B respectively, showing a significant (p<0.001) reduction in bleeding time in group B. Furthermore, the mean (± SD) clotting time was 6.37 ± 0.36 and 5.35 ± 0.29 minutes in group A and group B respectively, indicating a significant (p<0.001) decrease in clotting time in group B compared to group A.

Table-2: Comparison of Prothrombin time (PT), Activated partial thromboplastin time (APTT), Bleeding time (BT) and Clotting time (CT) in study subjects (N=70)

Variable	Group A (n=35)	Group B (n=35)	p-value
PT (Seconds)	13.07 ± 0.77 (12.00 - 14.30)	11.56 ± 0.87 (10.20 - 14.00)	<0.001***
APTT (Seconds)	31.50 ± 1.00	28.42 ± 2.00	<0.001***

Variable	Group A (n=35)	Group B (n=35)	p-value
	(30.00 - 33.50)	(24.00 - 31.00)	
BT (Minutes)	3.48 ± 0.36	2.40 ± 0.24	<0.001***
	(2.83 - 4.00)	(2.00 - 3.17)	
CT (Minutes)	6.37 ± 0.36	5.35 ± 0.29	<0.001***
	(5.50 - 7.17)	(4.83 - 6.00)	

The mean ($\pm$ SD) PT values were 13.07 ± 0.77, 12.09 ± 0.64, and 10.77 ± 0.46 seconds in groups A, B1, and B2 respectively, with a significant decrease observed in both B1 ($p<0.001$) and B2 ($p<0.001$) compared to A. Moreover, B2 exhibited a significantly lower PT value than B1 ($p<0.001$). Similarly, for activated partial thromboplastin time (APTT), mean ($\pm$ SD) values were 31.50 ± 1.00, 29.82 ± 0.67, and 26.31 ± 1.33 seconds in groups A, B1, and B2 respectively, with significant reductions seen in both B1 ($p<0.001$) and B2 ($p<0.001$) compared to A, and B2 displaying a significantly lower APTT than B1 ($p<0.001$). Bleeding time (BT)

demonstrated mean ($\pm$ SD) durations of 3.48 ± 0.36, 2.49 ± 0.25, and 2.27 ± 0.15 minutes in groups A, B1, and B2 respectively, with significant decreases observed in both B1 ($p<0.001$) and B2 ($p<0.001$) compared to A, but no statistically significant difference was found between B1 and B2. Furthermore, clotting time (CT) showed mean ($\pm$ SD) values of 6.37 ± 0.36, 5.45 ± 0.29, and 5.20 ± 0.24 minutes in groups A, B1, and B2 respectively, with significant reductions observed in both B1 ($p<0.001$) and B2 ($p<0.001$) compared to A, while no statistically significant difference was noted between B1 and B2.

Table-3a: Prothrombin time (PT), Activated partial thromboplastin time (APTT), Bleeding time (BT) and Clotting time (CT) in different groups (N=70)

Variable	Group A (n=35)	Group B_1 (n=21)	Group B_2 (n=14)
PT (Seconds)	13.07 ± 0.77	12.09 ± 0.64	10.77 ± 0.46
	(12.00 - 14.30)	(11.00 - 14.00)	(10.20 - 12.00)
APTT (Seconds)	31.50 ± 1.00	29.82 ± 0.67	26.31 ± 1.33
	(30.00 - 33.50)	(28.00 - 31.00)	(24.0 - 29.0)
BT (Minutes)	3.48 ± 0.36	2.49 ± 0.25	2.27 ± 0.15
	(2.83 - 4.00)	(2.00 - 3.17)	(2.00 - 2.50)
CT (Minutes)	6.37 ± 0.36	5.45 ± 0.29	5.20 ± 0.24
	(5.50 - 7.17)	(5.00 - 6.00)	(4.83 - 5.83)

Table-3b: Multiple comparison

	PT p value	APTT p value	BT p value	CT p value
A vs B_1 vs B_2	0.000***	0.000***	0.000***	0.000***
A vs B_1	0.000***	0.000***	0.000***	0.000***
A vs B_2	0.000***	0.000***	0.000***	0.000***
B_1 vs B_2	0.000***	0.000***	0.106ns	0.075ns

In this study, significant negative correlations were observed between prothrombin time (PT) and pack-year of smoking duration ($r = -0.618$, $p < 0.001$), as well as activated partial thromboplastin time (APTT) and pack-year ($r = -0.750$, $p < 0.001$), indicating that as smoking duration increased, both PT and APTT decreased significantly. Similarly, bleeding time (BT)

exhibited a significant negative correlation with pack-year ($r = -0.578$, $p < 0.001$), suggesting a decrease in BT with longer smoking duration. Moreover, clotting time (CT) demonstrated a significant negative correlation with pack-year ($r = -0.617$, $p < 0.001$), indicating reduced CT as smoking duration increased.

Table-4: Correlation of coagulation parameters with pack-year in study group (n=35)

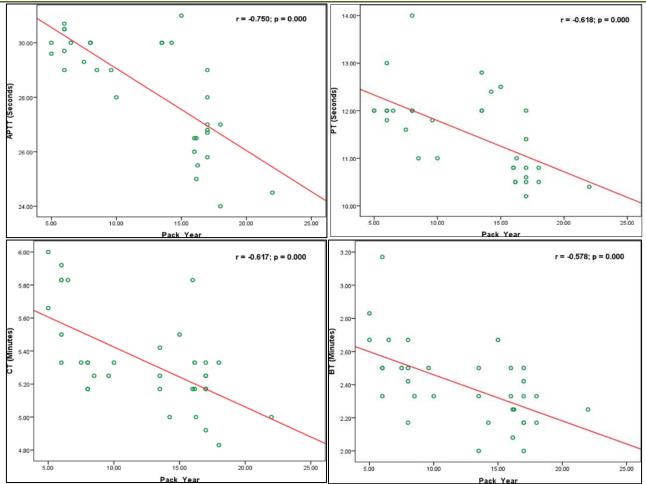|	r value	p value
PT (Seconds)	-0.618	0.000***
APTT (Seconds)	-0.750	0.000***
BT (Minutes)	-0.578	0.000***
CT (Minutes)	-0.617	0.000***

Figure-1a,1b,1c,1d: Correlation of prothrombin time (PT), activated partial thromboplastin time (APTT), bleeding time (BT), clotting time (CT) with pack-year in study group (n=35)

DISCUSSION

In this study, mean PT and APTT values were significantly (p< 0.001) lower among smokers in comparison to those of non-smokers. These findings were in agreement with that of other researchers [11, 15]. Moreover, when the smokers were further compared based on the duration of smoking, PT and APTT reduction were more pronounced in the smokers with > 15 pack-years of smoking history. These findings were in consistent with the study of other researchers [10, 14].

On the contrary, some other researchers found that mean PT and APTT values were significantly prolonged among smokers compared to those of non-smokers [2, 13]. This discrepancy might be due to the variation in the method of estimation, sample size and duration of smoking.

Whereas, some authors found no significant difference in the PT and APTT values between smokers and non-smokers [19, 20].

In this study, mean BT and CT values were significantly (p< 0.001) lower among smokers in comparison to that of non-smokers. Similar findings were reported by other researchers who reported shortened bleeding time after smoking high nicotine content cigarettes [21, 22]. However, when the smokers were further compared based on the duration of smoking no significant difference was observed in these values between two groups of smokers. No similar data was available for comparison.

However, some other researchers reported no immediate effect of cigarette smoking on bleeding time of habitual smokers [23]. This discrepancy might be due to variation in the study design and high nicotine content of cigarettes.

Whereas, no significant effect of cigarette smoking on clotting time was observed by some other investigators [24, 25].

In this study, PT and APTT values were negatively correlated with pack-year of smoking duration among the smokers. This relationship was

statistically (p<0.001) highly significant. These findings were in agreement with that of other studies [9, 11, 17].

On the contrary, some researchers found positive correlation between PT and APTT values of smokers with pack-year of smoking duration [2, 13].

In this study, BT and CT values were negatively correlated with pack- year of smoking duration among the smokers. This relationship was statistically (p<0.001) highly significant. This findings were in agreement with the other study [9].

CONCLUSION

This study found that PT, APTT, BT, and CT showed substantial reductions, especially in those who smoked for more than 15 pack-years, as compared to non-smokers. This trend may be attributed to the enduring inflammatory response generated by tobacco smoke, leading to the generation of inflammatory mediators that may contribute to the increased blood clotting associated with smoking cigarettes. The findings from this study along with evidences from the literature allow us to recommend that smokers who have smoked for more than 15 pack-years should be monitored for PT, APTT, BT, and CT which will be helpful to determine their altered coagulation status.

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