

Antioxidant status and smoking habits: relationship with diet

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ABSTRACT

Introduction: The present study was conducted to assess the association between smoking, dietary intake of antioxidants and plasma indices of oxidative stress and antioxidant defences in male smokers (cigarette and bidi smokers).

Methods: The study sample consisted of 100 healthy men, including 50 non-smokers and 50 smokers, who were subclassified into 25 cigarette smokers and 25 bidi smokers, aged 18–55 years. Erythrocyte superoxide dismutase and plasma ascorbic acid were measured as antioxidants and erythrocyte malondialdehyde as an oxidative stress index, by colorimetric methods.

Results: Smokers ate less fruits and vegetables than non-smokers, leading to them having a lower antioxidant level. Erythrocyte superoxide dismutase was significantly lower in cigarette smokers (0.193 U/mgP, p-value is less than 0.05) and bidi smokers (0.169 U/mgP, p-value is less than 0.001) as compared to non-smokers (0.231 U/mgP). Plasma ascorbic acid was also significantly lower in cigarette smokers (1.45 mg/100ml, p-value is less than 0.05) as well as in bidi smokers (1.38 mg/100ml, p-value is less than 0.001) as compared to non-smokers (1.73 mg/100ml). There was a significant increase in erythrocyte malondialdehyde concentration levels in cigarette smokers (171.47 $\mu\text{mol/gHb}$, p-value is less than 0.05) as well as in bidi smokers (231.04 $\mu\text{mol/gHb}$, p-value is less than 0.001) as compared to non-smokers (127.30 $\mu\text{mol/gHb}$).

Conclusion: These results provide enough evidence of increased oxidative stress and a compromised antioxidant defence system in smokers, and they are more profound in bidi smokers than in those smoking cigarettes. This study also revealed that the diet and nutrient intake of smokers are different from that of non-smokers.

Keywords: antioxidants, ascorbic acid, bidi

smokers, cigarette smokers, erythrocyte malondialdehyde, oxidative stress, superoxide dismutase

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INTRODUCTION

The adverse health effects of tobacco use were first reported centuries ago. Yet, it is only in recent decades that epidemiological studies have revealed the full extent of tobacco-related health disorders.⁽¹⁾ Cigarette smoking is the main risk factor for various chronic diseases, including cardiovascular disease, pulmonary disease and cancer.⁽²⁾ Similarly, a low consumption of fruits and vegetables is also associated with an increased risk of developing chronic diseases like cancer and atherosclerosis.⁽³⁻⁶⁾ It has been revealed that smokers have poorer diets than non-smokers.^(7,8) There are many toxic chemicals present in tobacco smoke such as nitric oxide and other oxidising radicals.⁽⁹⁾ These toxic chemicals cause damage to cellular functions.⁽¹⁰⁾ The low antioxidant status and increased oxidative stress in smokers have been clearly elucidated by the high oxidant content of smoke.⁽¹¹⁾ Therefore, smokers have been advised that they would benefit from increasing their consumption of antioxidants in their diet.⁽¹²⁾ Bidi smoking is extremely common in the countries of South Asia. Most Indian smokers, particularly those in the lower socioeconomic classes, smoke bidi. Bidis are small hand-rolled cigarettes wrapped in a piece of temburni leaf.

A number of studies have been undertaken to determine the amount of oxidative stress caused by cigarette smoking.⁽¹³⁻¹⁵⁾ However, to our knowledge, there is no such data about the extent of the effect caused by bidi smoking, which is more prevalent and widespread in India than cigarette smoking. The present study was conducted with the aim of describing the relations between smoking, dietary intake and blood indices of antioxidant defences, like superoxide dismutase (SOD) and ascorbic acid, as well as of oxidative stress, like malondialdehyde (MDA).

METHODS

The present study was carried out at the Department of Biochemistry, Gandhi Medical College, Bhopal, India. A total of 100 male subjects were included, 50 were individuals who had been smoking bidi or cigarettes for

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Table I. Demographics of the study population by smoking status.

Demographics	Non-smokers (n = 50)	Cigarette smokers (n = 25)	Bidi smokers (n = 25)	p-value
Age (years)	38.58 ± 12.08	32.80 ± 12.3	43.68 ± 8.49	< 0.001*
Age when smoking started (years)	–	18.3 ± 4.4	18.88 ± 5.73	≥ 0.05
Duration of smoking (years)	–	14.3 ± 12.4	24.8 ± 11.98	≤ 0.05
No. of cigarettes/bidis per day	–	7.6 ± 6.0	13.7 ± 11.44	< 0.05

Data is expressed as mean ± standard deviation.

*p < 0.001 (Student-Newmen-Keuls multiple-range test).

p < 0.05 is significant.

Table II. Dietary habits by smoking status.

Food group intake	Non-smokers (%) (n = 50)	Smokers* (%) (n = 50)	p-value (χ^2)
Milk	66	26	0.001
Egg	20	58	0.002
Fish	20	58	0.002
Meat	20	66	0.001
Cereals	100	96	0.247
Pulses	92	64	0.001
Fruits	74	18	0.001
Vegetables	82	54	0.002

*include cigarette smokers and bidi smokers.

p < 0.05 is significant (chi-square test).

more than one year (25 cigarette smokers, 25 bidi smokers), and 50 were age-matched non-smokers who acted as the controls. All subjects were in the age group of 18–55 years, and were healthy with no evidence of any chronic respiratory disease like asthma, tuberculosis, diabetes mellitus, chronic hepatitis or renal disease.

A questionnaire was offered to the subjects to obtain information regarding their smoking history, marital status, education, occupation, drinking habits, vitamin/mineral supplement and drug consumption. Their intake of various food items and frequency of consumption per day over the past one year were determined through interviews and with the help of a preformed food frequency questionnaire. The eligibility criteria regarding smoking status were as follows: smokers were eligible if they smoked ≥ 1 cigarette or bidi per day. Non-smokers were eligible if they had not smoked cigarettes or bidis for the last one year and had not been exposed to tobacco smoke at all. Subjects who had been taking ascorbic acid, α -tocopherol or multivitamin supplements before participating in the study were excluded. Exclusion criteria for participation in the study also included an intake of alcohol or tobacco in the past one year.

Venous blood samples were collected from the subjects in an ethylenediaminetetraacetic acid (EDTA) vial after an overnight fast. Plasma was separated for an estimation of the ascorbic acid level while haemolysate was prepared for SOD and MDA estimation. SOD was estimated using the

method described by Das et al.⁽¹⁶⁾ and ascorbic acid by the method described by Omaye et al.⁽¹⁷⁾ MDA was estimated by thiobarbituric acid assay, as described by Buege and Aust.⁽¹⁸⁾ Statistical analysis was performed with the Statistical Package for Social Sciences version 8.0 (SPSS Inc, Chicago, IL, USA). Blood parameters were analysed by analysis of variance (ANOVA) followed by a Student-Newmen-Keuls multiple-range test. Chi-square statistics were used to compare the dietary habits of smokers and non-smokers. A p-value ≤ 0.05 was used as a threshold of significance. Correlation coefficients (r) were calculated by Pearson's correlation analysis (two-tailed).

RESULTS

In our study, we found a significant difference in the mean age, duration of smoking and number of cigarettes/bidis puffed daily between cigarette and bidi smokers (Table I). After analysing the dietary intake according to the smoking status, we found a significant difference in the consumption of vegetarian, non-vegetarian and dairy products. The intake of non-vegetarian food items was lower in non-smokers than in smokers. On the other hand, smokers consumed less milk, pulses, fruits and vegetables than non-smokers and this difference was significant (p < 0.05), as depicted in Table II.

Erythrocyte SOD was significantly lower in cigarette smokers (0.193 U/mgP; p < 0.05) and bidi smokers (0.169 U/mgP; p < 0.001) as compared to non-smokers (0.231

Table III. Erythrocyte SOD, plasma ascorbic acid and erythrocyte MDA concentration levels by smoking status.

Parameters	Non-smokers (n = 50)	Cigarette smokers (n = 25)	Bidi smokers (n = 25)	F*	p-value
SOD (U/mgP)	0.231 ± 3.37	0.193 ^a ± 4.8	0.169 ^b ± 8.89	11.3	≤ 0.001
Ascorbic acid (mg/100ml)	1.73 ± 0.28	1.45 ^a ± 0.43	1.38 ^b ± 0.34	11.0	≤ 0.001
MDA (µmol/gHb)	127.30 ± 46.16	171.47 ^a ± 24.83	231.04 ^b ± 75.87	34.1	≤ 0.001

Data is expressed as mean ± standard deviation. Values indicated with different superscript letters are significantly different.

* One way analysis of variance (ANOVA).

p ≤ 0.05 (Student-Newmen-Keuls multiple-range test).

U/mgP). Plasma ascorbic acid was also significantly lower in cigarette smokers (1.45 mg/100ml; p < 0.05) and bidi smokers (1.38 mg/100ml; p < 0.001) as compared to non-smokers (1.73 mg/100ml) (Table III). The erythrocyte MDA concentration levels were significantly higher in cigarette smokers (171.47 µmol/gHb; p < 0.05) and bidi smokers (231.04 µmol/gHb; p < 0.001) as compared to non-smokers (127.30 µmol/gHb) (Table III).

A correlation analysis showed that there was a positive association of age with erythrocyte MDA concentration levels (r = 0.15), while plasma ascorbic acid and erythrocyte SOD showed a negative association (r = -0.15 and r = -0.01, respectively), although the correlation was not found to be significant. There was a significant positive correlation between the duration of smoking and erythrocyte MDA concentration level (r = 0.32, p < 0.05) (Table IV).

DISCUSSION

Our study results are similar to studies conducted in other countries.⁽¹³⁻¹⁵⁾ Some studies have reported that smokers have poorer dietary habits and consume significantly less ascorbic acid than non-smokers,⁽¹⁹⁻²²⁾ which may be the reason for an increased risk of cancer and cardiovascular diseases. The significant differences in food intake reported here between smokers and non-smokers (Table II) have also been shown in other communities, where smokers have a higher intake of non-vegetarian food items and a lower intake of fruits, vegetables and milk products.⁽²²⁾ According to a hypothesis proposed by Whichelow et al and Grunberg, smokers may find sweet foods, such as fruit juice, less palatable than fried foods,^(23,24) which was found to be the case in the present study. This may be due to the effect of smoking (via nicotine) on their sense of taste.⁽²⁵⁾

In our study, the extent of lipid peroxidation was found to be higher in smokers than in non-smokers, as shown by the significantly higher levels of erythrocyte MDA (Table III). Circulating erythrocytes are particularly susceptible to oxidative damage as they are exposed to a high partial pressure of oxygen, have membranes rich in polyunsaturated fatty acids and contain large amounts

Table IV. Correlation analysis between age, duration of smoking and number of items smoked/day with blood indices of oxidative stress.

	SOD	Ascorbic acid	MDA
Age	-0.15	-0.01	0.15
Duration of smoking	-0.19	0.02	0.32*
No. of cigarettes/bidis per day	-0.15	0.12	0.01

Correlation coefficients were calculated by Pearson's correlation analysis.

*correlation is significant at 0.05 level.

of iron that can potentiate a free radical reaction.⁽²⁶⁾ Our data confirms that cigarette smoking is associated with decreased SOD and ascorbic acid concentration levels, which suggests that with an increase in oxidative stress, there is a corresponding proportionate decrease in the antioxidant defence system (Table III).

Our study found a significant increase in erythrocyte MDA levels in bidi smokers as compared to cigarette smokers (Table III). Bidis have been reported to produce higher levels of carbon monoxide, nicotine and tar than cigarettes.^(27,28) One study has found that bidis produce approximately three times the amount of carbon monoxide and nicotine as well as approximately five times the amount of tar than cigarettes.⁽²⁹⁾ This may be reason for the significantly higher erythrocyte MDA levels in bidi smokers than cigarette smokers. It may be possible that these substances in bidis produce a greater amount of free radicals, which cause more deleterious effects on lipid peroxidation, resulting in higher erythrocyte MDA levels and lower concentrations of SOD and ascorbic acid activity due to the utilisation of these antioxidants for the scavenging of free radical generation.

As shown in this study, smoking results in a reduced supply of circulating antioxidants in the body, which may be due to the creation of an extra demand for antioxidants through oxidative stress. This effect is more marked in bidi smokers than in cigarette smokers. The diets of smokers usually contain lower amounts of antioxidant-rich foods. This results in a reduced antioxidant nutrient status of smokers. However, further investigations involving a large number of participants and analysis of other antioxidant

factors are required to confirm the extent of the free radical load generated by smoking and its effect on the antioxidant status.

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