

Serum and Salivary Immunoglobulins levels in Tobacco Smokers of Baghdad Governorate

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Abstract

It was shown that tobacco smoking was a major environmental factor which altered numerous changes in both antibody and cell mediated immune responses leading to increase the capability of the individual to develop various inflammatory and autoimmune diseases, thus the current study aimed to assess the levels of both serum and salivary Igs in 35 individuals who smoking tobacco and compared them to 35 non-smokers group. Results of this study revealed a slightly but non-significant increased ($p < 0.05$) in the serum and salivary levels of IgM in smokers as compared to non-smokers, while the serum and salivary levels of both IgG and IgA shown a significantly decreased ($p \leq 0.05$) in smokers as compared to non-smokers group. No positive correlation was found in the serum and salivary Igs levels in smokers. Body mass index (BMI) and smoking index (SI) were calculated for the smokers group, however, no significant differences were found in both serum and salivary Igs levels.

Keywords: Health, Smoking, Immunoglobulins (IgM, IgG and IgA); toxicity

Introduction

Tobacco smoking is one of the worldwide epidemic habits that associated with human beings death and morbidity⁽¹⁾. Various chronic diseases in the brain, respiratory tract and cardiovascular system are induced because of the antigenic, carcinogenic and mutagenic impacts of the toxic chemical compounds which found in tobacco⁽²⁾. The World Health Organization (WHO) expected that in 2030 tobacco smoking will be one of the main causes that lead to the death of about 8 million individuals especially in the developing countries⁽³⁾. According to Hussain and Sullivan study in Iraq, about 29-31% males and 3-4% females considered as active smokers⁽⁴⁾. Inhalation of tobacco smoking has many adverse effects on both humeral and cellular immune response, it implicated in the inhibition or the production of many pro-inflammatory and anti-inflammatory cytokines in human body⁽⁵⁾. Many studies confirmed that tobacco smoking has prospective effects on inflammation process and autoimmunity such as Chronic Obstructive Pulmonary Disease (COPD), Rheumatoid Arthritis (RA) and Systemic Lupus Erythematosus (SLE)⁽⁶⁾. Immunoglobulins (Igs) are glycoproteins found in serum, tissues and body fluids, synthesized by plasma cells in response to antigenic

stimulation⁽⁷⁾. Five classes of Igs have been established which include: IgM: plays an important role in primary immune response, IgG: considers as the main Ig in secondary immune response, IgA: protects the mucosal surfaces from infections, IgE: acts as anti-parasitic and anti-allergenic, IgD: acts as B-cell receptor^(8, 9). Igs that present in saliva play a very important role in oral cavity protection especially salivary IgA which inhibit the entry of infectious microorganisms⁽¹⁰⁾. The current study aimed to investigate whether there are differences in the serum and salivary Igs levels in tobacco smokers and non-smokers.

Materials and Method

The present study was carried out on 70 healthy males (with no clinical signs) lived in Bagdad Governorate, 35 individuals were smokers and 35 individuals were non-smokers, their age range from (18-40) years. Body mass index (BMI) was calculated by dividing the body weight (kg) on the square of the body height (m²) in both smokers and non-smokers as the followings: 7 smokers and 8 non-smokers had underweight BMI < 18.5 kg/m², 20 smokers and 22 non-smokers had normal BMI (18.5-24.9) kg/m², 5 smokers and 4 non-smokers had over weight BMI (25- 29.9) kg/m², while only 3 smokers

and 1 non-smokers had obese BMI >30 kg/m². Smoking index (SI) for smokers was calculated as the number of cigarettes per day multiplying by the years of smoking as the followings: 30 smokers had mild SI < 200 while only 5 smokers had moderate SI (200-600). Serum samples from both smokers and non-smokers were obtained after centrifugation of blood samples for 5 min. at 3000 rpm, then they were stored at -20°C until use. Unstimulated saliva samples were collected from the study subjects (whom prevented from eating and drinking for nearly 1 hr. prior to donation) after cleaning and rinsing their oral cavity. All saliva samples were centrifuged for 10 min. at 10000 rpm, then the supernatants were stored at -20°C until use. The levels of serum and salivary Igs were measured by using the turbidimetric immunoassay. In order to analyze the data of the current study which

were presented as mean ±Standard Error (S.E.), SPSS version 25.0 was used, one way ANOVA test has been dependent to find the differences between means which considered significant when the probability (P) value was ≤ 0.05⁽¹¹⁾.

Results

Serum Levels of Igs (IgM, IgG and IgA) in Smokers and Non-Smokers:

Results of the current study revealed that serum level of IgM showed non-significant increase (p>0.05) in smokers as compared to non-smokers, while the serum levels of both IgG and IgA were decreased significantly (p≤0.05) in smokers as compared to non-smokers (Table.1).

Table.1: Mean Serum Levels of Igs (IgM, IgG and IgA) in the Study Samples.

Samples	No.	Mean±S.E. IgM(mg/dl)	Mean±S.E. IgG(mg/dl)	Mean±S.E. IgA(mg/dl)
Smokers	35	123.188±5.88a	1075.76±33.112a	243.654±11.774a
Non-Smokers	35	119.385±2.99a	1235.58±41.014b	197.048 ±6.993b

*Similar letters=No significant difference (P> 0.05) between means.
*Different letters=Significant difference (P≤0.05) between means.

Salivary Levels of Igs (IgM, IgG and IgA) in the Study Samples.

According to the levels of Igs in saliva samples, salivary IgM in smokers showed slightly but non-significant increase (p>0.05) as compared to non-smokers; however the levels of salivary IgG and IgA in smokers were decreased significantly (p≤0.05) as compared to non-smokers (Table.2).

Table.2: Mean Salivary Levels of Igs (IgM, IgG and IgA) in the Study Samples.

Samples	No.	Mean±S.E. IgM(mg/dl)	Mean±S.E. IgG(mg/dl)	Mean±S.E. IgA(mg/dl)
Smokers	35	0.585±0.01a	1.869±0.06a	7.365±0.99a
Non-Smokers	35	0.56±0.07a	4.519±0.13b	16.05 ±0.56b

*Similar letters=No significant difference (P> 0.05) between means.
*Different letters=Significant difference (P≤0.05) between means.

Correlation between Serum Igs (IgM, IgG and IgA) in the Study Samples.

In smokers, negative correlation was found between serum IgM and IgG levels, no correlation was found between serum IgM and IgA levels as well as between serum IgG and IgA levels. In addition, no correlation was found among serum Igs in non-smokers (Table.3).

Table.3: Correlation between Serum Igs levels (IgM, IgG and IgA) in the Study Samples.

Serum Igs levels in Smokers		r	p	Significance
IgM(mg/dl)	IgG(mg/dl)	-0.01	0.956	Non-significant
	IgA(mg/dl)	0.103	0.557	Non-significant
IgG(mg/dl)	IgM(mg/dl)	-0.01	0.956	Non-significant
	IgA(mg/dl)	0.03	0.864	Non-significant
IgA(mg/dl)	IgM(mg/dl)	0.103	0.557	Non-significant
	IgG(mg/dl)	0.03	0.864	Non-significant
Serum Igs levels in Non-Smokers		r	p	Significance
IgM(mg/dl)	IgG(mg/dl)	0.299	0.081	Non-significant
	IgA(mg/dl)	0.387	0.092	Non-significant
IgG(mg/dl)	IgM(mg/dl)	0.299	0.081	Non-significant
	IgA(mg/dl)	0.163	0.35	Non-significant
IgA(mg/dl)	IgM(mg/dl)	0.387	0.092	Non-significant
	IgG(mg/dl)	0.163	0.35	Non-significant

Correlation between Salivary Igs (IgM, IgG and IgA) in the Study Samples.

In smokers, negative correlation was found between salivary IgM and IgG levels as well as between salivary IgM and IgA levels; however no correlation was found between salivary IgG and IgA levels. In addition, in non-smokers negative correlation was found between salivary IgM and IgA levels but no correlation was found between salivary IgM and IgG levels as well as between salivary IgG and IgA levels (Table.4).

Table.4: Correlation between Salivary Igs levels (IgM, IgG and IgA) in the Study Samples.

Salivary Igs levels in Smokers		r	p	Significance
IgM(mg/dl)	IgG(mg/dl)	-0.092	0.599	Non-significant
	IgA(mg/dl)	-0.197	0.256	Non-significant
IgG(mg/dl)	IgM(mg/dl)	-0.092	0.599	Non-significant
	IgA(mg/dl)	0.046	0.792	Non-significant
IgA(mg/dl)	IgM(mg/dl)	-0.197	0.256	Non-significant
	IgG(mg/dl)	0.046	0.792	Non-significant
Salivary Igs levels in Non-Smokers		r	p	Significance

Cont... Table.4: Correlation between Salivary Igs levels (IgM, IgG and IgA) in the Study Samples.

IgM(mg/dl)	IgG(mg/dl)	0.124	0.478	Non-significant
	IgA(mg/dl)	-0.43	0.807	Non-significant
IgG(mg/dl)	IgM(mg/dl)	0.124	0.478	Non-significant
	IgA(mg/dl)	0.08	0.649	Non-significant
IgA(mg/dl)	IgM(mg/dl)	-0.43	0.807	Non-significant
	IgG(mg/dl)	0.08	0.649	Non-significant

According to the BMI and SI in smokers, no significant differences were found in both serum and salivary Igs levels.

Discussion

Alteration in the immunological functions for both antibody and cell mediated immunity might be due to the inhalation of tobacco smoke^(12, 13). Several previous studies in other countries authorized that serum levels of Igs were lower in smokers by 10-20% as compared with non-smokers individuals⁽¹⁴⁾. Depending on the results of the current study, non-significant increase in terms of serum IgM was found in smokers which came in agreement with the results of Arinola *et al.*⁽¹⁵⁾ and Gonzalez-Quintela *et al.*⁽¹⁶⁾ who stated that this increased may be attributable to the ability of IgM in neutralizing the harmful toxins found in tobacco smoke by complement activation, conversely, Tarbiah *et al.*⁽¹³⁾ reported a significant increased while Olayanju *et al.*⁽¹⁷⁾ reported a significant decreased in the serum IgM in smokers as compared with non-smokers. In addition results of this research showed that both serum IgG and IgA levels were decreased significantly in smokers which were similar to the findings of Tarbiah *et al.*⁽¹³⁾ in terms of IgG and Aula and Fikry⁽¹⁸⁾ in terms of IgA level, however, IgG and IgA levels were found to be higher in smokers according to Prajapati and Jyoti study⁽¹⁹⁾. Various investigations have concentrated on the possible mechanisms by which tobacco smoke might alter B-cells (which responsible for the production of Igs after their differentiation into plasma cells) development, functioning and distribution, it was found that tobacco smoke might cause down regulation of murine marrow B220+CD34- pre-B cells and/or B220+CD34+ pro-B cells which they effect on B-cells development⁽²⁰⁾. In addition several molecular studies have stated that nicotinic receptors such as $\alpha 4$ and $\alpha 7$ subunits which

have essential roles in B-cells lines show elevated expression after long term exposure to nicotine which suppress B-cells secretions⁽²¹⁾. Furthermore, it was found that proliferative ability of T-cells and T-cells dependent antibody responses were decreased in smokers which subsequently suppress B-cells functions and causes reduction in the production of serum Igs⁽²²⁾. Moreover low number and impaired functions of regulatory B-cells in smokers infected by Helicobacter pylori had been improved by Li *et al.*⁽²³⁾. It was found that after smoking cessation the levels of serum IgM and IgG were increased significantly, however the level of IgA remained the same⁽²⁴⁾. Despite the fact that blood considered as an essential sample for disease diagnosis; saliva reveals the mucosal and systemic expression of many biomarkers in the human body, salivary IgA initiate from the plasma cells found in salivary glands, while salivary IgM and IgG are mainly originate from the serum⁽²⁵⁾. Regarding the results of the present study, salivary IgM revealed non-significant increase in smokers which analogs with the results of Barton *et al.*⁽²⁶⁾ but a significant decreased in salivary IgG and IgA which came in agreement with the results of Giuca *et al.*⁽²⁷⁾. It has been demonstrated that tobacco smoke has an adverse effect on the oral mucosa by altering the amounts of saliva secretions, enzymes and immune functions⁽²⁸⁾, so the reduction of salivary Igs which have important roles in mucosal defenses may increase the susceptibility of the smokers to develop several oral infections such as chronic periodontitis⁽²⁹⁾. Low level of salivary IgA which was found in this paper may be due to the perceived impact of tobacco smoke on the salivary glands or on other immunological cells that responsible for the production of salivary IgA. The absence of positive correlation in the smokers Igs levels in both serum and saliva in this study may be due to the smaller size of the smoking samples, in addition this study included only men because of the low prevalence

of smoking in females.

Conclusions

According to the data of the current study, tobacco smoking is strongly associated with the suppression of B-cell functions and thus alters the Igs production and levels in both serum and saliva samples of smokers individuals.

Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both MOH and MOHSER in Iraq

Conflict of Interest: Non

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