Evaluation of Salivary IgA in Diabetic and Non-Diabetic Patients: A Case-Control Study

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Abstract

Objective: Diabetes Mellitus as the most common metabolic disease of human has wide range of oral consequences such as oral infections which could make changes in the properties of saliva. The purpose of this study was to compare the amount of salivary IgA in diabetic and non-diabetic patients.

Materials and Methods: In this case-control study, the population consisted of 40 persons including 20 diabetic and 20 non-diabetic subjects. All of them had not eaten, drunk or smoked at least 90 minutes before sampling. Sampling launched and the salivary IgA was measured with nephelometric procedure with Minineph kit (Minineph TM Human Kit, Binding Site Ltd, Birmingham, UK).The data were statistically analyzed by T-test and chi-square using SPSS software.

Results: In 70% of the case group, salivary IgA was higher than the normal (>37.6mg/dl) but none were higher than normal in the non-diabetic (control) group and this difference was statically significant (p=0.009). IgA and age (p=0.303), and IgA and sex (p=0.0398) had no significant correlation on both case and control group.

Conclusion: Salivary IgA of diabetic patients is higher than the non-diabetics (p=0.009).

Keywords: Salivary, IgA, Diabetes

Introduction

Diabetes mellitus is a multifactorial metabolic disease characterized by increase in blood glucose and disturbances in the metabolism of carbohydrates, proteins and lipids (1). Increase in blood glucose is the main characteristic of diabetes which results from insufficient insulin secretion and hepatic gluconeogenesis (2,3). There are two major types of diabetes: type 1 or insulin-dependent diabetes and type 2 or non-insulin dependent diabetes. The worldwide prevalence of diabetes is predicted to rise from 180 millions in year 2000 to 320 millions in 2025 (4,5). Diabetes is a clinically complex disease and it is associated with many complications including neuropathy, retinopathy, nephropathy and cardiovascular diseases (6,7). This disease also has some oral complications which are dry mouth, tooth loss, periodontitis, gingivitis, dental abscesses and soft tissue lesions of the oral mucosa (6,8,9). Previous studies reported that the changes in

salivary compositions may affect the formation, signs and symptoms, and severity of oral manifestations in patients with diabetes. It has been demonstrated that detection of salivary components in patients with diabetes may be helpful in diagnosis, prevention and management of the oral manifestations in this population (10-12). The aim of this study was to evaluate salivary IgA levels in diabetic patients and to compare it with non-diabetes subjects.

Materials and Methods

The patients chosen to participate in this study consisted of 20 type 2 diabetic patients (11 women and 9 men) with the mean age of 60.1 years who were referred to Yazd diabetes research center (All the subject group had controlled diabetes according to their records). The control group consisted of 20 non-diabetic patients (12 women and 8 men) who were referred to oral medicine faculty of Yazd dental school. The age of the control group was matched with the case group. All patients were examined by a specialist for any oral lesions.

Inclusion criteria consisted of type 2 diabetic patients registered in Yazd diabetes research center and edentulous patients with no clinical signs of Candidiasis. Patients with teeth (for excluding periodontal disease and its effects) and/or with any underlying disease with or without diabetes mellitus were excluded from the study.

Saliva collection

Five milliliters of unstimulated whole salivary samples were collected by spitting method, in dry plastic vials while sitting in a relaxed position. Patients were prohibited to eat or drink anything at least 2 hours before sampling. The collected saliva samples were centrifuged (2000g for 10 minutes). The supernatants were stored at -70 centigrade until further analysis. All Samples were collected at 8-10 a.m. IgA was assessed in both patient and control groups.

Measurement of salivary IgA

IgA was assessed by the nephelometric method (Minineph TM Human Kit, Binding Site Ltd, Birmingham, UK). The determination of soluble antigen concentration by nephelometric methods involves a reaction with specific antiserum to form insoluble complexes. When light is passed through the suspension formed, a portion of light is scattered and detected by a photodiode. The amount of light scattered is directly proportional to the specific protein concentration in the test sample. Concentrations are automatically calculated by reference to a calibration curve stored within the instrument.

Statistical analysis

Statistical analysis was done by t-test and Chi-square test for unstimulated whole saliva IgA levels within both patient and control groups with significance defined as $p<0.05$. Data were expressed as mean±SD.

Results

Forty patients participated in this study including 11 women and 9 men in the case group, and 12 women and 8 men in the control group which were age-matched. Seventy percent of the case group had IgA levels higher than normal (>37.6mg/dl) but in the control group none were higher than normal which was statically significant ($p=0.009$) (Table 1). IgA and age ($p=0.303$) (Figure 1), and IgA and sex ($p=0.0398$)(Table 2) had no significant correlation on both case and control group.

Discussion

The most common biologic fluid for diagnosis and monitoring of the majority of the diseases is blood sample. Also, saliva has been studied frequently as an alternative for blood that may be helpful for diagnostic and surveillance purposes. Components of whole saliva are produced locally as well as components entering from serum that may be helpful for

Table 1. IgA levels in case and control groups.

Groups	Normal IgA n (%)	IgA higher than normal n (%)	p
Diabetic patients	6 (30)	14 (70)	0.009
Non-diabetic patients	20 (100)	0 (0)	

Table 2. Mean IgA levels according to sex in case group (mg/dl) (p=0.398)

Sex	Mean IgA level mean±SD	p
Men	76.3750 ±10.206	0.398
Women	60.0250 ±34.73586	

diagnosis of many systemic diseases and as well as understanding their oral manifestations better than before (14). There are many advantages to salivary assessment including non-invasive collection and being cheap for screening huge populations (14).

Mata et al (15) reported alterations in salivary compounds in patients with diabetes mellitus. These biologic alternations in whole saliva in patients with diabetes mellitus were not the same in one study to another; this could be due to the differences in sample collection and study design (16). In this study, we found significant differences in salivary IgA levels between diabetic patients and their matched controls. As we know, hyperglycemia is a major sign of diabetes mellitus and may reduce phagocytic function of granulocyte cells and facilitate colonization of specific microorganisms. Ketoacidosis, another sign of diabetes, may delay granulocytes migration to the site of injury and reduce phagocytic function. Neutrophils are also affected by hyperglycemia and chemostasis is also violated. Therefore this increase in IgA levels in diabetes patients may be due to presence of Candida species and humoral response of the immune system to this microorganism (17). Also compensatory mechanisms in the immune system will lead to increase in humoral response and increase in salivary IgA. Yavuzyilmaz et al. (10) demonstrated a significant increase in salivary IgA levels in patients with diabetes mellitus in comparison to matched controls. They suggested that it could be related to local factors such as calculus and higher bacterial plaque accumulation in these patients. In our study, the effect of periodontal disease was excluded by enrolling only edentulous patients. The findings of our study on salivary IgA levels were in contrast with other studies (18-20). These differences in results may be due to differences in the salivary collection methods,

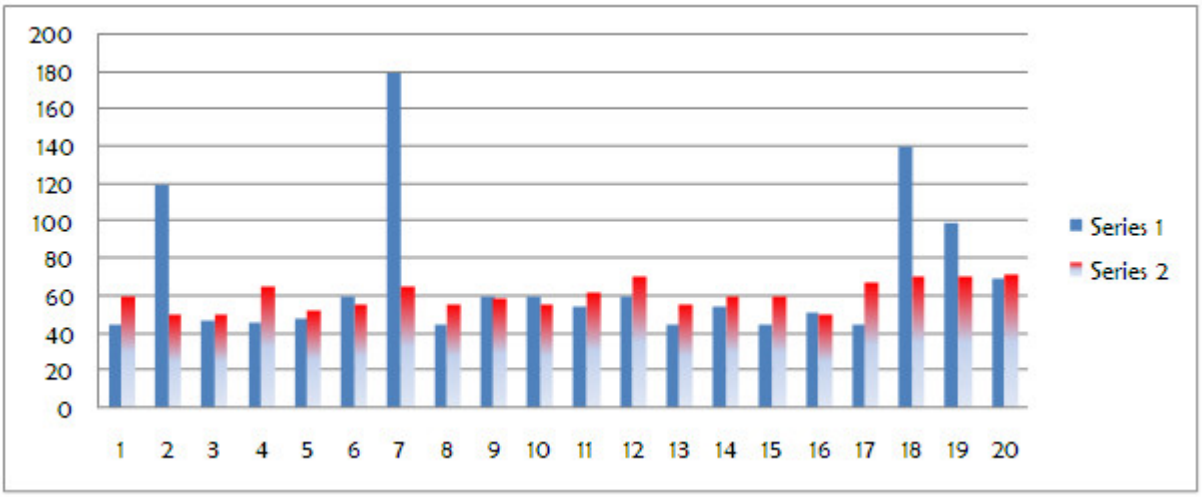

Figure 1. Correlation between age and IgA levels in case group (series1: IgA, series2: age)

the type of saliva collected (stimulated or unstimulated), the type of the disease (type 1 or type 2), and the diabetes control status. There was no significant difference between IgA levels by sex and age in our study which was consistent with other studies (21,22).

In a study by Wilson et al. (23) on diabetic patients with denture stomatitis, albumin and IgG was higher than normal, but salivary IgA levels were lower than the control group which could be related to the usage of local nystatin for relieving candidiasis and therefore reducing the immune response in the saliva. In a study by Silva-Boghossian et al. (24) in HIV positive patients there was a significant difference between salivary IgA in diabetic and non-diabetic patients which could be correlated to cellular and humoral immune systems dysfunction in these patients and the essential role of humoral immunity for the production of antibodies. So, greater caution should be taken about infections in diabetic patients, although it seems that mucocutaneous infections are not a problem because of the compensatory effect of salivary IgA. (23,24)

Conclusion

We conclude that diabetic patients have higher levels of IgA than non-diabetic patients and this may be due to compensatory response in the immune system of these patients.

References

1- Vaziri PB, Vahedi M, Abdollahzadeh SH, Abdolsamadi HR, Hajilooi M, Kasraee SH. Evaluation of Salivary Albumin in Diabetic Patients. Iranian Journal of Public Health 2009;38(3):54-9.

2- Conget I. Diagnosis, classification and cathogenesis of diabetes mellitus]. Rev Esp Cardiol 2002;55(5):528-35.

3- Alberti S, Spadella CT, Francischone TR, Assis GF, Cestari TM, Taveira LA. Exfoliative cytology of the oral mucosa in type II diabetic patients: morphology and cytomorphometry. J Oral Pathol Med 2003;32(9):538-43.

4- Schneider H, Shaw J, Zimmet P. Guidelines for the detection of diabetes mellitus- diagnostic criteria and rationale for screening. Clin Biochem Rev 2003 Aug;24(3):77-80.

5- Vernillo AT. Diabetes mellitus: Relevance to dental treatment. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2001;91(3):263-70.

6- Neville BW, Damm DD, Allen CM, Bouquot JE. Oral and maxillofacial pathology. Philadelphia: Saunders; 2009:842-5.

7- Batbayar B, Somogyi J, Zelles T, Fehér E. Immunohistochemical analysis of substance P containing nerve fibres and their contacts with mast cells in the diabetic rat's tongue. Acta Biol Hung 2003;54(3-4):275-83.

8- Madden TE, Herriges B, Boyd LD, Laughlin G, Chiodo G, Rosenstein D. Alterations in HbA1c following minimal or enhanced non-surgical, nonantibiotic treatment of gingivitis or mild periodontitis in type 2 diabetic patients: a pilot trial. J Contemp Dent Pract 2008 Jul 1;9(5):9-16

9- Lalla E, Cheng B, Lal S, Tucker S, Greenberg E, Goland R, et al. Periodontal changes in children and adolescents with diabetes: a case-control study. Diabetes Care 2006 Feb;29(2):295-9.

10- Yavuzyilmaz E, Yumak O, Akdoğanli T, Yamalik N, Ozer N, Ersoy F, et al. The alterations of whole saliva constituents in patients with diabetes mellitus. Aust Dent J 1996 Jun;41(3):193-7.

11- Dodds MW, Dodds AP. Effects of glycemic control on saliva flow rates and protein composition in non-insulin-dependent diabetes mellitus. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 1997 Apr;83(4):465-70.

12- Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Report of the expert committee on the diagnosis and classification of diabetes mellitus. Diabetes Care. 2003;26(1):5-20.

13- Navazesh M. ADA Council on Scientific Affairsand Division of Science. How can oral health care providers determine if patients have dry mouth? J Am Dent Assoc 2003;134(5):613-20.

14- Kaufman E, Lamster IB. The diagnostic applications of saliva--a review. Crit Rev Oral Biol Med 2002;13(2):197-212.

15- Mata AD, Marques D, Rocha S, Francisco H, Santos C, Mesquita MF, et al. Effects of diabetes mellitus on salivary secretion and its composition in the human. Mol Cell Biochem 2004 Jun;261(1-2):137-42.

16- Kimura I, Sasamoto H, Sasamura T, Sugihara Y, Ohgaku S, Kobayashi M. Reduction oincretinlike salivatin in saliva from patients with type 2 diabetes and in parotid glands of strept ozotocindiabetic

BALB/c mice. Diabetes Obes Metab 2001;3(4):254-8.
17- Little SW,Falace DA, Miller CS, Rhodus NL, Dental Management of the medically compromised patient 7th edition London. Mosby 2008;248-270
18- Gill CW, Bush WS, Burleigh WM, Cooke- Gomes D. Elevation of IgA levels in the noninsulin-dependent (type II) diabetic patient. Diabetes Care 1981 Nov-Dec;4(6):636-9.
19- Belazi MA, Galli-Tsinopoulou A, Drakoulakos D, Fleva A, Papanayiotou PH. Salivary alterations in insulin-dependent diabetes mellitus. Int J Paediatr Paediatr Dent 1998 Mar;8(1):29-33.
20- Dodds MW, Yeh CK, Johnson DA. Salivary alterations in type 2 (non-insulin-dependent) diabetes mellitus and hypertension. Community Dent Oral Epidemiol 2000;28:373-81.
21- Liberatore RR Jr et al, Is immunity indiabetic patients influencing the susceptibility to infections immunoglobulins complement and phagocytic function in children and adolescent with type 1 diabetes mellitus, pediatr diabetes 2005 Dec;6(4):206-12
22- Fukushima C et al, salivary IgA and oral candidiasis in asthematic patiens treated with in haled cortico steroid. J Asthema 2005:42(7):601-4
23- Wilson J, Wilton JM, Sterne JA. Comparison of secretory IgA and IgG isotype levels in palatal secretions of denture stomatitis patients with denture wearers having clinically healthy palates. The European journal of prosthodontics and restorative dentistry 2007;15(2):50-4
24- Silva Boghossian Cari, Castro GF, Teles RP, De Souza IP, Colombo AP. Salivary microbiota of HIVG-positive children and its correlation with HIV status, oral diseases, and total secretory IgA. International Journal of Paediatric Dentistry 2008;18(3):205-16.