



The Evaluation of Relationship between Salivary Cholesterol, Salivary Triglycerides and Dental Caries in People with Type 2 Diabetes

Anahita Ghorbani¹, Ozra Akha², Farshad Keshavarzi³, Mehran Ebrahimzadeh Hassanabadi⁴, Jaber Mosavi⁵, Maedeh Salehi^{1*}, Tahereh Molania^{1*}

¹DDS, MS of Oral and Maxillofacial Medicine, Assistant Professor, Department of Oral Medicine, Faculty of Dentistry, Mazandaran University of Medical Sciences, Sari, Iran

²MD, Endocrinologist, Associate Professor, Diabetes Research Center, Mazandaran University of Medical Sciences, Sari, Iran

³Post Graduate Student, Department of Periodontology, Babol University of Medical Sciences, Babol, Iran

⁴Dentistry Student, School of Dentistry, Student Research Committee, Mazandaran University of Medical Sciences, Sari, Mazandaran, Iran

⁵MD, MPH, Board of Community & Preventive Medicine, Assistant Professor, Department of Community & Preventive Medicine, Faculty of Medicine, Mazandaran University of Medical Sciences, Sari, Iran

DOI: 10.24896/jrmds.20186224

ABSTRACT

High levels of lipids in saliva are associated with high decay because they have adverse effects on the protective role of saliva. Our goal is to investigate the level of saliva cholesterol and triglyceride in type 2 diabetic patients and to determine the relationship between these saliva parameters and the status of dental decay. In this case-control study, a total 100 patients with diabetes, 50 with high triglycerides and cholesterol and 50 with normal range of these parameters (control) participated in the study. Both groups had at least 22 teeth. Non-stimulated saliva was collected from each individual for 5 minutes. All five tooth surfaces were examined for decay and DMFT of patients was recorded. According to the analysis, there was a significant relationship between serum cholesterol and saliva cholesterol, and also between serum triglyceride and saliva triglyceride. Carbohydrate, FBS, and HbA1c showed a relationship with years of having diabetes mellitus, but salivary cholesterol did not related to age of patients. Salivary triglyceride had a significant relationship only with years of having diabetes not with FBS, HbA1C and age of patients, also decay had a significant relationship with the age of the patients, and years of having diabetes, but it did not have relationship with blood glucose levels. In the study group, salivary cholesterol and DMFT showed a significant relationship, however, saliva triglyceride was not associated with DMFT in study and control group. In our study, it was found that there was a significant relationship between decay, missing, and filled teeth (DMFT) and salivary cholesterol, but this relationship was not found between salivary triglyceride and decay.

Keywords: Diabetes mellitus (DM), Hyperglycemia, Dyslipidemia

HOW TO CITE THIS ARTICLE: Anahita Ghorbani, Ozra Akha, Farshad Keshavarzi, Mehran Ebrahimzadeh Hassanabadi, Jaber Mosavi, Maedeh Salehi, Tahereh Molania, The Evaluation of Relationship between Salivary Cholesterol, Salivary Triglycerides and Dental Caries in People with Type 2 Diabetes, J Res Med Dent Sci, 2018, 6 (2): 154-159, DOI: 10.24896/jrmds.20186224

Corresponding author: Maedeh Salehi and Tahereh Molania

e-mail ✉ salehimaede1165@gmail.com; t_molania117@yahoo.com

Received: 25/08/2017

Accepted: 19/12/2017

INTRODUCTION

Diabetes mellitus is a metabolic disorder that is caused by hyperglycemia due to impaired insulin secretion or function [1]. This chronic disease

affects the metabolism of carbohydrates, proteins, fat, water and electrolytes [2].

Diabetes consists of two types: 1 and 2. A genetic component is involved in the onset of both types, but the effect of genetic on type 2 diabetes is much more than type 1 [3].

Type 2 diabetes usually occurs after age 40 and in obese people [3]. It is characterized by increased blood sugar and dyslipidemia due to the insulin resistance [4]. This kind of non-insulin-dependent diabetes is usually diagnosed in middle age, and the patient may have lived many years before diagnosis with this disease [5]. Type 2 diabetics are treated with oral antidiabetic drugs and diet [6]. Oral symptoms of diabetes include decreased salivation, difficulty in swallowing, and speech impairment, as well as increased probability of having oral infections. Candidiasis, dental decay, gingivitis, and mucositis also occur in these people [7].

Dental decay is a chronic infectious disease caused by factors such as decay causing microbial plaque on dental surfaces, plaque microbial activity, and carbohydrate intake, especially sucrose [8, 9].

It has been observed that systemic diseases cause decay through changing physiological activity in the mouth, and diabetes is one of these diseases [10, 11].

It has been shown that high levels of lipids in saliva are associated with high decay, dental plaque maturation, calculus, and periodontal diseases. Some salivary lipids, such as lysophosphatidylcholine, affect the activity of glycosyltransferase, which is associated with the activity of decay causing bacteria [12].

Studies have demonstrated that saliva can play an important role in the development and progression of enamel decay, and salivary lipids and proteins can have adverse effects on the protective role of saliva [13]. Previous studies have shown the relationship between salivary peptides and dental decay proteins, but there are few publications in terms of the relation between lipids, especially cholesterol and triglycerides with dental decay [7]. It seems that cholesterol concentrations of saliva are reflection of serum cholesterol concentrations in people with high cholesterol levels [14]. In many studies, there is

also a direct relationship between cholesterol and triglyceride levels in saliva and serum [15]. In a study on patients with Cystic fibrosis, high lipid levels of saliva have been proven that reflect the high levels of lipids in the serum of these people [12]. According to the study of subramaniam p and colleagues in 2014, both cholesterol and triglyceride of saliva in type 1 diabetic children showed higher amounts than the control group, and DMFT of diabetic children was more than the control group in this study. It was found that there is a significant relationship between saliva between triglycerides and dental decay in children [7]. Also, in a study by umiko *et al.*, it was found that lipids, free fatty acids, and triglycerides in people with decay probability are higher than the control group [13]. However, due to lack of information on the relationship between cholesterol and triglyceride and dental decay in diabetic people and because no study has been conducted on type 2 diabetes so far, hence our goal is to investigate the level of saliva cholesterol and triglyceride in type 2 diabetic patients and to determine the relationship between these saliva parameters and the status of dental decay.

MATERIALS AND METHODS

This study is a case-control type in which 50 patients with type 2 diabetes and higher triglycerides and cholesterol than normal range were considered as the experimental group and 50 patients with type 2 diabetes with the similar age and sex with the experimental group who had the normal range of triglyceride and cholesterol were considered as control group and were selected among the patients referred to the diabetes center of the late clinic of Mostafavian Imam Khomeini Hospital.

Individuals in both groups had at least 22 tooth and were almost equal in terms of socioeconomic and oral hygiene. The inclusion criteria for the experimental group was type 2 diabetic patients whose levels of FBS, HbA1C, and triglycerides and cholesterol were recorded in the last month and had triglycerides and cholesterol levels higher than normal ranges.

The inclusion criteria for the control group was diabetic patients whose blood glucose and cholesterol and triglyceride tests were recorded in the last month, as well as their cholesterol and triglycerides levels were in normal range.

Patients with any other systemic disease that affects the level of cholesterol and triglyceride, and smokers and alcohol users, or any drug that reduces salivation, were excluded.

At the beginning of the study, written consent was taken from the after they were informed.

In this study, non-stimulated saliva was collected from each individual in a sterilized graded container for 5 minutes. This non-stimulated saline was collected from the patient to reduce the effect of circadian rhythms at 9-11 am. Patients were requested one hour earlier than saliva collection to avoid eating, drinking and brushing, and any factor that stimulates saliva [7].

Saliva samples were stored at -4°C and stored in ice and immediately sent to a biochemical analysis laboratory [7]. Patients saliva were placed in a centrifuge device. All specimens were centrifuged at -27°C and were kept in a biochemistry laboratory to measure triglycerides and their cholesterol in one day, which would also reduce the errors of the spectrophotometric device.

Blank, standard and sample solutions were prepared according to Table 1 and the samples were incubated for 20 minutes at room temperature (20-25 ° C) or 10 minutes at 37 ° C. The standard optical absorption was measured in a spectrophotometer set at a wavelength of 546 nm (according to the kit instruction).

Table 1: Construction method of blank, standard, and sample solution

	Sample or standard	Blank
Sample or standard	10 microliters	—
Distilled water	—	10 microliters
reagent	1000 microliters	1000 microliters

Total triglyceride and total cholesterol were measured by cholesterol oxidase (CHOD) and glycerol 3 phosphate oxidase (GPD) in serum

cholesterol and triglyceride kits of the Kimia Paksh Company (7). Cholesterol levels higher than 4.64 mg and triglycerides higher than 3.78 mg/dL are considered as higher than normal (16). A flashlight and dental sterile mirror and catheter No. 23 were also used to check dental decay using the green_venrillam method (17). All five tooth surfaces were examined for decay and DMFT of patients was recorded on the chart. In the DMFT study, all the teeth were considered if only all patients had at least 22 teeth in their mouths.

The results were analyzed using SPSS 16 software. Data description was carried out through central index and dispersion and appropriate charts and tables. Parametric variables were analyzed by Student T test in two groups. Spearman correlation test was used to study the relationship between decay and saliva parameters. Also, P-value was considered less than 0.05.

RESULTS

In this study, 100 patients were examined from which 50 patients were in the experimental group and 50 in the control group. The patient group included people with type 2 diabetes that had triglycerides and cholesterol higher than the normal range, and the control group of type 2 diabetic patients had triglycerides and cholesterol in normal range (TG = 160 and Chol = 200).

The experimental group consisted of 31 males and 19 females (62% females and 38% males) between the ages of 21-69 years old, and the control group were at the same age and sex with the experimental group, so that the control group included 31 women and 19 males. The mean age of the group was 50.04 ± 11.52 and the mean age of the control group was 49.94 ± 11.89 years. The mean duration of the disease in the case group was 8.64 ± 4.19 years and in the control group, this average was 5.75 ± 5.5 (Table 2, 3).

Table 2: The characteristics of the control group

	FBS (Mg/dl)	Triglyceride level in serum(mg/dl)	Cholesterol level In serum(mg/dl)	Triglyceride level in saliva (mg/dl)	Cholesterol level in saliva (mg/dl)	DMFT	HbA _{1c} (%)
Average	150.40	127.66	163.46	6.78	2.85	13.88	7.66
Standard deviation	67.26	23.23	25.26	7.93	1.67	5.35	1.20
Lower bound	127	65	94	0	0	2	6.2
Upper bound	598	159	198	46.91	6.30	25	12

Table 3: The characteristics of the study group

	FBS (Mg/dl)	Triglyceride level in serum (mg/dl)	Cholesterol level In serum (mg/dl)	Triglyceride level in saliva (mg/dl)	Cholesterol level in saliva (mg/dl)	DMFT	HbA _{1c} (%)
Average	192.96	245.58	248.22	22.20	12.22	16.28	8.63
Standard deviation	65.17	225.3	43.5	22.79	12.98	5.82	1.30
Lower bound	127	161	200	0	0	2	6.3
Upper bound	426	1711	412	118.51	66	26	13

According to the analysis, there was a significant relationship between serum cholesterol and saliva cholesterol (P-value <0.001), as well as serum triglyceride and saliva triglyceride had also a significant association (P-value <0.05).

There was a significant correlation between carbohydrate, FBS, HbA_{1c}, and years of having diabetes mellitus in diabetic patients (P value <0.05). But there was not a strong association between saliva cholesterol and age of patients.

Regarding saliva triglyceride, only years of having diabetes had a significant relationship, but saliva triglyceride did not show any relationship with FBS, HbA_{1c} and age of patients.

The decay with salivary cholesterol showed a very strong and significant relationship (P-value=0.006), but in relation to saliva triglyceride, this relationship wasn't observed, and dentate decay did not show a relation with saliva triglyceride (P-value=0.14).

According to Table 4, decay had a significant relationship with the age of the patients, so that with increasing age, the DMFT level was increased in patients. There was also a significant relationship between decay and years of having diabetes (P-value<0.001) that means, patients who had more time with this disease showed higher DMFT, but there was no significant correlation between decay and blood glucose levels, meaning that decay with FBS and HbA_{1c} did not show a meaningful relationship.

According to Table 5, in the comparative analysis of two groups, the control and experimental groups and studying the results revealed that in the study group, the relationship between saliva cholesterol and DMFT showed a significant relationship (P-value=0.044); however, saliva triglyceride was not associated with DMFT (P-value=0.485).

In the study group, no relationship was found between salivary triglyceride and DMFT (P-value=0.908), and also saliva cholesterol did not show a correlation with DMFT in the control group (P value = 0.337).

Table 4: the relationship between DMFT and other studied variables

Study parameters	P-value
Saliva cholesterol and DMFT	0.006
Saliva Triglyceride and DMFT	0.144
Years of diabetes and DMFT	0.000
Patients' age and DMFT	0.000
FBS and DMFT	0.369
HbA _{1c} and DMFT	0.069

Table 5: Comparison the relationship of DMFT and saliva cholesterol and Triglyceride between study group and control group

	P value of Study Group	P value of control group
DMFT and salivary cholesterol	0.044	0.337
DMFT and salivary Triglyceride	0.485	0.908

DISCUSSION

Type 2 diabetes usually occurs after age 40 and in obese people [3]. It is characterized by increased blood sugar and dyslipidemia due to insulin resistance [4]. This kind of non-insulin-dependent diabetes is usually diagnosed in middle age, and the patient may have lived many years before diagnosis of diabetes [5]. Type 2 diabetics are usually treated with oral and hypoglycemic drugs and diet [6]. Systemic diseases have been reported to cause decay through changing physiological activity in the mouth, which diabetes is one of these diseases [10, 11].

Dental decay is a chronic infectious disease caused by factors such as decay microbial plaque on dental surfaces, plaque activity and carbohydrate intake, especially sucrose [8, 9]. Decay is a multifactorial disease that one of its risk factors is high lipid levels in saliva of people [18].

Increased lipophilic saliva lipids and low levels of glycolytic lipids are associated with maturation of the dental biofilm. Saliva lipids play a core role in initial plaque mineralization. High levels of saliva cholesterol lead to high lipid accumulation in plaque, which leads to slowing the release of lactic acid, which is one of the major causes of decay in diabetic people with lipid levels higher than normal range [7]. There are contradictory results between different publications about decay in diabetics and non-diabetics, since there are many risk factors in between [7].

In the present study, which was performed on saliva triglyceride and cholesterol, it was found that there was a significant relationship between decay, missing, and filled teeth (DMFT) and saliva cholesterol, but this relationship was not found between saliva triglyceride and decay.

Patients with higher saliva lipid levels have more decay, dental plaque, coliculus, and periodontal diseases. This is because the lipids change the bacterial adhesion by creating a water-repellent environment, which increases the decay range. A study by Bronislaw L and colleagues confirms this finding completely that patients with more saliva lipids have significant decay significantly [12].

Saliva lipids cause hydrophobic levels of bacteria, which help to bind bacteria to dental surfaces; the process of decay begins by sticking bacteria to the teeth [7].

In the study of Lopez me and colleagues on diabetic children, it was found that decay rate in these children is more than healthy controls. They studied saliva of diabetic children and found that the protein, glucose, and total urea saliva of these children were more than usual, while they stated that higher dmft rate of the diabetic group was related to this result, and salivary analysis of these children did not discuss the high lipid levels of diabetic children [19].

The total lipid level in saliva of people with higher decay (DMFT) was higher than those who showed less decay in their mouths. This study in the Umiko Tomita research showed that subjects with more decay than the control group (non-decay-teeth people) had a higher level of lipid, fatty acid, and triglyceride in their saliva [13]. In our study, cholesterol levels showed a significant correlation with decay, but there was no relation with saliva

triglyceride that according to our result, the cause of decay in type 2 diabetic patients is high cholesterol levels and saliva triglyceride of this people wasn't the cause of their decay.

In a study conducted in 2014 by Priya Subramaniam and colleagues on diabetic children, it was found that high levels of cholesterol and triglyceride in saliva were responsible for more decay in diabetic children than controls [7]. The study also explicitly stated that the rate of lipids is related to decays, but the disadvantage of this study is that people with diabetes have higher levels of saliva secretion, which is a high risk factor for glucose itself. For this reason, we selected both control and patient groups of diabetics to bring the two groups closer together and a closer examination of the relationship between cholesterol and triglyceride saliva with decays.

In our study, it was found that there is a significant relationship between the number of years of having diabetes and DMFT, which was consistent with Falk, Sandberg and Colin studies, and they concluded that the number of years of having diabetes mellitus increases the rate of decay [20-22].

CONCLUSION

In our study, it was found that there is a significant relationship between the years of having diabetes and DMFT. It was found that high levels of cholesterol and triglyceride in saliva were responsible for more decay in diabetic children than controls [7]. Also cholesterol levels showed a significant correlation with decay, but there was no relation with saliva triglyceride that according to our result, the cause of decay in type 2 diabetic patients is high cholesterol levels and saliva triglyceride of this people wasn't the cause of their decay.

REFERENCES

1. Matthews DC. The relationship between diabetes and periodontal disease. *Journal-Canadian Dental Association.* 2002; 68(3):161-64.
2. Hegde MN, Tahiliani D, Shetty S, Devadiga D. Salivary alkaline phosphatase and calcium in caries-active type II diabetes mellitus patients: an in vivo study. *Contemporary Clinical Dentistry.* 2014; 5(4):440-44.

3. Greenberg MS, Burkets GM. Oral medicine, diagnosis and treatment, 11 th ed. Spain: BC Decker Inc., 2008:509-511.
4. Javanbakht M, Baradaran HR, Mashayekhi A, Haghdoost AA, Khamseh ME, Kharazmi E, Sadeghi A. Cost-of-illness analysis of type 2 diabetes mellitus in Iran. *PloS one*. 2011; 6(10):e26864.
5. Walston J, Silver K, Bogardus C, Knowler WC, Celi FS, Austin S, Manning B, Strosberg AD, Stern MP, Raben N, Sorkin JD. Time of onset of non-insulin-dependent diabetes mellitus and genetic variation in the β 3-adrenergic-receptor gene. *New England Journal of Medicine*. 1995; 333(6):343-47.
6. Ciglar I, Šutalo J, Škaljac-Staudt G, Božikov J. Saliva as a risk factor for caries in diabetic patients. *Acta stomatologica Croatica*. 1991; 25(3):143-49.
7. Subramaniam P, Sharma A, Kaje K. Association of salivary triglycerides and cholesterol with dental caries in children with type 1 diabetes mellitus. *Special Care in Dentistry*. 2015; 35(3):120-22.
8. Swanljung O, Meurman JH, Torkko H, Sandholm L, Kaprio E, Mäenpää J. Caries and saliva in 12–18-year-old diabetics and controls. *European Journal of Oral Sciences*. 1992; 100(6):310-13.
9. Bokhout B, Hofman FX, Prahl-Andersen B. A sufficient cause model for dental caries. *Journal of Epidemiology and Biostatistics*. 2000; 5(3):203-08.
10. Närhi TO, Meurman JH, Ainamo A. Xerostomia and hyposalivation. *Drugs & Aging*. 1999; 15(2):103-16.
11. Sonis ST, Fazio RC, Fang L. Principles and practice of oral medicine. 2nd ed. Philadelphia: WB Saunders, 1995:32(5):145-151.
12. Slomiany BL, Murty VL, Slomiany A. Salivary lipids in health and disease. *Progress in Lipid Research*. 1985; 24(4):311-24.
13. Tomita Y, Miyake N, Yamanaka S. Lipids in human parotid saliva with regard to caries experience. *Journal of Oleo Science*. 2008; 57(2):115-21.
14. Karjalainen S, Sewon L, Soderling E, Larsson B, Johansson I, Simell O, Lapinleimu H, Seppänen R. Salivary cholesterol of healthy adults in relation to serum cholesterol concentration and oral health. *Journal of Dental Research*. 1997; 76(10):1637-43.
15. López-López J, Avila-Ortiz G. Dentistry and Diabetes: The Influence of Diabetes in Oral Diseases and Dental Treatments. *Journal of Diabetes Research*. 2016; Article ID 4372852, 15.
16. Singh S, Ramesh V, Oza N, Balamurali PD, Prashad KV, Balakrishnan P. Evaluation of serum and salivary lipid profile: A correlative study. *Journal of oral and maxillofacial pathology*. 2014; 18(1):4-8.
17. Khademi H, Kaviani NA, Mesripour M, Ahmadi SH. Dental damages in non-insulin dependent diabetes patients. *Bimonthly Journal of Hormozgan University of Medical Sciences*. 2006; 10(1):71-75.
18. IDF Diabetes Atlas-seventh edition 2015, <http://www.diabetesatlas.org>.
19. López ME, Colloca ME, Páez RG, Schallmach JN, Koss MA, Chervonagura A. Salivary characteristics of diabetic children. *Brazilian Dental Journal*. 2003; 14(1):26-31.
20. Sandberg GE, Sundberg HE, Fjellstrom CA, Wikblad KF. Type 2 diabetes and oral health: a comparison between diabetic and non-diabetic subjects. *Diabetes Research and Clinical Practice*. 2000; 50(1):27-34.
21. Falk H, Hugoson A, Thorstensson H. Number of teeth, prevalence of caries and periapical lesions in insulin-dependent diabetics. *European Journal of Oral Sciences*. 1989; 97(3):198-206.
22. Collin HL, Uusitupa M, Niskanen L, Koivisto AM, Markkanen H, Meurman JH. Caries in patients with non-insulin-dependent diabetes mellitus. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology and Endodontics*. 1998; 85(6):680-85.