ORIGINAL ARTICLE

Evaluation of Fasting Blood Sugar via Salivary Glucose in Type 2 Diabetes Mellitus

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Abstract

Objective: Diabetes mellitus (DM) as an ongoing metabolic disorder worldwide is a major public health concern. Diagnosis of DM is depended on clinical symptoms and laboratory findings. Fasting blood glucose (FBG) monitoring is currently the most common diagnostic method, which is an invasive and stressful procedure. Most recently, the use of non-invasive, convenient methods like fasting salivary glucose (FSG) has been highly regarded among researchers. The aim of present study was to evaluate the correlation of FSG with FBG in T2DM patients and healthy subjects.

Materials and Methods: In this cross sectional study, glucose level was measured using the glucose oxidase/peroxidase method in blood and unstimulated saliva in 50 T2DM patients and 50 non-diabetic subjects. After determining the normality of the data, Pearson's correlation coefficient was done to assess the correlation between FBG and FSG.

Results: The mean level of FBG, FSG and HbA1c of diabetic group were 161.00±5.6, 12.80±0.80 and 8.00±0.3 which were significantly higher than non-diabetic subjects (74.75±4.3, 6.5±0.75, 5.2±0.2.7, *P*-value: 0.001). The Pearson's correlation coefficient showed significantly strong relationship between FBS and FSG in both groups (*P*-value: 0.005).

Conclusion: This study demonstrated the presence of a significant correlation between FSG and FBG. Therefore, FSG level may be used as a non- invasive method to evaluate blood glucose in T2DM patients and healthy subjects.

Keywords: Salivary glucose, Type 2 diabetes mellitus, Blood glucose

Introduction

Type 2 diabetes mellitus (T2DM) is a syndrome of abnormal carbohydrate, fat and protein metabolism. T2DM is caused by absolute or relative lack of insulin and is a major public health concern (1). According to the last Diabetes International Federation (IDF) report, the rate of diabetic patients will reach to 438 million patients

worldwide in 2030 (2). Chronic hyperglycemia can threat the immune system, cardiovascular, renal and ophthalmic systems (3). To decrease the incidence of diabetes complications, screening and early diagnosis of T2DM is critical (4).

Laboratory diagnosis of T2DM is based on the invasive blood glucose level measurement in two situations: being in fasted state and not (random or causal glucose), in which the blood glucose level above 126 mg/dL is diagnosed as diabetes (5). Moreover, monitoring of hemoglobin A1c (HbA1c) provides an accurate measurement of glycemic control over the past three months (6). Recently, saliva-based tests for screening and monitoring of systemic diseases including DM have been developed appropriately (4,5,7).

Saliva collection by the patients at home is a non-invasive, safe, painless and performable method (1,8). According to the most salivary investigations in T2DM patients, there was a significant correlation between fasting blood glucose (FBG) and fasting salivary glucose (FSG) levels (9). This study was conducted to determine the correlation between FSG and FBG levels in T2DM patients compared to the non-diabetics.

Materials and Methods
Study Population
This cross sectional study was performed on 100 adults (sample size determined by previous studies(10)) between 21 to 55 years old. The studied samples were divided into two groups; diabetic group including 50 known T2DM patients and non-diabetic control group including 50 healthy subjects. In diabetic group, T2DM patients (FBS ≥147mg/dL and HbA1c ≥7.5%) were randomly chosen from Mostafavian health care unit of Sari, Iran. Control group were healthy individuals (age and sex matched) recruited at the Mazandaran University of Medical Sciences (MAZUMS) of Sari, Iran. Both case and control subjects had no systemic disease, no radiotherapy background at head and neck area, no alcohol use, no anti-

histamine and anti-cholinergic drug use. The only difference of these two groups was the diabetes mellitus in case group. The study was conducted according with MAZUMS ethical principles with ethical code of IR.MAZUMS.REC.94.1446. Also after being informed of the aim of the study, a written consent was received from each individual before his/her enrollment in the study.

The exclusion criteria were; smoking, systemic disease such as Sjogren's syndrome and cardiovascular disease.

Sampling and measurements
Saliva sampling was done between 9 and 11 AM after an over-night fasting. For the collection of saliva, the participant was asked to wash his/her mouth with water, sit in the coachman's position, head slightly down and not to swallow, stimulate saliva production (11). Whole saliva (approximately 3 mL) was collected in sterile graded containers. Saliva samples were centrifuged immediately to remove cell debris (× 1000 g for 10 min at 4°C). The supernatant was extracted and stored in small aliquots at - 80°C for biochemical analyses. Blood sample was taken between 8 and 10 AM after an over-night fasting. Whole blood was drawn from the median vein and was allowed to clot in the test tube, followed by centrifuging at 3000 rpm for 10 min and then the serum was separated.

Glucose analysis was done using GOD/POD (glucose oxidase-peroxidase) method, which is based on oxidation of glucose to gluconic acid and hydrogen peroxide in the presence of glucose oxidase. For measurement of glucose concentration in blood/saliva of the patients, 10 μL of the serum/saliva was taken in a test tube, to which 1000 μL of the glucose reagent was added (glucose quantitative measure kit, Pars Azmoon co, Iran), incubation for 10 min at 37°C, followed by a spectrophotometry measurement at 546 nm. HbA1c level was measured by the immunoturbidimetric method (HbA1C quantitative measure kit, Pars Azmoon co, Iran).

Statistical analysis

All data were analyzed with SPSS-24. After determining the normality of the data with the Kolmogorov-Smirnov test, statistical analysis was done by independent T-test for quantitative variables and Chi square test for qualitative variables between two groups, respectively. Pearson's correlation coefficient was done to assess the correlation between FBS level and FSG level within groups. A *P*-value< 0.05 was considered statistically significant.

Results

As shown in Table 1, there were no significant differences between two groups regarding to the demographic and medical parameters. The mean level of FBG, FSG and HbA1c of each group were shown in Table 1. The average level of all three mentioned variables in diabetic group was higher than non-diabetic subjects and this difference was statistically significant by independent T-test. According to statistical analysis, the Pearson's correlation coefficient between FBG and FSG was significant. There was a strong relationship between FBS and FSG in both groups (Table 2).

Discussion

Salivary parameters are changed by metabolic, nutritional and neurological abnormalities also with hydration status and medication. It is believed that FSG follows a threshold mechanism that means an increase in FBG leads to increase in FSG due to the "leakage" across the basement membrane of the glands into the saliva (12). According to Qureshi et al. (13), long term hyperglycemia leads to microvascular alterations in the blood vessels, as well as basement membrane changes in the salivary glands. This is an impetus increasing leakage of glucose from the ductal cells of the salivary glands, which demonstrates the abundance of glucose content in saliva (14).

Whole saliva is frequently studied as an alternative for blood and can be valuable for diagnostic purpose (15,16). In this study, we compared FBG and FSG concentrations in a sample of 100 Iranian subjects. The Pearson's correlation coefficient demonstrated a correlation between FBG and FSG concentrations in both groups (p-value: 0.005). Thus, according with this study results, FSG serves as a reliable indicator of FBG in the diabetic patients with elevated blood glucose levels. Shashikumar et al. (12) showed a statistically significant correlation between the salivary and the blood glucose levels, which is accordance with our present study results. However, in contrast to our study, the studies by Jurysta et al. (17) Carda et al. (18) and Vasconcelos et al. (14) failed to prove the

Table 1. Demographic and medical features in diabetic and non-diabetic patients

Variables	Diabetic (n = 50)	Non- diabetic (n = 50)	*P*–value
Age (mean ± SE)	49.3±32	50.65±41	0.25
Sex Male N (%)	18(32%)	19(38%)	0.70
Fasting blood glucose (mean ± SE) mg/dl	161.00±5.62	74.75±4.33	0.001
HbA1c (mean ± SE)	8.00±0.31	5.2±0.2.72	0.001
Fasting saliva glucose, (mean ± SE) mg/dL	12.80±0.80	6.5±0.75	0.001

Table 2. **Correlation between fasting blood glucose level and fasting saliva glucose in diabetic and non-diabetic groups**

Group	Variables	r-square	*P*–value
Diabetic	Fasting blood glucose, Fasting saliva glucose,	0.192	0.005
Non- diabetic	Fasting blood glucose, Fasting saliva glucose,	0.185	0.005

presence of a correlation between FSG and FBG and they assumed that salivary glucose is not directly affected by blood glucose level, therefore could not be used as a method to monitor glycemic control in diabetic patients. As saliva may be a potential substitute for blood in lab tests for the diagnosis of some illnesses (19), including DM, our study supports the use of saliva as a diagnostic fluid in T2DM and non-diabetics. Overall, it should be noted that a cross sectional study leads to correlations which cannot be called cause and reason relations. In addition, due to its prevalence testing ability, it would be rare to prove the incidence of high FSG and FBG and its correlation in a population.

Conclusions

The present findings showed that FSG has a significant positive correlation with FBG. Therefore, FSG could be a potentially practicable non-invasive tool to investigate glycaemic control in T2DM and non-diabetics.

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Conflict of Interest

The authors declare that there are no conflicts of interest.

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