



COMPARATIVE STUDY OF SALIVARY AND SERUM GLUCOSE AS DIAGNOSTIC BIOMARKERS IN DIABETIC AND NON DIABETIC PATIENTS

Biochemistry

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ABSTRACT

Aim & Objective: To estimate and compare of serum and salivary glucose level in diabetic and non-diabetic patient and saliva can be used as a non invasive diagnostic fluid in Diabetic Mellitus patients.

Material and method: This comparative cross-sectional study conducted in Department of Dentistry & orthopaedic, Government medical college Jalaun (Orai) U.P. included 100 subjects aged between 30-60 years including both genders (Male & Female), which were divided into two groups; Group- I (Control): 50 Non-Diabetic Mellitus patients Group-II (Study): 50 Diabetic Mellitus patients. Serum and saliva collected from all participants and analyzed by auto-analyzer, followed by standard solution and the readings were noted, noted reading used for tabulation master chart and statistical analysis done.

Results: One way ANOVA student t test with Tukey HSD applied and found highly significant value in all groups. Mean age was calculated in group I & II respectively ± 43.94 & ± 44.64 years. In group I, mean value of serum ± 90.1 mg/dl & in group II serum glucose level mean was 188.32 mg/dl and salivary glucose level in group I, ± 0.34 mg/dl and group II, ± 4.52 mg/dl. When comparison was done between group I and group II on the bases of Fasting Serum glucose level respectively 90.1 mg/dl and 188.32 mg/dl and salivary fasting glucose level 0.34 mg/dl and 4.52 mg/dl respectively.

Conclusion: On the behalf of present study saliva is non-invasive, cost effective and multiple sampling in one patient diagnostic biological fluid in diabetic mellitus, it can also use on large population, easy to collect and process.

KEYWORDS

INTRODUCTION

Oral cavity is mirror of whole body, this is a very wide line in our medical filed but it is hundred percent true. When any small and large changes occurs in our body either physiological or biochemical, that's reflection shown in our oral cavity. In our society, so many people suffering from life treating diseases, such as blood pressure, Diabetic mellitus, cardiovascular disease etc. Diabetic mellitus is very common metabolic disorders characterised by hyperglycaemia (elevated blood sugar) in middle as well as old age group due to lack of mainly physical activity in every social-economic groups. So many tests required for diagnosis as well as recordation of its controlling.

Saliva is a biological fluid containing highly complex, which is gaining popularity as a diagnostic tool. Saliva is a watery substance, which contains 99.5% of water and 0.5% of organic and inorganic substance. Salivary glands are located in all parts of mouth except gums and anterior part of the hard palate. 93% of saliva is produced by major salivary glands, and 7% of saliva is produced by minor salivary glands. Saliva production increases rapidly during meals. Saliva is a good indicator of plasma levels of various substances such as drugs and hormones, and it offers advantage over serum because it can be collected by non-invasive method. Saliva can be used as a diagnostic tool because it is a cost-effective approach for scanning large population.

AIMS AND OBJECTIVE:

To estimate and compare of serum and salivary glucose level in diabetic and non-diabetic patient and saliva can be used as a non invasive diagnostic fluid in Diabetic Mellitus patients.

MATERIALS AND METHOD:

This comparative cross-sectional study conducted in Department of Dentistry & orthopaedic with the collaboration of department of biochemistry, Government medical college Jalaun (Orai) U.P. In this study included 100 subjects, aged between 20-60 years including both genders, divided into two groups; Group- I: 50 Diabetic Mellitus patients (Type 1 and Type 2) Group-II: 50 normal and healthy individuals as controls, who visit and reported to regular outpatient department, from 14 July 2021 to 14 August 2021. The sample size was determined depending upon the average new dental & orthopaedic patients OPD presenting with medical conditions. The detailed case history and informed written consent was taken from each patients.

In our study included subjects with confirmed diagnosed cases of diabetic mellitus with the age group of 20 to 60 years and not suffering

from any other systemic disease except diabetic mellitus, those sample excluded who were under treatment and suffering from other disease, Salivary gland abnormality and surgeries, receiving radiotherapy, under long term local and systemic drug therapy except (Diabetes related drugs).

In case of serum glucose level, 2 ml fasting blood sample was withdrawn from the subjects under aseptic conditions from median cubital vein of forearm with a 5ml syringe and collected in sodium fluoride and potassium oxalate vial then after centrifuged at 2000 rpm for 5 minutes, when serum was separated for further analysis.

In same subject saliva was collected, before collection, advised to avoid food and fluid intake 2 hours prior to collection of saliva. The saliva was collected in early morning, resting position after rinsing the mouth with distilled water. The patient was instructed to spit in a sterile plastic container over a period of 5 minutes. The 2ml of un-stimulated whole salivary sample was collected from both groups, and sent to laboratory immediately. Collected saliva centrifuged at 2000 rpm for 5 minutes and then supernatants were obtained for analysis.

The serum and salivary glucose estimation was performed by auto-analyzer using glucose oxidase-peroxidase end-point method. Obtained reading was noted used for statistical analysis.

STATISTICAL ANALYSIS AND RESULTS:

A total of 100 subjects were selected in the age range of 20 – 60 years. In group 1 mean age 43.94 years and group 2 mean age value 44.64 years, both gender included (56 female and 44 male) in this study. Statistic calculation done by one way ANOVA student t test with Tukey HSD and found values as mentioned below tables.

Table1: Compare between Saliva and serum in group I

Comparison between Saliva & Serum in Group I (Control Group)			
	Serum	Saliva	Value
N	50	50	100
Mean	90.1	0.34	45.22
Std. Dev.	7.135	0.6263	45.3867
F value	7852.62524		
P value	<.00001 (Highly Significant)		

Table 2: Comparison between Saliva and serum in group II

Comparison between Saliva & Serum in Group II (Study Group)			
	Serum	Saliva	Value

N	50	50	100
Mean	188.32	4.52	96.42
Std. Dev.	70.9888	2.5891	105.0166
F value	334.73749		
P value	<.00001 (Highly Significant)		

Table 3: Comparison between Group I & Group II on the basis of Serum glucose Level in diabetic mellitus

SERUM GLUCOSE LEVEL IN DIABETIC MELLITUS			
	Group 1	Group 2	Value
N	50	50	100
Mean	90.1	188.32	139.21
Std. Dev.	7.135	70.98	70.396
F value	94.76005		
P value	<.00001 (Highly Significant)		

Table 4: Comparison between Group I & Group II on the basis of salivary glucose Level in diabetic mellitus

SALIVARY GLUCOSE LEVEL IN DIABETIC MELLITUS			
	Group 1	Group 2	Value
N	50	50	100
Mean	0.34	4.52	2.43
Std. Dev.	0.6263	2.5891	2.815
F value	123.11585		
P value	<.00001 (Highly Significant)		

DISCUSSION AND CONCLUSION:

Diabetes mellitus is a complex multi-system disorder characterized by a relative or absolute insufficiency of insulin secretion and/or concomitant resistance to the metabolic action of insulin. It is the most common endocrine disorder with potentially devastating complications that affects all age groups worldwide. Chronicity of the disease leads to its long term complications that are responsible for the high rates of morbidity and mortality.² Therefore, early diagnosis plays a substantial role in effective treatment. Currently, diagnosis of diabetes is achieved by evaluating plasma glucose levels. Monitoring blood glucose at frequent intervals causes discomfort and psychological trauma to patients; therefore, a much simpler and non-invasive technique for the diagnosis and monitoring of diabetes is desirable.³

In diabetic patients, the permeability of acinar cells of the major salivary glands increases. This provides an increase in the ultra-filtration of blood serum components such as glucose, amylase and blood proteins.⁴ Glucose is a small molecule that reaches saliva through the basement membrane of salivary glands by ultra filtration and also by way of the gingival sulcus and thus, glucose is expressed into the saliva.^{4,5}

Saliva is an organic bio-fluid that can indicate local and systemic alterations. The components of saliva can be related to the hormonal, immunologic, neurologic, nutritional and metabolic state of an individual.⁶

The increased level of glucose in saliva determines if an individual is diabetic. Therefore, saliva can be encouraged to be used as a diagnostic media.⁷ With the above background, this study was taken up to assess, if salivary glucose can serve as a reliable diagnostic indicator of diabetes mellitus. In this study, Mean age distribution in Group I was 43.94 years and that of group II was 44.64 years. This is in accordance with the study conducted by Sreedevi et al⁸ where similar mean age distribution was observed.

Estimation of mean fasting serum glucose and salivary glucose in group I, the observed mean value of fasting serum glucose level was 90.1 mg/dl and fasting salivary glucose was 0.34 mg/dl. (Table 1) The observed values were within the normal limits. Hence, were grouped as controls. After compare within the group 1, obtained <.00001 p value that's was <0.05 so it was highly significant.

Estimation of mean fasting serum glucose and salivary glucose in group II, the observed mean value of fasting serum glucose was 188.52 mg/dl and fasting salivary glucose was 4.52 mg/dl. (Table 2)

Fasting serum glucose – This is in accordance with Mirzaii-Dizgah I et al⁹, Patel BJ et al¹⁰, Shahbaz et al¹¹ where they conducted the study using fasting serum glucose. In contrast, Ponni V et al¹² conducted a

study to assess blood glucose level using gingival crevicular blood in diabetic patients. **Fasting salivary glucose** – this is in accordance with Ivanovski K et al⁴, Karki PC et al¹³, Abikshyeet et al¹⁴, Panchbhai et al¹⁵ where they also used fasting unstimulated saliva as a study sample. This study is in contrast with the study conducted by Jha SK et al³, Mirzaii-Dizgah I et al⁹ where fasting stimulated saliva was used as a study sample.

In diabetes mellitus, hyperglycemia is a result of absolute or relative lack of insulin and hepatic gluconeogenesis.⁴ Due to hyperglycemia, the advanced glycation end products (AGEs) cause changes in the microvasculature, basal membrane of salivary glands and oral mucosal tissues. This leads to easier diffusion of glucose from blood to saliva and also through gingival crevicular fluid. Hence, glucose is detected in saliva.^{14,16}

The detection of increased level of glucose in serum and saliva suggests that, the subjects are affirmed with suffer from diabetes mellitus.

Comparison of fasting serum glucose in group I and II, The mean fasting serum glucose in group I was 90.1 mg/dl and in group II was 188.52 mg/dl. The mean fasting serum glucose was found to be higher in group II than in group I. The observed difference was found to be highly statistically significant, ($P < 0.00001$). (Table 4) This is in accordance with the study conducted by Mirzaii-Dizgah I et al⁹, Patel BJ et al¹⁰, Shahbaz, et al¹¹ where they observed high serum glucose levels in the study group.

Comparison of fasting salivary glucose, the mean fasting salivary glucose in group I was 0.34 mg/dl and in group II was 4.52 mg/dl. The mean fasting salivary glucose was found to be higher in group II when compared to group I. The observed difference was found to be statistically highly significant, ($P < 0.00001$). (Table 3) This is in similar with the study conducted by Ivanovski K et al⁴, Karki PC et al¹³, Abikshyeet et al¹⁴, Panchbhai et al¹⁵ where they also found salivary glucose levels to be higher in diabetes compare to controls.

Saliva as a biological fluid has the potential to display substances existing in serum. Glucose, as a small molecule diffuses easily through semi-permeable membrane of the basement membrane of salivary glands, due to impaired permeability associated with diabetes thus, reaching the salivary secretion by ultra filtration. Glucose also diffuses easily through the membrane of the blood vessels, passing through the blood to the gingival fluid, by way of the gingival sulcus, and making its way to the saliva.^{4,5}

The presence of increased level of salivary glucose suggests that, the subjects are affirmed with diabetes. In such situation, this calls for further evaluation of the disease.

Correlation between fasting serum glucose and salivary glucose in group II, there was statistically significant correlation observed between fasting serum and fasting salivary glucose in group II, ($P < 0.00001$). (Table 2) This is supported by studies conducted by Abikshyeet et al¹⁴, Jha.S.K et al³, Agrawal RP et al¹⁷, Patel BJ et al¹⁰, Arora et al¹⁸, Shahbaz et al¹¹, where they also found significant correlation between fasting serum and salivary glucose. In contrast to our study Carolina et al and Forbat et al did not find significant correlation.¹⁹

Saliva as a biological fluid reflects blood constituents. Since, glucose is expressed both in serum and saliva of diabetic patients; one can use saliva as a sample in the diagnosis of diabetes mellitus. Diabetes mellitus (DM) is a metabolic disorder characterized by chronic hyperglycaemic condition resulting from defects in insulin secretion, insulin action or both. Diagnosis and monitoring of type II diabetes is achieved by evaluating blood glucose levels at frequent intervals causing discomfort and thus reduces the patient compliance. Hence, there is a splurge of interest in non-invasive diagnostic method which necessitates the need for other bodily fluids. Saliva reflects substances that are exist in blood serum. In diabetes mellitus, glucose molecule diffuses easily through semi-permeable membrane of the basement membrane of salivary glands, which occurs because of impaired permeability. Due to the effects of diabetic membranopathy, raised percolation of glucose occurs from blood to saliva. This expression of salivary glucose is a reflection of the existing blood glucose. Thus, saliva is prompted to be used as a diagnostic media in the diagnosis of diabetes mellitus.

This present study recommends the use of salivary glucose as a diagnostic aid to diagnose and monitor diabetes mellitus as it is noninvasive, easy to collect, expertise not required, economical, painless and also allowing multiple sampling. Therefore, we commend the use of salivary glucose as a reliable diagnostic indicator of diabetes mellitus.

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