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Original article:

Study of comparison of salivary glucose concentration, blood glucose and capillary blood glucose in diabetes mellitus cases in tertiary care hospital

Dr Charu Sharma[1], Dr R.J.Shendye[2] , Dr Vijendra Sharma[3], Dr Shubh Kiran Sharma[4]

[1]Dept of Biochemistry, BJGMC, Pune, currently working as Demonstrator in Dept of Biochemistry in GMC, Chandigarh

[2]Professor &HOD, Dept of Biochemistry, GMC, Aurangabad

[3, 4]PhD

Corresponding author: Dr Charu Sharma ; Email: drcharusharma05@gmail.com

Abstract:

Introduction: Monitoring blood glucose levels at frequent intervals can cause unnecessary discomfort and mental trauma to the patients, therefore a much simpler and non-invasive technique for diagnosis and monitoring of diabetes is quite desirable.

Material and methods: In the study group, we have selected 100 diabetic patients. We have also selected 100 healthy patients. As the patients came in the OPD, they were included in the study after carefully taking the history and according to the inclusion and exclusion criteria as controls. One hundred healthy individuals as control group and 100 individuals in study group who are known diabetics were included in study. Venous blood glucose and salivary glucose samples were processed in the department of Biochemistry. The capillary blood glucose estimation was done in the OPD itself by glucometer.

Results: In our study the mean salivary glucose levels in DM group was found to be 12.47±3.755 mg/dl and that of control group was 4.54±1.024mg/dl, ranging from a minimum value of 6.0 mg/dl to maximum of 21.0 mg/dl and median value was 11.8 mg/dl for DM group. The number of individuals in <40 years age bracket were 28 in number and in ≥ 40 years age bracket were 72 in number. All correlation "r" are highly significant as $p<0.001$

Conclusion: Saliva is one of the most abundant secretions in the human body, which can be obtained easily. In this study, we found a positive correlation between venous blood glucose and salivary glucose, as well as capillary blood glucose and salivary glucose. We also found that mean salivary glucose levels are higher in diabetes patients than that seen in the control group, and the higher levels are correlating with each other. Thus we propose that salivary glucose levels can be used as a non invasive modality for monitoring diabetes.

Keywords: Salivary glucose concentration, diabetes mellitus

Introduction:

Diabetes Mellitus is a group of metabolic disorders of carbohydrate metabolism in which glucose is underutilized at cellular level, producing hyperglycemia and glycosuria, reflecting a distortion in the equilibrium between utilization of glucose by the tissues and liberation of glucose by the liver. [1]The greatest increase in prevalence of diabetes is, occurring in developing countries including Asia and Africa, where most patients will probably be found by 2030. [2]This increase in new cases in the developing countries is because of the trend of urbanization and lifestyle changes, which includes increase in the sedentary lifestyles, less physically demanding work and the global nutrition transition, amounting to an increased intake of high energy-dense but nutrient-poor foods that are high in sugar and saturated fats (also called as western diet)[3]. The risk of getting type 2 diabetes has been found to be greatly associated with lower socio-economic position across countries [4]. According to the Indian

Heart Association, India is projected to harbor 109 million individuals with diabetes by 2035.[5]

The management of diabetes requires constant monitoring to maintain the optimal blood glucose levels in order to control the dreaded complications of the disease. The techniques conventionally used in monitoring glucose require venous/capillary blood sample and these procedures being invasive, elicit pain and discomfort which may even limit the frequency of testing. Therefore, noninvasive methods using other body fluid such as saliva has been of particular interest and can lead to tighter control on glucose levels. [6]

Saliva is one of the most abundant secretions in the human body and its collection is easy and noninvasive [7] Estimation of salivary glucose may provide an alternative, non-invasive approach in monitoring of glucose in diabetic patients.

The objective of this study was to find the correlation between the fasting blood sugar (both venous and capillary) and the fasting salivary level and to evaluate its role as a potential tool for monitoring of blood glucose.

Material and methods:

The present prospective study was carried out after the ethical approval from the Institutional Ethical Committee was received. In the study group, we have selected 100 diabetic patients and 100 healthy patients as controls.

The sample size was estimated using sample size estimation calculator.

As the patients came in the OPD, they were included in the study after carefully taking the history, informed consent and according to the inclusion and exclusion criteria.

Inclusion criteria:

1. 100 healthy individuals as control group
2. 100 individuals in study group who are known diabetics

Exclusion criteria:

1. Patients suffering from any salivary gland disease.
2. Patients who have underwent salivary gland surgery.
3. Patients on radiotherapy.
4. Patients with oral cancer or with the previous history of oral cancer.
5. Patients with Xerostomia
6. Patients with any other oral pathology.

Venous blood glucose and salivary glucose samples were processed in the department of Biochemistry. The capillary blood glucose estimation was done in the OPD itself using glucometer.

The venous blood glucose samples were processed in the Department of Biochemistry on XL640 Autoanalyzer. The samples were collected in the collection centre in OPD from fasting patients. The samples were centrifuged in order to separate the plasma. After the proper separation of plasma the sample were processed in the autoanalyzer within 2 hours of sample collection.

Capillary samples were processed by method of dry chemistry using Dr Morepan glucometer.

Salivary Estimation:

Subjects were asked to rinse the mouth with water and wait for 5 minutes. They were asked to spit saliva into the container vial. Around 2 ml of non stimulated saliva sample was taken from each subject. It was confirmed prior that the subjects were fasting. The samples were collected in the OPD. The samples were centrifuged within 30 minutes of the collection. The samples were processed on Erba Chem-5 Plus v2 semi autoanalyzer, within 2 hours of the collection.

Salivary glucose was estimated using following formula:

$$\text{Salivary Glucose} = \frac{\text{absorbance of test}}{\text{absorbance of the standard}} \times \text{concentration of standard (mg/dl)}$$

The unit of measurement is mg/dl

The statistical analysis was carried out on SPSS software version 19.

Results:

A total number of 200 subjects were studied, out of which 100 were known diabetics and 100 were non-diabetics treated as control. The number of females in DM group was 37 and that of control group was 41. The number of males in DM group was 63 and that in control group were 59.

The minimum age in both the groups was 18 years and maximum age in DM group was 84 years while that in control group was 73 years. The average age in DM group was 50.95 years with standard deviation of 15.282. The mean age in control group was 39.37 years with standard deviation of 14.212 (table 1).

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Table1. Age distribution for diabetic and non diabetic

Age in yrs	Mean age in yrs	SD	Unpaired t	P
DM group	50.95	15.282	5.549	<0.001 HS
Control group	39.37	14.212		

The unpaired‘t’ test value is 5.549. All values were found to be highly significant as the p value is <0.001.

A. Fasting venous blood glucose levels (mg/dl)

The average fasting venous blood glucose found in the DM group was 168.53mg/dl with standard deviation of 46.850. The average fasting venous blood glucose in control group was 86.51mg/dl with SD of 9.667 (table2). The unpaired ‘t’ test value was 16.152.

Table 2. Fasting venous blood glucose levels (mg/dl)

Venous blood glucose	Mean	SD	Unpaired t	P
DM group	168.53	49.850	16.152	<0.001 HS
Control group	86.5	9.667		

The minimum value of fasting venous blood glucose was 80 mg/dl and the maximum value estimated was 349 mg/dl in the DM group. While the minimum value of fasting venous blood glucose in the control group was estimated to be 64 mg/dl and maximum value was estimated to be 109mg/dl. The median values (middle point of a number set) for the DM group was 155.5mg/dl and that for control group was 88.0 mg/dl.

B. Capillary blood glucose in diabetics and non diabetics:

The average fasting capillary blood glucose found in the DM group was 176.50mg/dl with standard deviation of 50.371. The average fasting capillary blood glucose in control group was 93.73 mg/dl with SD of 9.359 (table3). The combined P value was <0.001 which is highly significant.

Table 3. Capillary blood glucose in diabetics and non diabetics

Capillary blood glucose	Mean	SD	Unpaired ‘t’	P
DM group	176.50	50.371	16.156	<0.001 HS
Control group	93.73	9.359		

The minimum value of fasting capillary blood glucose was 87 mg/dl and the maximum value estimated was 356 mg/dl in the DM group. The minimum value of fasting capillary blood glucose in the control group was 72 mg/dl and maximum value was estimated to be 113 mg/dl. The median value for the DM group was 163.5 mg/dl and that for control group was 94.0 mg/dl

C. Salivary glucose (mg/dl) in diabetics and non diabetics:

The mean fasting salivary glucose estimated in the DM group was 12.47 mg/dl with standard deviation of 3.755. The average fasting salivary glucose in control group was 4.54 mg/dl with SD of 1.024 (table 4). The P value was <0.001 which is highly significant.

Table 4 Glucose(mg/dl) in diabetics and non diabetics

Salivary Glucose	Mean	SD	Unpaired T	P
DM group	12.47	3.755	20.365	<0.001 HS
Control group	4.54	1.024		

The minimum value of fasting salivary glucose in the control group was 2.0 mg/dl and maximum value estimated was 6.8 mg/dl. The minimum value of fasting salivary glucose was 6.0 mg/dl and the maximum value observed was 21.0 mg/dl in the DM group. The median value (central value) for the DM group was 11.8 mg/dl and that for control group was 4.8 mg/dl.

D.Correlation between venous blood glucose, capillary blood glucose and salivary glucose:

Table 4 Correlation between venous blood glucose, capillary blood gluose and salivary glucose

All cases	Venous glucose Vs Capillary glucose	Venous glucose vs Salivary glucose	Capillary glucose Vs Salivary glucose
DM gp	0.996	0.925	0.926
Control gp	0.930	0.579	0.598

The coefficient of correlation “r” for venous blood glucose and capillary blood glucose for DM group was r=0.996, while that for control group was r=0.930. The coefficient of “r” for venous blood glucose and salivary glucose for DM group is equal to 0.925 and that for the control group is r=0.579. The correlation between Capillary blood glucose and salivary glucose in the study was r=0.926 and that of

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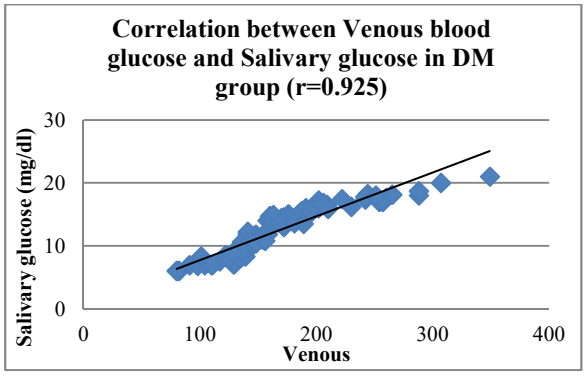
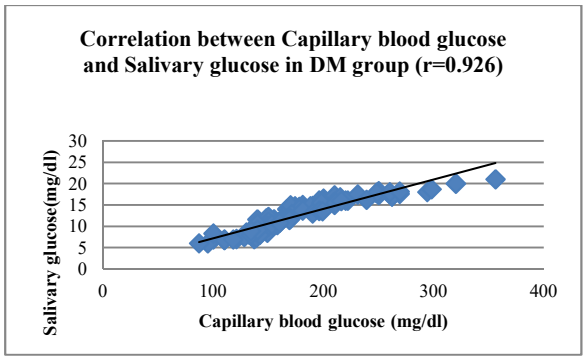
control group was r= 0.598. All correlation "r" are highly significant as p<0.001



Graph 1 shows the correlation between venous blood glucose and salivary glucose (r=0.925)



Graph 2 denotes the correlation between capillary blood glucose and salivary glucose in the DM group where the coefficient of correlation was seen to be r=0.926. All correlation were found to be highly significant as p<0.001

E. Correlation between venous blood glucose, capillary blood glucose and salivary glucose in age group <40 years age and ≥40 years age in dm group

Table 5 Correlation between venous blood glucose, capillary blood glucose and salivary

	Venous blood glucose Vs Capillary blood glucose	Venous blood glucose Vs Salivary glucose	Capillary blood glucose Vs Salivary glucose
DM gp (n=28) **Age <40 yrs**	0.989	0.943	0.935
DM gp (n=72) **Age ≥40 yrs**	0.997	0.928	0.930

The table 5 shows us the correlation between the venous blood glucose, capillary blood glucose and salivary glucose in subjects confirming in <40 years age and ≥40 years age in both DM group. The number of individuals in <40 years age bracket were 28 in number and in ≥ 40 years age bracket were 72 in number.

All correlation "r" are highly significant as p<0.001.

Discussion:

Monitoring the glucose is an integral part of management of diabetes and preventing the complications associated with it. The modes of diagnosis and management of diabetes have evolved over the years from urine sugar testing to various POCT (point of care testing) devices like glucometer that measure capillary blood glucose. Various studies have shown the significant correlations between venous and capillary blood glucose.

Glucose can be found in various biological fluids other than blood, like CSF and saliva. Normally the saliva contains glucose in small quantities. Primary objective of this study was to evaluate whether salivary glucose is correlating with venous/ capillary glucose and whether it can be used as POCT.

In our study the mean salivary glucose levels in DM group was found to be 12.47±3.755 mg/dl and that of control group was 4.54±1.024 mg/dl, ranging from a minimum value of 6.0mg/dl to maximum of 21.0 mg/dl and median value was 11.8 mg/dl for DM group. The median values for control group was 4.8 mg/dl.There was a significant rise in mean salivary glucose in patients of DM as compared to salivary glucose levels of control group (p<0.001). This indicates that salivary glucose in patients with diabetes is higher than that compared to the salivary glucose in the control group. Gupta Shruti et al in 2015 [8], undertook a study to estimate glucose levels of saliva and to assess its correlation between serum and salivary glucose levels with regard to the duration of diabetes, age, gender, serum and salivary glucose of 200 subjects of which 100 were diabetics and 100 were control group. The study revealed that a significant correlation is observed between salivary glucose level and gender or age in both diabetic and non-diabetic subjects and duration of diabetes in the diabetic group. The study

conducted by Sener et al in 2009[9] revealed that salivary glucose concentration in diabetic patients is higher than in control group. Our results coincided well with the works of Sener et al (2009); Gupta Shruti et al (2015).[8,9]

Further we found a significant correlation between venous blood glucose and salivary glucose in both DM group as well as controls. The coefficient of correlation between venous blood glucose and salivary glucose for DM group was r=0.925 and that of the control group was r=0.579, although the coefficient of correlation for venous blood glucose and salivary glucose is not as high for the control group as seen with the DM group, the p value was <0.001, which shows the correlation to be highly significant. Azizi et al in 2013[60] convened the study to find a relation between blood glucose and saliva glucose level in diabetic patients. The study was undertaken on 75 diabetic patients as case and 75 healthy individuals as control. A significant correlation of blood glucose in diabetic and control subjects with salivary glucose was found in the study. The result of our study are in consonance with the results of Satish et al (2013)[10]; Panchbhai et al(2012)[11]. However study by Forbat et al[12] concluded that blood glucose levels do not correlate well with the salivary glucose levels. Gupta Anjali et al (2015)[8] conducted a study with a total number of 250 patients. The study was conducted to evaluate correlation between blood glucose and salivary glucose levels in diabetic individuals and healthy individuals revealing the p value to be 0.247 confirming the results to be non significant which are not in consonance with our results.

We also evaluated the capillary blood glucose and the salivary glucose for DM group as well as the control group and estimated the correlation for the same. The coefficient of correlation (r) between capillary blood glucose and salivary glucose in DM group was 0.926 and in the control group was observed 0.598. This shows a positive correlation between the capillary blood glucose and the salivary glucose in both the groups and the p value is <0.001 making it highly significant. Our results supports the study convened by Puttaswamy et al[13] .

In the present study, we found a positive correlation between venous blood glucose and salivary glucose, as well as the capillary blood glucose and venous blood glucose in the DM group as well as control group. The coefficient of correlation (r) between venous blood glucose and salivary glucose in DM group was 0.925 and in the control group was 0.579. We also found that mean salivary glucose levels are higher in diabetics than that of control group. The coefficient of correlation (r) between venous blood glucose and capillary blood glucose in DM group was 0.996 and in the control group was 0.930 (Table5)

Conclusion:

Saliva is one of the most abundant secretions in the human body, which can be obtained easily. In this study, we found a positive correlation between venous blood glucose and salivary glucose, as well as capillary blood glucose and salivary glucose. We also found that mean salivary glucose levels are higher in diabetes patients than that seen in the control group, and the higher levels are correlating with each other. Thus we propose that salivary glucose levels can be used as a non invasive modality for monitoring diabetes. An appropriate device, if developed for this purpose monitoring of salivary glucose levels can be used as point of care testing technique.

Study Limitations:

The present study was done on the subjects attending the OPD. Sufficient data is not available regarding the critically ill patients. The flow of saliva, sampling of saliva and the value of salivary glucose in critically ill patients still remains unexplored. Further studies and analysis needs to be done to see the correlation of blood and salivary glucose in critically ill patients.

The mean salivary glucose levels in various studies have been found to be less than 20 mg/dl. Till the time an appropriate device for estimation of salivary glucose, such as glucometer used for capillary blood glucose (glucometer is not sensitive below 20 mg/dl), is made, this technique may not be freely used as point of care testing.

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