Gingival crevicular blood glucose in chronic periodontitis patients

Sunisa Intaranonvilai[1], Ananya Promsudthi[2]

[1] Department of Dentistry, Thatakiab Hospital, Chachoengsao, Thailand

[2] Department of Oral Medicine and Periodontology, Faculty of Dentistry, Mahidol University, Bangkok, Thailand

Abstract

Objective: To evaluate the potential use of gingival crevicular blood for glucose measurement in comparison to capillary blood glucose and fasting plasma glucose in chronic periodontitis patients with and without diabetes (DM).

Materials and methods: Chronic periodontitis patients with DM (CP+DM) and without DM (CP+nonDM), n=26 in each group, participated in this study. Venous bloods from participants were analyzed for fasting plasma glucose (FPG) in laboratory. Gingival crevicular blood glucose (GCBG) and capillary blood glucose (CBG) measurements were obtained by glucometer. Each FPG measurement was converted to corrected venous glucose (cFPG) and used in statistical analysis. Analysis of agreement by Bland and Altman was used to analyze agreement between GCBG-CBG and GCBG-cFPG measurements.

Results: In the CP+DM and CP+nonDM groups, the limits of agreement were considerably wide for GCBG-CBG (-0.70, 1.77 and -0.87, 1.49) and GCBG-cFPG (-1.21, 1.78 and -0.90, 1.30) measurements, respectively. The results suggested that there were discrepancies between GCBG and CBG measurements as well as GCBG and FPG measurements in both groups.

Conclusions: Within the limitations of the present study, a good agreement between GCBG-CBG and GCBG-FPG measurements was not observed. Gingival crevicular blood may not be suitable for blood glucose evaluation in dental office in untreated chronic periodontitis patients.

Key words: bleeding on probing, chronic periodontitis, dental office, diabetes, gingival crevicular blood, glucose level

How to cite: Intaranonvilai S, Promsudthi A. Gingival crevicular blood glucose in chronic periodontitis patients. M Dent J 2016; 36: 317-327.

Corresponding author:
Ananya Promsudthi
Department of Oral Medicine and Periodontology, Faculty of Dentistry, Mahidol University,
6 Yothi Street, Rajthevi, Bangkok 10400, Thailand
Tel: 02-2007841
Fax: 02-2007840
email: ananya.pro@mahidol.ac.th

Received: 14 July 2016
Accepted: 21 November 2016

Introductions

Chronic periodontitis is an infectious disease resulting in inflammation within the supporting tissues of the teeth, progressive attachment loss, and bone loss and is characterized by pocket formation and/or recession of the gingival.[1] Diabetes is a risk factor of chronic periodontitis.[2, 3] The prevalence, severity, and progression of periodontal disease among periodontitis patients with diabetes are higher than patients without diabetes. In addition, the prevalence and severity of periodontal disease of poorly controlled diabetes are higher than patients with well controlled diabetes.[4, 5] The mean percentage of teeth with at least one site with a probing depth of ⩾ 5 mm was significantly higher in persons with diabetes/ prediabetes than persons with normoglycemia.[6] Patients with diabetes have an increased risk of periodontal tissue destruction comparing to patients without diabetes when measured by attachment loss and bone loss with an odds ratio of 2.81 and 3.43 respectively. In other words, diabetes patients have a higher risk of developing periodontitis.[7]

Analysis of data from the National Health and Nutrition Examination Survey (NHANES) of the United States in 2003-2004 found that a total of 62.9 percent of persons without periodontitis and 93.4 percent of those with periodontal disease met the American Diabetes Association (ADA) guidelines for diabetes screening. Of those at risk of diabetes with periodontal disease, 33.9 percent had seen a dentist in the past 6 months, 50 percent in the past year, and 60.4 percent in the past 2 years. It can be concluded that many at risk of diabetes persons with periodontal disease recently visited a dentist.[8] Besides assessing perioperative glucose level to decrease the risk of adverse hypoglycemic events, Mealey suggested that the evaluation of capillary blood glucose (CBG) in the dental clinic has the potential for screening patients at risk of diabetes or prediabetes. However, dentists should be knowledgeable about diabetes and be able to correctly interpret the results.[9]

During periodontal examination, probing into inflamed gingival sulcus can cause blood oozing from the sulcus, so called gingival crevicular blood. Sites with inflamed gingival tissue and bleeding on probing (BOP) are the source of gingival crevicular blood which has been suggested to be used for blood glucose measurement.[10] However, the volume of the gingival crevicular blood must be sufficient upon probing and the blood should be free of contamination from contact with the teeth and gingiva.[11] Obtaining gingival crevicular blood is less traumatic and invasive than obtaining capillary blood by using sharp object to puncture the finger tip.[12, 13] Study found that patients preferred gingival crevicular blood glucose (GCBG) measurement (51 percent) to capillary blood glucose (CBG) measurement (31.4 percent). Most of the correspondents indicated that the collection of gingival crevicular blood felt like a routine dental cleaning.[14] Beikler et al. examined GCBG and CBG in moderate to severe periodontitis patients with diabetes and without diabetes and found that glucose levels obtained by the two methods had high intrapatient correlation (r = 0.98).[12] In addition, high positive correlations between GCBG and CBG in groups of patients with and without type 2 diabetes have been reported.[15, 16] Also, a study conducted in patients with diabetes found high correlations between GCBG and CBG, GCBG and corrected fasting plasma glucose (cFPG), and CBG and cFPG.[17] Therefore, the gingival crevicular blood has been suggested to be utilized as a source of blood for blood glucose level measurement.[12, 15-17]

In contrast, Muller and Behbehani found low agreement between GCBG and CBG in gingivitis and periodontitis patients with diabetes

and without diabetes.[18] Sarlati et al. reported sufficient agreement only between GCBG and CBG in persons with diabetes and failed to prove sufficient agreement between the paired methods of GCBG, CBG, and FPG in persons without diabetes. Gingival crevicular blood was suggested to be used for blood glucose measurement during periodontal examination only in persons with diabetes.[19] However, another study did not support the application of gingival crevicular blood for blood glucose measurement in either persons with or without diabetes since there was a significant difference between GCBG and CBG in both groups.[20] Controversy remains over the utilization of gingival crevicular blood for blood glucose measurement. Certainly, the collection of gingival crevicular blood during periodontal examination is non-invasive and painless. Simple non-invasive screening test, available in dental clinic, would be preferred especially in needle phobia patients and might provide early diabetes diagnosis. Chronic periodontitis patients could benefit from diabetes screening during periodontal examination if gingival crevicular fluid is suitable for blood glucose measurement. This study aimed to evaluate the potential use of gingival crevicular blood for glucose reading in comparison to capillary blood glucose and fasting plasma glucose in chronic periodontitis patients with and without diabetes.

Materials and methods

Study population

The study population was recruited from persons attending the Dental Department and/or the Diabetic Clinic, Bangkruai Hospital, Thailand. Persons diagnosed as chronic periodontitis according to the 1999 classification system of the American Academy of Periodontology (AAP)[21] after full mouth periodontal examinations. Persons with type 2 diabetes were diagnosed by the physicians according to the American Diabetes Association.[22] Persons without diabetes were persons who had fasting plasma glucose test results less than 7 mmol/L and without any diabetic symptoms. The following exclusion criteria were also applied: history of periodontal treatment within 6 months of study entry, any indications for antibiotic premedication, abnormal low or high hematocrit such as polycythemia vera, anemia, dialysis, systolic blood pressure < 90 mmHg, intake ascorbic acid in 2 days or acetaminophen in 3 days, Parkinson's disease, severe systemic disease such as severe cardiovascular, renal, hepatic, and immunologic disorders, type 1 diabetes and gestational diabetes. Participants were aged match and divided into two groups: patients with chronic periodontitis and type 2 diabetes (CP+DM) and patients with chronic periodontitis and no diabetes (CP+nonDM).

Using Altman's nomogram and standard deviation of difference from previous study,[19, 23] it was decided to have at least 25 persons in each group in order to have a 85% chance of detecting a 1 mmol/L difference in means between glucometer tested blood glucose at the 5% level of significance. The study protocol was approved by the Faculty of Dentistry/Faculty of Pharmacy, Mahidol University, Institutional Review Board (MU-DT/PY-IRB 2014/DT009). Participant information sheets were provided and explained to all persons. All participants provided written informed consent.

Data collection

Participants' age, gender, and body mass index (BMI; weight divided by the square of height, kg/m^2) were collected.

Periodontal examination

All participants received a full mouth periodontal examination by one examiner (S.I.) and the following clinical periodontal parameters were measured using calibrated periodontal

probe (PCPUNC15; Hu-Friedy, Chicago, IL, USA) and recorded on six sites per tooth: probing depth, clinical attachment level, and BOP. Probing depth was measured (to the nearest millimeter) from the gingival margin to the bottom of the pocket. Clinical attachment level was defined as the distance from cemento-enamel junction to the bottom of pocket. Evaluation for BOP was performed by gentle probing along the buccal and lingual gingival crevices, sites that bled within 10 s were recorded as BOP-positive (BOP+).[24]

Determination of plasma/blood glucose

Fasting plasma glucose (FPG)

Venous blood samples were collected from vein at antecubital fossa and assayed for FPG using hexokinase assay[25] at the Bangkruai Hospital laboratory.

Gingival crevicular blood glucose (GCBG)

Gingival crevicular blood was collected from a site with gingival inflammation, probing depth more than 3 mm and no suppuration. The tooth was polished with pumice, rinsed and dried. Gauze was used to isolate the tooth from lip and tongue to prevent contamination of gingival crevicular blood with saliva. Periodontal probe was penetrated into gingival crevice, and then the oozing gingival crevicular blood was collected with the test strip inserted in glucometer (Fora POCT S10; ForaCare Inc., Moorpark, CA, USA) for GCBG measurement. The GCBG of the same sites were measured twice. The GCBG measurements were done within 30 minutes of venous blood collection.

Capillary blood glucose (CBG)

Capillary blood was collected from the finger-tip using single-use disposable lancet. The first drop of blood was discarded; the second drop of blood was applied to the test strip inserted in glucometer for CBG measurement. Also, the CBG measurements were done within 30 minutes of venous blood collection.

Statistical analysis

Data were analyzed using a software program (SPSS for Windows, version16; SPSS Inc., Chicago, IL, USA). Descriptive statistics was used to present patient's characteristics. Intraexaminer reliability of GCBG measurement was determined by Intraclass Correlation Coefficient (ICC). The independent *t*-test was used for comparison of clinical periodontal parameters between two groups.FPG was converted to corrected venous glucose by using the following formula: corrected venous glucose (cFPG) = laboratory (mmol/L) × [1.0 - (0.0024 × Hct)] + 0.2 mmol/L,[13, 17] and used in further statistical analysis. Relationship between GCBG-CBG and GCBG-cFPG was displayed with scatterplot and line of equality. Analysis of agreement by Bland and Altman was used to analyze agreement between the measurements of GCBG-CBG and GCBG-cFPG.[26]

Results

Fifty two patients, consisting of 26 patients in each group, participated in this study, 12 males and 14 females in the CP+DM group and 5 males and 21 females in the CP+nonDM group. No statistically significant difference in the mean age between 2 groups has been observed. CP+DM group had mean age 57 years with FPG in the range of 4.61 to 26.00 mmol/L. Diabetic control of this group consisted of oral medications in 23 patients, oral medications and insulin injection in 2 patients, and exercise and diet control in 1 patient. CP+nonDM group had mean age and standard deviation of 57.15 ± 8.67 years with FPG ranging from 4.83 to 6.83 mmol/L. The mean, the standard deviation and the range of GCBG, CBG and FPG of CP+DM and CP+nonDM groups are shown in Table 1. Table 2 shows BMI and clinical periodontal parameters in CP+DM and CP+nonDM groups. CP+DM had

Table 1 The mean, standard deviation and range of glucose level assessment (mmol/L)

	CP+DM (*n*=26)			CP+nonDM (*n*=26)		
	GCBG	CBG	FPG	GCBG	CBG	FPG
Mean	9.05	8.51	9.34	5.45	5.15	5.55
Standard Deviation	3.85	3.75	4.23	0.59	0.54	0.47
Minimum	4.33	4.06	4.61	3.58	4.50	4.83
Maximum	23.58	22.61	26.00	6.72	6.89	6.83

GCBG, gingival crevicular blood glucose; CBG, capillary blood glucose; FPG, fasting plasma glucose; CP+DM, chronic periodontitis with diabetes group; CP+nonDM, chronic periodontitis with no diabetes group

Table 2 Body mass index (BMI) and clinical periodontal parameter (mean±SD) in chronic periodontitis with diabetes (CP+DM) groupand chronic periodontitis with no diabetes (CP+nonDM) group

	CP+DM (n=26)	CP+nonDM (n=26)	P-value*
BMI (kg/m2)	26.67 ± 6.32	24.99 ± 4.34	N/A
Probing depth (mm)	3.54 ± 0.85	3.13 ± 0.51	0.039
Clinical attachment level (mm)	4.76 ± 1.68	3.93 ± 0.70	0.025
Bleeding on probing (%)	80.88 ± 14.59	74.13 ± 19.43	0.16

* *t*-test

statistically significant deeper probing depth and clinical attachment level than CP+nonDM (P = 0.039 and P = 0.025, respectively). There was reliability of GCBG measurement as shown by intraclass correlation coefficient, which was 0.993 with 95% CI (0.985, 0.997, P = 0.00) in CP+DM group, and 0.773 with 95% CI (0.556, 0.892, P = 0.00) in CP+nonDM group.

Figure 1. shows the scatterplot of GCBG and CBG, and GCBG and cFPG with the line of equality in CP+DM and CP+nonDM groups. From the illustration, the majority of GCBG values were under the line of equality except for the plot of GCBG and cFPG in CP+nonDM group. Figures 2. and 3. show Bland and Altman plot demonstrating plot of differences against means of GCBG-CBG measurements and GCBG-cFPG measurements with lines of means and upper and lower limits of agreement in CP+DM and CP+nonDM groups. In the CP+DM group, the mean difference and standard deviation of GCBG and CBG measurements was 0.54 ± 0.62mmol/L with the limits of agreement of -0.70 and 1.77. The mean difference and standard deviation of GCBG and cFPG measurements was 0.29 ± 0.75mmol/L with the limits of agreement of -1.21 and 1.78 (Table 3). In the CP+nonDM group, the mean difference and standard deviation of GCBG and CBG measurements was 0.31 ± 0.59mmol/L with the limits of agreement of -0.87 and 1.49. The mean difference and standard deviation of GCBG and cFPG measurements was 0.20 ± 0.55 with the limits of agreement of -0.90 and 1.30 (Table 3). The maximum negative and positive differences of each analysis of agreement were shown in Table 3.

Discussion

The objective of this study was to evaluate the potential use of gingival crevicular blood for blood glucose measurement by comparing GCBG measurement with CBG and FPG measurements in CP+DM and CP+nonDM persons.

This study was designed to control factors influencing blood glucose measurements, and therefore, blood collections from veins, capillaries and gingival crevicular crevices were done within thirty minutes to minimize error from glucose metabolism. Also, the FPG were assayed in single laboratory.

The CP+DM group had mean BMI of 26.67 kg/m^2 which is considered as obesity, while CP+nonDM group had mean BMI of 24.99 kg/m^2 which is classified as overweight for Asian populations.[27] CP+DM group had deeper probing depth and more attachment loss compared with age-matched CP+nonDM group. These results are in accordance with a previous study that reported that patients with diabetes had more attachment loss than patients without diabetes.[7, 28]

Previous studies reported high correlation between GCBG and CBG in patients with and/or without diabetes[12, 15-17] and between GCBG and cFPG.[17] However, high correlation does not mean acceptable agreement between measurement methods. Agreement between measurement

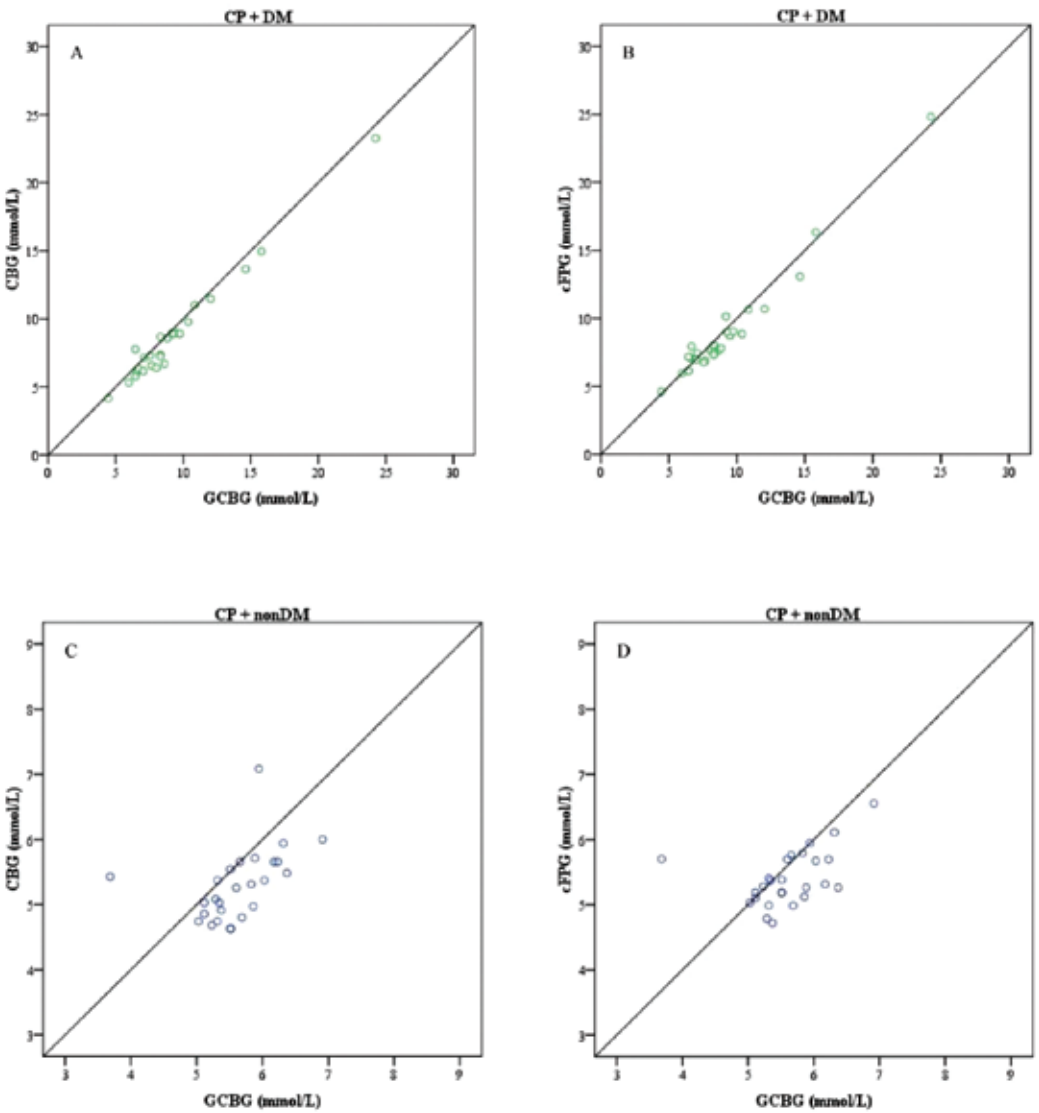

Figure 1 Scatterplots of GCBG and CBG (A), GCBG and cFPG(B) with the line of equality in chronic periodontitis with diabetes (CP+DM) group and scatterplots of GCBG and CBG (C), GCBG and cFPG(D) with the line of equality in periodontitis with no diabetes (CP+nonDM) group.

methods means the values from two methods of measurement are equal and should be on the line of equality which has slope equal to one and no interception. However, it is very unlikely that the different measurement methods will give identical result. Therefore we look at how much the difference between methods is. Most of the differences should lie between mean difference ± 1.96 standard deviation of the difference (limits of agreement). The differences within the limits of agreement should not be clinically important in order to use the two measurement methods interchangeably.[26] While high correlation between values means strong linear relationship between the values of measurements, the values lie along any straight

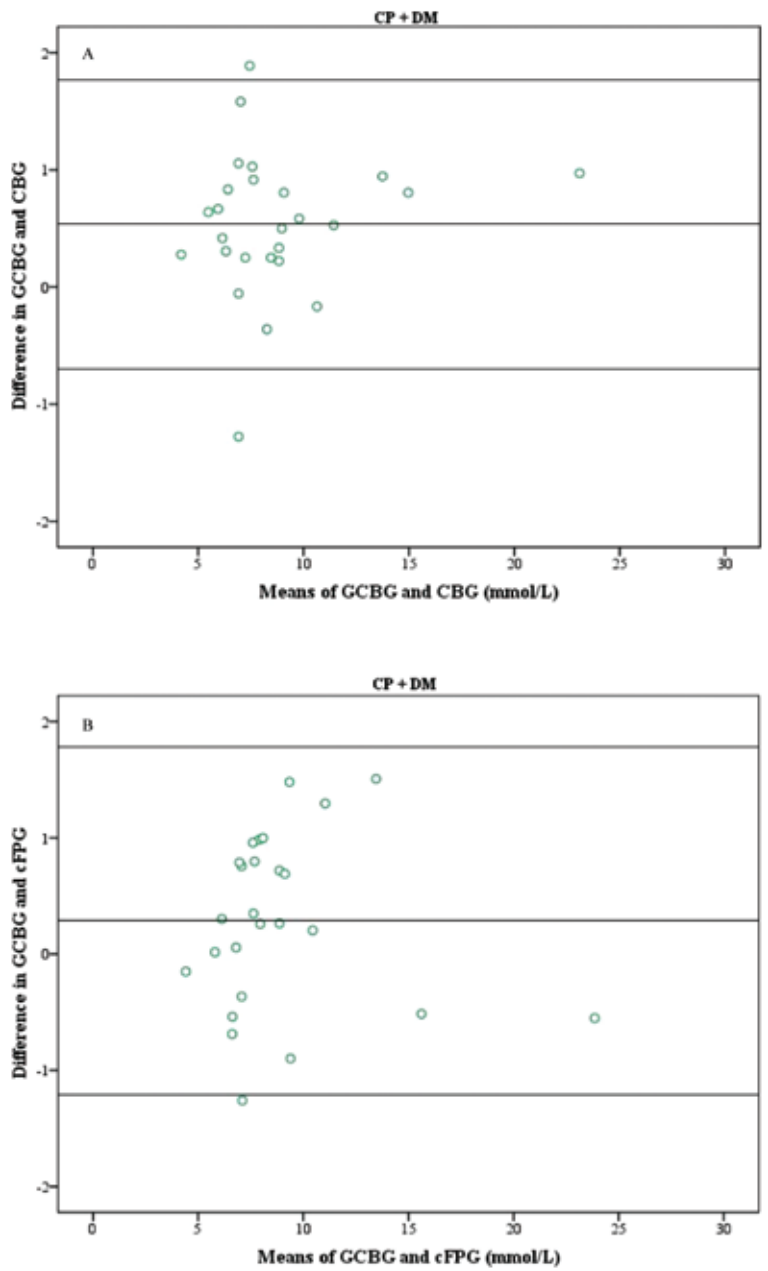

Figure 2 Difference against mean of GCBG and CBG (A) and GCBG and cFPG (B) with lines representing the mean difference and the upper and lower limits of agreement in chronic periodontitis with diabetes (CP+DM) group.

line with any slope, and the values from two methods of measurement can be unequal. The high correlation can conceal considerable lack of agreement between the two methods of measurement.[26] Therefore, in order to evaluate the potential use of gingival crevicular blood for glucose level assessment interchangeably with capillary blood, we used Bland and Altman analysis of agreement to analyze the data in the present study.

The mean differences between GCBG-CBG as well as GCBG-cFPG in CP+DM group were 0.54 and 0.29 mmol/L respectively, and in CP+nonDM were 0.31 and 0.20 mmol/L respectively. Even though the mean differences were not much when the limits of agreement

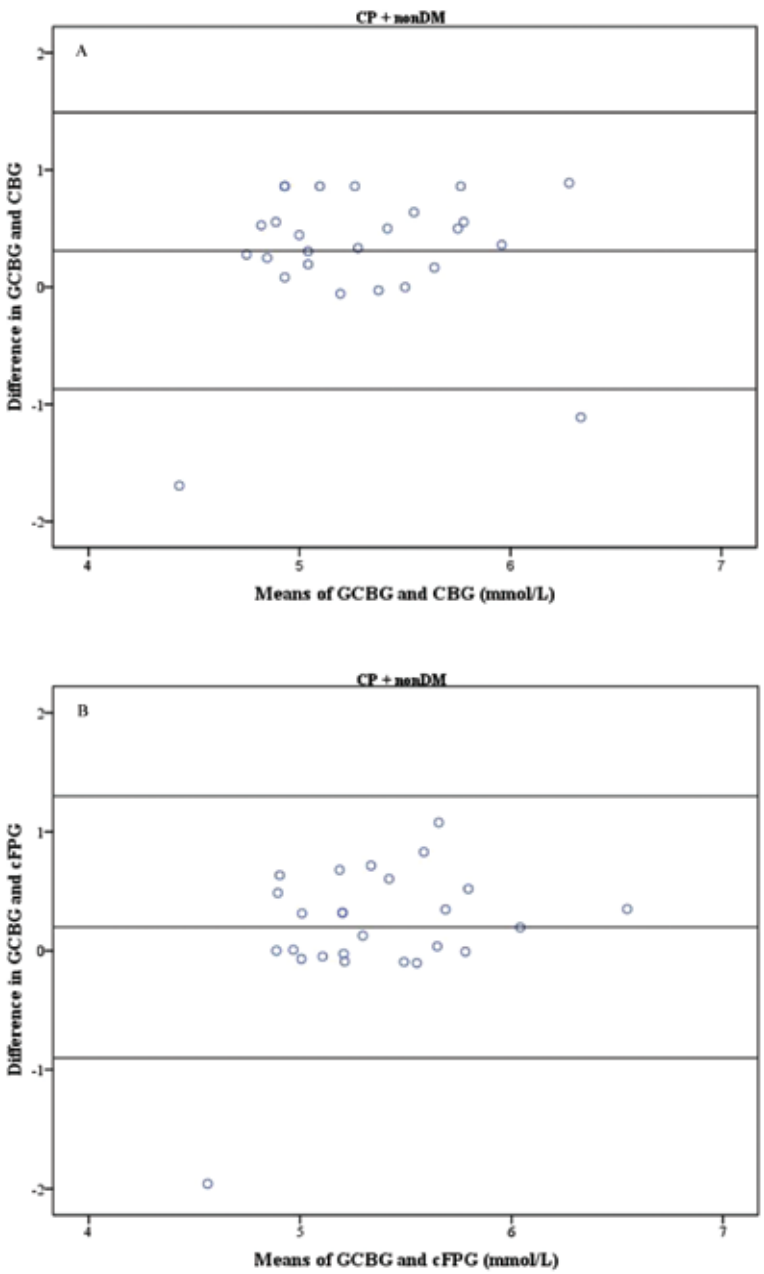

Figure 3 Difference against mean of GCBG and CBG (A) and GCBG and cFPG (B) with lines representing the mean difference and the upper and lower limits of agreement in chronic periodontitis with no diabetes (CP+nonDM) group.

of GCBG-CBG as well as GCBG-cFPG in both groups were considered, the intervals were considerably wide (-0.70, 1.77 mmol/L and -1.21, 1.78 mmol/L respectively in CP+DM group and -0.87, 1.49 mmol/L and -0.90, 1.30 mmol/L respectively in CP+nonDM group). The degrees of agreement are not acceptable due to wide interval (2.2 – 3.0 mmol/L) which could have clinical impact. The results suggested that there were discrepancies between GCBG and CBG measurements as well as GCBG and FPG measurements in both groups.

Considering the wide interval (2.2 – 3.0 mmol/L) of the limits of agreement of GCBG-CGB as well as GCBG-cFPG in both groups, it is unacceptable to use GCBG interchangeably with CGB or FPG. Gingival crevicular blood may not be suitable for blood glucose measurement even though the collection of gingival crevicular blood is a non-invasive and painless.

Further statistical analysis found that there was statistically significant higher mean GCBG than mean CBG in CP+DM and CP+nonDM groups (paired t test, P=0.00 and P=0.013, respectively). Similar result has been reported by previous study.[20] Persons with chronic periodontitis have increased gingival crevicular fluid compared to persons with gingivitis or healthy gingiva[29, 30] due to higher inflammation of the periodontium. Gingival crevicular fluid is an inflammatory exudate which reflects the composition of serum. Previous study reported that glucose level was detected in gingival crevicular fluid; moreover, glucose concentration in gingival crevicular fluid of persons with diabetes was higher than that of persons without diabetes.[31] Therefore, gingival crevicular fluid may contribute to the discrepancies between GCBG and CBG due to the combination of glucose in gingival crevicular fluid and gingival crevicular blood in the CP+DM and CP+nonDM groups. Because gingival crevicular fluid increases with severity of gingival inflammation, patients with more severe periodontitis may have greater discrepancies between GCBG and CBG than patients with less severe periodontitis. Initial periodontal treatments can eliminate gingival inflammation with significant decrease in gingival crevicular fluid.[32, 33] Whether or not there is significant difference between GCBG and CBG after initial periodontal therapy has not been reported.

In order to assess agreement between two methods of measurement, usually the measurements of different methods should be made in random order,[26] however, blood collections for glucose measurements in the present study were not done in random order. This posed a limitation of the present study since the venous blood was collected first and followed by the collections of gingival crevicular blood and capillary blood.

Table 3 Analysis of agreement of blood glucose levels (mmol/L)

	CP+DM		CP+nonDM	
	GCBG-CBG	GCBG-cFPG	GCBG-CBG	GCBG-cFPG
Maximum negative difference	-1.28	-1.26	-1.69	-1.96
Maximum positive difference	1.89	1.51	0.89	1.08
Mean difference±SD	0.54 ± 0.62	0.29 ± 0.75	0.31 ± 0.59	0.20 ± 0.55
95% limits of agreement	-0.70, 1.77	-1.21, 1.78	-0.87, 1.49	-0.90, 1.30

GCBG, gingival crevicular blood glucose; CBG, capillary blood glucose; FPG, fasting plasma glucose; CP+DM, chronic periodontitis with diabetes group; CP+nonDM, chronic periodontitis with no diabetes group; SD, standard deviation.

Within the limitations of the present study, a good agreement between GCBG-CBG measurements and GCBG-FPG measurements was not observed. Gingival crevicular blood may not be suitable for blood glucose evaluation in the dental clinic in untreated chronic periodontitis patients.

Acknowledgements

This study was supported by a Faculty of Dentistry, Mahidol University research grant.

Funding: Faculty of Dentistry, Mahidol University research grant
Competiting interests: None declared
Ethical approval: Faculty of Dentistry / Faculty of Pharmacy, Mahidol University, Institutional Review Board (MU-DT/PY-IRB 2014/DT009)

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