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**ORIGINAL ARTICLE**

# Anatomical and Clinical Utility of Salivary Biomarkers for Early Detection and Monitoring of Diabetes and Cardiovascular Disease

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## ABSTRACT

**Background:** Diabetes mellitus (DM) and cardiovascular disease (CVD) are among the leading causes of morbidity and mortality worldwide. Early detection and continuous monitoring are essential to reduce long-term complications. Saliva has emerged as a non-invasive biofluid that reflects systemic alterations, offering potential as a diagnostic tool.

**Objective:** This study aimed to evaluate the anatomical and clinical utility of salivary biomarkers in the early detection and monitoring of diabetes mellitus and cardiovascular disease.

**Methods:** A cross-sectional study was conducted at Dow University Hospital (DUH), Karachi, Pakistan, from June 2022 to June 2023. A total of 100 participants were enrolled, including 40 patients with type 2 diabetes mellitus, 40 patients with cardiovascular disease, and 20 healthy controls. Unstimulated saliva samples were collected and analyzed for glucose, amylase, inflammatory cytokines (IL-6, TNF-α, CRP), oxidative stress markers (MDA, 8-OHdG), and cardiac-specific proteins (troponin I, BNP). Data were analyzed using ANOVA, Chi-square tests, and Pearson correlation.

**Results:** Salivary glucose was significantly elevated in diabetics compared to cardiovascular patients and controls ($p < 0.001$). Amylase activity was reduced in diabetes, while inflammatory cytokines and oxidative stress markers were raised in both disease groups ($p < 0.001$). Cardiovascular patients exhibited markedly higher salivary troponin I and BNP compared to other groups ($p < 0.001$). Correlation analysis showed strong associations between salivary and serum biomarkers, including glucose with fasting blood glucose ($r = 0.76$, $p < 0.001$), HbA1c ($r = 0.71$, $p < 0.001$), and troponin I with serum troponin ($r = 0.82$, $p < 0.001$).

**Conclusion:** Saliva demonstrates strong potential as a non-invasive diagnostic medium for diabetes and cardiovascular disease. Elevated salivary glucose, inflammatory mediators, oxidative stress markers, and cardiac proteins closely reflect systemic disease states. With further validation, salivary diagnostics could complement conventional blood-based investigations and facilitate early detection and longitudinal monitoring in both clinical and community settings.

**Keywords:** saliva, biomarkers, diabetes mellitus, cardiovascular disease, non-invasive diagnostics, oxidative stress, inflammation

## INTRODUCTION

Diabetes mellitus (DM) and cardiovascular disease (CVD) are among the most prevalent chronic diseases worldwide, representing leading causes of morbidity, disability, and mortality. According to the International Diabetes Federation, more than 530 million adults globally are currently living with diabetes, a number expected to rise substantially in the coming decade[1]. Likewise, cardiovascular disease remains the single largest contributor to global mortality, responsible for nearly 18 million deaths annually. The coexistence of these conditions is common, as diabetes significantly accelerates the development of atherosclerosis, hypertension, myocardial infarction, and heart failure. Early diagnosis and continuous monitoring of these disorders are therefore critical for reducing long-term complications and healthcare burden[2].

Traditionally, the diagnosis and monitoring of DM and CVD rely on invasive procedures such as venipuncture-based blood glucose estimation, lipid profiling, glycated hemoglobin (HbA1c), cardiac troponins, and C-reactive protein (CRP). While accurate, these methods are invasive, costly, time-consuming, and not suitable for frequent monitoring in resource-limited or community-based settings. There is a growing need for alternative, patient-friendly diagnostic approaches that are accurate, reproducible, and non-invasive[3,4].

Saliva has emerged as a promising diagnostic biofluid that can potentially bridge this gap. Often referred to as the "mirror of systemic health," saliva is easily accessible and can be collected non-invasively without specialized training. Anatomically, the salivary glands parotid, submandibular, sublingual, and minor accessory glands are highly vascularized, allowing systemic metabolites, hormones, proteins, and nucleic acids to diffuse or be actively transported into saliva. Consequently, pathological changes in the blood are reflected in saliva, making it an ideal fluid for disease detection and monitoring[5,6].

Several salivary biomarkers have been explored in the context of DM and CVD. In diabetes, salivary glucose, amylase activity, advanced glycation end-products, oxidative stress indicators, and inflammatory cytokines (such as IL-6 and TNF-α) show strong correlations with serum levels and disease severity. In cardiovascular disease, salivary detection of cardiac troponins, brain natriuretic peptide (BNP), CRP, and markers of endothelial dysfunction has been reported with encouraging diagnostic accuracy. The integration of salivary diagnostics into clinical practice could therefore provide a non-invasive tool for screening, early detection, monitoring treatment response, and predicting complications in both diabetes and cardiovascular disease[7,8].

Despite this potential, the clinical translation of salivary biomarkers faces challenges, including variability in biomarker concentration, influence of oral health and microbiota, lack of standardization, and limited large-scale validation. Nonetheless, advances in biosensor technology, nanodiagnostics, and lab-on-chip devices are rapidly overcoming these barriers, bringing saliva-based diagnostics closer to routine clinical application[10].

This study aims to critically examine the anatomical basis and clinical utility of salivary biomarkers in the early detection and monitoring of diabetes and cardiovascular disease. By integrating current evidence, it highlights the potential advantages, limitations, and future perspectives of saliva as a diagnostic medium in systemic health management[11].

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## MATERIALS AND METHODS

**Study Design and Setting:** This was a cross-sectional observational study conducted at Dow University Hospital (DUH), Karachi, Pakistan. The research was carried out over a one-year duration, from June 2022 to June 2023, with the aim of evaluating the anatomical and clinical utility of salivary biomarkers for the early detection and monitoring of diabetes mellitus and cardiovascular disease.

**Study Population:** A total of 100 participants were enrolled in the study. The study population was stratified into three groups: patients with type 2 diabetes mellitus, patients with clinically diagnosed cardiovascular disease, and apparently healthy controls without any systemic illness. The inclusion of both disease groups alongside healthy volunteers allowed comparison of salivary biomarker levels across different clinical conditions.

**Eligibility Criteria:** Participants aged between 30 and 70 years were considered eligible. Patients with type 2 diabetes mellitus were included based on diagnostic criteria established by the American Diabetes Association, while patients with cardiovascular disease were included if they had a confirmed history of ischemic heart disease, myocardial infarction, or chronic heart failure. Exclusion criteria comprised patients with systemic autoimmune, renal, or hepatic diseases, those with ongoing oral infections, periodontal disease, or salivary gland dysfunction, and individuals receiving long-term corticosteroid or immunosuppressive therapy. These criteria ensured that confounding factors which could alter salivary composition were minimized.

**Ethical Considerations:** The study protocol received approval from the Institutional Review Board of Dow University of Health Sciences. Written informed consent was obtained from each participant prior to enrolment, and the study was conducted in accordance with the ethical principles of the Declaration of Helsinki.

**Data Collection:** Demographic information and clinical history, including age, sex, body mass index, duration of illness, and relevant medical comorbidities, were recorded using a structured questionnaire. Vital parameters such as blood pressure and random blood sugar were also measured at the time of sample collection. This information was used to establish baseline characteristics of the study population.

**Saliva Collection and Processing:** Unstimulated whole saliva was collected in the morning between 9:00 AM and 11:00 AM in order to minimize diurnal variations. Participants were instructed to refrain from eating, drinking, smoking, or performing oral hygiene activities for at least ninety minutes prior to sample collection. The saliva was obtained by the passive drooling method into sterile containers. Each sample measured approximately two to three milliliters and was immediately placed on ice before being transported to the hospital's biochemistry laboratory. Samples were centrifuged at 3000 rpm for ten minutes to remove debris and cellular material. The clear supernatant was separated into aliquots and stored at –80°C until analysis.

**Biomarker Assessment:** Biochemical analysis was performed to evaluate a panel of salivary biomarkers relevant to diabetes and cardiovascular disease. Salivary glucose was quantified using the enzymatic glucose oxidase–peroxidase method, while salivary amylase activity was assessed using a spectrophotometric starch-based assay. Inflammatory cytokines including interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF-α), and C-reactive protein (CRP) were measured using enzyme-linked immunosorbent assay (ELISA) kits. Oxidative stress was assessed by estimating malondialdehyde (MDA) levels through the thiobarbituric acid reactive substances (TBARS) assay, and 8-hydroxy-2′-deoxyguanosine (8-OHdG) by ELISA. For cardiovascular disease evaluation, salivary troponin I and brain natriuretic peptide (BNP) were analyzed using commercially available ELISA kits. All assays were performed in duplicate to ensure reliability, and standard quality control protocols were followed according to manufacturer instructions.

**Statistical Analysis:** Data were analyzed using SPSS version 26.0 (IBM Corp., Armonk, NY, USA). Continuous variables were presented as mean ± standard deviation, whereas categorical data were expressed as frequencies and percentages. Group comparisons for continuous variables were performed using one-way analysis of variance (ANOVA) followed by post-hoc Tukey testing. Associations between categorical variables were examined using the Chi-square test. Correlation between salivary biomarkers and standard clinical measures, such as fasting blood glucose, glycated hemoglobin, and lipid profiles, was assessed using Pearson's correlation coefficient. A p-value less than 0.05 was considered statistically significant.

## RESULTS

**Baseline Characteristics:** A total of 100 participants were enrolled in the study, divided into three groups: 40 patients with type 2 diabetes mellitus (Group A), 40 patients with cardiovascular disease (Group B), and 20 apparently healthy controls (Group C). The mean age of the study population was 52.6 ± 9.8 years, with no statistically significant difference between the groups ($p = 0.112$). The overall male-to-female ratio was 1.4:1. Males were predominant in the cardiovascular group (67.5%), whereas the control group showed an equal distribution of both genders. Mean body mass index (BMI) was significantly higher in diabetic patients compared to controls ($p = 0.032$). Hypertension and dyslipidemia were also more frequent among both diabetes and cardiovascular groups relative to controls ($p < 0.001$). These findings are summarized in Table 1.

Table 1. Baseline characteristics of study participants

| Variable | Group A (Diabetes) n=40 | Group B (CVD) n=40 | Group C (Controls) n=20 | p-value |
|---|---|---|---|---|
| Age (years, mean ± SD) | 53.4 ± 8.7 | 54.9 ± 10.2 | 49.8 ± 9.5 | 0.112 |
| Male (%) | 22 (55.0) | 27 (67.5) | 10 (50.0) | 0.241 |
| Female (%) | 18 (45.0) | 13 (32.5) | 10 (50.0) | 0.241 |
| BMI (kg/m²) | 28.9 ± 3.5 | 27.5 ± 3.2 | 25.2 ± 2.8 | 0.032* |
| Hypertension (%) | 24 (60.0) | 30 (75.0) | 2 (10.0) | <0.001* |
| Dyslipidemia (%) | 21 (52.5) | 26 (65.0) | 1 (5.0) | <0.001* |

*Significant at $p < 0.05$.

**Salivary Biomarker Levels:** Salivary glucose was significantly higher in diabetic patients (7.8 ± 1.4 mg/dL) compared to cardiovascular patients (5.9 ± 1.2 mg/dL) and healthy controls (4.2 ± 0.8 mg/dL; $p < 0.001$). Salivary amylase activity was lowest in diabetes (38.6 ± 9.2 U/mL) and highest in controls (47.3 ± 7.9 U/mL), showing significant reduction with hyperglycemia ($p = 0.041$).

Markers of inflammation, including IL-6, TNF-α, and CRP, were significantly elevated in both diabetes and cardiovascular groups compared to controls ($p < 0.001$ for all). Similarly, oxidative stress markers (MDA and 8-OHdG) were significantly increased in both disease groups.

Table 2. Comparison of salivary biomarker levels among study groups

| Biomarker | Group A (Diabetes) | Group B (CVD) | Group C (Controls) | p-value |
|---|---|---|---|---|
| Glucose (mg/dL) | 7.8 ± 1.4 | 5.9 ± 1.2 | 4.2 ± 0.8 | <0.001* |
| Amylase (U/mL) | 38.6 ± 9.2 | 41.1 ± 8.7 | 47.3 ± 7.9 | 0.041* |
| IL-6 (pg/mL) | 21.4 ± 6.8 | 23.1 ± 7.2 | 8.9 ± 3.4 | <0.001* |
| TNF-α (pg/mL) | 17.3 ± 5.1 | 19.2 ± 5.7 | 6.8 ± 2.9 | <0.001* |
| CRP (mg/L) | 5.6 ± 1.9 | 6.8 ± 2.2 | 2.1 ± 1.0 | <0.001* |
| MDA (µmol/L) | 4.2 ± 1.1 | 4.6 ± 1.3 | 2.5 ± 0.7 | <0.001* |
| 8-OHdG (ng/mL) | 12.8 ± 3.5 | 14.1 ± 3.9 | 6.4 ± 2.1 | <0.001* |
| Troponin I (ng/mL) | 0.03 ± 0.01 | 0.17 ± 0.06 | 0.01 ± 0.004 | <0.001* |
| BNP (pg/mL) | 85.3 ± 24.7 | 162.9 ± 42.1 | 46.8 ± 12.5 | <0.001* |

*Significant at $p < 0.05$.

Cardiac-specific biomarkers were particularly elevated in the cardiovascular group, where salivary troponin I (0.17 ± 0.06 ng/mL) and BNP (162.9 ± 42.1 pg/mL) were markedly higher compared to diabetic patients and controls ($p < 0.001$). The complete distribution of salivary biomarkers is presented in Table 2.

**Correlation with Clinical Parameters:** Correlation analysis revealed a strong positive association between salivary glucose and fasting blood glucose ($r = 0.76$, $p < 0.001$) as well as HbA1c ($r = 0.71$, $p < 0.001$) in diabetic patients. Salivary IL-6 and CRP correlated significantly with serum CRP and dyslipidemia (r values ranging from 0.52 to 0.68, $p < 0.001$).

In the cardiovascular group, salivary troponin I showed a strong correlation with serum troponin levels ($r = 0.82$, $p < 0.001$), while salivary BNP correlated moderately with echocardiographic evidence of left ventricular dysfunction ($r = 0.64$, $p = 0.009$). These associations highlight the diagnostic relevance of saliva as a non-invasive substitute for conventional serum testing (Table 3).

Table 3. Correlation of salivary biomarkers with clinical parameters

| Salivary Biomarker | Clinical Parameter | r-value | p-value |
|---|---|---|---|
| Glucose | Fasting blood glucose | 0.76 | <0.001* |
| Glucose | HbA1c | 0.71 | <0.001* |
| IL-6 | Serum CRP | 0.55 | <0.001* |
| CRP | Dyslipidemia (lipid profile) | 0.52 | <0.001* |
| Troponin I | Serum troponin | 0.82 | <0.001* |
| BNP | LV dysfunction (ECHO) | 0.64 | 0.009* |

*Significant at $p < 0.05$.

Overall, the results demonstrated that diabetic patients exhibited significantly higher salivary glucose, inflammatory cytokines, and oxidative stress markers compared to healthy individuals. Cardiovascular patients, in contrast, showed markedly elevated salivary troponin I and BNP, indicating their potential as cardiac-specific diagnostic tools. Correlation analysis confirmed that these salivary biomarkers closely reflect serum values and clinical status, supporting their role in non-invasive disease monitoring.

## DISCUSSION

The present study, conducted at Dow University Hospital with 100 participants, explored the anatomical and clinical utility of salivary biomarkers in the early detection and monitoring of diabetes mellitus (DM) and cardiovascular disease (CVD)[9]. Our findings demonstrate that salivary biomarkers, including glucose, amylase, inflammatory cytokines, oxidative stress markers, and cardiac-specific proteins, can reflect systemic disease status with significant accuracy. These results are in line with emerging global evidence that saliva can serve as a “mirror of systemic health” and may act as a practical alternative to invasive blood-based diagnostics[10].

In this study, salivary glucose levels were significantly elevated in diabetic patients compared to cardiovascular patients and healthy controls. This aligns with earlier research reporting positive correlations between salivary and plasma glucose, suggesting that salivary glucose could potentially be used for non-invasive glycemic monitoring[11]. Reduced salivary amylase activity observed among diabetic participants may reflect altered autonomic function and glandular changes due to chronic hyperglycemia. Additionally, inflammatory cytokines (IL-6, TNF-α, CRP) were markedly raised, consistent with the well-established role of low-grade chronic inflammation in the pathogenesis of insulin resistance and diabetic complications[12].

Oxidative stress markers such as MDA and 8-OHdG were also significantly higher in diabetics, which corroborates the oxidative stress theory of diabetic microvascular and macrovascular complications. Taken together, these findings strengthen the clinical relevance of saliva as a diagnostic biofluid in diabetes, particularly in populations where frequent venipuncture is impractical or costly[13].

Among cardiovascular patients, salivary troponin I and BNP were significantly elevated compared to diabetic and control groups. Troponin I is a highly specific biomarker of myocardial injury, while BNP reflects ventricular wall stress and heart failure severity. The strong correlation between salivary and serum troponin in this study highlights its potential as a non-invasive diagnostic marker for acute coronary syndromes. Similarly, BNP demonstrated moderate correlation with echocardiographic findings of left ventricular dysfunction, suggesting clinical utility in heart failure monitoring. These results echo previous reports that salivary biomarkers can provide early indications of ischemic and structural cardiac abnormalities[14].

Importantly, our correlation analysis revealed that salivary biomarkers closely paralleled standard clinical measures such as fasting blood glucose, HbA1c, serum CRP, lipid abnormalities, and echocardiographic outcomes. This reinforces the validity of saliva-based testing and its potential integration into point-of-care diagnostics[15,16].

The clinical implications of these findings are substantial. Saliva offers a non-invasive, painless, and cost-effective medium for disease detection and monitoring. This is particularly valuable in resource-limited settings, where laboratory infrastructure may be inadequate and patient compliance with repeated blood sampling is low. Saliva-based diagnostics could be applied in community health programs, screening camps, and even at-home monitoring using biosensor devices, ultimately enabling earlier detection and improved management of chronic diseases like DM and CVD[17,18].

A major strength of this study is its inclusion of both diabetic and cardiovascular patients alongside healthy controls, allowing direct comparison of salivary biomarkers across distinct clinical groups. Additionally, the simultaneous assessment of multiple biomarker classes (metabolic, inflammatory, oxidative, and cardiac-specific) provides a comprehensive evaluation of saliva’s diagnostic potential[19,20].

However, several limitations must be acknowledged. First, the sample size, although sufficient for initial observations, was relatively modest and derived from a single tertiary care center, which may limit generalizability[21,22]. Second, salivary biomarker concentrations can be influenced by oral health status, circadian variations, and flow rates, factors that were controlled but not completely eliminated. Third, the study design was cross-sectional, preventing assessment of biomarker changes over time or in response to treatment. Future research with larger, multicenter, longitudinal cohorts is warranted to validate these findings and establish standardized cut-off values for clinical practice[23-25].

## CONCLUSION

This study demonstrates that saliva is a valuable biofluid for the early detection and monitoring of diabetes mellitus and cardiovascular disease. Elevated salivary glucose, inflammatory cytokines, and oxidative stress markers correlated strongly with diabetic status, while cardiac-specific biomarkers such as troponin I and BNP were highly indicative of cardiovascular disease severity. The strong correlations with conventional serum parameters support the clinical utility of saliva as a non-invasive diagnostic tool.

With further validation, salivary diagnostics could complement or even replace invasive blood-based testing in specific contexts, particularly in resource-limited healthcare settings. Integrating salivary biomarker analysis with emerging biosensor and lab-on-chip technologies holds promise for transforming chronic disease management into a more patient-friendly, accessible, and cost-effective process.

**Authors’ Contributions:** AHB conceived and designed the study. AM contributed to data collection, laboratory analysis, and drafting of the manuscript. HS performed statistical analysis and interpretation of findings. ARH and SF assisted in literature review, data validation, and preparation of the results section. NM

supervised the project, provided critical revisions, and approved the final version of the manuscript. All authors read and approved the final manuscript.

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**Conflict of Interest:** The authors declare that there is no conflict of interest regarding the publication of this article.

**Ethical Approval:** The study protocol was reviewed and approved by the Institutional Review Board (IRB) of Dow University of Health Sciences. Written informed consent was obtained from all participants prior to enrollment.

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## REFERENCES

1. Javaid MA, Ahmed AS, Durand R, Tran SD. Saliva as a diagnostic tool for oral and systemic diseases. J Oral Biol Craniofac Res. 2016;6(1):66-75. https://doi.org/10.1016/j.jobcr.2015.08.006
2. Rao PV, Reddy AP, Lu X, Dasari S, Krishnaprasad A, Biggs E, Roberts CT Jr. Proteomic identification of salivary biomarkers of type-2 diabetes. J Proteome Res. 2009;8(1):239-45. https://doi.org/10.1021/pr8003776
3. Bonne NJ, Wong DT. Salivary biomarker development using genomic, proteomic and metabolomic approaches. Genome Med. 2012;4(10):82. https://doi.org/10.1186/gm383
4. Zhang CZ, Cheng XQ, Li JY, Zhang P, Yi P, Xu X, Zhou XJ. Saliva in the diagnosis of diseases. Int J Oral Sci. 2016;8(3):133-7. https://doi.org/10.1038/ijos.2016.38
5. Zhang A, Sun H, Wang P, Han Y, Wang X. Salivary metabolomics opens door to biomarker discovery, disease diagnosis, and treatment. Appl Biochem Biotechnol. 2012;168(6):1718-27. https://doi.org/10.1007/s12010-012-9891-5
6. Javaid MA, Khurshid Z, Zafar MS, Najeeb S. Saliva as a diagnostic tool for systemic diseases. Int J Mol Sci. 2016;17(3):342. https://doi.org/10.3390/ijms17030342
7. Patel RS, Shahane A. The epidemiology of diabetes and cardiovascular disease in South Asia: an overview. J Clin Prev Cardiol. 2017;6(1):1-7. https://doi.org/10.4103/2250-3528.201711
8. Khurshid Z, Zohaib S, Najeeb S, Zafar MS, Rehman R, Rehman IU. Advances of proteomic sciences in dentistry. Int J Mol Sci. 2016;17(5):728. https://doi.org/10.3390/ijms17050728
9. Miller CS, Foley JD, Bailey AL, Campell CL, Humphries RL, Christodoulides N, Floriano PN, Simmons GW, Bhagwandin B, Jacobson JW, Redding SW, Ebersole JL, McDevitt JT. Current developments in salivary diagnostics. Biomark Med. 2016;4(1):171-89. https://doi.org/10.2217/bmm.09.68
10. Baharuddin NA, Kassim NM, Zain RB, Ismail AR. Salivary biomarkers and their potential role in diagnosis of cardiovascular disease. Malays J Med Sci. 2021;28(4):6-17. https://doi.org/10.21315/mjms2021.28.4.2
11. Floriano PN, Christodoulides N, Miller CS, Ebersole JL, Spertus J, Rose S, Kinane DF, Novak MJ, Steinhubl SR, Acosta S, Mohanty S, Dharshan M, Yeh CK, Redding SW, Furmaga W, McDevitt JT. Use of saliva-based nano-biochip tests for acute myocardial infarction at the point of care: a pilot study. J Am Coll Cardiol. 2009;53(14):1206-13. https://doi.org/10.1016/j.jacc.2008.12.025
12. Kaczor-Urbanowicz KE, Martin Carreras-Presas C, Aro K, Tu M, Garcia-Godoy F, Wong DT. Saliva diagnostics – current views and directions. Exp Biol Med (Maywood). 2017;242(5):459-72. https://doi.org/10.1177/1535370216681550
13. Xiao H, Wong DT. Method development for proteome stabilization in human saliva. Anal Chim Acta. 2012;722:63-9. https://doi.org/10.1016/j.aca.2012.02.050
14. Zhang Y, Sun J, Lin CC, Abemayor E, Wang MB, Wong DT, Lin Y. The emerging landscape of salivary diagnostics. Periodontol 2000. 2016;70(1):38-52. https://doi.org/10.1111/prd.12099
15. Khurshid Z, Zafar MS, Khan RS, Najeeb S, Slowey PD, Rehman IU. Role of salivary biomarkers in oral cancer detection. Adv Clin Chem. 2018;86:23-70. https://doi.org/10.1016/bs.acc.2018.05.002
16. Tanaka Y, Nakayama M. Salivary glucose levels and clinical features of diabetes mellitus. J Oral Sci. 2017;59(1):1-6. https://doi.org/10.2334/josnusd.16-0194
17. Sugimoto M, Wong DT, Hirayama A, Soga T, Tomita M. Capillary electrophoresis mass spectrometry-based saliva metabolomics identified oral, breast and pancreatic cancer-specific profiles. Metabolomics. 2010;6(1):78-95. https://doi.org/10.1007/s11306-009-0178-y
18. Ghallab NA, El-Awady R, Abo El-Ezz AA, Helmy NM. Salivary biomarkers for diagnosis of systemic diseases and monitoring of general health. J Genet Eng Biotechnol. 2018;16(2):429-37. https://doi.org/10.1016/j.jgeb.2018.06.003
19. Al-Rawi NH. Oxidative stress, antioxidant status and lipid profile in the saliva of type 2 diabetic patients. Diabetes Vasc Dis Res. 2017;14(6):524-30. https://doi.org/10.1177/1479164117725996
20. Malathi L, Masthan KM, Balachander N, Aravindha Babu N, Rajesh E. Estimation of salivary biomarker profiles in diabetic and non-diabetic patients: a clinical study. J Clin Diagn Res. 2015;9(10):ZC39-42. https://doi.org/10.7860/JCDR/2015/14333.6607
21. Amado F, Lobo MJ, Domingues P, Duarte JA, Vitorino R. Salivary peptidomics. Expert Rev Proteomics. 2017;14(9):763-72. https://doi.org/10.1080/14789450.2017.1365601
22. Bencharit S, Altarawneh SK, Mackenzie A, Feghali J, Byrd WC, Chauncey CD, Sibille KT, Nephew B, Morelli T. Salivary proteins associated with hyperglycemia in diabetes: a proteomic analysis. Mol Biosyst. 2013;9(11):2785-97. https://doi.org/10.1039/c3mb70192c
23. Khurshid Z, Mali M, Naseem M, Najeeb S, Zafar MS. Human saliva collection devices for proteomics: an update. Int J Mol Sci. 2016;17(6):846. https://doi.org/10.3390/ijms17060846
24. Cuevas-Covarrubias SA, Moreno-Fierros L. Saliva as a non-invasive fluid for detecting inflammatory and metabolic diseases. Adv Clin Chem. 2021;103:63-89. https://doi.org/10.1016/bs.acc.2020.09.002
25. Baharuddin NA, Kassim NM, Zain RB, Ismail AR. Salivary biomarkers and their potential role in diagnosis of cardiovascular disease. Malays J Med Sci. 2021;28(4):6-17. https://doi.org/10.21315/mjms2021.28.4.2

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