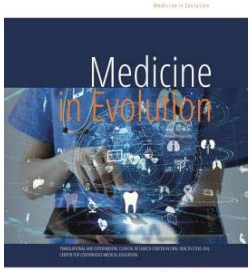
Exploring Salivary Biomarkers and Health Parameters in Type 2 Diabetic and Non-diabetic Patients



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Abstract

1.Background/Objectives: Diabetes mellitus (DM) is a chronic, non-communicable disease with significant systemic and oral health complications. Early, non-invasive diagnostic approaches, such as salivary biomarker analysis, could improve management and reduce associated complications. Salivary alpha-amylase has emerged as a promising biomarker due to its role in glycemic control and stress-related sympathetic activation. This study aims to evaluate salivary alpha-amylase levels as potential biomarkers for type 2 diabetes mellitus (T2DM) and explore their correlation with clinical parameters.2. Methods: A cross-sectional study was conducted involving 40 participants (20 T2DM patients and 20 controls). Saliva samples were collected and analyzed for alpha-amylase using ELISA. Data on oral health, body mass index (BMI), glycated hemoglobin (HbA1c), and total cholesterol were also collected.3. Results: Salivary alpha-amylase levels were significantly higher in T2DM patients (mean: 177.86 µ/L) compared to controls (mean: 90.98 µ/L, p = 0.001). Poorer oral health, higher BMI, and increased cholesterol levels were observed in diabetic participants. HbA1c levels revealed suboptimal glycemic control in a subset of patients.4. Conclusions: Salivary alpha-amylase shows potential as a non-invasive biomarker for T2DM diagnosis and monitoring. The findings emphasize the need for integrated medical and dental management to address systemic and oral health challenges in diabetes. Further research is needed to validate and standardize salivary diagnostic tools.

Keywords: diabetes, salivary amylase, oral health, saliva, biomarker

INTRODUCTION

Diabetes mellitus is a chronic, non-communicable disease that is progressing at an accelerated rate worldwide. The morbidity and mortality associated with diabetes are due to the persistent increase in hyperglycemia and the manifestation of associated complications, such as cardiovascular disorders, nephropathy, retinopathy, neuropathy, and lower limb amputations. Regular testing for prevention, early diagnosis, and decisive management of diabetes have become critical to reducing the global incidence of the disease. Additionally, people with diabetes are at a significantly higher risk of developing infections, which greatly reduces life expectancy. In the oral cavity, complications such as xerostomia, gingivitis, periodontitis, dental caries, and delayed healing of all oral lesions are commonly observed. According to recent data from the International Diabetes Federation (IDF), 463 million adults are living with diabetes. This represents 9.3% of the global adult population. The total number is projected to reach 578 million by 2030 and 700 million by 2045 [1].

The intricate connections between oral health and systemic diseases, including diabetes mellitus, emphasize the critical role of maintaining periodontal health for overall well-being. Diabetes, as a chronic and rapidly escalating global condition, exacerbates the risk of periodontal disease by altering the oral environment through mechanisms such as impaired immune response, increased inflammation, and delayed healing. Factors such as age, systemic conditions, and tobacco use further compound this vulnerability, intensifying both periodontal and systemic disease manifestations. These shared risk factors also create common pathways linking periodontal disease with major noncommunicable diseases, including cardiovascular disease, cancer, chronic respiratory disease, and diabetes itself. Recognizing the oral cavity as a "window to general health" underscores the undeniable relationship between oral and systemic health. Improving oral health literacy and promoting preventive behaviors are essential for mitigating oral diseases and their systemic implications. Given the shared risk factors between oral and systemic diseases, studying salivary biomarkers offers valuable insights into health parameters, particularly in populations affected by Type 2 diabetes [2].

Blood analysis is considered the only conventional method for evaluating biological control. However, blood sampling is invasive, which can lead to complications such as psychological stress and anxiety for most patients. Currently, research is focusing on non-invasive techniques, such as diagnosing type II diabetes mellitus using saliva. Saliva collection is a safe and low-risk method that reduces the potential for virus transmission. As a result, saliva stands out as an innovative, non-invasive, and simple tool for disease diagnosis. It holds the promise of becoming a practical alternative to conventional diagnostic methods, such as serum or urine tests, in the future [3]. Saliva analysis offers several advantages: it is cost-effective, easy to collect, transport, and store. Saliva is a heterogeneous oral fluid composed of numerous constituents that play a crucial role in maintaining oral health homeostasis. Studies show that both the composition and function of saliva are influenced by local and systemic changes. Therefore, salivary molecules could serve as strong indicators for predicting, monitoring, and diagnosing systemic and local disorders. Early diagnosis can prevent or delay long-term health complications in individuals with type II diabetes. If undiagnosed and/or untreated, diabetes significantly impacts quality of life. Consequently, various biochemical parameters of saliva can be investigated, such as glucose, salivary amylase, and immunoglobulin A [4].

The validation of salivary biomarkers in terms of accuracy, sensitivity, specificity, and reliability is essential for advancing personalized approaches to the prevention and treatment of systemic comorbidities, such as type 2 diabetes mellitus (T2DM). This validation could also

impact oral health and host response parameters. T2DM is a chronic metabolic disorder associated with various oral manifestations, including poor periodontal health and changes in salivary composition. Altered protein concentrations detected in the saliva of individuals with diabetes have been proposed as useful tools for identifying T2DM. However, it remains unclear whether salivary protein biomarkers can reliably differentiate between a periodontally healthy T2DM individual and one with periodontitis as part of routine clinical diagnosis [5].

Traditional methods for monitoring glycemic levels, such as measuring blood glucose and glycated hemoglobin (HbA1c), rely on invasive techniques like venous puncture and capillary blood sampling. While accurate, these methods are time-consuming, potentially stressful, and less practical for frequent or immediate use. Point-of-care (POC) testing offers a faster alternative by enabling specimen analysis directly at or near the site of patient care, reducing delays in clinical decision-making. However, invasive procedures may still induce stress, particularly in younger populations or those with neuropsychiatric disorders, which can exacerbate hyperglycemia. Non-invasive approaches, such as salivary diagnostics, provide a promising solution for overcoming these limitations. Salivary glucose levels, typically ranging from 0.5 to 1 mg/dL, increase after food intake and correlate well with blood glucose and HbA1c levels under both stimulated and unstimulated conditions. Other salivary markers, such as fructosamine glycated proteins and salivary amylase, also show strong associations with glycemic control. Salivary amylase, a key enzyme in starch digestion, plays an important role in postprandial glucose regulation. Higher salivary amylase activity is linked to lower blood glucose levels following starch intake, likely due to increased insulin secretion. Furthermore, salivary amylase is sensitive to stress, serving as a marker of sympathetic nervous system activation, and may reflect stress-related glycemic changes in diabetes patients. Despite these advantages, salivary glucose testing can be influenced by factors such as oral bacterial flora, hydration, and certain medications, potentially affecting accuracy. This underscores the need for further exploration of alternative salivary biomarkers for diabetes management. Advances in salivary diagnostics hold significant potential for non-invasive, accessible, and efficient monitoring of glycemic control, but additional research is required to validate these methods and address their current limitations [6].

Aim and objectives

This study aims to explore the potential of salivary alterations as a diagnostic tool for type 2 diabetes mellitus by analyzing the composition and characteristics of saliva in individuals with diabetes and comparing these findings to a control group. The primary objective is to assess salivary amylase levels to evaluate their potential as biomarkers for type 2 diabetes diagnosis, while secondary objectives include correlating salivary markers with clinical and laboratory parameters such as glycated hemoglobin, body mass index (BMI), and total cholesterol. By integrating demographic and clinical data collected through questionnaires, the study also investigates the influence of lifestyle factors and medical history on salivary changes. This research aspires to propose a non-invasive, efficient alternative to traditional blood glucose testing, advancing accessible diagnostic methods and improving clinical management to reduce complications associated with diabetes.

MATERIAL AND METHODS

The investigation of salivary amylase alterations in patients with type 2 diabetes was conducted as part of a comprehensive research project at the Oral Health Clinic, Faculty of Dentistry, Timişoara, between 2023 and 2024. To ensure the scientific integrity and ethical compliance of the study, the research protocol underwent a thorough review by the

University's Ethics Committee. This cross-sectional study was carried out at the Translational and Experimental Clinical Research Center for Oral Health, within the Clinic of Preventive, Community Dentistry and Oral Health at the "Victor Babeş" University of Medicine and Pharmacy, Timişoara, Romania. The study involved the collection of saliva samples from 40 participants under standardized conditions.

The study adhered to the ethical guidelines set forth in the Declaration of Helsinki (1964) by the World Medical Association. Ethical approval was granted by the Ethics Committee of the "Victor Babeş" University of Medicine and Pharmacy, Timişoara, Romania (approval number 34/2018). Participation in the study was entirely voluntary, and informed consent was obtained from all participants. As the study involved the completion of a questionnaire containing personal data and the collection of biological samples, written consent was obtained from all participants before their inclusion in the research.

Initially, a group of 53 patients was selected for inclusion in the study. Subsequently, after applying the appropriate exclusion criteria, the number of participants was reduced to 40, who served as the source of data for the present study. Careful consideration was given to selecting subjects within the same age group and from the same geographical area to ensure the consistency of the data analyzed (Figure 1).

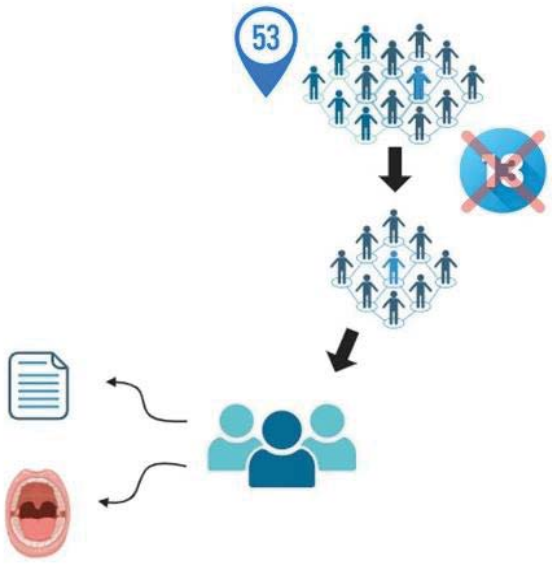

Figure 1. Schematic diagram of the workflow

The inclusion and exclusion criteria were established in accordance with the objectives and purpose of the research. To be eligible for inclusion in the study, participants had to meet the following inclusion criteria: they needed to be within a predefined age range, diagnosed with type 2 diabetes mellitus according to relevant international criteria, and willing to provide informed consent for participation in the study.

The exclusion criteria included the presence of other metabolic or endocrine disorders that could affect salivary composition, the use of medications that might influence salivary secretion, or any other conditions that could interfere with the collection and interpretation of salivary data.

By applying these criteria, it was ensured that the participants selected for the study represented a homogeneous group, suitable for the evaluation of salivary changes associated with type 2 diabetes.

To collect general information about the subjects, such as age, sex, type of diabetes diagnosis, and disease duration, a questionnaire was used in the first stage of the study. Additionally, the presence of other systemic conditions, ongoing treatments, and the subjects' smoking history was investigated, thus supplementing the obtained data with relevant information regarding their lifestyle and general oral health status. Furthermore, body mass index (BMI) was determined and blood analyses were performed to assess glycated hemoglobin, blood glucose levels, and total cholesterol. These additional data allow for a more comprehensive understanding of the participants' health status and the potential correlations between the various variables investigated.

In the second stage, the experimental phase, saliva samples were collected to determine salivary amylase levels. The collection took place between 06:00 and 08:00 in the morning, before performing oral hygiene and without prior consumption of food or liquids. Special containers, called Salivettes, were used for saliva collection, with a minimum sample volume of 1 ml required for each sample. The collection process involved opening the Salivette container to expose the internal absorbent pad, which was not touched by hand (Figure 2). The pad was then placed directly in the oral cavity by gently tilting the container and chewed lightly for 2 minutes to allow it to become saturated with saliva. Afterward, the pad was placed back into the container without being touched and the container was securely sealed. Subsequently, the samples are stored in a cryogenic environment at low temperatures before being transported to a specialized laboratory for analysis. Saliva was then analyzed using the ELISA method, a biochemical test that employs a solid-phase enzyme immunoassay to detect a ligand in a liquid sample, utilizing antibodies specific to the protein to be measured. ELISA is widely used as a diagnostic tool in medicine.

Figure 2. Salivette – containers for saliva sample collection

The data were entered into a computer (MS Office 2010, Excel spreadsheet) and subjected to statistical analysis using the SPSS statistical software package (version 23). To measure statistically significant differences between the two groups, the independent samples t-test and the Chi-square test were used.

RESULTS

The study cohort consisted of 40 participants, divided equally into two groups: 20 individuals with type 2 diabetes and 20 non-diabetic controls. Within the diabetic group, 12

participants were women, with an average age of 45.3 years, and 8 were men, with an average age of 41.7 years. In comparison, the non-diabetic group included 11 women, with an average age of 44.4 years, and 9 men, with an average age of 45.1 years. The age distribution between the two groups was relatively similar, with no significant differences in average age, ensuring comparability for further analyses. The balanced gender distribution and similar age ranges across both groups helped minimize demographic variability, allowing for a more focused assessment of the specific parameters being studied. This alignment strengthens the validity of the comparisons drawn between diabetic and non-diabetic individuals, particularly when examining salivary biomarkers, clinical metrics, and other related health parameters. The demographic characteristics of the diabetic and control groups, based on responses collected from questionnaires, are presented in Table 1.

The duration since diagnosis of diabetes was 11 ± 8 years. Blood glucose levels were 261 ± 131 mg/dL. Data obtained from the diabetic patients were compared with a control (non-diabetic) group consisting of 20 subjects, including 11 women and 9 men. The mean (±SD) blood glucose levels in this group were 92 ± 9 mg/dL.

Table 1 shows the distribution of study subjects, indicating that 20 subjects with diabetes and 20 subjects in the normal group were included.

Table 1. Distribution of study subjects in the Diabetes and Non-Diabetes group

No.	Study Group	Type 2 Diabetes	Average Age	Non-diabetes	Average Age
1.	Women	12	45.3	11	44.4
2.	Men	8	41.7	9	45.1
Total		20		20	

In the studied sample, two distinct groups were analyzed: one consisting of individuals with diabetes and the other of individuals without diabetes. The average age of the diabetic group is 43.5 years, with a standard deviation of 1.8 years, indicating moderate variability in ages within this group. In contrast, the non-diabetic group recorded a slightly higher average age of 44.7 years, but with a significantly smaller standard deviation of only 0.35 years. This suggests greater homogeneity in terms of age within the non-diabetic group. These data indicate that, although the average age is similar between the two groups, the age variability is considerably lower among individuals without diabetes (Table 2).

Table 2. Descriptive table with age and standard deviation for the studied sample

No.	Sample	Average Age	Standard deviation
1.	DIABETIC	43,5	1,8
2.	NON DIABETIC	44,7	0,35

Table 3 presents the comparison of salivary alpha-amylase levels between diabetic patients and healthy subjects, showing a statistically significant difference between the two groups ($p < 0.05$).

Table 3. Comparison of salivary alpha-amylase levels between diabetic patients and healthy subjects using the independent samples t-test

No.	Sample	Mean	Standard Deviation	*p* Value
1.	DIABETIC	177.86	72.17	0.001*
2.	NON-DIABETIC	90.98	44.17	0.001*

Another important factor that was analyzed based on the questionnaire was the oral health. The results show that diabetic patients mostly visit the dentist 2-3 times, or even 4 times a year, while healthy subjects typically visit the dentist 1-2 times a year. This suggests

that oral health is poorer among diabetic patients. One reason for this is the reduced salivary flow, which consequently increases the incidence of dental caries. Regarding the frequency of dental hygiene, the majority of diabetic patients brush their teeth once a day; 4 out of 20 reports brushing twice a day. In contrast, among the healthy subjects, 10 brush once a day, and 10 brush twice a day. All diabetic patients, as well as healthy subjects, use fluoride toothpaste and auxiliary hygiene tools, either a water flosser or dental floss.

As is well known, type 2 diabetes is associated with certain oral manifestations, such as dry mouth, halitosis, dental mobility, spontaneous or brushing-induced gum bleeding, fungal infections, and taste alterations. Based on the responses from diabetic patients, we find that 11 of them report experiencing dry mouth and bad breath, 6 have dental mobility, and 16 patients complained of gum bleeding, with the majority occurring during brushing, and only a few spontaneously (Figure 3). When comparing these responses with those of healthy subjects, we find that only 7 out of 20 healthy subjects report minor gum bleeding, which is exclusively caused by tooth brushing. Among healthy subjects, xerostomia (dry mouth) is present in 2 individuals, while dental mobility is absent (Figure 4).

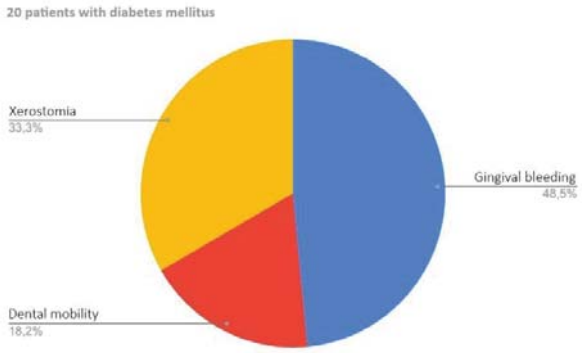

Figure 3. Description of the diabetic patient sample based on oral manifestations

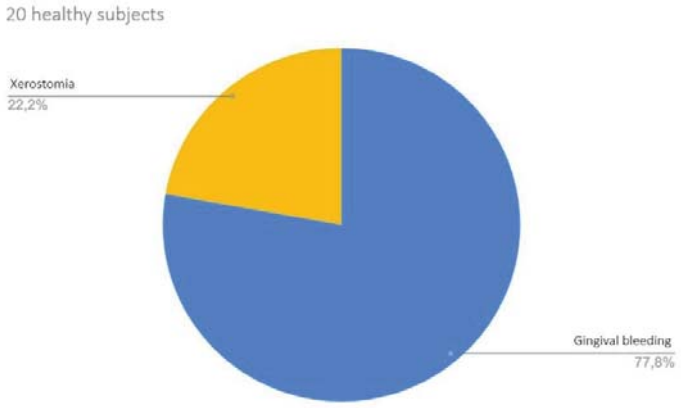

Figure 4. Description of the non-diabetic patient sample based on oral manifestations

The age, weight, and height of both diabetic patients and healthy subjects were recorded, allowing for the calculation of Body Mass Index (BMI) for each participant. A graph was generated to facilitate a comparative analysis of the results. The data revealed a significant difference in BMI values between the groups. Notably, none of the diabetic patients had a BMI below 25, which is considered within the normal range. Within the BMI range of 25-30, six individuals were identified, comprising four women and two men. The 30-35 BMI range showed the highest prevalence, with 13 individuals classified as overweight,

including eight women and five men. Only one individual was observed with a BMI exceeding 35 (Figure 5).

Among the healthy subjects, a distinct distribution of BMI values was observed, highlighting notable lifestyle differences compared to diabetic patients. In the BMI category below 25, 50% of the healthy subjects, totaling 10 individuals, were classified. Within the BMI range of 25-30, nine individuals were identified, while one subject fell within the 30-35 BMI range. No individuals in the healthy group had a BMI exceeding 35 (Figure 5).

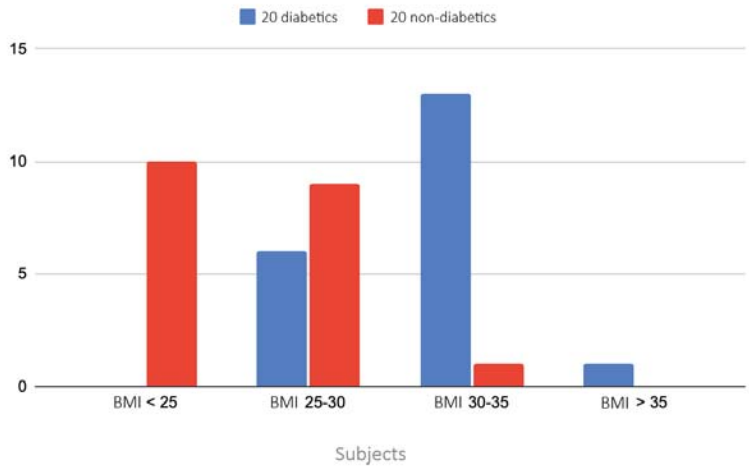

Figure 5. Description of the diabetic and non-diabetic patient sample based on body mass index

Glycated hemoglobin was analyzed in diabetic patients, as this test is routinely conducted as part of their clinical management. Glycated hemoglobin serves as a critical marker for assessing long-term glycemic control in individuals with diabetes. Normal values, indicative of well-controlled diabetes, range between 6-7%. Values exceeding 7% suggest inadequate glycemic control, potentially due to factors such as non-adherence to prescribed antidiabetic medication or dietary recommendations. A comparative graph was generated based on the results obtained from the diabetic patients. The analysis revealed that the majority of patients (17 individuals) had glycated hemoglobin levels within the target range of 6-7%. Conversely, three patients were found to have glycated hemoglobin levels exceeding 7%, reflecting suboptimal glycemic control (Figure 6).

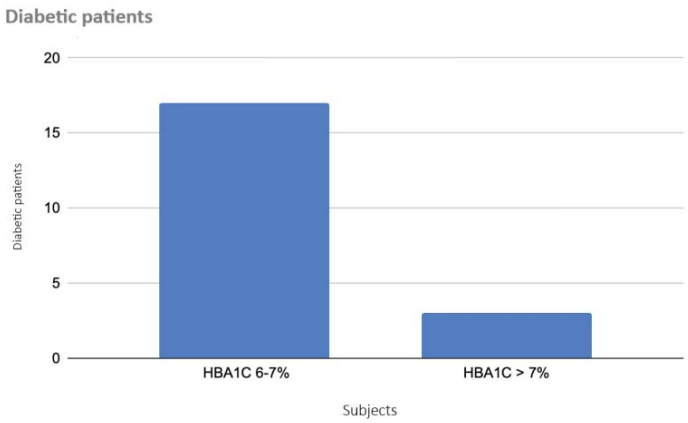

Figure 6. Description of the diabetic patient sample based on glycated hemoglobin levels

Total cholesterol levels were analyzed in both diabetic patients and healthy controls to compare values between the groups and explore the potential influence of type 2 diabetes on this parameter. The aim was to assess whether diabetes impacts cholesterol levels and to correlate these findings with health status. Among the diabetic patients, six individuals were found to have total cholesterol levels within the range of 200-240 mg/dL, which is considered borderline normal. However, 14 patients exhibited cholesterol levels exceeding 240 mg/dL, indicating hypercholesterolemia. In contrast, the healthy control group presented markedly different results: five individuals had cholesterol levels in the range of 200-240 mg/dL, while the majority, 15 individuals, had cholesterol levels below 200 mg/dL, considered optimal. These findings highlight a significant disparity in cholesterol levels between diabetic and healthy individuals.

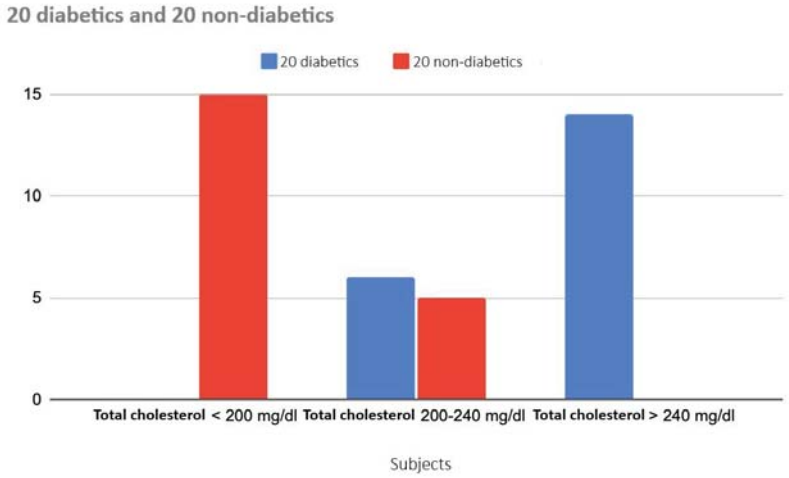

Figure 7. Description of the diabetic and non-diabetic patient sample based on total cholesterol levels

DISCUSSIONS

This study highlights the potential of salivary diagnostics as a non-invasive, cost-effective, and practical approach to evaluating type 2 diabetes mellitus (T2DM) and associated complications. The analysis of salivary alpha-amylase levels revealed significantly higher concentrations in diabetic patients compared to healthy controls, supporting the hypothesis that salivary biomarkers can serve as reliable indicators of glycaemic status. These findings align with previous research, demonstrating that salivary amylase levels correlate with blood glucose and glycated haemoglobin (HbA1c) levels, thereby offering a non-invasive alternative to traditional blood-based diagnostic methods [7,8]. Importantly, the observed increase in salivary amylase levels among diabetics underscores its potential utility in routine clinical monitoring and early detection of diabetes.

Salivary amylase begins the hydrolysis of starch in the mouth, accounting for no more than 30% of the total hydrolysis of starch. Since salivary amylase is inactivated by an acidic pH, no significant carbohydrate hydrolysis occurs in the stomach. Acinar cells, which produce salivary amylase, are innervated by both sympathetic and parasympathetic pathways. Activation of the sympathetic nervous system increases amylase synthesis, thereby increasing the concentration of amylase in saliva, while parasympathetic activity increases the rate of salivary flow with little or no effect on amylase synthesis. Salivary amylase is associated with the autonomic system and is involved in glycemic digestion, making it a promising biomarker for evaluating and monitoring diabetes mellitus [9]. Blood glucose

levels after starch consumption are influenced by genetic differences in salivary amylase, an enzyme that breaks down dietary starch. Higher salivary amylase activity is associated with lower blood glucose levels. In fact, individuals with high salivary amylase concentrations have had significantly lower blood glucose responses after starch ingestion compared to individuals with low enzyme concentrations, with this difference being mediated by increased plasma insulin concentrations in individuals with high enzyme levels [9].

Salivary alpha-amylase has been identified as a marker of sympathetic nervous system activity and stress. Its activity increases under sympathetic autonomic stimulation, making it a potential indicator for evaluating glycemic control in pathological conditions like diabetes mellitus (DM), where stress plays a role in glycemic fluctuations. Measuring salivary amylase provides a non-invasive method for monitoring stress-mediated changes in diabetic patients [10,11]. Key techniques for measuring salivary amylase include enzyme-linked immunosorbent assays (ELISA) and colorimetric methods, with the Phadebas® method being particularly accurate and user-friendly. Saliva collection methods can significantly influence the results, as factors like diurnal variations, saliva collection schedules, and techniques (e.g., cotton swabs vs. flow collection) affect amylase measurements. Studies have reported conflicting results regarding salivary amylase levels in diabetic patients, with some showing reduced levels due to autonomic neuropathy and microvascular complications, while others found increased levels linked to altered glucose regulation mechanisms. These discrepancies may stem from variations in sample collection, disease duration, comorbidities, and medication use. Hyperglycemia impacts salivary gland function, reducing salivary flow and altering composition, leading to oral health issues such as dry mouth, infections, caries, and mucosal changes. Saliva's biochemical profile, including glucose, proteins, electrolytes, and enzymes like amylase, reflects systemic changes in diabetes and can be used for disease diagnosis and management [12-16].

The present study observed higher salivary alpha-amylase levels in diabetic patients compared to healthy controls, supporting its potential as a biomarker. The increase is likely due to glucose regulation mechanisms involving the pancreas and salivary glands, highlighting the interconnectedness of systemic and oral health in diabetes. Further research is needed to standardize methodologies and validate salivary biomarkers for clinical use. In this study, the mean total salivary alpha-amylase level in diabetic subjects was 177.86 µ/L, while in normal subjects it was 90.98 µ/L, a difference that was statistically significant ($p=0.001$). These results are consistent with the study conducted by Panchabhai AS and colleagues, who measured salivary glucose, salivary alpha-amylase, total salivary proteins, and salivary flow rate in diabetic patients in India. The results of that study showed a decrease in salivary alpha-amylase levels in patients with controlled diabetes who were under treatment, compared to normal subjects [17]. In the present study, an increase in salivary alpha-amylase levels was found in patients newly diagnosed with diabetes mellitus. This finding could explain why an increase in salivary alpha-amylase levels was observed in our study. The results are in line with those of the study by Pal and colleagues, who demonstrated a significant positive correlation between salivary alpha-amylase levels and total proteins in diabetic patients. Furthermore, the present study aligns with the research conducted by Malathi and colleagues, who investigated salivary alpha-amylase as a diagnostic tool for early-stage diabetic patients. In that study, the average salivary alpha-amylase level in diabetic patients was 2739.48 µ/L, compared to 1740.38 µ/L in the normal group. In our study, newly diagnosed diabetic patients were evaluated for salivary alpha-amylase levels, and the results showed that the level of salivary alpha-amylase was higher in men than in women [16].

Salivary alpha-amylase is a component of saliva whose level does not change with age. Ben-Aryeh and colleagues found lower levels of alpha-amylase in older individuals,

while others observed no significant differences or even reported elevated levels of this enzyme. Salivary alpha-amylase is primarily produced by the parotid glands and is considered a marker of parotid saliva. It has been reported that these glands can maintain their secretory function throughout human life, which may explain the results obtained in our study, where no significant differences in alpha-amylase levels were found between younger and older individuals. Differences in results may be attributed to varying methodologies, different age groups, and/or whether saliva was collected in a stimulated or unstimulated state. Another factor that may influence results is stress, which is inherently present in dental practice and can be induced in patients by routine dental procedures or even by regular check-ups. It is known that stress increases salivary alpha-amylase levels [18].

The replacement of blood tests with other non-invasive samples, such as saliva, is increasingly being proposed for a variety of pathologies and is especially useful for patients with neurocognitive disorders or children, for whom blood collection is very stressful. This is primarily due to the fact that saliva tests are cheaper than blood tests, they are non-invasive and easy to store. Additionally, saliva is less infectious than blood, easier to handle in diagnostic procedures, and does not coagulate [19].

The study also explored the broader implications of T2DM on oral health. Diabetic participants reported significantly higher rates of oral manifestations, such as xerostomia, gum bleeding, halitosis, and dental mobility, compared to their non-diabetic counterparts. These symptoms are consistent with the well-documented impact of hyperglycemia on salivary gland function and periodontal health, which predispose individuals with diabetes to oral infections and delayed wound healing [20,21]. The reduced salivary flow observed in diabetic patients contributes to an increased risk of dental caries and periodontal disease, further emphasizing the interconnectedness of systemic and oral health. Notably, the frequent dental visits among diabetic patients, as opposed to the healthy controls, highlight the heightened need for oral healthcare interventions in this population.

BMI and cholesterol levels provided additional insights into the systemic health disparities between diabetic and non-diabetic individuals. None of the diabetic participants had a BMI below 25, with the majority falling within the overweight or obese categories. This contrasts sharply with the non-diabetic group, where a substantial proportion maintained a normal BMI. These results underscore the strong association between obesity and diabetes, with obesity acting as a significant risk factor for insulin resistance and glycemic dysregulation [22]. The cholesterol analysis revealed a similar trend, with a high prevalence of hypercholesterolemia among diabetic patients, further corroborating the increased cardiovascular risk associated with diabetes. This highlights the need for comprehensive management strategies that address not only glycemic control but also lipid profile optimization and weight management [23].

Glycated hemoglobin analysis provided a valuable measure of long-term glycemic control among diabetic participants. While most individuals achieved HbA1c levels within the target range of 6-7%, a subset exhibited levels exceeding 7%, indicative of suboptimal glycemic control. This finding points to potential challenges in treatment adherence, dietary regulation, or medication efficacy, necessitating a personalized approach to diabetes management. The significant variability in glycemic control observed in the study population underscores the importance of early detection and continuous monitoring of diabetes to mitigate long-term complications [24].

Oral manifestations of type 2 diabetes can be prevented through several approaches, including ensuring proper brushing and flossing behaviors, encouraging patients to visit the dentist for routine check-ups, and controlling blood glucose levels. Many patients with diabetes are unaware of the relationship between diabetes and oral health. There is a lack of awareness about the importance of maintaining among diabetic patients. Furthermore, only a

small percentage of patients diagnosed with diabetes visit the dentist for regular periodontal checks. It is assumed that every diabetic patient is at risk for periodontal disease and should be referred for periodontal screening and educated about the importance of oral health and regular dental visits. It has been reported that more than 90% of patients with diabetes mellitus (DM) experience oral manifestations due to the lack of regular dental check-ups. It has been suggested that individuals with higher educational levels are more concerned about the prevention and control of the disease. Therefore, providing education will increase awareness, which will help prevent oral complications of diabetes [25,26]. Involvement of oral health professionals in the strategies for identifying individuals at risk of diabetes mellitus will strengthen the preventive and screening efforts needed to prevent oral diseases. Treatment outcomes can be improved if dental practitioners are aware of the dental implications and risk factors of diabetes mellitus. Diabetic patients should be encouraged to visit the dentist to reinforce and educate them on oral health information through diabetes and dental care centers. Systemic health is closely linked to oral health, especially in diabetic individuals, which increases the need for collaborative dental and medical management of the patient. To improve the general and oral health of diabetic patients, a collaborative relationship should be developed between patients, physicians, and dentists [27,28].

Finally, the study demonstrated the utility of integrating demographic, clinical, and lifestyle data to provide a holistic understanding of the interplay between systemic and oral health in diabetes. The use of questionnaires to collect data on oral hygiene practices, medical history, and systemic health parameters further enriched the analysis, providing a comprehensive picture of the multifaceted impact of T2DM. The promising results obtained from salivary diagnostics suggest a shift toward non-invasive diagnostic modalities, which could improve patient compliance and accessibility, particularly in resource-limited settings [29].

Further research is warranted to validate these findings across larger populations and diverse settings, as well as to address potential limitations, such as the influence of oral bacterial flora and hydration status on salivary marker accuracy.

CONCLUSIONS

In conclusion, this study supports the viability of salivary biomarkers as diagnostic and monitoring tools for T2DM while highlighting the broader systemic and oral health challenges faced by individuals with diabetes. The findings underscore the importance of a multidisciplinary approach to diabetes management, integrating medical, dental, and lifestyle interventions to improve patient outcomes.

Conflicts of Interest

The authors declare no conflict of interest.

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