

# **Scientific potential of the salivary proteome in clinical management of Diabetes Mellitus**

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## **Introduction**

Human saliva is abundant in proteins, peptides and other components like amylase, cystatins, hormones, lysozyme, lactoferrin, mucin, lipase, peroxidase and growth factors with variable concentrations in numerous pathological processes located not only in the mouth, but also throughout the whole body (Shah, 2018). Efforts are being made towards its application in the laboratory diagnosis and monitoring of various diseases as well as evaluating therapeutic responses. Replacing traditionally used serum markers with newer ones originating from the saliva could enable less risky and stressful sampling procedure and faster access to various therapeutic interventions. Additionally, this biomaterial is suitable for certain population groups such as pediatric patients, owing to the enhanced compliance.

## **Aim of the study and methodology**

This research encompassed a comprehensive literature survey which was conducted using the PubMed database. The keywords: salivary proteome, proteomics, salivary markers and diabetes were applied as keywords and publication within the past 5 years was used to filter the results. All papers that met these criteria along with some suggested in the section "similar articles" were reviewed with an aim to summarize the current status and identify future directions for research in this field.

## **Discussion**

Improperly regulated glucose concentration can have a profound effect on the salivary proteome causing changes in the expression of various proteins. This was confirmed in the observational study conducted by a group of scientists in Beijing, China (Jia et al., 2021). Among the 5,721 proteins that were quantified, significant association with Diabetes Mellitus type 2 (DM 2) was demonstrated for 40 proteins. The proteins involved in oxidative stress-related processes were up-regulated, while those associated with saliva secretion were down-regulated. In addition, the three proteins XNDC17, ZG16B and FAM3D enable distinction between states of elevated versus decreased glucose levels. Interestingly, this study is one of the few to use LC – MS / MS relying on the data-independent acquisition (DIA) method to investigate salivary composition. Namely, DIA is a recently-developed global proteomics strategy based on MS, that offers better understanding of pharmacokinetics, pharmacodynamics and facilitates drug development (Jia et al., 2021).

A group of scientists and clinicians from Athens, Greece, detected for the first time approximately 2000 salivary proteins, involved in the pathology of DM 1 by conducting a cohort study with more than 36 pediatric patients. S100-A7, Beta defense 4A, HPSE, AMBP, ALB, A2M, APOA2, LPO, SERPIN, S100A2 were down-regulated among the patients and the following bioinformatics analysis of these proteins suggested their application in preventive therapeutic approach. On the other hand, proteins such as: phospholipid transport

ATPase IF, DENR, KRT7, SPRR1A, CASP4, S100A10, PSMB7 were upregulated (Pappa et al., 2018).

Since the inflammatory mediators are directly associated to the pathology of DM, numerous salivary inflammatory proteins can be used in the clinical management. Thus, a cohort study was performed on 20 subjects diagnosed with DM 2, which identified the adipokines such as resistin, visfatin, and ghrelin as key salivary biomarkers, considering that inflammatory adipokines could be used for early diagnosis in patients with high risk of developing DM 2 (Srinivasan et al., 2018). A study conducted by Agho in 2021 found a significantly increased level of salivary C-reactive protein-CRP, as an indicator of inflammation in patients with DM versus healthy controls. CRP measured by ELISA can be used as a diagnostic and monitoring marker, because of the positive correlation with HbA1C values (Agho et al., 2021). The study by Zhang and the collaborators, evaluated the association of Common Salivary Protein 1 levels with DM, paving the way for further research that could result in replacement of the traditional laboratory analysis of DM using serum as a material with an advanced method. According to their data which were later also confirmed by Fouani, the levels of Common Salivary Protein were significantly increased compared to the controls. This is extremely important, given the fact that rapid diabetes screening is required in order to start the treatment early and thus avoid complications associated with the disease (Fouani et al., 2021; Zhang et al., 2020).

Inadequately controlled glucose concentration is directly related to changes in the oral cavity as well as other complications of DM. Thus, changes in sodium-glucose cotransporter 1-(GLT1) protein and aquaporins (AQP), are associated with xerostomia as a complication of DM. This is particularly noticeable in AQ5 in which increased expression is observed as a result of oxidative stress caused by hyperglycaemia. Along with AQP, nitric oxide synthase- tetrahydrobiopterin protein (NOS-BH4) is a key promoter of hyposalivation in the pathology of xerostomia. This finding can be useful in the development of new therapies for this complication. In addition to these proteins associated with xerostomia, the study of Fouani also pointed salivary amylase as a marker for monitoring DM progression (Fouani et al., 2021). The same year 2021, a research group examined the salivary proteome in adolescents as the most common population group that would benefit from this type of diagnostic approach. The identified proteins such as proline-rich proteins, mucins, histatins, cystatins, and statins protect the tooth surface, attract calcium ions, and promote remineralization. Decreased salivary secretion in children with DM 1 causes changes in salivary protein composition and is significantly associated with caries prevalence. The

increased incidence of caries in this study population is clarified by the decreased regulation of the most differentially expressed proteins, that are involved in the protective mechanisms of caries (Pappa et al., 2021).

## Conclusion

The published literature undoubtedly highlights the saliva potential for performing rapid and accurate diagnosis of DM even in outpatient setting. The pathology and chronic presentation of the disease itself, warrant periodic monitoring which can be conveniently achieved using saliva specimens. This review identified several directions for further research in this field with well-designed studies involving larger cohorts for validation of the proposed biomarkers or identification of others and more importantly standardization of the methodology for sampling and analyzing the saliva with proteomic approach.

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