Review Article

Salivary Biomarkers: A New Wave in Early Disease Detection and Diagnosis

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ABSTRACT

Salivary biomarkers have emerged as a revolutionary tool in the field of medical diagnostics, offering a non-invasive, accessible, and cost-effective approach for early disease detection and monitoring. This review provides a comprehensive examination of salivary biomarkers, highlighting their diverse roles in diagnosing various health conditions. It delves into the composition of saliva and its diagnostic potential, underscoring the advantages of using saliva over other biological fluids and addressing the challenges associated with saliva collection and analysis. Significant advancements in technology that enhance the detection and analysis of salivary biomarkers are discussed, along with breakthroughs in identifying specific biomarkers for diseases such as cancer, cardiovascular diseases, diabetes, autoimmune disorders, and infectious diseases. Ethical and privacy considerations, particularly concerning consent and the implications of early disease detection, are critically examined.

Key Words: Salivary Biomarkers, Non-Invasive Diagnostics, Early Disease Detection, Technological Advancements in Healthcare, Biomarker Sensitivity and Specificity

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INTRODUCTION

Salivary biomarkers are biological molecules found in saliva that can be indicative of various health conditions or diseases. They include a wide range of substances such as enzymes, hormones, antibodies, and genetic materials.[1] The significance of these biomarkers lies in their potential to provide non-invasive, easily accessible, and cost-effective means for early disease detection and monitoring.[2] Unlike blood tests, which require venipuncture and can be more invasive and stressful for patients, saliva collection is simple and can be done without the need for specialized medical personnel or equipment.[3] This aspect of salivary biomarkers holds great promise in transforming healthcare by enabling more widespread and frequent monitoring of health conditions, particularly in settings where traditional medical resources are limited.[4]

The present review paper is particularly important as it consolidates current knowledge and advances in the field of salivary biomarkers. It aims to highlight the molecular mechanisms underlying these biomarkers, their clinical relevance, and the technological advancements that have enabled their detection and analysis. Furthermore, this review will discuss the challenges and prospects in the field, paving the way for further research and development. By providing a comprehensive overview of salivary biomarkers, this paper seeks to underscore their potential in reshaping diagnostic practices and contributing to the evolution of personalized medicine.

METHODS

To conduct a literature survey on Salivary Biomarkers**,** a search was conducted from July 2023 till December 2023 across various electronic databases, including PubMed, SCOPUS, EMBASE, COCHRANE library, and Science Direct. The search utilized MeSH terms/keywords such as "Saliva," "Biomarkers," "Diagnosis," and "Detection." In addition to the electronic search, cross-references and textbooks were manually searched for relevant articles. The inclusion criteria included articles published in the English language from July 2000 to July 2023 that fulfilled the study objectives. The article selection process involved assessing the inclusion and exclusion criteria, as well as conducting a quality assessment. Out of the initial 932 articles identified, 128 were selected based on their titles and abstracts. After evaluating the full texts and applying the inclusion and exclusion criteria, 33 articles were chosen for the review, meeting the study's criteria.

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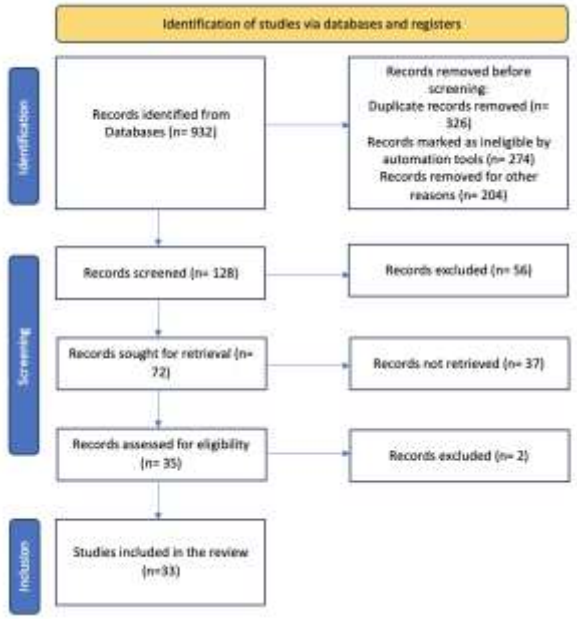

Figure No.1: Flowchart showing the step-by-step identification of the studies via databases

SALIVA AS A DIAGNOSTIC FLUID

Saliva is a multifaceted biofluid that plays a significant role beyond its traditional functions in digestion and oral health.[5] Its complex composition includes proteins, enzymes, hormones, antibodies, nucleic acids, and cells shed from the oral mucosa, as well as electrolytes and other small molecules.[6] These components make saliva a reflective medium of the body's physiological and pathological states, offering a rich source of biomarkers for disease detection and monitoring.[7] For instance, specific proteins in saliva can be indicators of oral health conditions and systemic diseases, while genetic material in saliva can reveal genetic predispositions or the presence of infectious agents.[8] This comprehensive molecular profile underscores the diagnostic potential of saliva, positioning it as a critical tool in non-invasive health assessments.[9]

Advantages of Using Saliva over Other Biological Fluids: Saliva presents several advantages over other biological fluids such as blood in the context of diagnostic applications.[10] Primarily, the collection of saliva is non-invasive, painless, and can be performed without the need for specialized medical staff or equipment, thereby reducing patient discomfort and the risk of infection.[11] This simplicity and ease of collection facilitate frequent sampling, which is crucial for monitoring health conditions over time or assessing the effectiveness of treatments.[12] Additionally, saliva collection is less costly compared to blood draws, making it a more accessible option for regular health monitoring in various settings, including remote or resource-limited environments.[13]

Table No.1: Composition and Diagnostic Potential of Saliva

Component	Description	Diagnostic Potential	Examples
Proteins	Enzymes, immunoglobulins, and other proteins.	Indicators of oral health, inflammatory conditions, and systemic diseases like cancer.	Cyclin D1 and CD44 in saliva has been linked to oral cancer
Hormones	Various hormones secreted into saliva.	Can indicate endocrine function and stress-related conditions.	Elevated levels of cortisol in saliva can indicate stress responses or disorders of the hypothalamic-pituitary-adrenal axis.
Antibodies	Immune proteins that respond to pathogens.	Useful in detecting infections and immune response status.	Presence of HIV antibodies and viral RNA in saliva
Nucleic Acids	DNA and RNA, including microbial genetic material.	Can reveal genetic predispositions, presence of pathogens, and cancer biomarkers.	Detection of HPV DNA in saliva can indicate an increased risk for certain types of oral cancers. Additionally, salivary RNA markers like miR-125a and miR-200a have been associated with oral cancer.
Electrolytes	Salts and minerals such as sodium, potassium, and calcium.	Reflect hydration status and electrolyte balance, potentially indicating metabolic disorders.	Elevated sodium levels in saliva may suggest dehydration, while abnormal potassium levels can indicate adrenal disorders such as Addison's disease.
Cells from Oral Mucosa	Cells shed from the lining of the mouth.	Can be used to detect local oral diseases, and potentially, systemic diseases.	Presence of abnormal or dysplastic cells in saliva can indicate oral cancers or precancerous conditions.
Metabolites	Products of cellular metabolic processes.	Can indicate metabolic conditions and provide insight into overall health.	Elevated levels of certain metabolites like lactate in saliva can indicate tissue hypoxia or intense physical exertion. Salivary uric acid, on the other hand, can be a marker for gout or kidney disorders.

Challenges in Saliva Collection and Analysis: Despite its advantages, saliva collection and analysis present unique challenges. The composition of saliva can be influenced by various factors such as circadian rhythms, food intake, oral hygiene, and individual variability, which may affect the consistency and reliability of biomarker measurements.[14] Additionally, the lower concentration of certain biomarkers in saliva compared to blood requires more sensitive and advanced analytical techniques for accurate detection and quantification.[15] Ensuring the stability of biomarkers during collection, storage, and transport is also critical, as enzymatic activity and other biochemical processes in saliva can alter biomarker profiles. Addressing these challenges is essential for the reliable use of saliva as a diagnostic fluid in clinical settings.

ADVANCES IN SALIVARY BIOMARKERS

In recent years, significant strides have been made in harnessing the power of salivary biomarkers, driven by technological advancements and scientific discoveries.[16] This has opened new avenues for non-invasive, cost-effective, and patient-friendly diagnostic methods, challenging the traditional reliance on blood and other invasive specimens.[17] The following sections will delve into the latest technological developments in salivary diagnostics and the remarkable breakthroughs in identifying specific biomarkers for a variety of diseases. This exploration not only highlights the progress made but also sets the stage for future innovations that could revolutionize the way health conditions are diagnosed and managed.

Table No.2: Recent technological developments in salivary diagnostics

Technological Development	Description	Impact on Salivary Diagnostics
Nanotechnology-Based Sensors	Utilizes nanoscale materials to enhance sensitivity in detecting biomarkers in low concentrations.[18]	Detection of early-stage diseases by identifying minute quantities of biomarkers.[19]
Point-of-Care Devices	Compact, portable devices that facilitate on-site saliva testing, providing immediate results.[20]	Rapid, convenient, and frequent monitoring of health conditions.
High-Throughput Omics Technologies	Advanced techniques in genomics, proteomics, and metabolomics for comprehensive biomarker profiling.[21]	Discovery of new biomarkers and provides a deeper understanding of disease mechanisms.
Microfluidic Technologies	Miniaturized devices for precise fluid handling with minimal sample volumes, improving analysis efficiency and cost.[22]	Enhances the feasibility of salivary diagnostics by reducing sample size requirements and streamlining the analysis process.
Digital and Mobile Health Integration	Incorporating salivary diagnostics with digital platforms, including mobile health apps for data tracking and analysis.[23]	Promotes patient engagement and personalized monitoring, enabling real-time health management.

Significant breakthroughs in identifying specific biomarkers for diseases: To the esteemed medical community, recent breakthroughs in the field of salivary diagnostics have heralded a new era in disease detection and monitoring.[24] Salivary biomarkers have emerged as critical tools in the non-invasive diagnosis of a spectrum of diseases, providing a window into the body's pathological state through a medium that is easily accessible and patient-friendly.[9] This advancement is particularly significant in oncology, where specific salivary biomarkers have been identified for early cancer detection, including but not limited to oral, breast, and pancreatic cancers.[25] In the realm of infectious diseases, saliva-based diagnostics have proven invaluable for rapid and non-intrusive screening, particularly evidenced in the detection of HIV and COVID-19. Additionally, the identification of salivary biomarkers in autoimmune diseases such as Sjogren's syndrome and systemic lupus erythematosus offers a less invasive diagnostic alternative, enhancing patient comfort and compliance.[26] The implications in neurology are equally promising, with salivary biomarkers opening new pathways for monitoring neurological disorders like Alzheimer's and Parkinson's disease.[27] These breakthroughs not only underscore the immense potential of saliva as a diagnostic fluid but also align with the ongoing shift towards more personalized, patient-centred healthcare practices.

SALIVARY BIOMARKERS IN VARIOUS DISEASES

Biomarkers for Periodontal Disease: Periodontal disease can be effectively monitored using salivary biomarkers. Researchers have identified various biomarkers in saliva that are indicative of periodontal disease.[28] These include inflammatory cytokines such as interleukin-1β (IL-1β) and tumour necrosis factor-alpha (TNF-α), enzymes like matrix metalloproteinases

(MMPs), and bacterial by-products.[29] The presence and levels of these biomarkers correlate with the severity of periodontal disease, providing a useful tool for early detection, monitoring disease progression, and evaluating treatment responses.[30]

Markers for Oral Cancer: In the realm of oral cancer, salivary diagnostics have shown promising potential in early detection and monitoring.[31] Several biomarkers have been identified in saliva that are associated with oral cancer, including specific proteins, DNA mutations, and RNA molecules.[32] For instance, the overexpression of proteins such as cyclin D1 and CD44, as well as alterations in the levels of certain microRNAs, have been linked to oral cancer. Additionally, the presence of tumour-derived DNA in saliva offers a non-invasive means of detecting genetic alterations associated with malignancy.[33] These salivary biomarkers not only facilitate early detection of oral cancer but also aid in monitoring disease progression and response to treatment.

Table No.3: Salivary biomarkers and Systemic diseases

Category	Disease	Salivary Biomarkers	Clinical Relevance
Systemic Diseases	Cardiovascular Diseases	Biomarkers such as C-reactive protein, myoglobin, and troponin.	Early detection and monitoring of cardiovascular events
	Diabetes	Glucose levels, inflammatory cytokines, and glycoproteins.	Monitoring glucose control and detecting complications related to diabetes.
	Autoimmune Diseases	Autoantibodies and inflammatory markers specific to each condition.	Diagnosing and monitoring diseases like Sjogren's syndrome, rheumatoid arthritis, and systemic lupus erythematosus.
Infectious Diseases	HIV/AIDS	HIV antibodies and viral RNA.	Early detection and monitoring of HIV infection
	Hepatitis	Antigens and antibodies specific to hepatitis viruses.	Non-invasive screening and monitoring of hepatitis infections.
	Other Viral and Bacterial Infections	Pathogen-specific antigens and antibodies.	Rapid and non-invasive diagnosis of various infectious diseases.

CHALLENGES AND LIMITATIONS

Variability in Salivary Composition: One of the primary challenges in utilizing salivary biomarkers for diagnostic purposes is the inherent variability in saliva composition. This variability can be influenced by numerous factors including age, gender, diet, circadian rhythms, and overall health status of an individual. For instance, hormonal fluctuations can significantly alter saliva composition, potentially impacting the concentration of certain biomarkers. Furthermore, conditions like dehydration or salivary gland dysfunction can affect saliva flow and composition, thereby influencing biomarker levels.

Sensitivity and Specificity of Biomarkers: Another significant challenge in salivary diagnostics is ensuring high sensitivity and specificity of biomarkers. Sensitivity refers to the ability of a test to correctly identify those with the disease (true positive rate), while specificity relates to the test's ability to correctly identify those without the disease (true negative rate). Many salivary biomarkers, while promising, still face challenges in achieving the levels of sensitivity and specificity that are standard in more established diagnostic methods like blood tests. This limitation can lead to false positives or negatives, impacting the clinical utility of these tests.

Standardization of Collection and Analysis Methods: The standardization of saliva collection and analysis methods is crucial for the reliability and reproducibility of results. Currently, there is a lack of standardized protocols for saliva collection, which can affect the integrity and concentration of biomarkers. Factors such as the time of day, method of stimulation, and collection technique can all influence the results. Additionally, the methodologies used in the analysis of saliva samples can vary significantly, leading to inconsistencies in data interpretation. Developing standardized, universally accepted protocols for saliva collection and analysis is essential to ensure the accuracy and comparability of salivary diagnostic tests across different settings and populations.

FUTURE DIRECTIONS

The future of salivary biomarkers holds great promise for transforming personalized medicine and integrating seamlessly with digital health technologies, revolutionizing individualized treatment plans, enhancing patient outcomes, and necessitating focused research, substantial funding, and collaborative efforts across academia, healthcare, and industry to translate innovations into clinical practice.

ETHICAL AND PRIVACY CONSIDERATIONS

Consent and Privacy in Biomarker Testing: The use of salivary biomarkers for disease detection and monitoring necessitates ensuring informed consent,

comprehensive patient education, and robust privacy protections to safeguard against unauthorized access or misuse of personal health information, thereby upholding ethical standards and fostering patient trust in healthcare.

Ethical Implications of Early Disease Detection: The ethical implications of early disease detection using salivary biomarkers encompass concerns regarding the psychological impact, potential stigma, and discrimination, necessitating comprehensive counselling and support for patients, as well as discussions about the right to choose not to know one's genetic risks, particularly in cases where effective treatments or preventive measures are unavailable.

CONCLUSION

The review highlights the potential of salivary biomarkers for early disease detection and monitoring, emphasizing technological advancements, challenges such as variability and ethics, and the promise of integration into personalized medicine and digital health for improved patient care.

Author's Contribution:

Concept & Design of Study:	Faraj Alotaiby
Drafting:	Faraj Alotaiby
Data Analysis:	Faraj Alotaiby
Revisiting Critically:	Faraj Alotaiby
Final Approval of version:	Faraj Alotaiby

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