Review Article

Salivary biomarkers and temporomandibular disorders: A systematic review

ABSTRACT

Temporomandibular disorders (TMD) are a common condition affecting the musculoskeletal group evoking clinical signs such as pain, restricted mouth opening, and disability in the temporomandibular joint (TMJ), masticatory musculature, and the osseous structures in the surroundings. Saliva is a strong proponent of a diagnostic and prognostic tool for TMDs. Hence, a systematic review was undertaken to answer the research question "What is the role of salivary biomarkers in the identification of TMD?" A thorough literature search was performed in databases of PubMed, Embase, and Google Scholar till February 2022. Every included study was characterized by Study ID, location, sample size, demographic information, biomarker analysis, assessment method, and results. Newcastle-Ottawa scale was used to assess the methodological quality of all qualifying research. A total of eight articles were included for the review after screening the titles, abstracts, and full-text articles. The review included articles of observational design with a control group. TMD disorders were confirmed both clinically and radiographically in the study of Shoukri *et al.* TMDs are commonly prevalent in maxillofacial conditions. Despite the availability of various diagnostic techniques, certain limitations are remarkable. The researchers are yet to ascertain a gold standard biomarker to identify TMD.

Keywords: Diagnosis, maxillofacial, pain, saliva, TMD

INTRODUCTION

Temporomandibular disorders (TMD) are a common condition affecting the musculoskeletal group evoking clinical signs such as pain, restricted mouth opening, and disability in the temporomandibular joint (TMJ), masticatory musculature, and the osseous structures in the surroundings.[1,2] It affects around 5–12% of the population[3] with enhanced occurrence in adolescents, is pronounced in the mid-age, and diminishes with progressing age.[4,5]

TMD does not attribute to a single anatomical cause; as a result of TMJ degeneration, disk displacement, and pain within the masticatory muscle.[6] Degeneration can occur due to several reasons such as degenerative joint pathology, osteoarthritis (OA), autoimmune arthritis, and exacerbation of mechanical stressors. Stimulation of nociceptors results in elevated levels of neuropeptides, inflammatory mediators, and hypoxia; this results in pain and functional issues with consequent degeneration of joints and muscles and induces stress. Hence, TMD is considered a condition exhibiting heterogeneous pathologies.

TMJ-related problems not only pose problems in their diagnosis but also in establishing the best interventional course. Additionally, the variations in TMD findings between individuals at various times make TMD diagnosis more difficult.[7] Sufficient knowledge is necessary to frame a suitable treatment for the established diagnosis.

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Literature shows saliva as a potential diagnostic aid for TMD because of the exchange of substances from the blood.[8] The transfer of compounds from the saliva by active transport, diffusion across the cell membrane, or passive diffusion is made possible by a thin layer of epithelial cells isolating the salivary ducts from the circulatory system.[8] Furthermore, certain disease-specific biomarkers are found only in saliva and not in blood. Salivary samples are much easier to collect, are non-invasive, and are also cost-effective.[9] Considering all these, saliva is a strong proponent as a diagnostic and prognostic tool for TMDs. Hence, a systematic review was undertaken to answer the research question "What is the role of salivary biomarkers in the identification of TMD?"

P—Human population affected with TMD
C—Human subjects without TMD
O—Expression of salivary biomarkers
S—Observational, case–control study

METHODOLOGY

Data sources and literature search strategy

A thorough literature search was performed in databases of PubMed, Embase, and Google Scholar till February 2022. The search strategy included articles published only in the English language and was based on keywords such as "Biomarkers OR prognostic OR marker OR diagnostic" AND "Temporomandibular disorders OR Temporomandibular diseases." A manual search of the unpublished literature along with the references of the included citation was also conducted. The present systematic review is as per the recommendations of the preferred reporting items for systematic reviews and meta-analyses checklist.[10]

Eligibility criteria

The inclusion of studies for the review was based on (1) original studies published in English with full-text available, (2) research with temporomandibular disorders, (3) the expression of salivary biomarkers in TMD.

Animal studies, conference proceedings, reviews, letter to the editor, and chapters in the book were excluded. Studies not related to the topic's context or those with insufficient data were not included.

Study selection and data extraction

Endnote software was used to remove duplicate records. After screening the titles and abstracts independently, eligible articles were identified by two researchers. In case of any discrepancy, it was sorted out by a third investigator. Every included study was characterized by Study ID, location, sample size, demographic information, biomarker analysis, assessment method, and results.

Outcome assessment

The recruited studies were checked for the score of salivary biomarkers between the case and control groups.

Quality assessment (Risk of bias)

Newcastle-Ottawa scale (NOS)[11] was used to assess the methodological quality of all qualifying research. NOS scores each article between 0 and 9 based on three evaluation criteria: selection, comparability, and exposure or outcome. Studies of good quality were defined as articles having NOS scores above 6. Any differences of opinion between investigators were settled by constructive agreement.

RESULTS

A total of eight articles were included for the review after screening the titles, abstracts, and full-text articles [Figure 1]. Study characteristics of included studies are shown

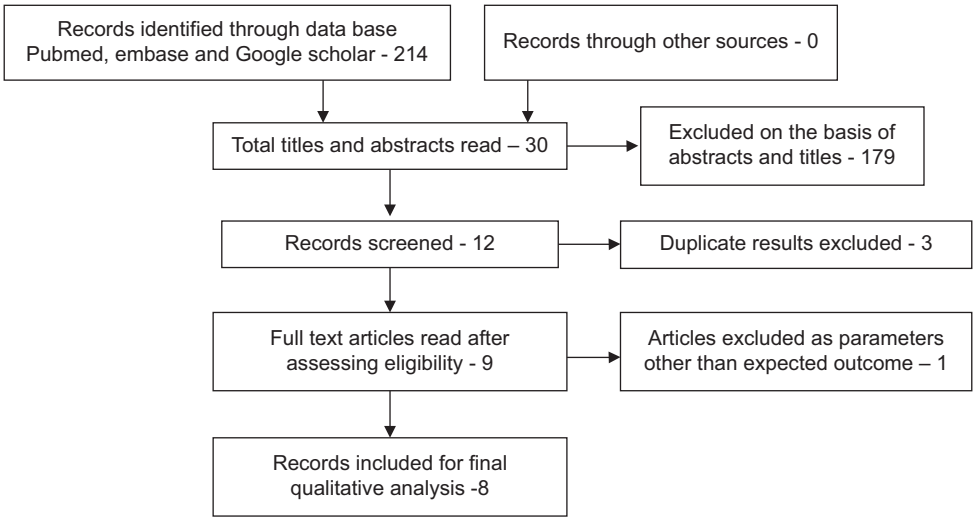

Figure 1: Flow chart diagram for article inclusion

in Table 1.[12-19] When checked for the methodological quality of studies, all the studies scored 7 suggesting a good low risk of bias as seen in Table 2.

The review included articles of observational design with a control group. TMD disorders were confirmed both clinically and radiographically in the study of Shoukri *et al*.[15] The studies of Hajer Jasim *et al*.,[13] Kobayashi *et al*.,[17] and Lalue Sanches *et al*.[12] diagnostically confirmed TMD by a calibrated researcher. Hajer Jasim *et al*.[13] and Kobayashi *et al*.[17] also tested for the diurnal variation of saliva on TMD. The study of Shoukri *et al*.[15] correlated the detected biomarkers of inflammation with the morphological presentation of condyles using artificial intelligence among patients affected with TMJ osteoarthritis.

A clear female predilection was noted in the study of Hajer Jasim *et al*.[13] Lalue Sanches[12] and Cê *et al*.[16] conducted their study with only female recruits. A significant correlation of biomarkers with TMD was noted in seven studies, but not in the study of Kobayashi *et al*.[17]

Table 1: Characteristics of studies included

Study ID	Location	Sample population	Demographical population	Biomarkers assessed	Method of assessment	Study findings
Monique Lalue Sanches *et al*. 2020[12]	Sao Paulo	26 females with TMD and 27 controls	Mean age of 41.0+12.14 years	Phenylacetate, dimethylamine, maltose, acetoin, and isovalerate	Metabolomic analysis through H-NMR (nuclear magnetic resonance) spectroscopy	The metabolomic profile of the case group prior to any intervention differed significantly from the control group at $P<0.002$
Hajer Jasim *et al*. 2020[13]	Sweden	39 cases reporting with TMD myalgia were compared with adequately matched 39 controls	Cases had a mean age of 28.8+7.4 years; 32 females and 7 males in each group	Glutamate, serotonin (5-HT), nerve growth factor (NGF), brain derived neurotrophic factor (BDNF), and substance *P* (SP)	Glutamate was analyzed through ISCUSS analyzer, NGF, and BDNF via multiplex electrochemiluminescence spray, SP, and 5-HT through enzyme immunoassay kit	Salivary glutamate levels were higher in cases (40.22±13.23 ∝mol/L) than in controls (33.24±11.27 ∝mol/L)
Khayamzadeh *et al*. 2019[14]	Iran	32 cases of TMD and 32 controls	Not mentioned	Cortisol and SAA	ELISA testing for cortisol and Photometric method for SAA	Cortisol and SAA levels were significantly elevated in cases at $P=0.011$ and $P=0.044$
Shoukri *et al*. 2019[15]	USA	17 patients affected with TMJ osteoarthritis for <10 years were compared with age and gender matched the same number of controls	Mean age is 39.9+11.7 years	6Ckine, ANG, BDNF, CXCL16, ENA-78, GM-CSF, IFN- , IL-1 , IL-6, MMP-3, MMP-7, PAI-1, TGF- 1, TIMP-1, TNF- , VE-cadherin, and VEGF	Custom human quantibody protein microassay	MMP-3, VE-Cadherin, 6C-Kine, and PAI-1 were positively expressed with significant correlation to condylar variation
Cê *et al*. 2018[16]	Brazil	18 females were affected with TMD as cases and 27 in the control group	In the age range of 18–84 years	Salivary Interleukin-1 (IL-1)	ELISA	IL-1 levels were higher in TMD group
Kobayashi *et al*. 2017[17]	Brazil	38 children diagnosed with TMD and 38 without	10.63+1.68 years in both case and control groups	Salivary stress biomarkers - Cortisol and salivary alpha amylase (sAA)	Cortisol was assessed via ELISA technique while sAA levels were through an automated technique	Salivary stress biomarkers were not significantly associated with TMD
Lawaf *et al*. 2015[18]	Iran	Group 1–28 cases of TMD having pain; Group 2–28 cases of TMD without pain; Group 3–28 controls	29.50±3.8 years; 14 females and 14 males	Total Antioxidant capacity (TAC)	Spectrophotometric analysis	No significant difference was noted between the three groups
Rodriguez de Sotillo *et al*. 2011[19]	Canada	20 female patients with TMD and 10 females as controls	Mean age of cases 40.50±15.53 years	ELISA	8-hydroxydeoxyguanosine (8-OHdG), Malondialdehyde, and total antioxidant status	Mean values of 8-OHdG were higher in cases than controls, significant at $P<0.001$ and $P=0.002$, respectively

Table 2: Newcastle-Ottawa rating for included studies

Study ID	Is the case design adequate	Selection			Comparability	Ascertainment of exposure	Outcome	Non-response	Total score
		Case representativeness	Control selection	Definition of controls	Comparability of cases and controls		Same method of ascertainment		
Monique Lalue Sanches *et al.* 2020	*	*	*	*	*	*	*	-	7
Hajer Jasim *et al.* 2020	*	*	*	*	*	*	*	-	7
Mina Khayamzadeh *et al.* 2019	*	*	*	*	*	*	*	-	7
Shoukri *et al.* 2019	*	*	*	*	*	*	*	-	7
Patricia S Ce *et al.* 2018	*	*	*	*	*	*	*	-	7
Kobayashi *et al.* 2017	*	*	*	*	*	*	*	-	7
Lawaf *et al.* 2015	*	*	*	*	*	*	*	-	7
Rodriguez de Sotillo *et al.* 2011	*	*	*	*	*	*	*	-	7

DISCUSSION

Systematic reviews rank higher in evidence-based decision-making when conducted with discrete methodological details with the inclusion of high-quality studies. This review aimed to evaluate the diagnostic reliability of salivary biomarkers in TMD assessment. TMD is a diverse group of diseases. TMJ-related problems are difficult to diagnose, and there is debate regarding the best course of action. Additionally, the variations in TMD findings among people at various times add to the difficulties in TMD diagnosis. To create a suitable treatment in response to the established diagnosis, sufficient knowledge is necessary.

Biomarker refers to "a characteristic that is objectively measured and evaluated as an indicator of normal biological process, pathogenic processes, or pharmacologic responses to a therapeutic intervention."[20] An ideal biomarker should have a number of key tenets, including the necessity of being present in all patients diagnosed (e.g., high sensitivity and specificity), disease specificity, detection before overt clinical symptoms are visible, and reversibility following the administration of the appropriate therapy. The optimal biomarkers should also allow for a cut-off value with little overlap between a healthy condition and disease, information demonstrating the cumulative history of the illness, and not simply reflecting the severity of the illness. Additionally, it is anticipated that a biomarker-based optimal diagnostic strategy will lower the overall cost of the diagnosis. The expenses associated with measurement and incorrect diagnosis would be combined to provide the economic value in this situation.[21]

A unique set of disease biomarkers would contribute to context-relevant, approachable, and greatly simplified methods of TMD diagnosis once they had achieved sufficient specificity and sensitivity. However, there is no panel of appropriate and direct disease indicators for TMDs that is often employed in clinical settings. Based on the previously covered cumulative data, there is sufficient progress, indicating the attempts to conduct a full appraisal of this topic and future research objectives.

A varied array of biomarkers was tested in the current review. Though the majority of the studies do show a significant association of biomarkers in TMD, none of them are validated at the genetic or molecular level with validation still remaining in infancy. Salivary cortisol was assessed in two studies[14,17] as a stress biomarker of TMD. The hormone secreted by hypothalamus–pituitary–adrenal axis is correlated with stressful periods and also with increased masticatory muscle activity.[22] Four

studies (Cê *et al.*, Rodríguez de Sotillo *et al.*, Lawaf *et al.*, and Hayer Jasim *et al.*) in the review evaluated pain biomarkers in TMD. In addition to inflammatory cytokines, neurotransmitters, and neuropeptides have also been found to be biological indicators of long-standing TMDs. It has been shown that neuropeptide and neurotransmitter status might change as a result of peripheral tissue injury or nerve malfunction, which frequently results in chronic sensitization of peripheral pathways. Additionally, the function of these molecular neurotransmitters and peptides in the mechanism of central sensitization, which is the aberrant amplification of ascending peripheral input or the dysregulation of the inhibitory modulatory system, has been identified.[23] Summatively, seven studies showed a significant correlation between salivary biomarkers and temporomandibular disorders suggesting their probable role in diagnostics.

To the best of the author's knowledge, no other systematic review of literature is published in this context. All the studies included had adequately matched controls thus combating the presence of confounding factors, which if present could have distorted the review results. The inclusion of all high-quality articles (NOC score = 7) reduced the possibility of skewed results. The sample size in the included studies was small and no multicentric trials were identified in the search; coupled with heterogeneity in clinical presentation, validation of these studies as conclusive evidence remains unclear. It is still unknown whether these putative biomarkers can meet reliability and practical requirements (such as cost and speed) to close the implementation-to-validation gap.

CONCLUSION

TMDs are commonly prevalent in maxillofacial conditions. This systematic review aims to answer the research question—What is the role of salivary biomarkers in the identification of TMD? The data reviewed in the present paper highlight the role of various biomarkers that showed an association with TMDs. Despite the availability of various diagnostic techniques, certain limitations are remarkable as only seven studies showed an association of salivary biomarkers with TMD. The researchers are yet to ascertain a gold standard biomarker to identify TMD.

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Conflict of interest

There are no conflicts of interest.

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