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PRECISION DIAGNOSTICS IN ORAL SQUAMOUS CELL CARCINOMA: THE EMERGING ROLE OF MICRORNA-375: A SYSTEMATIC REVIEW

Systematic Review

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ABSTRACT

Background: Oral squamous cell carcinoma (OSCC) is a prevalent malignancy of the oral cavity, frequently diagnosed at advanced stages due to the lack of reliable early detection biomarkers. MicroRNA-375 (miR-375), a known tumor suppressor, has emerged as a potential non-invasive diagnostic biomarker owing to its consistent downregulation in malignant tissues and body fluids. Its regulatory role in tumor progression through interaction with oncogenic pathways underscores its clinical significance.

Objective: This systematic review aimed to evaluate the diagnostic accuracy and expression profile of miR-375 in OSCC across different biological samples, including tissue, saliva, plasma, and oral cytology specimens.

Methods: A comprehensive search of PubMed, Scopus, and Google Scholar was conducted from June 1 to June 30, 2025, following PRISMA guidelines. Boolean search strings combining "miR-375," "oral squamous cell carcinoma," and "diagnostic biomarker" were used. Studies published between 2011 and 2025 were included if they assessed differential miR-375 expression in human OSCC and reported diagnostic metrics such as AUC, sensitivity, and specificity. Screening and data extraction were performed independently by two reviewers, and risk of bias was evaluated using the QUADAS-2 tool.

Results: Out of 66 initial records, 11 studies met inclusion criteria. All reported significant miR-375 downregulation in OSCC relative to controls. Diagnostic accuracy was high across specimen types, with AUC values ranging 0.90–0.96, sensitivity 80–100%, and specificity 64–100%. One study identified miR-375 as a marker differentiating progressive from non-progressive premalignant lesions. Mechanistic analyses revealed that reduced miR-375 expression promotes oncogenesis via deregulation of MYC and CIP2A pathways.

Conclusion: miR-375 is consistently underexpressed in OSCC and demonstrates robust diagnostic potential across tissue and liquid biopsy platforms. Standardized, multicenter prospective studies are needed to validate its application in clinical screening and early detection.

Keywords: biomarker, diagnostic accuracy, early detection, liquid biopsy, microRNA-375, oral squamous cell carcinoma, systematic review.

INTRODUCTION

Oral squamous cell carcinoma (OSCC) remains one of the most prevalent and aggressive malignancies of the oral cavity, contributing substantially to global morbidity and mortality. Despite advancements in surgical and adjuvant therapeutic interventions, the overall prognosis for OSCC patients remains poor, primarily due to the disease being diagnosed at advanced stages. This delayed diagnosis is largely attributed to the absence of reliable, sensitive, and specific biomarkers capable of detecting early pathological changes in oral mucosal tissues (1,2). Consequently, a major research focus has emerged within precision oncology to identify molecular determinants that can enhance the diagnostic accuracy, prognostic prediction, and individualized treatment strategies for OSCC. Among the molecular entities under investigation, microRNAs (miRNAs) have gained significant attention for their post-transcriptional regulatory roles in gene expression. These small non-coding RNA molecules have been implicated in multiple biological processes, including cell proliferation, differentiation, apoptosis, and metastasis (3-5). Their stability in biological fluids and tissue specificity make them promising noninvasive biomarkers in cancer diagnostics. Among them, microRNA-375 (miR-375) has emerged as a particularly relevant candidate in oral cancer biology due to its tumor-suppressive properties and consistent deregulation in malignancies of epithelial origin (6-8). Accumulating evidence suggests that miR-375 expression is markedly reduced in OSCC tissues when compared with normal oral mucosa, and this downregulation correlates with advanced tumor grade, increased metastatic potential, and poorer patient survival outcomes (9,10).

Functionally, miR-375 inhibits key oncogenic pathways by targeting molecules such as platelet-derived growth factor-A (PDGF-A) and insulin-like growth factor 1 receptor (IGF-1R), thereby suppressing tumor cell proliferation, invasion, and migration (11). This mechanistic insight underscores its potential clinical utility not only as a diagnostic and prognostic marker but also as a therapeutic target in precision cancer management. Furthermore, the clinical implications of miR-375 extend beyond its tumor-suppressive role. Its detection in noninvasive biological samples such as saliva and serum has opened new avenues for early diagnosis and risk stratification among OSCC patients (12-15). These developments suggest that miR-375 could play a pivotal role in bridging the gap between molecular research and clinical practice, particularly within the evolving paradigm of precision medicine. Given the expanding body of evidence, there is a compelling need to systematically consolidate and evaluate current research on miR-375 to better define its diagnostic, prognostic, and therapeutic significance in OSCC. Therefore, the objective of this systematic review is to integrate existing literature to elucidate the molecular role of miR-375 in the pathogenesis of OSCC and to rationalize its potential as a precision biomarker for early detection, prognosis, and targeted therapeutic intervention in oral cancer.

METHODS

This systematic review was conducted following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines to ensure methodological transparency and reproducibility (10). As registries that do not include health outcomes are currently ineligible for registration on PROSPERO, this review was not prospectively registered. The study adhered to established principles for evidence synthesis, focusing on diagnostic accuracy studies related to microRNA-375 (miR-375) in oral squamous cell carcinoma (OSCC). A comprehensive and systematic search strategy was developed to identify all relevant studies evaluating the diagnostic significance of miR-375 in OSCC. Electronic databases including PubMed, Scopus, and Google Scholar were searched between June 1, 2025, and June 30, 2025. Boolean operators and Medical Subject Headings (MeSH) terms were employed to enhance search sensitivity and precision. The search strategy integrated keywords such as “microRNA-375,” “miR-375,” “oral squamous cell carcinoma,” “oral cancer,” “OSCC,” “diagnosis,” “diagnostic,” and “biomarker.” The detailed database-specific strategy and number of results retrieved are summarized in Table 1. To minimize bias, filters were applied to restrict results to studies published between 2011 and 2025 in English. The review was guided by a clearly defined PICO framework. The population included human participants with histopathologically confirmed OSCC. The index test was the expression analysis of microRNA-375 (miR-375) in biological specimens such as tumor tissue, plasma, serum, or saliva. Comparators included healthy individuals, adjacent non-cancerous tissue, or patients with premalignant oral conditions or lesions. The outcomes of interest were diagnostic performance measures (e.g., sensitivity, specificity, and area under the ROC curve) and significant differential expression levels of miR-375 ($p < 0.05$) between OSCC and control groups. Eligible study designs included original observational research such as case–control, cross-sectional, and cohort studies.

INSIGHTS-JHR
INSIGHTS-JOURNAL OF HEALTH AND REHABILITATION

Studies were excluded if they were conducted on non-human models (animal or in vitro), focused solely on prognostic or therapeutic roles of miR-375 without diagnostic evaluation, or were reviews, case reports, conference abstracts, letters, or editorials. Articles not published in English or lacking accessible full texts were also excluded to ensure methodological rigor and reproducibility. The selection process involved a two-phase screening approach to ensure transparency and reliability. In the first phase, titles and abstracts were independently screened by two primary reviewers using predefined inclusion and exclusion criteria. In the second phase, the full texts of potentially eligible articles were retrieved and reviewed collaboratively by the lead and supervisory reviewers. All discrepancies were discussed until full consensus was achieved, and no conflicts were reported during the process. Reference management and duplicate removal were performed using EndNote software. The PRISMA flow diagram was employed to illustrate the selection and screening process, documenting the number of studies identified, excluded, and finally included for qualitative synthesis. Data extraction was conducted manually by the primary reviewer and cross-verified by the supervisory reviewer to ensure accuracy. No automation tools, digitizing software, or translation services were employed at any stage. Extracted data included the first author, year of publication, country, study design, sample size, patient demographics, type of biological specimen, detection method (e.g., RT-qPCR), normalization technique, and main diagnostic outcomes such as expression levels, sensitivity, specificity, ROC analysis, and area under the curve (AUC) values. In cases where multi-report studies were encountered, inclusion decisions were made by consensus among the reviewing team. When reporting was incomplete, interpretations were made conservatively based on available methodological details.

The primary outcome of interest was the diagnostic expression profile of miR-375 in OSCC across different biological matrices, with special emphasis on studies analyzing circulating miR-375 as a noninvasive biomarker. Secondary characteristics such as presence of control groups, source of study funding, and reporting transparency were also recorded to contextualize the findings. The methodological quality and potential risk of bias of included studies were evaluated using the Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2) tool, following the recommendations of the Cochrane Handbook for Systematic Reviews of Diagnostic Test Accuracy (11). Each study was independently assessed by two reviewers across four domains: patient selection, index test, reference standard, and flow and timing. Judgments of “low risk,” “high risk,” or “unclear risk” were assigned based on consensus, and results of the quality assessment are presented in Table 2. Overall, the methodology of this review was designed to ensure comprehensive coverage of the literature, minimize bias, and provide a robust qualitative synthesis of the diagnostic role of miR-375 in OSCC, thereby establishing an evidence-based foundation for its potential integration into precision diagnostic frameworks.

Table: Comprehensive Database Search Strategy for Identification of Studies Assessing Diagnostic Role of microRNA-375 (miR-375) in Oral Squamous Cell Carcinoma (OSCC)

Data-base	Search no:	Search Component	Search Terms	Search Field / Operators	Filters Applied	Results Returned
Pubmed	1	microRNA-155 terms	("microRNA-375" OR "miR-375")	All fields / Boolean operators	Filters: Year: 2011–2025	1,358 results
	2	Oral cancer terms	("oral squamous cell carcinoma" OR "oral cancer" OR "OSCC")	All fields / Boolean operators	Filters: Year: 2011–2025	24,896 results
	3	Diagnostic terms	(Diagnosis OR diagnostic OR biomarker)	All fields / Boolean operators	Filters: Year: 2011–2025	6,205419 results
	4	Final combined search	("microRNA-375" OR "miR-375") AND ("oral squamous cell carcinoma" OR "oral cancer" OR "OSCC") AND (diagnosis OR diagnostic OR biomarker)	All fields / Boolean operators (combined search components 1–3)	Filters: Year: 2011–2025	25 results
Scopus	1		TITLE-ABS-KEY ("microRNA-375" OR "miR-375")	All fields / Boolean operators	Filters: Year:	41

INSIGHTS-JHR
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Data-base	Search no:	Search Component	Search Terms	Search Field / Operators	Filters Applied	Results Returned
			AND TITLE-ABS-KEY ("oral squamous cell carcinoma" OR "oral cancer" OR "OSCC") AND TITLE-ABS-KEY (diagnosis OR diagnostic OR biomarker)		2011-2025	
Google Scholar	1		("miR-375" OR "microRNA-375") "oral squamous cell carcinoma" diagnosis filetype:pdf		Filters: English Year: 2011–2025	140 results

RESULTS

A total of 66 studies were initially identified through the electronic database search—25 from PubMed and 41 from Scopus—using the structured Boolean strategy designed for this review. After the removal of duplicates and preliminary screening of titles and abstracts, 11 studies were shortlisted for full-text assessment based on relevance to the diagnostic role of microRNA-375 (miR-375) in oral squamous cell carcinoma (OSCC). During the second round of screening, two studies were excluded: one by Jia et al. (12), written in Japanese and therefore ineligible due to translation limitations, and another by Gissi et al. (13), which excluded miR-375 from analysis owing to undetectable expression in 62% of samples and lack of diagnostic data. Consequently, 11 studies met the inclusion criteria and were incorporated into the qualitative synthesis. Google Scholar was searched as a supplementary source to ensure comprehensiveness, but no additional eligible studies were retrieved. The included studies, conducted between 2011 and 2024, represented diverse geographical regions including China, Japan, the United States, India, Italy, Denmark, Taiwan, and Ukraine, reflecting broad international interest in the diagnostic potential of miR-375. Most studies employed case-control or observational designs, while a few utilized experimental or translational approaches. Sample sizes ranged from 10 to 61 participants, with biological specimens including fresh-frozen tissue, saliva, serum, plasma, oral cytology, and formalin-fixed paraffin-embedded (FFPE) tissue. The most commonly used detection techniques were reverse transcription quantitative polymerase chain reaction (RT-qPCR), TaqMan assays, microarray profiling, and digital PCR, ensuring methodological consistency across datasets.

A consistent pattern of miR-375 downregulation in OSCC was reported across the majority of studies, underscoring its tumor-suppressive role. Notably, Zhang et al. (6) and Tsai et al. (16) observed significantly reduced miR-375 expression in tumor tissues compared with adjacent normal mucosa ($p < 0.01$), while Jung et al. (17) reported a progressive decrease in miR-375 expression from early to advanced tumor stages, demonstrating an inverse correlation with oncogenic CIP2A expression ($r = 0.53$, $p = 0.03$). Similarly, Narasimhan et al. (1) identified significant differential expression between OSCC and paired controls ($p < 0.05$), although no formal ROC analysis was presented. In contrast, Burtyn (14) reported an upregulation of miR-375 in saliva samples of OSCC patients, suggesting potential biological variability linked to sample type or tumor microenvironment. Several studies also provided quantitative diagnostic performance data, highlighting the clinical relevance of miR-375 as a biomarker. Saika et al. (7) demonstrated an area under the ROC curve (AUC) of 0.942, with 80% sensitivity, 100% specificity, and 90% diagnostic accuracy in distinguishing early OSCC cases with latent lymph node metastasis. Qianting He (18) reported similar diagnostic strength, with AUC values of 0.90 (cytology) and 0.91 (tissue), alongside sensitivities and specificities exceeding 80% in most comparisons. Crimi et al. (19) validated these findings through plasma-based assays, reporting an AUC of 0.96, sensitivity of 80%, and specificity of 100%, supporting the potential of circulating miR-375 as a noninvasive diagnostic biomarker. Furthermore, Lajer et al. (20) integrated miR-375 into an eight-miRNA classifier achieving an AUC of 0.94, with 91% sensitivity and 83% specificity, indicating its robust predictive power when combined with other miRNAs.

In terms of demographic and clinical characteristics, the included studies predominantly involved adults diagnosed with histopathologically confirmed OSCC, with age distributions typically ranging from 30 to 70 years. While gender distribution was variably reported, most studies did not stratify findings by sex or tumor site, representing a minor limitation in the comparative analysis.

INSIGHTS-JHR
INSIGHTS-JOURNAL OF HEALTH AND REHABILITATION

Nonetheless, the inclusion of diverse sample types—particularly tissue and saliva—highlighted the growing exploration of miR-375 as both an invasive and noninvasive biomarker. The risk of bias and applicability concerns of the included studies were assessed using the QUADAS-2 tool, as summarized in Table 3. Overall, most studies demonstrated a low risk of bias across all domains, indicating strong methodological reliability. Specifically, studies by Zhang et al. (6), Saika et al. (7), Tsai (18), and Lajer et al. (20) were rated as low risk in patient selection, index test, and reference standard domains. Minor concerns arose in studies by Crimi et al. (19) and Narasimhan et al. (1), primarily due to small sample sizes and unclear blinding procedures, while a few studies showed uncertainty in flow and timing domains. No significant applicability concerns were identified, suggesting that the evidence was consistent with the review objectives. Collectively, the synthesis of results reinforces that miR-375 is consistently under-expressed in OSCC tissues relative to normal or premalignant controls and exhibits promising diagnostic potential, as reflected by multiple studies reporting high AUC values and significant discriminatory ability. Although minor methodological heterogeneity was observed in sample types and assay protocols, the collective evidence strongly supports the role of miR-375 as a candidate biomarker for early detection and molecular stratification of OSCC within the framework of precision diagnostics.

Table 1: Summary of Included Studies Assessing Expression Patterns and Diagnostic Performance of microRNA-375 (miR-375) in Oral Squamous Cell Carcinoma (OSCC)

First Author (Year)	Country	Study type	Sample Type	OSCC Sample Size	Control Type (ACF/normal)	Control Size (n)	Detection Method	Expression Pattern of miR_375	Diagnostic Findings
Burtyn (2024) (14)	Ukraine	Observational clinical study	Saliva	61	Normal healthy volunteers	10	RT-PCR (TaqMan assay)	Upregulated (2.84× higher vs. control, $p<0.05$)	miR-155 was significantly elevated in OSCC saliva; positively correlated with tumor size (T index, $r = 0.75$) and nodal metastasis ($r = 0.71$). miR-155 levels were higher in resistant tumors and associated with poor NACT response ($\chi^2 = 3.894$, $p = 0.09$).
Zhang et al. (2017) (6)	China	Experimental (in vitro & ex vivo)	Fresh frozen tissue & SCC-4 cell line	44 patients	Normal oral epithelium	44 normal tissues	qRT-PCR, luciferase reporter assay	Downregulated in OSCC	miR-375 was significantly downregulated in OSCC vs. adjacent normal tissue ($P<0.001$).

First Author (Year)	Country	Study type	Sample Type	OSCC Sample Size	Control Type (ACF/normal)	Control Size (n)	Detection Method	Expression Pattern of miR_375	Diagnostic Findings
									Low miR-375 expression was associated with lymph node metastasis and poorer overall survival (P=0.0076).
Saika et al. (2024) (7)	Japan	Case-control study	Primary eOSCC tissue	eOSCC tissue 30 (15 with latent LNM, 15 without)	Adjacent normal mucosa	Not explicitly stated	Microarray, RT-qPCR, Digital PCR	Downregulated in eOSCC with latent cervical LNM	AUC = 0.942; Sensitivity = 80%, Specificity = 100%, Accuracy = 90%. miR-375-3p significantly downregulated in cases with latent LNM; potential predictive biomarker for occult metastasis
Harrandah (2016) (15)	USA	Case-control using FFPE archival tissues	FFPE tissue (progressive lesions and OSCC)	31 OSCC (paired with progressive premalignant lesions)	Non-progressive premalignant lesions	6	RT-qPCR (TaqMan assays)	Downregulated in OSCC vs. progressive lesions; downregulated in progressive vs. non-progressive lesions	miR-375 significantly distinguished progressive from non-progressive lesions (p=0.0004); ROC AUC not reported; strong prognostic potential suggested
Qianting He(2016) (16)	China / USA	Diagnostic case-control	Oral cytolog	19 cytology, 10 + 12	Normal healthy subjects	20 (cytology), 10	qRT-PCR (TaqMan), Deep	Downregulated in	AUC (cytology) = 0.90; AUC

First Author (Year)	Country	Study type	Sample Type	OSCC Sample Size	Control Type (ACF/normal)	Control Size (n)	Detection Method	Expression Pattern of miR_375	Diagnostic Findings
			y & tissue	tissue (training + TCGA)	(cytology) + Adjacent normal tissues (tissue)	+ 12 (tissue)	sequencing (TCGA)	OSCC vs. normal	(tissue) = 0.91; Sensitivity = 100%, Specificity = 64% (cytology); Sensitivity = 83%, Specificity = 83% (tissue); miR-375 combined with miR-21 showed high diagnostic potential
Jung et al. (2013) (17)	USA	Experimental	Fresh-frozen tissue & cell lines	17 tumors (6 early, 11 advanced)	Normal oral tongue tissue	5	q RT-PCR	Significantly underexpressed in OSCC; further decreased in advanced tumors	Significant inverse correlation between miR-375 and CIP2A expression (R = 0.53, p = 0.03); miR-375 overexpression suppressed proliferation, migration, invasion; supports diagnostic and prognostic potential
Tsai (2017) (18)	Taiwan	Case-control	Tissue (paired)	39	Adjacent Normal	39	qRT-PCR (TaqMan)	Significantly downregulated	Significantly reduced in OSCC vs. normal tissue (P<0.01); suggested as potential diagnostic and prognostic biomarker

INSIGHTS-JHR
INSIGHTS-JOURNAL OF HEALTH AND REHABILITATION

First Author (Year)	Country	Study type	Sample Type	OSCC Sample Size	Control Type (ACF/normal)	Control Size (n)	Detection Method	Expression Pattern of miR_375	Diagnostic Findings
Crimi et al. (2020) (19)	Italy	Case-control pilot study	10 OSCC patients, 10 healthy controls	Plasma (liquid biopsy)	ddPCR	hsa-miR-375-3p, hsa-miR-133a-3p	Histopathology and clinical diagnosis	miR-375-3p: AUC 0.96, Sensitivity 80%, Specificity 100%	Small sample size; validated computational findings; confirmed miR-375-3p downregulation in OSCC
Lajer (2011) (20)	Denmark	Observational (Translational study)	Fresh-frozen tissue biopsies	51	Normal tissue	37	Microarray (Agilent), validated by RT-qPCR	Downregulated in OSCC vs normal	miR-375 was part of an 8-miRNA SVM classifier with AUC 0.94, sensitivity 91%, specificity 83%
Narasimhan et al,(1) (2023)	India & UAE	Case-control observational study	Tumor tissue	22	Adjacent normal mucosa (paired controls from contralateral site)	22	qRT-PCR (SYBR Green, U6 as reference gene)	Significantly downregulated in OSCC (68.1% cases; mean fold change ≈ 83.9; $p < 0.05$)	Demonstrated significant differential expression of miR-375 between OSCC and controls; suggested diagnostic potential but no formal ROC/AUC analysis reported.
Shi et al.(21) (2015	China	Observational, case–control	FFPE tissue	15	Adjacent normal oral mucosa	15	Microarray + qRT-PCR validation	Significantly **downregulated** in progression from normal → OLP → OSCC	Differential expression of miR-375 demonstrated; no sensitivity/specificity reported, but findings support its role

First Author (Year)	Country	Study type	Sample Type	OSCC Sample Size	Control Type (ACF/normal)	Control Size (n)	Detection Method	Expression Pattern of miR_375	Diagnostic Findings
									as a potential early-stage biomarker for OSCC.

Table 2: Quality Assessment of Included Studies Using the QUADAS-2 Tool for Risk of Bias and Applicability Concerns in Evaluating Diagnostic Accuracy of microRNA-375 (miR-375) in Oral Squamous Cell Carcinoma (OSCC)

Study risk of bias concerns about applicability							
Study ID / Author (Year)	Patient Selection	Index Test	Reference Standard	Flow and Timing	Patient Selection	Index Test	Reference Standard
Burtyn (2024)	L	L	L	L	L	L	L
Zhang et al. (2017)	L	L	L	L	L	L	L
Saika et al. (2024)	L	L	L	L	L	L	L
Harrandah (2016)	L	L	L	L	L	L	L
Qianting He(2016)	L	L	L	L	L	L	L
Jung et al. (2013)	L	L	L	L	L	L	L
Tsai (2017)	L	L	L	L	L	L	L
Crimi et al. (2020)	H	U	L	H	L	L	L
Lajer et al., (2011)	L	U	L	L	L	L	L
Narasimhan et al., (2021)	U	L	L	L	L	L	L
Shi et al., 2015	U	L	L	L	L	L	L

DISCUSSION

The findings of this systematic review underscore the significant diagnostic and prognostic relevance of microRNA-375 (miR-375) in oral squamous cell carcinoma (OSCC). Across multiple included studies, a consistent pattern of miR-375 downregulation was evident in OSCC tissues and biological fluids, confirming its role as a tumor-suppressive molecule involved in oral carcinogenesis. Lower expression levels of miR-375 were found to be strongly correlated with poorer overall survival among patients with oral and laryngeal malignancies (HR 1.23, 95% CI 1.10–1.37), reinforcing its potential as a prognostic biomarker for disease progression and therapeutic outcomes (18,19). Quantitative PCR-based analyses further demonstrated that decreased miR-375 levels were associated with aberrations in critical tumor-suppressor pathways, including those regulated by p53, suggesting its functional contribution to the molecular pathogenesis of OSCC (20,21). Mechanistically, miR-375 inhibits oncogenesis by directly targeting cancerous inhibitor of protein phosphatase 2A (CIP2A), an oncoprotein that stabilizes MYC through suppression of the tumor-suppressor PP2A. Downregulation of CIP2A by miR-375 restores PP2A activity, leading to MYC dephosphorylation and subsequent degradation, thereby reducing tumor cell proliferation and invasion (22,23). The loss of miR-375 expression observed in OSCC therefore indicates disruption of this regulatory axis, contributing to increased oncogenic potential and metastatic behavior. The collective evidence from experimental

and clinical studies thus supports miR-375 as an integral component of oral tumor biology, with diagnostic and therapeutic implications. Comparative evaluation of biological sample types revealed concordant findings across different matrices, including tissue, saliva, plasma, and cytology (24). Tissue-based analyses consistently reported significant downregulation of miR-375 compared with adjacent normal epithelium, with strong associations with lymph node metastasis and adverse clinical outcomes. Studies employing liquid biopsies demonstrated comparable diagnostic accuracy, with plasma-derived miR-375 achieving high discriminative power (AUC 0.96; sensitivity 80%; specificity 100%) in distinguishing OSCC patients from healthy controls. Similarly, the use of oral cytology samples revealed high diagnostic accuracy (AUC 0.90; sensitivity 100%; specificity 64%), supporting the potential of miR-375 as a noninvasive biomarker for early detection (25,26). Notably, the combination of miR-375 with other miRNAs, such as miR-21, further improved diagnostic precision, indicating that multiplex biomarker panels may enhance clinical applicability.

In parallel, evidence from studies comparing miR-375 and miR-155 expression emphasized the complex molecular landscape of OSCC. While miR-375 was consistently downregulated, miR-155 was frequently upregulated, and its elevated expression correlated positively with tumor size and nodal involvement. This contrast highlights the necessity of contextual interpretation and supports the rationale for multi-marker diagnostic models rather than reliance on a single biomarker. Furthermore, the consistent under-expression of miR-375 in premalignant lesions and its ability to discriminate progressive from non-progressive cases suggest its potential role in early risk stratification and preventive oncology. The synthesis of available literature highlights the strength of miR-375 as a reliable diagnostic and prognostic candidate. Its reproducible downregulation across independent cohorts and biological matrices reflects robust biological consistency. The diversity of populations and methodologies also enhances the external validity of the findings. Moreover, mechanistic insights into its regulation of oncogenic pathways strengthen the biological plausibility of its diagnostic value. The incorporation of advanced quantitative techniques, such as droplet digital PCR and high-sensitivity qRT-PCR, further enhances assay precision and clinical translation potential. Nevertheless, several methodological limitations were identified across studies. Heterogeneity in study designs, sample collection methods, normalization strategies, and assay platforms introduces variability in reported expression levels and diagnostic metrics. Sample sizes varied widely, with several studies based on small patient cohorts (10–30 participants), limiting statistical power and generalizability. The lack of standardization in control selection—often confined to healthy individuals or adjacent normal tissues without inclusion of benign or inflammatory oral lesions—may overestimate diagnostic specificity in real-world clinical contexts. Additionally, most included studies were retrospective and observational, limiting causal inference and precluding definitive validation of diagnostic thresholds.

Publication bias could not be excluded due to the English-language restriction, which may have favored the inclusion of studies with positive outcomes. Access limitations to subscription-based databases such as Embase and institutional constraints may have further restricted the retrieval of potentially relevant literature. Furthermore, variability in the reporting of normalization genes and internal controls may have affected comparability of quantitative data across studies. Despite these limitations, the collective body of evidence demonstrates strong and biologically consistent downregulation of miR-375 in OSCC, corroborating its role as a tumor-suppressor microRNA with diagnostic and prognostic utility. Future research should prioritize large-scale, multicenter prospective trials with standardized pre-analytical and analytical protocols, inclusion of clinically relevant control groups such as premalignant oral conditions, and exploration of combinatorial miRNA panels incorporating miR-375, miR-21, and miR-155. Integration of miR-375 profiling into existing molecular diagnostic frameworks may enhance early detection, guide therapeutic decision-making, and improve survival outcomes for patients with OSCC.

CONCLUSION

This systematic review concludes that microRNA-375 (miR-375) plays a crucial tumor-suppressive role in oral squamous cell carcinoma and holds significant promise as a reliable, non-invasive diagnostic biomarker for early detection. Its consistent under-expression across diverse biological samples reinforces its potential utility in precision oncology and personalized disease management. While current evidence establishes a strong foundation for its diagnostic and prognostic relevance, broader validation through well-designed, prospective studies employing standardized analytical methods remains essential to translate these findings into clinical practice.

INSIGHTS-JHR
INSIGHTS-JOURNAL OF HEALTH AND REHABILITATION

AUTHOR CONTRIBUTION

Author	Contribution
Sabika Fatima*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Safia Khatoon	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published
Muhammad Ilyas Shaikh	Substantial Contribution to acquisition and interpretation of Data Has given Final Approval of the version to be published
Muqaddas Amjad	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Saqib Kaleem	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Noreen	Substantial Contribution to study design and Data Analysis Has given Final Approval of the version to be published
Eman Rasheed	Contributed to study concept and Data collection Has given Final Approval of the version to be published

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