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Assessment of LNC-NORAD Expression Changes in Tumor and Surgical Margin Samples of Oral Squamous Cell Carcinoma

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ABSTRACT

Introduction: Oral squamous cell carcinoma is a biologically aggressive malignancy in which molecular alterations may extend beyond visible tumor tissue into surgical margins. Long non-coding RNAs, particularly LNC-NORAD, may influence genomic stability, stress responses, and tumor progression. This study aimed to assess LNC-NORAD expression changes in tumor and surgical margin samples of OSCC.

Material and methods: This descriptive cross-sectional study was conducted at Tabriz University of Medical Sciences in 2024. Using convenience sampling, 100 specimens comprising 50 oral squamous cell carcinoma tissues and 50 matched healthy surgical margins were collected from 50 patients. LNC-NORAD expression was quantified by real-time PCR and evaluated alongside age, sex, smoking status, tumor site and size, histological grade, invasion characteristics, TNM stage, lymph node involvement, and margin status.

Results: LNC-NORAD expression was significantly higher in OSCC tumor tissues than in matched healthy surgical margins (3.18 ± 1.07 vs. 1.24 ± 0.43; $P<0.001$). Increased tumoral expression was associated with smoking, larger tumor size, poorer histological differentiation, deeper invasion, perineural invasion, advanced TNM stage, and lymph node metastasis. By contrast, no significant associations were observed with sex, anatomical tumor site, or lymphovascular invasion.

Conclusion: LNC-NORAD is markedly upregulated in OSCC tissues compared with matched surgical margins and is closely associated with several indicators of tumor aggressiveness. These findings suggest that this long non-coding RNA may serve as a useful molecular marker for identifying biologically advanced and clinically high-risk oral squamous cell carcinoma.

Introduction

Oral squamous cell carcinoma (OSCC) is one of the most prevalent malignancies of the head and neck region and remains a major public health burden worldwide, particularly in populations with high exposure to tobacco, alcohol, betel quid, and poor oral hygiene. Despite progress in surgical techniques, radiotherapy, and systemic therapies, the overall survival rate of patients with OSCC has improved only modestly over recent decades.

Local recurrence, regional metastasis, and treatment resistance continue to compromise outcomes, indicating that conventional clinicopathological parameters alone are insufficient to fully explain the biological behavior of this disease [1].

The development of OSCC is a multistep biological process driven by the accumulation of genetic and epigenetic alterations within the oral epithelium.

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These alterations disrupt normal cellular homeostasis, resulting in uncontrolled proliferation, resistance to apoptosis, enhanced invasive capacity, and immune evasion. In this context, molecular profiling has become increasingly important for identifying biomarkers that may improve early diagnosis, refine prognostic assessment, and uncover novel therapeutic targets. Among the diverse classes of regulatory molecules implicated in oral carcinogenesis, long non-coding RNAs have emerged as particularly influential modulators of tumor biology [2].

Long non-coding RNAs (lncRNAs) are RNA transcripts longer than 200 nucleotides that lack protein-coding potential but exert broad regulatory effects at transcriptional, post-transcriptional, and epigenetic levels. They participate in chromatin remodeling, transcriptional control, RNA splicing, and the modulation of messenger RNA stability and translation. Over the past decade, dysregulated lncRNA expression has been recognized as a hallmark of many human malignancies, including cancers of the oral cavity. Their tissue-specific expression patterns and diverse biological functions have positioned them as promising candidates for molecular diagnosis and targeted intervention in oncology [3].

Among the lncRNAs implicated in cancer, non-coding RNA activated by DNA damage, commonly known as NORAD, has attracted growing scientific interest because of its role in maintaining genomic stability. NORAD is highly conserved and is induced in response to cellular stress, particularly DNA damage. It functions, at least in part, by interacting with PUMILIO proteins and regulating the expression of genes involved in chromosome segregation, DNA replication, and mitotic progression. Through these mechanisms, NORAD contributes to genomic integrity, a process that is critically disrupted during malignant transformation [4].

The biological significance of NORAD in cancer appears to be complex and context dependent. In some malignancies, NORAD has been reported to act as an oncogenic lncRNA by promoting proliferation, migration, invasion, and resistance to apoptosis. In other settings, its relationship with genome maintenance suggests a more nuanced role that may vary according to tumor type, molecular background, and microenvironmental stress. This duality makes NORAD especially relevant for investigation in OSCC, a cancer characterized by marked genomic instability, chronic inflammatory exposure, and diverse environmental risk factors [5].

OSCC arises in an anatomic and biological environment that is constantly exposed to chemical, microbial, and mechanical insults. Chronic exposure to carcinogens such as tobacco-derived compounds and alcohol metabolites induces persistent oxidative stress, DNA damage, and inflammatory signaling within the oral mucosa. These events create a fertile setting for malignant transformation and clonal expansion. Because NORAD is closely linked to cellular responses to DNA damage and stress adaptation, altered expression of this lncRNA may reflect fundamental mechanisms involved in the initiation and progression of OSCC [6].

An important challenge in the management of OSCC is the high risk of local recurrence even after apparently complete surgical excision. Histopathological examination of resection margins remains the standard approach for assessing surgical adequacy, yet it may fail to detect molecular alterations in tissues that appear microscopically normal. The concept of field cancerization provides a useful framework for understanding this problem, proposing that genetically or epigenetically altered epithelial fields may extend beyond the visible tumor. Molecular evaluation of surgical margins may therefore reveal early oncogenic changes not captured by routine morphology alone [7].

Within this framework, the analysis of lncRNA expression in both tumor tissue and corresponding surgical margins offers a valuable strategy for exploring the molecular extent of disease. Comparing these two tissue compartments can provide insight into whether a candidate biomarker is confined to malignant tissue or also altered in adjacent fields exposed to the same carcinogenic environment. If NORAD expression differs significantly between tumors and surgical margins, such a finding could deepen our understanding of OSCC biology and potentially support the use of lncRNA-based markers in surgical and prognostic assessment [8].

Previous studies have shown that lncRNAs such as HOTAIR, MALAT1, NEAT1, and H19 are dysregulated in OSCC and are associated with proliferation, epithelial-mesenchymal transition, metastasis, and poor survival. These findings have strengthened the rationale for examining additional lncRNAs that may contribute to oral tumorigenesis through distinct molecular pathways. NORAD is of particular interest because, unlike many lncRNAs studied primarily in relation to transcriptional regulation, it is directly connected to genome surveillance and the cellular response to genotoxic stress, both of which are central features of oral carcinogenesis [9].

Emerging evidence from other cancer types suggests that aberrant NORAD expression may influence malignant behavior through its interaction with microRNAs and downstream signaling pathways. By functioning as a competing endogenous RNA, NORAD may modulate the availability of tumor-suppressive microRNAs and thereby affect the

expression of genes involved in cell-cycle progression, invasion, stemness, and chemoresistance. Such mechanisms may be highly relevant in OSCC, where aggressive local invasion and therapeutic resistance remain defining clinical challenges. However, the expression pattern and significance of NORAD in OSCC, particularly in relation to surgical margins, remain insufficiently characterized [10].

Another reason to investigate NORAD in OSCC lies in the heterogeneity of this malignancy. Tumors arising in the oral cavity differ substantially in their anatomical location, differentiation status, exposure history, and molecular architecture. This heterogeneity often translates into unpredictable clinical behavior, even among cases with similar histological staging. Molecular markers capable of capturing biologically meaningful differences may therefore improve patient stratification. Evaluating NORAD expression across tumor and margin tissues may help clarify whether this lncRNA is linked to local aggressiveness, early field alterations, or both [11].

From a translational perspective, identifying lncRNAs associated with OSCC could have implications beyond diagnosis. Because lncRNAs are detectable by sensitive molecular techniques and may be measurable in tissue and body fluids, they have considerable appeal as biomarkers for screening, monitoring, and therapeutic targeting. If NORAD is consistently dysregulated in tumor samples relative to surgical margins, it may represent a candidate biomarker for distinguishing malignant from non-malignant tissue or for identifying patients at greater risk of residual molecular disease after resection. Such possibilities justify closer investigation in clinicopathological studies [12].

The study of paired tumor and surgical margin samples is especially valuable because it reduces interindividual variability and allows more direct interpretation of molecular differences within the same biological context. In OSCC, where exposure to carcinogens often affects the entire mucosal field, the use of matched specimens can provide a clearer picture of tumor-specific dysregulation. This design may also help identify whether NORAD changes represent a sharp transition associated with malignant transformation or a gradient of alteration extending into adjacent tissues. Such distinctions are important for both biological understanding and surgical oncology [13].

Although interest in lncRNAs has expanded rapidly, many potentially important transcripts remain underexplored in oral cancer research. NORAD is one such molecule whose known links to genomic stability, stress responses, and regulatory RNA networks make it a compelling target for study. A better understanding of its expression pattern in OSCC could contribute to the growing body of evidence that non-coding RNAs are not passive transcriptional byproducts but active determinants of tumor behavior. This is particularly relevant in cancers of the oral cavity, where accessible tissue sampling offers a practical opportunity for translational molecular investigation [14].

Given these considerations, the present study was designed to assess LNC-NORAD expression changes in tumor and surgical margin samples of oral squamous cell carcinoma. By comparing paired specimens obtained from patients with OSCC, this work seeks to determine whether NORAD expression differs between malignant tissue and adjacent margins and to provide a molecular perspective on tumor-associated alterations within the oral mucosal field. Clarifying this relationship may improve current understanding of OSCC pathogenesis and support future efforts to develop lncRNA-based biomarkers in oral oncology [15].

Material and methods

Design of the study:

This descriptive cross-sectional study was conducted at Tabriz University of Medical Sciences, Tabriz, Iran, during 2022. The study was designed to evaluate the association between first-postoperative-day C-reactive protein levels and the development of chronic post-mastectomy pain in women undergoing mastectomy. All eligible patients were assessed within the defined study period, and the required clinical, laboratory, and follow-up data were collected according to a standardized research protocol.

Sampling

Sample size was estimated using the single-group descriptive cross-sectional sample size formula, commonly expressed as the Cochran formula for estimating a proportion: $n=Z^2P(1-P)/d^2$. Considering a 95% confidence level ($Z=1.96$), an assumed proportion of 0.50 to yield the maximum sample size, and a precision of approximately 0.14, the calculated sample size was about 49 participants; therefore, 50 patients were ultimately included in the study. Participants were recruited using a convenience sampling method from eligible patients referred to the study setting during the study period.

Inclusion/exclusion criteria

Women aged 18 years or older who underwent mastectomy for breast cancer at the study center during 2022, were willing to participate, and provided written informed consent were included in the study. Patients were required to have the ability to understand study procedures and respond to pain assessments during follow-up. Exclusion criteria included documented acute infection, chronic

inflammatory or autoimmune disease, severe hepatic or renal dysfunction, known metastatic disease at the time of surgery, reoperation during the immediate postoperative period, major postoperative complications likely to substantially alter inflammatory markers, current long-term corticosteroid or immunosuppressive therapy, pre-existing chronic pain syndromes unrelated to breast disease, regular opioid use before surgery, major psychiatric or cognitive disorders interfering with reliable assessment, incomplete medical records, or loss to follow-up.

Procedure

A venous blood sample was obtained from each participant on the first postoperative day, approximately 24 hours after surgery, to measure serum C-reactive protein. CRP levels were quantified in the hospital laboratory using a standard immunoturbidimetric assay and were recorded in mg/L. In addition to CRP, demographic and clinical variables were collected from patient interviews and medical records, including age, body mass index, marital status, smoking status, comorbidities, menopausal status, tumor laterality, type of mastectomy, axillary intervention, duration of surgery, type of anesthesia, postoperative analgesic regimen, length of hospital stay, and relevant oncologic history.

Chronic post-mastectomy pain was evaluated at least 3 months after surgery through structured follow-up assessment. Pain was defined as persistent pain in the chest wall, axilla, shoulder, or ipsilateral upper arm lasting beyond the normal healing period and not attributable to infection or tumor recurrence. Pain intensity was assessed using the Visual Analog Scale, with patients asked to rate their average pain severity over the preceding period on a 0 to 10 scale. Additional measured variables included the presence or absence of chronic pain, pain location, analgesic use during follow-up, history of adjuvant chemotherapy or radiotherapy, and postoperative complications such as seroma, hematoma, or wound-related events.

Data analysis

Because the study variables were normally distributed, data were analyzed using parametric statistical methods in SPSS software. Quantitative variables were presented as mean and standard deviation, and qualitative variables were expressed as frequency and percentage. The association between first-postoperative-day CRP level and chronic post-mastectomy pain was assessed using the independent-samples test for comparison between patients with and without chronic pain. Pearson correlation analysis was used to examine the relationship between CRP level and pain intensity score. In addition, multivariable linear or logistic regression analysis was performed, as appropriate, to evaluate the independent association of CRP with pain outcomes after adjustment for potential confounding variables. A p value of less than 0.05 was considered statistically significant.

Ethical considerations

This study was approved by the Ethics Committee of Tabriz University of Medical Sciences under the ethics code IR.TBZMED.REC.1401.485. Written informed consent was obtained from all participants before enrollment. All data were collected confidentially, anonymized before analysis, and used solely for research purposes. Participants were assured that refusal to participate or withdrawal from the study at any stage would not affect their medical care or treatment process.

Results

A total of 100 tissue specimens were obtained from 50 patients with oral squamous cell carcinoma at Imam Reza Hospital in Tabriz. The specimens consisted of 50 tumor tissues and 50 matched histologically healthy surgical margins, representing equal proportions of the tissue sample set. The patients had a mean age of 59.36 ± 11.42 years, and most were male. The demographic and clinicopathological characteristics of the patients, together with the distribution of the paired tissue specimens, are summarized in Table 1.

Table 1. Demographic and clinicopathological characteristics of patients with oral squamous cell carcinoma and distribution of the collected tissue samples

Variable	Category	Value, n (%)
Patients	Total	50 (100.0)
Tissue samples	Total	100 (100.0)
-	Tumor tissue	50 (50.0)
-	Matched healthy surgical margin	50 (50.0)
Age (years)	Mean ± SD	59.36 ± 11.42
-	Range	34-81
Sex	Male	32 (64.0)
-	Female	18 (36.0)
Smoking status	Smoker	27 (54.0)

-	Non-smoker	23 (46.0)
Tumor site	Tongue	16 (32.0)
-	Buccal mucosa	9 (18.0)
-	Floor of the mouth	7 (14.0)
-	Gingiva/alveolar ridge	11 (22.0)
-	Other oral sites	7 (14.0)
Tumor size	≤2 cm	14 (28.0)
-	>2-4 cm	23 (46.0)
-	>4 cm	13 (26.0)
Histological grade	Well differentiated	19 (38.0)
-	Moderately differentiated	22 (44.0)
-	Poorly differentiated	9 (18.0)
Depth of invasion	≤10 mm	28 (56.0)
	>10 mm	22 (44.0)
Lymph vascular invasion	Absent	35 (70.0)
-	Present	15 (30.0)
Perineural invasion	Absent	31 (62.0)
-	Present	19 (38.0)
TNM stage	Stage I/II	24 (48.0)
-	Stage III/IV	26 (52.0)
Lymph node status	N0	29 (58.0)
-	N+	21 (42.0)
Surgical margin status	Histologically negative	50 (100.0)
-	Histologically positive	0 (0.0)

LNC-NORAD expression was significantly higher in OSCC tissues than in the corresponding histologically healthy surgical margins (3.18 ± 1.07 vs. 1.24 ± 0.43; P<0.001). Higher tumoral expression was associated with smoking, larger tumor size, poorer histological differentiation, greater depth of invasion, perineural invasion, advanced TNM stage, and lymph node involvement. No statistically significant differences were detected according to sex, anatomical site, or lymph vascular invasion, as detailed in Table 2.

Table 2. Comparison of LNC-NORAD expression between tumor and matched healthy surgical margin tissues and its association with clinicopathological characteristics

Variable	Category	n	Tumor LNC-NORAD expression, Mean ± SD	Margin LNC-NORAD expression, Mean ± SD	P-value*
Tissue comparison	Tumor vs. matched margin	50 pairs	3.18 ± 1.07	1.24 ± 0.43	<0.001†
Sex	Male	32	3.26 ± 1.09	1.27 ± 0.45	0.438
-	Female	18	3.01 ± 1.04	1.19 ± 0.39	
Smoking status	Smoker	27	3.49 ± 1.02	1.30 ± 0.45	0.028
-	Non-smoker	23	2.82 ± 1.03	1.17 ± 0.40	
Tumor site	Tongue	16	3.37 ± 1.08	1.21 ± 0.41	0.684
-	Buccal mucosa	9	2.96 ± 0.91	1.29 ± 0.46	-
-	Floor of the mouth	7	3.25 ± 1.19	1.15 ± 0.38	-
-	Gingiva/alveolar ridge	11	3.09 ± 1.12	1.32 ± 0.48	-
-	Other oral sites	7	3.06 ± 1.13	1.22 ± 0.42	-
Tumor size	≤2 cm	14	2.77 ± 0.87	1.16 ± 0.36	0.047
-	>2–4 cm	23	3.16 ± 1.01	1.28 ± 0.45	-
-	>4 cm	13	3.66 ± 1.19	1.27 ± 0.47	-
Histological grade	Well differentiated	19	2.83 ± 0.92	1.19 ± 0.39	0.031
-	Moderately differentiated	22	3.22 ± 0.99	1.28 ± 0.44	-

-	Poorly differentiated	9	3.83 ± 1.26	1.25 ± 0.49	-
Depth of invasion	≤10 mm	28	2.89 ± 0.94	1.21 ± 0.41	0.036
-	>10 mm	22	3.55 ± 1.12	1.28 ± 0.46	-
Lymph vascular invasion	Absent	35	3.00 ± 1.01	1.22 ± 0.42	0.091
-	Present	15	3.60 ± 1.13	1.29 ± 0.47	-
Perineural invasion	Absent	31	2.94 ± 0.97	1.20 ± 0.40	0.044
-	Present	19	3.57 ± 1.15	1.31 ± 0.47	-
TNM stage	Stage I/II	24	2.76 ± 0.88	1.18 ± 0.39	0.006
-	Stage III/IV	26	3.57 ± 1.10	1.30 ± 0.46	-
Lymph node status	N0	29	2.89 ± 0.95	1.19 ± 0.40	0.021
-	N+	21	3.58 ± 1.13	1.31 ± 0.47	-

Figure 1 shows a clear increase in LNC-NORAD expression in oral squamous cell carcinoma tissues compared with the matched healthy surgical margins. The mean expression level in tumor samples was 3.18 ± 1.07, whereas the corresponding value in margin tissues was 1.24 ± 0.43, indicating nearly a 2.6-fold elevation in the malignant specimens. This difference was statistically significant ($P<0.001$), supporting the view that LNC-NORAD is markedly upregulated in tumor tissue and may be linked to the molecular events underlying oral squamous cell carcinoma progression.

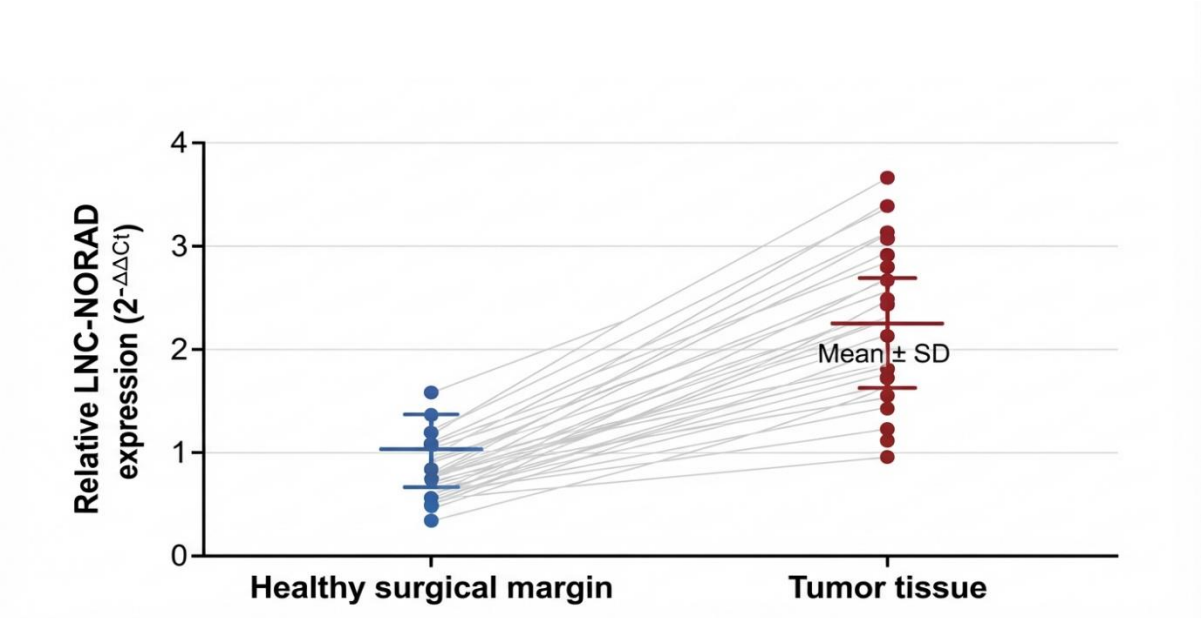

Figure 1. Relative LNC-NORAD Expression in Tumor and Matched Healthy Surgical Margin Tissues of Oral Squamous Cell Carcinoma

Figure 2 demonstrates that LNC-NORAD expression rises with advancing disease stage in oral squamous cell carcinoma. The mean tumoral expression level was 2.76 ± 0.88 in patients with Stage I/II disease and increased to 3.57 ± 1.10 in those with Stage III/IV disease. This difference was statistically significant ($P=0.006$), indicating that higher LNC-NORAD expression is associated with more advanced tumor burden and may reflect a stronger biological role for this long non-coding RNA in tumor progression and clinical aggressiveness.

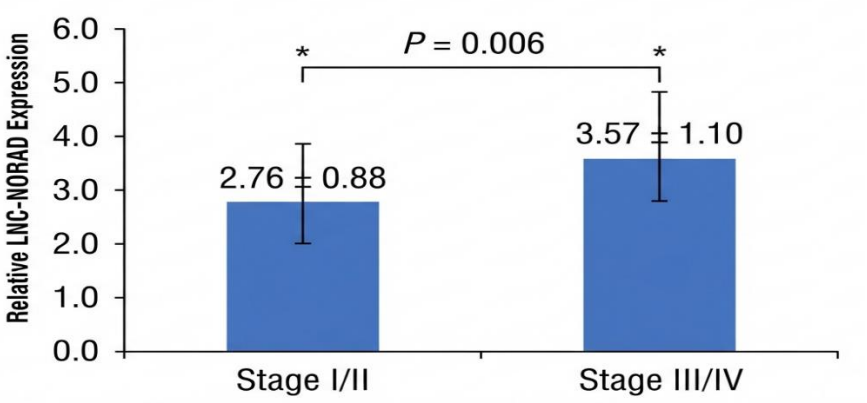

Figure 2. LNC-NORAD Expression According to TNM Stage in Oral Squamous Cell Carcinoma

Discussion

The present study was designed to assess changes in LNC-NORAD expression in tumor and surgical margin samples of oral squamous cell carcinoma and to clarify whether this transcript may be linked to the inflammatory and biological behavior of the disease. This objective is clinically meaningful because OSCC develops within a complex microenvironment shaped not only by genetic instability and uncontrolled proliferation, but also by chronic inflammation, stromal interaction, and progressive local invasion. Examining LNC-NORAD in both malignant tissue and matched margins therefore offer a more informative view of molecular alteration than studying tumor tissue alone. [15]

In summary, our findings showed that LNC-NORAD expression was markedly elevated in OSCC tissues compared with matched healthy surgical margins, with mean values of 3.18 ± 1.07 and 1.24 ± 0.43, respectively, yielding a highly significant difference ($P<0.001$). Higher tumoral expression was also associated with smoking, larger tumor size, poorer histological differentiation, deeper invasion, perineural invasion, advanced TNM stage, and lymph node metastasis. By contrast, no significant relationships were observed with sex, anatomical tumor site, or lymph vascular invasion. Together, these results suggest that LNC-NORAD overexpression is not merely present in OSCC, but is enriched in clinically aggressive disease. [16]

The most immediate interpretation of the primary finding is that LNC-NORAD is actively involved in oral carcinogenesis rather than being an incidental byproduct of malignant transformation. The clear separation between tumor and margin expression supports the view that this lncRNA participates in pathways that become amplified as normal mucosa evolves into invasive carcinoma. Long non-coding RNAs are now recognized as central regulators of transcriptional control, RNA stability, and protein signaling networks, and NORAD in particular has been linked to genomic maintenance and stress adaptation. Its increased expression in tumor tissue may therefore reflect an adaptive survival mechanism that allows malignant cells to tolerate oxidative, replicative, and inflammatory stress. [17]

The difference between tumor and margin tissues is also important from a field-cancerization perspective. Histologically negative margins may appear normal under the microscope, yet they often retain subtle molecular alterations caused by shared carcinogenic exposure, especially in the oral cavity. In this setting, the relatively low expression of LNC-NORAD in margin tissue compared with overt tumor suggests that although the surrounding mucosa may be exposed to the same environmental insults, full malignant activation of this transcript occurs predominantly within transformed tissue. This pattern strengthens the argument that LNC-NORAD could help distinguish biologically aggressive tumor compartments from adjacent tissue that is morphologically intact but molecularly less disturbed. [18]

The association between higher LNC-NORAD expression and smoking is biologically plausible. Tobacco smoke introduces reactive oxygen species, DNA-damaging compounds, and chronic inflammatory stimuli into the oral epithelium, all of which can reshape non-coding RNA expression. Smoking-related oxidative stress may promote the transcription of lncRNAs involved in cellular defense and genomic stability, including NORAD, as malignant cells attempt to survive in a hostile environment. At the same time, chronic tobacco exposure sustains inflammatory cytokine release and epithelial injury, which may further stimulate pathways that favor tumor persistence and progression. The higher LNC-NORAD level observed in smokers therefore fits well with the established molecular consequences of tobacco-driven oral carcinogenesis. [19]

The link between LNC-NORAD overexpression and larger tumor size, advanced stage, and nodal

involvement suggests that this transcript may contribute to tumor expansion and dissemination. As tumors enlarge and progress, they encounter hypoxia, metabolic stress, immune pressure, and structural barriers to invasion. Molecules that help cells maintain proliferation under these conditions are often overexpressed in advanced cancers. NORAD may support this process by stabilizing stress-response pathways, preserving cellular fitness, or modulating downstream targets that influence growth and migration. The higher expression seen in Stage III/IV tumors and node-positive cases therefore implies that LNC-NORAD is associated not only with tumor presence, but with the capacity of OSCC to acquire a more aggressive clinical phenotype. [20]

The relationship between elevated expression and poor histological differentiation provides additional support for a role in aggressive tumor biology. Poorly differentiated tumors usually show greater genetic disorganization, weaker epithelial maturation, and a stronger tendency toward invasion and recurrence. LncRNAs are increasingly recognized as regulators of epithelial-mesenchymal transition, stemness, and dedifferentiation, all of which are features of high-grade malignancy. The progressive increase in LNC-NORAD from well-differentiated to poorly differentiated tumors in this study may indicate that it participates in the molecular reprogramming that accompanies loss of normal squamous architecture. In practical terms, this makes LNC-NORAD a potentially informative marker of biological severity rather than a simple diagnostic signal. [21]

The significant associations with greater depth of invasion and perineural invasion are especially notable because these features are strong indicators of local aggressiveness in OSCC. Deeper invasion reflects the ability of tumor cells to remodel extracellular matrix, interact with stromal fibroblasts, and evade tissue boundaries, while perineural invasion indicates a highly adaptive and destructive pattern of spread. Both processes are influenced by inflammatory mediators, growth factors, and signaling cascades that can be regulated by non-coding RNAs. It is therefore reasonable to propose that increased LNC-NORAD expression may facilitate communication between tumor cells and the surrounding microenvironment, enhancing invasive behavior through inflammation-linked and matrix-remodeling mechanisms. [22]

The lack of significant association with sex or anatomical site is not surprising and may actually reinforce the specificity of the observed pattern. Biological aggressiveness in OSCC is more directly shaped by molecular dysregulation, carcinogen exposure, and host-tumor interaction than by patient sex alone or by broad anatomical classification. Likewise, the absence of a significant relationship with lymph vascular invasion should be interpreted cautiously rather than negatively. This variable may require a larger sample to detect meaningful differences, particularly when the prevalence of positive cases is limited. It is possible that the current study had adequate power to identify major expression gradients related to invasion and stage, but less power to resolve weaker associations. [23]

Another important dimension of these findings is the potential relationship between LNC-NORAD and inflammation. OSCC often arises in a background of persistent mucosal irritation, immune activation, and cytokine-driven tissue remodeling. Chronic inflammation can sustain DNA damage, promote angiogenesis, suppress antitumor immunity, and create conditions favorable to malignant progression. If LNC-NORAD is responsive to inflammatory stress, its overexpression in tumor tissue may reflect not only intrinsic cancer-cell activity but also a broader inflammatory ecosystem surrounding the lesion. This interpretation aligns with the present study objective and may explain why higher expression is concentrated in tumors with more invasive and advanced clinicopathological features. [24]

From a translational standpoint, these data suggest that LNC-NORAD may have value as a biomarker for disease stratification in OSCC. Because its expression differed sharply between tumor and margin tissues and tracked with several adverse pathological variables, it may help identify patients with biologically aggressive disease who require closer surveillance or more intensive treatment planning. The paired design used here is a particular strength because it reduces interindividual variability and allows each patient's margin tissue to serve as an internal comparator. Even so, the results should be interpreted within the context of a cross-sectional study and should ideally be validated in larger cohorts using survival, recurrence, and treatment-response endpoints. [25]

Overall, the present findings support the view that LNC-NORAD is overexpressed in OSCC and that this overexpression is closely linked to features of tumor progression, invasion, and advanced clinical behavior. The observed pattern is biologically consistent with the known functions of long non-coding RNAs in genomic stress adaptation, inflammatory signaling, and malignant evolution. The lower expression in matched healthy margins further underscores its relevance to the malignant state. Future research should determine whether LNC-NORAD is simply a marker of aggressive biology or an active driver of it, and whether targeting its regulatory network could open new avenues for prognostic assessment and molecularly

informed therapy in oral squamous cell carcinoma. [26]

Conclusion

The present findings indicate that LNC-NORAD is significantly overexpressed in oral squamous cell carcinoma relative to histologically healthy surgical margin tissue, supporting its possible involvement in oral tumorigenesis. Its significant associations with smoking, tumor size, histological grade, depth of invasion, perineural invasion, TNM stage, and nodal status further suggest that elevated LNC-NORAD expression reflects a more invasive and advanced disease phenotype. Taken together, these results highlight the potential diagnostic and prognostic relevance of LNC-NORAD in the molecular evaluation of OSCC.

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Authors' Contributions

All authors contributed to data analysis, drafting, and revising of the paper and agreed to be responsible for all the aspects of this work.

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