http://www.jrdms.dentaiau.ac.ir
e(ISSN): 2383 -2754
p(ISSN):2588-4166
JRDMS Journal of Research in Dental and Maxillofacial Sciences

Clinical Significance of Salivary Biomarkers in Oral Squamous Cell Carcinoma: A Review

Manzarpour M[1], Homaie M[1] ,Farhadi S[2]

[1] *Student of Dentistry, Dental Branch of Tehran, Islamic Azad University, Tehran, Iran*
[2] *Assistant Professor, Oral & Maxillofacial Pathology Dept, Dental Branch of Tehran, Islamic Azad University, Tehran, Iran*

ARTICLE INFO

Article Type
Review Article

Article History
Received: Apr 2017
Accepted: May2017
ePublished: June 2017

Keywords:
Oral Squamous Cell Carcinoma, Biomarkers, Diagnosis, Prognosis, Treatment Outcome

ABSTRACT

Background and Aim: Oral squamous cell carcinoma (OSCC) accounts for approximately 3% of all cancers worldwide, and if diagnosed early, it has a five-year survival rate of around 85%; however, a late diagnosis may decrease the survival rate to 50%. Aberrant expression of several genes is associated with the hallmarks of OSCC including uncontrolled cell proliferation, poor differentiation, invasion, metastasis, and angiogenesis. The potential of molecular biomarkers for the diagnosis, prognosis, or monitoring of the treatment efficacy in OSCC has been extensively explored during the last decades. This study aimed to review the significance of salivary biomarkers in the treatment outcome of OSCC.

Materials and Methods: The articles in scientific databases including Google Scholar, ScienceDirect, Medline, and PubMed, published between 2004 and 2017, were searched by using relevant keywords including OSCC, biomarkers, diagnosis, prognosis and treatment outcome. Thirty-four articles were reviewed in this study.

Results: According to the findings of the reviewed studies, several salivary biomarkers including subcutaneous adipose tissue (SAT), interleukin-8 (IL-8), Cyfra 21-1, 8-hydroxy-2-deoxyguanosine, malondialdehyde, lactate dehydrogenase (LDH), Annexin A8, ErbB2, carcinoembryonic antigen (CEA), C-reactive protein (CRP), and salivary proteomic biomarkers might be used as indicators for the detection of oral cancer and premalignant oral disease (PMOD) and as a potential marker in the prognosis of OSCC.

Conclusion: Salivary biomarker analysis seems to be a major advancement in the diagnosis of OSCC, and it is a fast-developing field in scientific research. The results indicate that salivary biomarkers can be useful diagnostic and prognostic tools in OSCC.

Review Article

Please cite this paper as: Manzarpour M, Homaie M, Farhadi S. Clinical Significance of Salivary Biomarkers in Oral Squamous Cell Carcinoma: A Review. J Res Dent maxillofac Sci.2017;2(3):22-30.

***Corresponding author:**
Farhadi S
dr.sfarhadi@gmail.com

Introduction:

Oral squamous cell carcinoma (OSCC) accounts for approximately 3% of all cancers worldwide.[1] OSCC comprises more than 90% of all oral cancers, [2,3] and if diagnosed early, it has a five-year survival rate of about 85%.[4] Nowadays, even with various scientific advancements being explored and discovered every day, the worldwide prognosis of oral cancer has diminished rather than improved. This is a recognized public health care problem with a high mortality and morbidity rate.[5]

A wide range of molecules is involved in oral carcinogenesis. Aberrant expression of several genes is linked with the hallmarks of OSCC including uncontrolled cell proliferation, defective apoptosis, poor differentiation, epithelial/mesenchymal transition, metastasis, and angiogenesis.[6] The potential of molecular biomarkers for the diagnosis, prognosis, or monitoring of the treatment outcome in OSCC has been vastly explored during the last decades. In the era of personalized medicine, immunohistochemical detection and/or mRNA expression profiling of molecular biomarkers are expected to significantly contribute to the development of new screening tests with a high sensitivity and specificity and tailor-made therapies for OSCC.[7]

Saliva contains a large number of proteins and peptides, which are used as biomarkers for the detection of different oral and systemic diseases. Saliva is one of the most candid tools for the diagnosis of OSCC because of its direct contact with oral cancerous lesions.[8] Salivary biomarkers have been proven as cost-effective adjuncts in the diagnosis of oral and oropharyngeal carcinoma.[8] In their review on salivary genomics in oral cancer, Shah et al have insisted that saliva is a potential biomarker for OSCC as it contains locally expressed proteins including alpha-amylase, lactoferrins, lysozymes, proline-rich proteins, mucins, histatins, cystatins, and transferrin.[8] Markopoulos et al stated that the molecular diagnosis of OSCC can be done at three levels.[9] Alterations in the cellular DNA causes variations in mRNA transcripts and intra-cellular or extracellular protein levels. Salivary biomarkers can be genetic, protein, or metabolomic markers.[10]

Considering the wide range of salivary biomarkers, this study aimed to review the related recent studies in this field to summarize and help significance s of OSCC.

Methods and Materials:

The literature over the past 13 years (2004-2017) were searched through Google Scholar, ScienceDirect, Medline and PubMed databases by using relevant keywords including OSCC, biomarkers, diagnosis, prognosis, and treatment outcome. Finally, 34 articles were reviewed in this study.

Results:

The findings of the reviewed studies indicated that the majority of salivary biomarkers including subcutaneous adipose tissue (SAT), interleukin-8 (IL-8), Cyfra 21-1, 8-hydroxy-2-deoxyguanosine, malondialdehyde, lactate dehydrogenase (LDH), Annexin A8, ErbB2, carcinoembryonic antigen (CEA), C-reactive protein (CRP), and salivary proteomic biomarkers might be used as indicators for the detection of oral cancer and premalignant oral disease (PMOD) and as a potential marker in the prognosis of OSCC. Table 1 demonstrates the summary of the reviewed studies.

Table 1- Summary of the reviewed studies

No	Title	Authors/Date of publication	Sample volume	Biomarkers/methods	Conclusions
1	Chronic periodontitis can affect the levels of potential oral cancer salivary mRNA biomarkers.(11)	2017 Cheng et al	105 human subjects: OSCC, periodontitis, and healthy controls	S100P mRNA/pre-amplification reverse transcription-quantitative PCR	Salivary S100P mRNA could be a reliable biomarker for OSCC detection, regardless of the presence of chronic periodontitis.
2	Development of a Multiplexed Liquid Chromatography Multiple-Reaction-Monitoring Mass Spectrometry (LC-MRM/MS) Method for Evaluation of Salivary Proteins as Oral Cancer Biomarkers. (12)	2017 Chen et al	109	56 salivary proteins/simple and robust multiple reaction monitoring (MRM)-based targeted proteomics approach incorporating liquid chromatography with mass spectrometry detection (LC-MRM/MS)	The results clearly showed a significant elevation of most targeted proteins in the salivary samples obtained from OSCC patients compared to the controls.
3	Metabolomic analysis of the saliva of Japanese patients with oral squamous cell carcinoma. (13)	2017 Ohshima et al	43 (22 Japanese patients with OSCC and 21 healthy controls)	25 metabolites/comprehensive quantitative metabolomic analysis using CE-MS.	The findings suggest the usefulness of metabolites as salivary biomarkers in Japanese patients with OSCC.
4	Salivary IL-8, IL-6 and TNF-α as Potential Diagnostic Biomarkers for Oral Cancer. (14)	2017 Sahibzada et al		Review article	Chemokines are held responsible, amongst many other proinflammatory cytokines, for inducing oral cancer.
5	Serum and salivary levels of chemerin and MMP-9 in oral squamous cell carcinoma and oral premalignant lesions. (15)	2017 Ghallab and Shaker	45 (15 healthy controls, 15 patients with oral premalignant lesions (OPMLs), and 15 patients with early-stage OSCC)	Chemerin and Matrix metallopeptidase-9 (MMP-9) in serum and saliva samples/ELISA.	Chemerin and MMP-9 might be considered as salivary diagnostic biomarkers for OPMLs, for early detection of OSCC, and for detecting early cancerization of OPMLs.
6	Salivary biomarkers as tools for oral squamous cell carcinoma diagnosis: A systematic review. (16)	2017 Stuani et al		Review article	The lack of methodological criteria within studies and the absence of a consensus on marker choice are obstacles for future studies.
7	Advances of Salivary Proteomics in Oral Squamous Cell Carcinoma (OSCC) Detection: An Update. (17)	2016 Sannam Khan et al		Review article	Due to the easily obtained characteristics of saliva, the focus on OSCC has and will be more shifted to serum and saliva analysis instead of tissue analysis.
8	Salivary biomarkers in oral squamous cell carcinoma - An insight. (18)	2016 Radhika et al		Review article	Salivary biomarker analysis for OSCC is a major advancement in diagnosis.
9	Salivary mRNA markers having the potential to detect oral squamous cell carcinoma segregated from oral leukoplakia with dysplasia. (19)	2016 Michailidou et al	85 (34 patients with primary OSCC, 20 patients with oral leukoplakia and dysplasia, 15 patients with mild dysplasia, 5 patients with severe dysplasia/in situ carcinoma, and 31 matched healthy controls)	IL-1B, IL-8, OAZ and SAT mRNA/sequence-specific primers and real-time RT-PCR	SAT and IL-8 mRNAs have a good discriminatory ability for OSCC patients only, but not for patients with oral leukoplakia, dysplasia or PMOD.

No	Title	Authors/Date of publication	Sample volume	Biomarkers/methods	Conclusions
10	Salivary Biomarkers for Detection of Oral Squamous Cell Carcinoma in a Taiwanese Population. [20]	2016 Gleber-Netto et al	180 (60 OSCC patients, 60 controls, and 60 PMOD patients)	Seven transcriptomic markers (IL8, IL1β, SAT1, OAZ1, DUSP1, S100P, and H3F3A)/qPCR, and two proteomic markers (IL8 and IL1β) were evaluated by ELISA.	The combination of transcriptomic and proteomic salivary markers is of great value for oral cancer detection and differentiation from PMOD patients and controls.
11	Correlation of Cyfra 21-1 levels in saliva and serum with CK19 mRNA expression in oral squamous cell carcinoma. [21]	2016 Malhotra et al	50	Cyfra 21-1 in serum and salivary ECLIA and CK19 messenger RNA (mRNA) expression in tissue by fluorescent quantitative RT-PCR.	We advocate salivary Cyfra 21-1 as a better diagnostic marker over serum Cyfra 21-1 as well as a potential marker in the prognosis of OSCC.
12	A non-invasive study to estimate and compare salivary sialic acid level as tumor marker in patients with pre-cancer and oral cancer. [22]	2016 Jacob et al	60 (20 healthy patients, 20 cases of oral pre-cancer patients, and 20 cases of OSCC)	Salivary TSA levels/unstimulated whole salivary samples were collected from patients and were evaluated.	The glycoprotein metabolism is significantly altered in the saliva of patients with oral pre-cancer lesions and OSCC.
13	A review on oral cancer biomarkers: Understanding the past and learning from the present. [23]	2016 Santosh et al		Review article	Development of clinically valid candidate biomarkers with greater clinical utility values for oral cancer screening is highly recommended.
14	Salivary 8-hydroxy-2-deoxyguanosine, malondialdehyde, vitamin C, and vitamin E in oral pre-cancer and cancer: diagnostic value and free radical mechanism of action. [24]	2016 Kaur et al	160 (40 OSCC, 40 oral lichen planus lesions, 40 oral leukoplakia, 40 oral submucous fibrosis, and a control group)	Salivary 8-OHdG, MDA, and vitamins C and E were measured in unstimulated saliva.	Presence of oxidative DNA and lipid damage in pre-cancerous and OSCC patients. Detection of salivary 8-OHdG, MDA, vitamin C, and vitamin E can act as suitable diagnostic biomarkers of oral pre-cancer and cancerous lesions.
15	Salivary Lactate Dehydrogenase (LDH)- A Novel Technique in Oral Cancer Detection and Diagnosis. [25]	2016 Lokesh et al	50 (30 OSCC patients and 20 healthy controls)	LDH values/unstimulated salivary samples collected from the selected patients were centrifuged and processed.	LDH values were significantly higher in patients with OSCC. Furthermore, the levels significantly correlated with the histopathological grade of the tumor.
16	Annexin A8 is a novel molecular marker for detecting lymph node metastasis in oral squamous cell carcinoma. [26]	2016 Oka et al	13	KRT19 and ANXA8 mRNA/microarray analysis	Both KRT19 and ANXA8 mRNA may be useful markers for detecting lymph node metastases in OSCC patients.
17	Salivary Levels of ErbB2 and CEA in Oral Squamous Cell Carcinoma Patients. [27]	2016 Honarmand et al	53	ErbB2, CEA/ELISA	The role of ErbB2 as a tumor marker in patients with OSCC must still be regarded as controversial. In contrast, the salivary level of CEA may find application for early detection of patients.
18	Altered serum and salivary C-reactive protein levels in patients with oral premalignant lesions and oral squamous cell carcinoma. [28]	2016 Metgud and Bajaj	60 (20 healthy individuals, 20 patients with OPMLs, and 20 OSCC patients)	Serum and salivary CRP levels were evaluated in salivary and blood samples.	Mean CRP levels were higher in patients with oral premalignant lesions compared to the controls. CRP levels in OSCC patients were elevated and were associated with advanced tumor stages.

No	Title	Authors/Date of publication	Sample volume	Biomarkers/methods	Conclusions
19	Potential Salivary Proteomic Markers of Oral Squamous Cell Carcinoma. [29]	2016 Gallo et al	75 (45 OSCC patients and 30 healthy controls)	Saliva proteome/SELDI-TOF mass spectrometry of saliva	The saliva proteome presents significant modifications in OSCC patients and may be useful for knowledge advancement regarding oral carcinogenesis and definition of diagnostic and prognostic biomarkers.
20	Insights into immune responses in oral cancer through proteomic analysis of saliva and salivary extracellular vesicles. [30]	2015 Winck et al	58	The proteome of whole saliva and salivary extracellular vehicles (EVs) from patients with OSCC and healthy individuals were analyzed by LC-MS/MS and label-free protein quantification.	The saliva proteome analysis revealed that immune processes are related to the presence of OSCC and indicated that proteomics data can contribute to determining OSCC prognosis.
21	Salivary microRNAs in oral cancer. [31]	2015 Zahran et al	100 (20 healthy controls, 40 PMDs, 20 biopsy-confirmed OSCC, and 20 recurrent aphthous stomatitis (RAS) as controls)	MicroRNA Isolation Kit (Qiagen, UL)/miRNA expression analysis was performed using qRT-PCR (Applied Biosystems).	Salivary determination of the miRNAs (miRNA-21, miRNA-184, and miRNA-145) might furnish a noninvasive and rapid adjunctive aid for revealing malignant transformations in oral mucosal lesions, particularly miRNA-184.
22	Salivary and serum level of CYFRA 21-1 in oral precancer and oral squamous cell carcinoma. [32]	2015 Rajkumar et al	100 (premalignant subjects and normal healthy subjects)	Serum and salivary CYFRA 21-1 levels/ELISA. Appropriate statistical tests.	The outcome of this study suggests that salivary CYFRA 21-1 can be utilized as a biomarker for early detection of oral cancer.
23	Salivary Immunosuppressive Cytokines IL-10 and IL-13 are Significantly Elevated in Oral Squamous Cell Carcinoma Patients. [33]	2015 Aziz et al	63 (30 OSCC patients and 33 age- and gender-matched healthy controls)	Immunosuppressive cytokines including IL-4, IL-10, IL-13, and IL-1RA were evaluated in each sample using Luminex multianalyte profiling (xMAP) technology on the BioPlex instrument.	Salivary levels of immunosuppressive cytokines, IL-4, IL-10, IL-13, and IL-1RA, could prove to be potential biomarkers of OSCC and can be further investigated as markers of early detection and disease progression.
24	Investigation and identification of potential biomarkers in human saliva for the early diagnosis of oral squamous cell carcinoma. [34]	2014 Wang et al	60 (30 OSCC patients and 30 apparently healthy individuals)	Ultra-performance liquid chromatography-mass spectrometry (UPLC-MS) in hydrophilic interaction chromatography mode.	Salivary metabolite biomarkers for the early diagnosis of OSCC were verified in this study and can be applied as a potential technique of preclinical screening of OSCC.
25	Salivary RNA Signatures in Oral Cancer Detection. [35]	2014 Panta and Venna		Quantitative polymerase chain reaction (qPCR) and microarrays (proven gold standard) followed by qPCR are the principal methods used in salivary RNA analysis.	Protein phosphatase 1, H3 Histone, Family 3A, Interleukin 1 Beta, Interleukin 8, ornithine decarboxylase antizyme 1, Spermidine N1-Acetyltransferase 1, S100 calcium binding protein P, and miR-31 are upregulated and the levels of miR-125a and miR-200a are downregulated in oral cancer patients.
26	Salivary auto-antibodies as noninvasive diagnostic markers of oral cavity squamous cell carcinoma. [36]	2014 Wu et al	348	Multiplexed bead-based platform was used to simultaneously detect auto-antibodies (auto-Abs) in salivary samples.	Auto-Abs were effective for distinguishing the well-differentiated OSCC from other types.

No	Title	Authors/Date of publication	Sample volume	Biomarkers/methods	Conclusions
27	Oral Squamous Cell Carcinoma Detection By Salivary Biomarkers in a Serbian Population. [37]	2012 Brinkmann et al	86	Six transcriptome (DUSP1, IL8, IL1B, OAZ1, SAT1, S100P) and three proteome (IL1B, IL8, M2BP) biomarkers were tested on 18 early-stage and 17 late-stage OSCC patients and 51 healthy controls using quantitative PCR and ELISA.	Four transcriptome (IL8, IL1B, SAT1, S100P) and all proteome biomarkers were significantly elevated in OSCC patients. Patient-based salivary diagnostics is a highly promising approach for OSCC detection.
28	Salivary lncRNA as a potential marker for oral squamous cell carcinoma diagnosis. [38]	2012 Tang et al	20	Review article	The findings suggest that the detection of lncRNAs in saliva may be used as a noninvasive and rapid diagnostic tool for the diagnosis of oral cancer.
29	Salivary analytes in patients with oral squamous cell carcinoma. [39]	2011 Fuchs et al	48	Salivary magnesium, calcium, copper, chloride, phosphate, potassium, sodium, total proteins, and amylase were evaluated. Sodium, potassium, and chloride were determined by indirect potentiometry, whereas copper, magnesium and phosphate were determined by atomic absorption spectrophotometry.	In patients with OSCC, increased salivary sodium and chloride might reflect their overall dehydration status due to alcohol consumption rather than a consequence of OSCC itself.
30	Salivary zinc finger protein 510 peptide as a novel biomarker for detection of oral squamous cell carcinoma in early stages. [40]	2011 Jou et al	77 (47 OSCC patients and 30 healthy donors)	A total of 77 salivary samples from both groups were analyzed using MALDI-TOF MS technology.	Identifying 24-mer ZNF510 peptide as OSCC-related salivary biomarkers via proteomic approach proved useful in adjunct diagnosis for early detection rather than specific diagnostic marker for progression of OSCC patients.
31	Tumor and salivary matrix metalloproteinase levels are strong diagnostic markers of oral squamous cell carcinoma. [41]	2011 Stott-Miller et al	73 (primary OSCC, oral dysplasia, and control subjects)	Evaluation of the expression of MMP1, MMP3, MMP10, and MMP12 in oral epithelial tissue using Affymetrix U133 2.0 Plus GeneChip arrays, followed by quantitative reverse transcription-PCR (qRT-PCR) for MMP1, and determination of MMP1 and MMP3 concentrations in saliva.	Tumor and salivary MMPs are robust diagnostic biomarkers of OSCC.
32	Salivary proteomics for oral cancer biomarker discovery. [42]	2008 Hu et al	128 (64 OSCC patients and 64 healthy subjects)	Shotgun proteomics	Patient-based saliva proteomics is a promising approach to searching for OSCC biomarkers.
33	Concomitant analysis of salivary tumor markers--a new diagnostic tool for oral cancer [43]	2006 Nagler et al	21 tongue OSCC patients	Carbohydrate antigens CA125 and CA19-9, TPS, CEA, SCC, and Cyfra 21-1 circulatory tumor markers in the saliva were evaluated.	The increase reported in salivary tumor markers may be used as a diagnostic tool, especially when a concurrent analysis for significantly increased markers is done.
34	Salivary transcriptome diagnostics for oral cancer detection. [44]	2004 Li et al	64 (32 primary T1/T2 OSCC patients and 32 normal subjects)	RNA isolation was done from the saliva supernatant, followed by two-round linear amplification with T7 RNA polymerase.	The utility of salivary transcriptome diagnostics is successfully demonstrated in this study for oral cancer detection.

Discussion:

Salivary proteomics is developing through the identification of unique biomarkers for early detection approaches including metabolomics, proteomics, genomics, and bioinformatics.[17] The salivary biomarkers are used because saliva encompasses a wide range of compounds,[45] it is easily accessible,[46] and the sampling process is comfortable for patients.[47] The method is non-invasive and safe with low chances of pathogen transmission in comparison with blood samples. [48] Also, saliva is easy to store [49] and does not coagulate. The systematic analysis of salivary proteomic biomarkers and screening of saliva provides an efficient diagnostic tool to turn salivary diagnostics into clinical reality for oral cancer. Because of cellular and molecular heterogeneity of OSCC, several genes are involved in oral carcinogenesis; therefore, the use of several potential biomarkers renders a more precise diagnosis than the use of any marker alone.[17] In Addition, due to the characteristics of saliva, such as its proximity with the oral cavity and noninvasive sampling, the focus on OSCC has been changed to serum and saliva analysis as an alternative to tissue analysis.[17] Salivary biomarkers of OSCC can be used for cancer detection and monitoring non-cancerous activities.[17] The salivary biomarkers discussed in this study can be used for the detection of altered mRNA transcripts and changed intra-cellular or extra-cellular protein levels, and they can be genetic, protein, or metabolomic biomarkers,[45] The detection of these changes by the use of new technologies such as mass spectrometry, gel electrophoresis, chromatography, microarrays, high performance liquid chromatography (HPLC), polymerase chain reaction (PCR), and enzyme-linked immunosorbent assay (ELISA) is a developing field associated with point-of-care technologies, electrochemical detection, RNA sequencing, and liquid biopsy.[17] Furthermore, primary screening by the use of saliva could be an excellent choice considering the advancements in proteomics and genomics.

The extensive sighting of biomarkers and their authentication will change the field of diagnosis for oral cancers and non-cancerous activities. This review provides a comprehensive assessment of the developing diagnostic proteomic tools and biomarkers for the early detection and diagnosis of OSCC by the use of saliva.

Conclusion:

According to the reviewed studies, salivary biomarker analysis seems to be a major development in the diagnosis of OSCC, and it is a fast-growing field in scientific research. The results show that salivary biomarkers can be a useful diagnostic and prognostic tool in OSCC.

References:

1. Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, et al. Cancer incidence and mortality worldwide: sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer 2015 Mar 1;136(5):E359-86.

2 . Attar E, Dey S, Hablas A, Seifeldin IA, Ramadan M, Rozek LS, et al. Head and Neck Cancer in a Developing Country: A Population-Based Perspective Across 8 Years. Oral Oncol 2010 Aug; 46(8):591-6.

3.Bagan J, Sarrion G, Jimenez Y. Oral Cancer: Clinical Features. Oral Oncol 2010 Jun;46(6):414-7.

4.Abbasi F, Farhadi S, Esmaili M. Efficacy of Pilocarpine and Bromhexine in Improving Radiotherapy-induced Xerostomia. J Dent Res Dent Clin Dent Prospects. 2013 May;7(2):86-90.

5 Ford PJ, Farah CS. Early detection and diagnosis of oral cancer: Strategies for improvement. J Cancer Policy 2013 Mar-Jun;1(1-2):e2-e7.

6. Shahsavari F, Farhadi S, Sadri D, Sedehi M. Evaluation of Microvascularity by CD34 Expression in Esophagus and Oral Squamous Cell Carcinoma. J Contemp Dent Pract 2015 Jun 1;16(6):458-62.

7. Sargolzaei S, Farhadi S, Kazemi B, Bandehpour M, Kharazifard MJ. The correlation between p16 expression and INK4a locus mutation with grades and stages in oral squamous cell carcinoma. Indian J Pathol Microbiol. 2014 Jan-Mar;57(1):24-30.

8. Shah FD, Begum R, Vajaria BN, Patel KR, Patel JB, Shukla SN, et al. A review on salivary genomics and proteomics biomarkers in oral cancer. Indian J Clin Biochem. 2011 Oct;26(4):326–334.

9. Markopoulos AK, Michailidou EZ, Tzimagiorgis G. Salivary markers for oral cancer detection. Open Dent J 2010 Aug;4:172-8.
10. Bano S, David MP, Indira AP. Salivary biomarkers for oral squamous cell carcinoma: an overview. IJSS Case Rep Rev. 2015;1(8):39-45
11. Cheng YL, Jordan L, Chen HS, Kang D, Oxford L, Plemons J, et al. Chronic periodontitis can affect the levels of potential oral cancer salivary mRNA biomarkers. J Periodontal Res 2017 Jun;52(3):428-37.
12. Chen YT, Chen HW, Wu CF, Chu LJ, Chiang WF, Wu CC, et al. Development of a Multiplexed Liquid Chromatography Multiple-Reaction-Monitoring Mass Spectrometry (LC-MRM/MS) Method for Evaluation of Salivary Proteins as Oral Cancer Biomarkers. Mol Cell Proteomics 2017 May;16(5):799-811.
13. Ohshima M, Sugahara K, Kasahara K, Katakura A. Metabolomic analysis of the saliva of Japanese patients with oral squamous cell carcinoma. Oncol Rep 2017 May;37(5):2727-34.
14. Sahibzada HA, Khurshid Z, Khan RS, Naseem M, Siddique KM, Mali M, et al. Salivary IL-8, IL-6 and TNF-α as Potential Diagnostic Biomarkers for Oral Cancer. Diagnostics (Basel) 2017 Apr 9;7(2). pii: E21.
15. Ghallab NA, Shaker OG. Serum and salivary levels of chemerin and MMP-9 in oral squamous cell carcinoma and oral premalignant lesions. Clin Oral Investig 2017 Apr;21(3):937-47.
16. Stuani VT, Rubira CM, Sant'Ana AC, Santos PS. Salivary biomarkers as tools for oral squamous cell carcinoma diagnosis: A systematic review. Head Neck 2017 Apr;39(4):797-811.
17. Sannam Khan R, Khurshid Z, Akhbar S, Faraz Moin S. Advances of Salivary Proteomics in Oral Squamous Cell Carcinoma (OSCC) Detection: An Update. Proteomes 2016 Dec 15;4(4). pii: E41.
18. Radhika T, Jeddy N, Nithya S, Muthumeenakshi RM. Salivary biomarkers in oral squamous cell carcinoma – An insight. J Oral Biol Craniofac Res 2016 Nov; 6(Suppl 1): S51–S54.
19. Michailidou E, Tzimagiorgis G, Chatzopoulou F, Vahtsevanos K, Antoniadis K, Kouidou S, et al. Salivary mRNA markers having the potential to detect oral squamous cell carcinoma segregated from oral leukoplakia with dysplasia. Cancer Epidemiol 2016 Aug;43:112-8.
20. Gleber-Netto FO, Yakob M, Li F, Feng Z, Dai J, Kao HK, et al. Salivary Biomarkers for Detection of Oral Squamous Cell Carcinoma in a Taiwanese Population. Clin Cancer Res. 2016 Jul 1;22(13):3340-7.
21. Malhotra R, Urs AB, Chakravarti A, Kumar S, Gupta VK, Mahajan B. Correlation of Cyfra 21-1 levels in saliva and serum with CK19 mRNA expression in oral squamous cell carcinoma. Tumour Biol 2016 Jul;37(7):9263-71.
22. Jacob TV, Ramesh M, Murali S, Ramesh K, Sanjay PR, Abraham P. A non-invasive study to estimate and compare salivary sialic acid level as tumor marker in patients with pre-cancer and oral cancer. J Cancer Res Ther 2016 Apr-Jun;12(2):634-9.
23. Santosh AB, Jones T, Harvey J. A review on oral cancer biomarkers: Understanding the past and learning from the present. J Cancer Res Ther. 2016 Apr-Jun;12(2):486-92.
24. Kaur J, Politis C, Jacobs R. Salivary 8-hydroxy-2-deoxyguanosine, malondialdehyde, vitamin C, and vitamin E in oral pre-cancer and cancer: diagnostic value and free radical mechanism of action. Clin Oral Investig 2016 Mar;20(2):315-9.
25. Lokesh K, Kannabiran J, Rao MD. Salivary Lactate Dehydrogenase (LDH)- A Novel Technique in Oral Cancer Detection and Diagnosis. J Clin Diagn Res 2016 Feb;10(2):ZC34-7.
26. Oka R, Nakashiro K, Goda H, Iwamoto K, Tokuzen N, Hamakawa H. Annexin A8 is a novel molecular marker for detecting lymph node metastasis in oral squamous cell carcinoma. Oncotarget 2016 Jan 26;7(4):4882-9.
27. Honarmand MH, Farhad-Mollashahi L, Nakhaee A, Nehi M. Salivary Levels of ErbB2 and CEA in Oral Squamous Cell Carcinoma Patients. Asian Pac J Cancer Prev 2016;17(S3):77-80.
28. Metgud R, Bajaj S. Altered serum and salivary C-reactive protein levels in patients with oral premalignant lesions and oral squamous cell carcinoma. Biotech Histochem. 2016;91(2):96-101.
29. Gallo C, Ciavarella D, Santarelli A, Ranieri E, Colella G, Lo Muzio L, et al. Potential Salivary Proteomic Markers of Oral Squamous Cell Carcinoma. Cancer Genomics Proteomics 2016 Jan-Feb;13(1):55-61.
30.Winck FV, Prado Ribeiro AC, Ramos Domingues R, Ling LY, Riaño-Pachón DM, Rivera C, et al. Insights into immune responses in

oral cancer through proteomic analysis of saliva and salivary extracellular vesicles. Sci Rep 2015 Nov 5;5:16305
31. Zahran F, Ghalwash D, Shaker O, Al-Johani K, Scully C. Salivary microRNAs in oral cancer. Oral Dis 2015 Sep;21(6):739-47.
32. Rajkumar K, Ramya R, Nandhini G, Rajashree P, Ramesh Kumar A, Nirmala Anandan S. Salivary and serum level of CYFRA 21-1 in oral precancer and oral squamous cell carcinoma. Oral Dis 2015 Jan;21(1):90-6.
33. Aziz S, Ahmed SS, Ali A, Khan FA, Zulfiqar G, Iqbal J, et al. Salivary Immunosuppressive Cytokines IL-10 and IL-13 Are Significantly Elevated in Oral Squamous Cell Carcinoma Patients. Cancer Invest 2015;33(7):318-28.
34.Wang Q, Gao P, Wang X, Duan Y. Investigation and identification of potential biomarkers in human saliva for the early diagnosis of oral squamous cell carcinoma. Clin Chim Acta 2014 Jan 1;427:79-85.
35. Panta P, Venna VR. Salivary RNA Signatures in Oral Cancer Detection. Anal Cell Pathol 2014;2014:450629.
36. Wu CC, Chang YT, Chang KP, Liu YL, Liu HP, Lee IL, et al. Salivary auto-antibodies as non-invasive diagnostic markers of oral cavity squamous cell carcinoma. Cancer Epidemiol Biomarkers Prev 2014 Aug;23(8):1569-78.
37. Brinkmann O, Kastratovic DA, Dimitrijevic MV, Konstantinovic VS, Jelovac DB, Antic J, et al. Oral Squamous Cell Carcinoma Detection by Salivary Biomarkers in a Serbian Population. Oral Oncol 2011 Jan;47(1):51-5.
38. Tang H, Wu Z, Zhang J, Su B. Salivary lncRNA as a potential marker for oral squamous cell carcinoma diagnosis. Mol Med Rep 2013 Mar;7(3):761-6.
39. Fuchs PN, Rogić D, Vidović-Juras D, Susić M, Milenović A, Brailo V, et al. Salivary analytes in patients with oral squamous cell carcinoma. Coll Antropol 2011 Jun;35(2):359-62.
40. Jou YJ, Lin CD, Lai CH, Tang CH, Huang SH, Tsai MH, et al. Salivary zinc finger protein 510 peptide as a novel biomarker for detection of oral squamous cell carcinoma in early stages. Clin Chim Acta 2011 Jul 15;412(15-16):1357-65.
41. Stott-Miller M, Houck JR, Lohavanichbutr P, Méndez E, Upton MP, Futran ND, et al. Tumor and salivary matrix metalloproteinase levels are strong diagnostic markers of oral squamous cell carcinoma. Cancer Epidemiol Biomarkers Prev 2011 Dec;20(12):2628-36.
42. Hu S, Arellano M, Boontheung P, Wang J, Zhou H, Jiang J, et al. Salivary proteomics for oral cancer biomarker discovery. Clin Cancer Res 2008 Oct 1;14(19):6246-52.
43. Nagler R, Bahar G, Shpitzer T, Feinmesser R. Concomitant analysis of salivary tumor markers--a new diagnostic tool for oral cancer. Clin Cancer Res 2006 Jul 1;12(13):3979-84.
44. Li Y, St John MA, Zhou X, Kim Y, Sinha U, Jordan RC, et al. Salivary transcriptome diagnostics for oral cancer detection. Clin Cancer Res 2004 Dec 15;10(24):8442-50.
45. Spielmann N, Wong DT. Saliva: diagnostics and therapeutic perspectives. Oral Dis 2011 May;17(4):345–54
46. Mishra A, Verma M. Cancer biomarkers: are we ready for the prime time? Cancers (Basel) 2010 Mar 22;2(1):190-208.
47. Tanaka T, Tanaka M, Tanaka T. Oral carcinogenesis and oral cancer chemoprevention: A review. J Pathol Res Int 2011;2011:431246.
48. Yoshizawa JM, Schafer CA, Schafer JJ, Farrell JJ, Paster BJ, Wong DT. Salivary biomarkers: toward future clinical and diagnostic utilities. Clin Microbiol Rev 2013 Oct;26(4):781-91.
49. Cheng YS, Rees T, Wright J. A review of research on salivary biomarkers for oral cancer detection. Clin Transl Med 2014 Feb 24;3(1):3.