ORIGINAL RESEARCH

Association of Stathmin (Op18) with TNM Staging and Grading of Oral Squamous Cell Carcinoma and Its Role in Tumor Progression

Purnima Vadla[1], Gaddam Deepthi[2], Vaishnavi Julakanti[3], Divya Jahagirdar[4], Swetha Meruva[5], Swapnika Tantravahi[6]

Abstract

Aim: The purpose of the study was to evaluate the expression of stathmin in different histological grades and tumor, node, metastasis (TNM) staging of Oral carcinoma and various grades of oral dysplasia. The study also aims at observing the stathmin expression with respect to lymph node metastasis.

Materials and methods: A total of 90 histopathologically confirmed tissue sections were acquired, of which 30 sections of oral dysplasia, 30 oral squamous cell carcinoma (OSCC) and 30 normal tissue sections were stained immunohistochemically with stathmin. The tissue sections, were categorized into different grades of oral dysplasia and OSCC based on histopathological examination. For estimation of stathmin expression, manual examination of 300 cells was done in a minimum of five different areas of tissue section and a mean proportion of positive-stained cells were determined. The statistical analysis of the results was done using ANOVA test.

Results: A statistically significant increase in mean staining scores of stathmin in OSCC group compared to dysplasia and control groups. A statistically significant difference was observed in different grades of dysplasia and OSCC groups. Stage III and stage IV OSCC tissue sections showed high intensity staining scores of stathmin expression.

Conclusion: An increased expression of stathmin was detected in various grades of OSCC and also with respect to staging of oral cancer. Half the cases of OSCC with lymph node metastasis showed high intensity scores of stathmin. Based on the above facts, stathmin expression was indicated as a potential tool for predicting the outcome of oral cancer patients with lymph node metastasis and its expression was increased in the group with poor prognosis.

Clinical significance: Any damage/mutation to stathmin can result in defects in cell division resulting in aneuploidy and in turn cancers. In this study, the results showed that there is a differential expression of stathmin in the early and the advanced grades and different TNM stages of OSCC. A high expression of stathmin was observed in all the cases with lymph node metastasis. These observations prove that stathmin has an important role in the progression, tumorigenicity, and prognosis of the oral cancer.

Keywords: Biomarker, Leukoplakia, Oral squamous cell carcinoma, Prognosis, Stathmin.

The Journal of Contemporary Dental Practice (2022): 10.5005/jp-journals-10024-3342

Introduction

Stathmin or oncoprotein 18/Op18, is a microtubule destabilizer protein which are vital for intracellular transport, mitosis, maintaining cell shape and motility.[1,2]

Microtubule dynamics and assembly are under the regulation of Op18. These mitotic spindle is formed as a result of polymerization of these microtubules during mitosis, the structure that is essential for accurate chromosome segregation and cell division. At the molecular level, the polymerization and depolymerization activity of microtubules is under the control of stathmin. At the onset of mitosis, the depolymerizing activity is suppressed by phosphorylation to permit the microtubule polymerization to form the microtubule assembly. Stathmin dephosphorylation is initiated at the exit of the mitosis and before cells enter an interphase.[3]

Obligatory expression or inhibition of stathmin results in various mitotic spindle defects. Overexpression leads to abnormalities in or a complete lack of mitotic spindle and cells may get arrested in the initial stages of mitosis. Inhibition of stathmin expression results in abnormalities in mitotic spindle which might result in accumulation of cells in the middle of mitosis. Thus, stathmin plays an important role in the mitotic spindle dynamics for the timely entry and exit of cell from mitosis.[4]

A variety of human cancers such as esophageal cancer,[5] breast cancer,[6] endometrial cancer,[7] colorectal cancer,[8] hepatocarcinoma,[9] prostate cancer,[10] ovarian cancer,[11] etc. has a close association of stathmin with TNM staging, cancer cell differentiation, lymph node

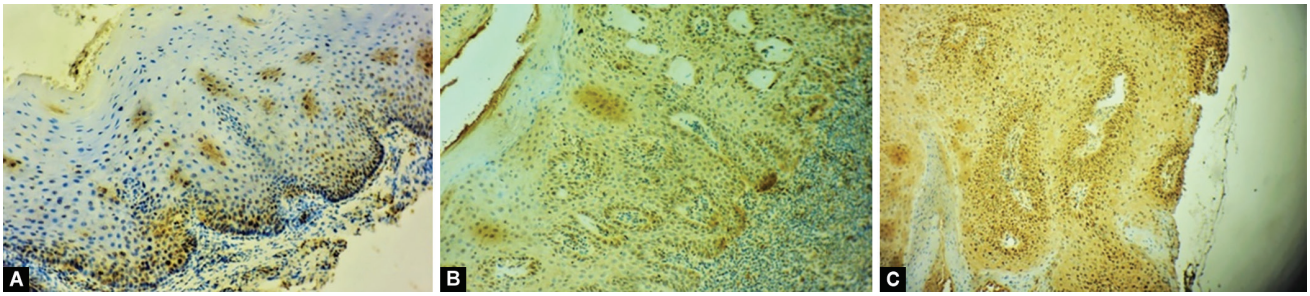
[1]Department of Oral Pathology and Microbiology, Mamata Dental College, Khammam, Telangana, India
[2]Department of Oral and Maxillofacial Pathology, Kamineni Institute of Dental Sciences, Narketpally, Telangana, India
[3]Department of Oral Pathology, Mamata Dental College, Khammam, Telangana, India
[4]General Dentistry Department, Gandhi Hospital, Hyderabad, Telangana, India
[5]Health Informatics, George Mason University, Fairfax, Virginia
[6]General Dentistry Department, Mallareddy Dental College for Women, Hyderabad, India

Corresponding Author: Gaddam Deepthi, Department of Oral and Maxillofacial Pathology, Kamineni Institute of Dental Sciences, Narketpally, Telangana, India, Phone: +91 7799357588, e-mail: drdeepthireddy999@gmail.com

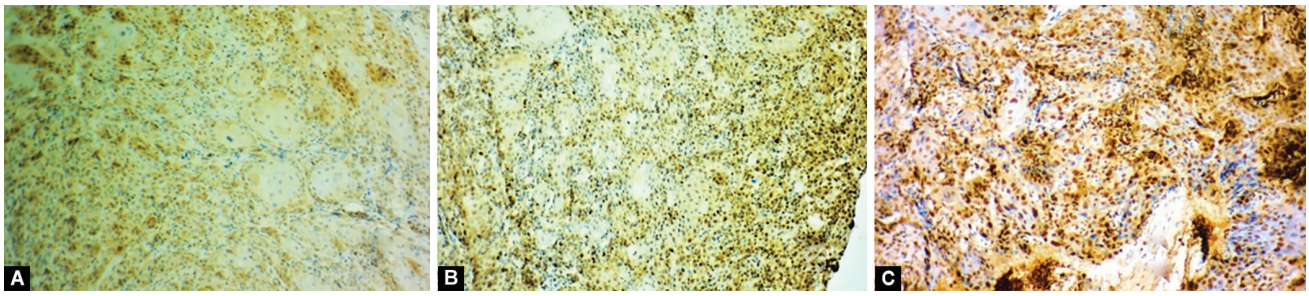
How to cite this article: Vadla P, Deepthi G, Julakanti V, *et al*. Association of Stathmin (Op18) with TNM Staging and Grading of Oral Squamous Cell Carcinoma and Its Role in Tumor Progression. J Contemp Dent Pract 2022;23(5):497–502.

Source of support: Nil

Conflict of interest: None



Figs 1A to C: Photomicrograph of stathmin immunoexpression in (A) Mild dysplasia (10×); (B) Moderate dysplasia (10×); and (C) Severe dysplasia (10×)



Figs 2A to C: Photomicrograph of stathmin immunoexpression in (A) Well-differentiated OSCC (10×); (B) Moderately differentiated OSCC (10×); and (C) Poorly differentiated OSCC (10×)

metastases, and poor prognosis. These studies suggested that stathmin is a downstream target of p53 and a positive regulator of motility resulting in the growth and motility of the cancer cells. Hence, overexpression of stathmin may play a role in tumor formation and development and also can be used as an tumor marker and molecular target for cancer therapy.[12] Based on the above facts, the purpose of this study is to correlate the biologic behavior of OSCC, TNM staging, and grading and oral leukoplakia and its various grades with respect to expression of stathmin immunohistochemically.

Materials and Methods

A retrospective analysis of 90 formalin fixed, paraffin embedded, histologically diagnosed tissue sections were retrieved from the Oral Pathology Department, Mamata Dental College, Khammam, Telangana, India. Out of 90 sections, 30 tissue sections were histologically diagnosed different grades of oral leukoplakia (oral dysplasia) (Fig. 1) and 30 sections of histologically diagnosed different grades of OSCC (Fig. 2) along with a control group consisting of 30 normal healthy mucosa. The distribution of groups is considered as listed in Table 1. The study was carried out only after obtaining the institutional ethical clearance. The histopathological grading for oral leukoplakia was done according to World Health Organization (WHO) grading of oral epithelial dysplasia, grading of OSCC was done using Broder's system.[13,14] Hemotoxylin and eosin stained tissue sections were used to verify the tissue sections histopathologically and categorized into different groups before continuing with the immunohistochemistry (IHC) procedure. The positive control used was brain tissue and normal oral mucosal tissue was taken as negative control for each batch of staining.

Furthermore, 3-µm thick sections were taken onto silanized slides. These sections were passed through a series of procedures such as deparaffinization, rehydration through two changes of xylene, and through grades of alcohol followed by distilled water for 30 seconds.

Table 1: Distribution of groups

Group	*Grading*	*Number*	*Staging*	*Number*
Dysplasia (*n* = 30)	Mild	16	–	–
	Moderate	10	–	–
	Severe	4	–	–
OSCC (*n* = 30)	Well-differentiated	14	–	–
	Moderately differentiated	10	–	–
	Poorly differentiated	6	–	–
	–	–	Stage I	5
	–	–	Stage II	7
	–	–	Stage III	11
	–	–	Stage IV	7

Antigen retrieval was done by placing the slides in a Tris-EDTA buffered saline solution and were heated for four times at 100°C for 5 minutes using microwave oven. Following this, the slides were brought to room temperature. Prior to immunostaining procedure, all the reagents in the IHC kit stored in the refrigerator were brought to room temperature. The immunostaining stathmin 1 antibody reagents were obtained from Novus Biologicals super-sensitive detection system. During the entirety of the IHC procedure, the drying of tissue sections is avoided. The procedure starts with the washing of sections gently with phosphate buffered saline (PBS) 3 times with a time gap of 30 seconds for each wash and excess of buffer solution was tapped off. This PBS wash was done after each step. Followed by covering of the tissue sections with the peroxide block (3% H_2O_2) for 10 minutes and then by power block for 5 minutes at room temperature and the excess power block was drained off. Peroxide block inhibits endogenous enzyme activity

prior to staining. Power block exposes the retrieved antigens and keeps them in position. In the next step, the all the tissue sections were treated with pre-diluted stathmin primary antibody excluding the negative control. Following this, 1 hour incubation of the slides at 21°C is done. A super-enhancer was added to the tissue sections which acts as a link between primary antibody and antigen. The sections were then covered with substrate chromogen solution for 10 minutes and then counterstained using Mayer's hematoxylin for 2 minutes, followed by washing the slides gently under running tap water for bluing. Dehydration was done by passing through various grades of alcohol for 5 minutes each. Clearing of tissue sections by xylene and later mounted using DPX and observed under the microscope. On microscopic examination, visibility of a brown colored end product at the site of target antigen indicates positive immunoreactivity. Subsequently, all the tissue sections were observed and graded them as either positive or negative.

The examination of slides was done and a total of 300 cells were examined manually in each tissue section and in at least 5 areas and determination of mean percentage of positive-stained slides was done. Then, the following staining scores were assigned for each tissue section: Less than 10%, 0; 11–25%, 1; 26–50%, 2; 51–75%, 3; 76–90%, 4; and 91–100%, 5. These interpretations were done by two observers to exclude the interobserver bias. The obtained data was subjected to statistical analysis using statistical package for social sciences (SPSS) software, version 20.

Results

Comparison between OSCC, Oral Dysplasia and Normal Groups

Oral Leukoplakia (Oral Dysplasia) Group

Among the 30 tissue sections, 3 sections showed a staining score of 0 (10%), 15 sections showed 1 (50%), 6 sections showed 2 (20%), 2 sections showed 3 (6.66%), and 4 sections showed 4 (13.33%).

The OSCC Group

Among the 30 sections in carcinoma group, the staining score was found to be 0 in only 1 section (3.33%), 7 sections showed scoring of 1 (23.33%), 9 sections showed a scoring of 2 (30%), 7 sections showed a scoring of 3 (23.33%), 2 sections showed a scoring of 4 (6.66%), and 4 sections showed a scoring of 5 (13.33%). The normal control group showed a staining score of 0 in all 30 sections (100%).

Kruskal–Wallis analysis of variance (ANOVA) test was performed to compare the staining scores between OSCC, dysplasia, and normal groups, and the results showed a statistically significant difference with a $p = 0.0001$ (Fig. 3).

Comparison of Levels of Stathmin Expression between Various Grades of Dysplasia

Among 30 oral dysplasia sections, 16 sections were histologically diagnosed as mild dysplasia, 10 were moderate dysplasia, and 4 were severe dysplasias. Among the 16 mild dysplasia sections, the stathmin staining score was found to be 0 in 3 sections (18.75%), and 13 sections showed a score of 1 (81.25%). Out of 10 moderate dysplasia sections, 2 sections showed a staining score of 1 (20%), 6 sections showed score of 2 (60%), 2 sections showed a score of 3 (20%). All of the 4 severe dysplasia sections showed a staining score of 4 (100%). Kruskal–Wallis ANOVA test showed a statistically significant difference among various histopathological grades of dysplasia with a $p = 0.0001$ for stathmin immunoexpression (Fig. 4).

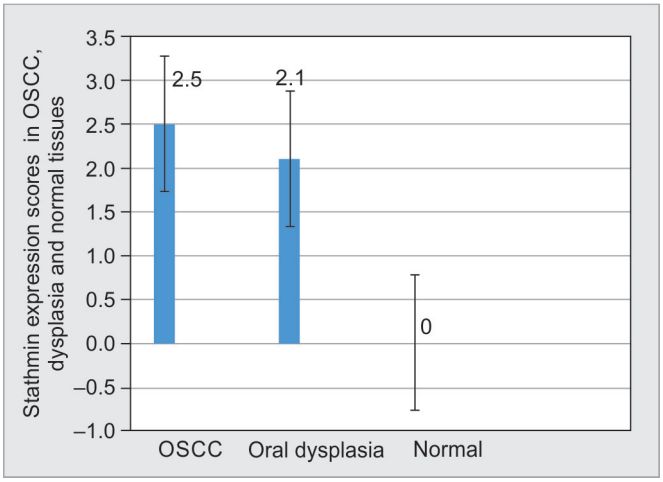


Fig. 3: Mean intensity staining scores of stathmin expression of normal, dysplasia, and carcinoma groups

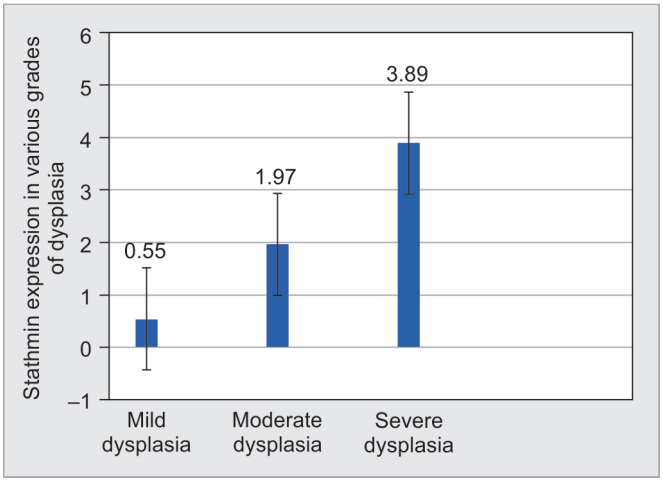


Fig. 4: Comparison of various histological grades of dysplasia (mild, moderate, and severe) for stathmin expression with respect to staining intensity scores

Comparison of Stathmin Immunoexpression between Various Grades of OSCC

Among 30 OSCC sections, 14 cases were histologically diagnosed as well-differentiated OSCC (WDSCC), 10 sections were moderately differentiated squamous cell carcinoma (MDSCC), and 6 sections were poorly differentiated squamous cell carcinoma (PDSCC). Out of 14 WDSCC, one case showing the staining score of 0 (7.14%), 7 sections showed the staining score of 1 (50%), and 6 sections showed the staining score of 2 (42.85%). Out of 10 MDSCC, the staining score was 2 in 3 sections (30%), and 3 in 7 sections (70%). In 6 poorly differentiated sections, the staining score was found to be 4 in 2 sections (33.33%), and 5 in 4 sections (66.66%). A statistically significant difference between various histological grades of OSCC was found using Kruskal–Wallis ANOVA test with respect to stathmin immunoexpression with a $p = 0.0001$ (Fig. 5).

Comparison of Stathmin Immunoexpression among Different Stages of OSCC (TNM Staging)

Among 30 cases of OSCC, 5 cases were staged as stage I, 7 cases in stage II, 11 cases in stage III, and 7 cases in stage IV. Among

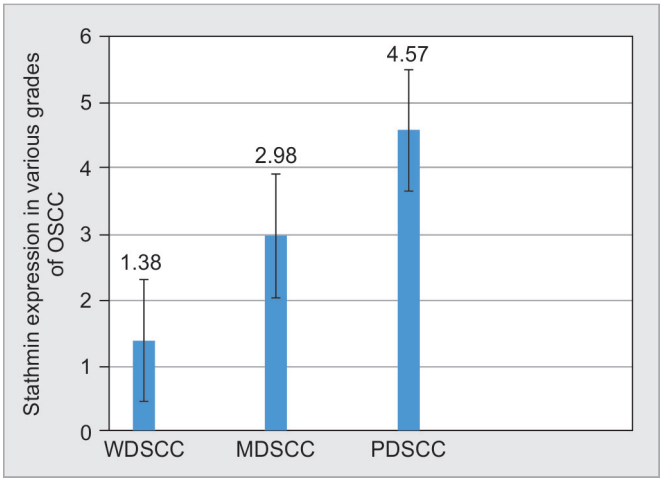


Fig 5: Comparison of various histological grades of OSCC for stathmin expression with respect to staining intensity scores

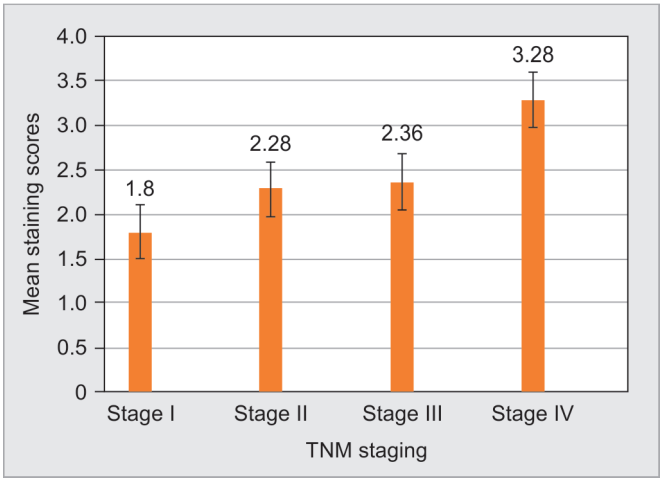


Fig. 6: Graph showing mean staining intensity scores in different stages of OSCC (TNM staging)

the 5 cases in stage I, the staining intensity score was 1 in 2 cases (40%), score 2 in 2 (40%) cases, and score 3 in 1 (20%) case. Among the 7 cases in stage II, the staining intensity score was 1 in 1 case (14.2%), score 2 in 5 cases (71.4%), and score 5 in 1 case (14.2%). In 11 cases of stage III, the staining intensity score was 0 in 1 case (9.1%), score 1 in 3 cases (27.2%), score 2 in 2 cases (18.2%), score 3 in 2 cases (18.2%), score 4 in 2 cases (18.2%), and score 5 in 1 case (9.1%). In 7 cases of stage IV, the staining intensity score was 1 in 1 case (14.2%), score 3 in 4 cases (57.1%), and score 5 in 2 cases (28.6%). The statistical analysis was done using one-way ANOVA test, the *p*-value was found to be 0.2935, which is found to be insignificant at $p < 0.05$ (Fig. 6).

Lymph Node Metastasis

Upon checking for lymph node metastasis, out of the 30 cases of OSCC, 15 cases showed lymph node metastasis with high staining intensity scores, that is, a score 5 in 3 cases (20%), a score 4 in 2 cases (13.33%), and a score 3 in 5 cases (33.33%) compared to the 15 cases which did not show nodal metastasis, that is, a score of 5 in only 1 case (6.66%) and a score of 3 in only 2 cases (13.33%) indicating that stathmin has a role in progression of nodal metastasis.

A statistically significant increase in the stathmin expression was found in OSCC group compared to oral dysplasia and normal control groups. The stathmin expression was found to be high in poorly differentiated OSCC cases with high staining intensity scores of 4 and 5 compared to other grades of OSCC. Likewise, the staining intensity of stathmin was as high as 4 in severe dysplasia compared to other grades of dysplasia.

Discussion

Stathmin otherwise called as oncoprotein 18/Op18, p18, p19, stathmin1, or metablastin is a cytosolic phosphoprotein primarily identified in neuroendocrine cells.[15,16] These are microtubule regulating proteins, that is, in microtubule assembly and dynamics. Stathmin is made of 149 amino acids, which are organized into four domains (I–IV), and the core region acts as a site for tubulin interaction with the additional requirement of either an N- or C-terminal extension. The C-terminal is interaction domain of stathmin. The N-terminal region of stathmin is the regulatory domain of stathmin, which shows four phosphorylation domains, designated as Ser 16, 25, 38, and 63. These phosphorylation domains closely correlate with the kinases involved in major intracellular regulatory cascades.[16]

Expression of stathmin is tightly controlled both during development and in adult tissues and that it is invariably induced by pro-mitogenic stimuli. The studies conducted also demonstrated that its expression is almost restricted to proliferating cells of all tissues with exception of glial cells, neurons and anterior pituitary cells. The levels of stathmin protein parallel the levels of its mRNA, suggesting stathmin's expression predominantly at transcription levels.[17]

Numerous reports have suggested a higher expression of stathmin in cancerous tissue than the normal tissue. The mechanism behind the higher expression is very little understood. There are few studies conducted which detailed about the association of stathmin with mutant p53 gene in breast cancer cell lines and hepatocellular carcinoma.[18,19] Around 50% of the tumor cells in cancers exhibits loss of function of p53 and the p53 gene has dominant negative mutations that suppresses p53 protein function.[20–22] Wild type p53 seems to preserve a diploid number of chromosomes through repression of stathmin and derepression of stathmin in mutant p53 cells which result in aneuploidy.[6]

A link has been suggested correlating the stathmin expression with the phosphorylation and regulation of proliferation of cells. Various studies revealed that the downward expression of stathmin can result in cell cycle arrest at G2/M phase in esophageal cancer cells and pancreatic carcinoma cells, down regulation of expression of NF-κB and decrease in cellular proliferation and invasion in lung cancer cells. Evidence also showed that stathmin has a role in tumor progression by binding to CDK inhibitors p27 and p21, and can result in controlling the early phase of G1 to S transition.[23–26]

The enigma behind the overexpression and inhibition of stathmin expression results in mitotic arrest was resolved years later by independent identification of stathmin as a cytosolic factor involved in the regulation of microtubule assembly and dynamics.[15] A wide range of human cancers showed overexpression of stathmin such as breast carcinoma, lung adenocarcinoma, Hodgkin's lymphoma, ovarian carcinoma, Wilms' tumor, and adenoid cystic carcinoma of the salivary glands.[16,27–31]

The purpose of this study was to compare and assess the expression of stathmin in OSCC, oral leukoplakia, and normal

tissues. The statistical analysis revealed a statistically significant increase in stathmin expression in OSCC [(mean staining score = 2.50) compared to oral dysplasia (2.11)] and normal groups (0.00).

An increase in the expression of stathmin was found to increase from mild dysplasia (mean staining score = 0.55), to moderate dysplasia (1.97) to severe dysplasia (3.89).

Comparison within different grades of OSCC revealed a statistically significant increase in the stathmin expression from WDSCC (mean staining score = 1.38) to MDSCC (2.98) to PDSCC (4.67). The OSCC cases under different stages (TNM staging) were compared for stathmin expression, which revealed that cases under stage IV had high stathmin immunoexpression in the tissues compared to other stages (I, II, and III). According to the research conducted by Kouzu et al., stathmin was overexpressed in OSCC and primary tumors with respect to advanced stages and poor diseases free survival. This study also showed a substantial correlation to clinical staging with a significantly differed expression in stage I/II and stage III/IV signifying its role in tumor progression and aggressiveness.[32] The sathmin protein status expression was also correlated with the disease free survival signifying its role in tumor prognosis/progression.[33] Cheng et al. in their study identified an overexpression of stathmin in primary nasopharyngeal carcinomas, its recurrence and higher grades of tumor. This study is in concordance with this study with respect to stathmin expression in advanced stages and higher grade tumours.[33] Nakashima et al. demonstrated overexpression of stathmin in adenoid cystic carcinoma reported using 2D differential in-gel electrophoresis.[31]

Among the 30 cases of OSCC, almost 15 cases showed lymph node metastasis which also showed high intensity staining score (5) of stathmin in the tissues. Similar observations were found in a study conducted by Jeon et al., which revealed stathmin was an independent predictor of shorter recurrence-free survival, and associated with lymph node metastasis and high grade stages in a cohort of 226 gastric cancer patients.[34] The stathmin expression was indicated as a potential tool for predicting the outcome of breast cancer patients with lymph node metastasis and its expression was increased in the group with poor prognosis.[35] These observations were similar to that of this study.

Marafioti et al. in their study of immunophenotyping of stathmin in large case series of follicular lymphomas, observed that a correlation of stathmin with higher grades of tumor existed and concluded that stathmin could be used as a unique IHC and a diagnostic marker.[36] The major limitation of this study includes smaller sample size and lack of comparison between expression of stathmin with the treatment options, outcome, and prognosis. This study can be made as an adjuvant with other molecular techniques such as qRT-PCR, etc., to elaborate the role of stathmin in various cancers to direct us for the target-based therapy of cancer.

Conclusion

Based on the above facts, stathmin is not only overexpressed in OSCC but also there is a differential expression in early and advanced grades and different TNM stages of OSCC. A high expression of stathmin was observed in all the cases with lymph node metastasis. These observations point us that stathmin has an important role in the tumorigenicity, progression, and prognosis of the oral cancer.

References

1. Hailat N, Strahler JR, Melhem RF, et al. N-myc gene amplification in neuroblastoma is associated with altered phosphorylation of a proliferation related polypeptide (Op 18). Oncogene 1990;5(11):1615–1618. PMID: 2267130.
2. Tian X, Tian Y, Sarich N, et al. Novel role of stathmin in microtubule-dependent control of endothelial permeability. FASEB J 2013;26(9):3862–3874. DOI: 10.1096/fj.12-207746.
3. Sobel A. Stathmin: A relay phosphoprotein for multiple signal transduction? Trends Biochem Sci 1991;16:301–305. DOI: 10.1016/0968-0004(91)90123-d.
4. Rubin CI, Atweh GF. The role of stathmin in the regulation of the cell cycle. J Cell Biochem 2004;93(2):242–250. DOI: 10.1002/jcb.20187.
5. Liu F, Sun YL, Xu Y, et al. Expression and phosphorylation of stathmin correlate with cell migration in esophageal squamous cell carcinoma. Oncol Rep 2013;29(2):419–424. DOI: 10.3892/or.2012.2157.
6. Brattsand G. Correlation of oncoprotein 18/stathmin expression in human breast cancer with established prognostic factors. Br J Cancer 2000;83(3):311–318. DOI: 10.1054/bjoc.2000.1264.
7. Trovik J, Wik E, Stefansson IM, et al. Stathmin overexpression identifies high risk patients and lymph node metastasis in endometrial cancer. Clin Cancer Res 2011;17(10):3368–3377. DOI: 10.1158/1078-0432.CCR-10-2412.
8. Hsieh SY, Huang SF, Yu MC, et al. Stathmin1 overexpression associated with polyploidy, tumor-cell invasion, early recurrence, and poor prognosis in human hepatoma. Mol Carcinog 2010;49(5):476–487.
9. Zheng P, Liu YX, Chen L, et al. Stathmin, a new target of PRL-3 identified by proteomic methods, plays a key role in progression and metastasis of colorectal cancer. J Proteome Res 2010;9(10):4897–4905. DOI: 10.1021/pr100712t.
10. Varambally S, Yu J, Laxman B, et al. Integrative genomic and proteomic analysis of prostate cancer reveals signatures of metastatic progression. Cancer Cell 2005;8(5):393–406. DOI: 10.1016/j.ccr.2005.10.001.
11. Alaiya AA, Franzen B, Fujioka K, et al. Phenotypic analysis of ovarian carcinoma: polypeptide expression in benign, borderline and malignant tumors. Int J Cancer 1997;73(5):678–683. DOI: 10.1002/(sici)1097-0215(19971127)73:5<678::aid-ijc11>3.0.co;2-2.
12. Karst AM, Levanon K, Duraisamy S, et al. Stathmin 1, a marker of PI3K pathway activation and regulator of microtubule dynamics, is expressed in early pelvic serous carcinomas. Gynecol Oncol 2011;123(1):5–12. DOI: 10.1016/j.ygyno.2011.05.021.
13. Fleskens S, Slootweg P. The histological grading of oral leukoplakia was done based on World Health Organization criteria. Grading systems in head and neck dysplasia: Their prognostic value, weaknesses and utility. Head Neck Oncol 2009;1(1):11–19. DOI: 10.1186/1758-3284-1-11.
14. Broders AC. The microscopic grading of cancer. Surg Clin North Am 1941;21:947–962.
15. Belletti B, Baldassarre G. Stathmin: A protein with many tasks. New biomarker and potential target in cancer. Expert Opin Ther Targets 2011;15(11):1249–1266. DOI: 10.1517/14728222.2011.620951.
16. Biaoxue R, Xiguang C, Hua L, et al. Stathmin-dependent molecular targeting therapy for malignant tumor: The latest 5 years' discoveries and developments. J Transl Med 2016;14(1):279. DOI: 10.1186/s12967-016-1000-z.
17. Rowlands DC, Williams A, Jones NA, et al. Stathmin expression is a feature of proliferating cells of most, if not all, cell lineages. Lab Invest 1995;72(1):100–113. PMID: 7837783.
18. Alli E, Yang JM, Hait WN. Silencing of stathmin induces tumor-suppressor function in breast cancer cell lines harboring mutant p53. Oncogene 2007;26(7):1003–1012. DOI: 10.1038/sj.onc.1209864.
19. Yuan RH, Jeng YM, Chen HL, et al. Stathmin overexpression cooperates with p53 mutation and osteopontin overexpression, and is associated with tumour progression, early recurrence, and poor prognosis in hepatocellular carcinoma. J Pathol 2006;209(4):549–558. DOI: 10.1002/path.2011.

20. Khan W, Augustine D, Rao RS, et al. Stem cell markers SOX-2 and OCT-4 enable to resolve the diagnostic dilemma between ameloblastic carcinoma and aggressive solid multicystic ameloblastoma. Adv Biomed Res 2018;7:149. DOI: 10.4103/abr.abr_135_18. eCollection 2018.
21. Augustine D, Sekar B, Murali S, et al. Expression of inducible nitric oxide synthase in carcinomas and sarcomas affecting the oral cavity. South Asian J Cancer 2015;4(2):78–82. DOI: 10.4103/2278-330X.155686.
22. Singh P, Augustine D, Rao RS, et al. Interleukin-1beta and caspase-3 expression serve as independent prognostic markers for metastasis and survival in oral squamous cell carcinoma. Cancer Biomark 2019;26(1):109–122. DOI: 10.3233/CBM-190149.
23. Wang S, Akhtar J, Wang Z. Anti-STMN1 therapy improves sensitivity to antimicrotubule drugs in esophageal squamous cell carcinoma. Tumour Biol 2015;36(10):7797–7806. DOI: 10.1007/s13277-015-3520-1.
24. Lu Y, Liu C, Cheng H, et al. Stathmin, interacting with Nf-kappaB, promotes tumor growth and predicts poor prognosis of pancreatic cancer. Curr Mol Med 2014;14(3):328–339. DOI: 10.2174/1566524014666140228120913.
25. Nie W, Xu MD, Gan L, et al. Overexpression of stathmin 1 is a poor prognostic biomarker in non-small cell lung cancer. Lab Invest 2015;95(1):56–64. DOI: 10.1038/labinvest.2014.124.
26. Berton S, Pellizzari I, Fabris L, et al. Genetic characterization of p27 (kip1) and stathmin in controlling cell proliferation *in vivo*. Cell Cycle 2014;13(19):3100–3111. DOI: 10.4161/15384101.2014.949512.
27. Askeland C, Wik E, Finne K, et al. Stathmin expression associates with vascular and immune responses in aggressive breast cancer subgroups. Sci Rep 2020;10(1):2914. DOI: 10.1038/s41598-020-59728-3.
28. Jia, W, Lin Z, Wen J, et al. SEPTIN2 and STATHMIN regulate CD99-mediated cellular differentiation in Hodgkin's lymphoma. PLOS One 2015;10(5):e0127568. DOI: 10.1371/journal.pone.0127568.
29. Price DK, Ball JR, Bahrani–Mostafavi Z, et al. The phosphoprotein Op18/stathmin is differentially expressed in ovarian cancer. Cancer Invest 2000;18(8):722–730. DOI: 10.3109/07357900009012204.
30. Takahashi M, Yang XJ, Lavery TT, et al. Gene expression profiling of favorable histology Wilms tumors and its correlation with clinical features. Cancer Res 2002;62(22):6598–6605. PMID: 12438255.
31. Nakashima D, Uzawa K, Kasamatsu A, et al. Protein expression profiling identifies maspin and stathmin as potential biomarkers of adenoid cystic carcinoma of the salivary glands. Int J Cancer 2005;118(3):704–713. DOI: 10.1002/ijc.21318.
32. Kouzu Y, Uzawa K, Koike H, et al. Overexpression of stathmin in oral squamous cell carcinoma: correlation with tumour progression and poor prognosis. Br J Cancer 2006;94(5):717–723. DOI: 10.1038/sj.bjc.6602991.
33. Cheng AL, Huang WG, Chen ZC, et al. Identification of novel nasopharyngeal carcinoma biomarkers by laser capture microdissection and proteomic analysis. Clin Cancer Res 2008;14(2):435–445. DOI: 10.1158/1078-0432.CCR-07-1215.
34. Jeon TY, Han ME, Lee YW, et al. Overexpression of stathmin1 in the diffuse type of gastric cancer and its roles in proliferation and migration of gastric cancer cells. Br J Cancer 2010;102(4):710–718. DOI: 10.1038/sj.bjc.6605537.
35. Oishi Y, Nagasaki K, Miyata S, F et al. Functional pathway characterized by gene expression analysis of supraclavicular lymph node metastasis-positive breast cancer. J Hum Genet 2007;52(3):271–279. DOI: 10.1007/s10038-007-0111-z.
36. Marafioti T, Copie–Bergman C, Calaminici M, et al. Another look at follicular lymphoma: immunophenotypic and molecular analyses identify distinct follicular lymphoma subgroups. Histopathology 2013;62(6):860–875.