

PI3K/AKT/MTOR SIGNALING PATHWAY – A KEY PLAYER IN THE MOLECULAR LANDSCAPE OF MALIGNANT TRANSFORMATION OF ORAL LESIONS

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ABSTRACT

Aberrant PI3K/AKT/mTOR/ pathway have been considered one of the starting points of various types of human cancer, including the oral squamous cell carcinoma (OSCC). The important role played by this signaling pathway in oncogenesis and cancer evolution, oriented the researchers' attention toward studies of AKT and the PI3K/AKT/mTOR axis in the context of malignant transformation of oral precancerous lesions. Consequently, this article's aim is an update of the possible key role played by the PI3K /AKT/mTOR axis on the oral lesions' malignant transformation stage.

Oral potentially malignant disorders are defined as lesions that will most probably progress to malignant lesions of the oral cavity. These lesions are mainly represented by leukoplakia, erythroplakia, actinic cheilitis and lichen planus.

PI3K/AKT/mTOR signaling cascade should be regarded as a crucial subject for additional research, focusing not only on the early identification of malignant transformations of oral lesions, but also on opening new therapeutic ways in order to prevent malignant transformation and OSCC evolution.

Key words: PI3K/AKT/mTOR pathway, leukoplakia, erythroplakia, lichen planus, oral potentially malignant disorders

INTRODUCTION

Currently, the clinical and pathological stage at the time of diagnosis, still remains the most important influencing factor of prognosis in oral squamous cell carcinoma (OSCC). (1-3) Considering the high mortality rate, the anticipation of malignant transformation of the oral potentially malignant disorders and the early detection of oral malignancy will ensure a better prognosis and increased survival rates. (4,5) The most important OSCC risk factors include tobacco use (including smokeless tobacco), alcohol consumption, oral mucosal trauma caused by teeth and prosthetic devices, chronic mucosal inflammation, HPV infection, ultraviolet radiation (mainly UV-B) exposure. (6-8) Considering that most of the risk factors can be avoided, oral cancer may be regarded as a possible preventable disease. However, OSCC occurrence in patients outside the

risk groups is continuously reported. (9,10) In these patients case an important role should be assigned to the oral potentially malignant disorders. (11,12)

Oral potentially malignant disorders (OPMD) are defined as morphologically altered tissues, more likely able to develop cancer. (13) The potentially malignant oral epithelial lesions represent the manifestations of a group of oral pathologies, present before the onset of OSCC.

In the literature, over time these lesions have been referred to by terms such as “precancerous”, “precancerous/premalignant lesions” and “intraepithelial neoplasia”. The term “potentially malignant disorders” was adopted by the World Health Organization because there is no certainty that all precancerous lesions will eventually develop oral cancer. (14)

A broad spectrum of lesions and conditions such as leukoplakia, proliferative verrucous leukoplakia, erythroplakia, actinic cheilitis, oral lichen planus, oral submucous fibrosis, palatal mucosal changes in reverse smokers, lupus erythematosus and some hereditary conditions (epidermolysis bullosa and dyskeratosis congenita) are considered oral potentially malignant disorders. There is sufficient evidence for an increased risk of oral cancer also among patients presenting with "oral lichenoid lesions" as well as among those diagnosed with oral manifestations of chronic graft-versus-host disease, both of which have recently been added to the list of oral potentially malignant disorders (OPMD) by the WHO Collaborating Centre for Oral Cancer UK (2020) following an updated report on OPMD nomenclature and classification. (14)

Dealing with patients with oral potentially malignant disorders is difficult to manage. A proportion of patients will develop malignancies, so determining the patient's cancer risk is key to making management decisions. However, understanding the process of malignant transformation of lesions is not fully elucidated, and a precision approach based on the integration of numerous predictive markers has not yet been validated. (15-17)

Lesions with malignant potential have a statistically increased risk of progressing to cancer, but the risk varies depending on a number of factors related to the patient or lesion. (18)

There are many favoring factors associated with the development of potentially malignant lesions and the risk of malignant transformation of the lesion. (18,19).

Independent evolution of malignant or premalignant lesions is considered when they are result of separate genetic events triggers. (20) Recent theories reveal that transformed cells may migrate to adjacent cells, resulting in multiple lesions on various mucosal area. (21) For example, in gastrointestinal cancer, cancer cells seem to

be widespread in crypt or gland fission. (22) Migration can also be a possible mechanism for closely spaced oral lesions. However, Poh *et al.* study that included multiple pre-oral cancer lesions such as hyperplasia, dysplasia, highlighted that most of these multiple lesions occurred from clonally independent cells. (23) Moreover, the recent study of Bakker *et al.* reported that in widely isolated head and neck cancers, the lesions are usually not clonally associated, p53 mutations playing key roles. (24) Given the complex molecular relationship between the PI3K/AKT/mTOR axis and the tumor suppressor p53, a special attention should be assigned to the AKT/mTOR signaling pathway in the context of oral lesions malignant transformations.

Aberrant PI3K/AKT/mTOR pathway have been considered one of the starting points of various types of human cancer, including the oral squamous cell carcinoma (OSCC). (25,26) Consequently, this article's aim is an update of the possible key role of the AKT/mTOR signaling on the oral lesions' malignant transformation stage.

MATERIALS AND METHODS

In our research, we conducted the literature from databases such as PubMed, Science Direct, Google Scholar, Scopus and Web of Science using the following search terms: "PI3K/AKT/mTOR pathway", "oral squamous cell carcinoma", "leukoplakia", "erythroplakia", "lichen planus", and "oral potentially malignant disorders".

RESULTS AND DISCUSSIONS

The key cause of late-stage cancer diagnosis is delayed diagnosis, so achieving the goal of improving oral cancer prognosis means reducing the delay in diagnosis. The first goal that should be achieved is for clinicians to know precisely the clinical signs and symptoms that lead to the suspicion that a patient has oral cancer at an early stage or oral potentially malignant disorders (OPMD). (27)

1. Oral potentially malignant disorders (OPMD), characterized by an increased, variable risk of developing oral carcinoma, are mainly represented by:

a) Leukoplakia is the most common oral potentially malignant disorder. It is defined as predominant white lesion of the oral mucosa that cannot be characterized as another definable lesion. (28-31) Oral leukoplakia is more common found in elderly population, between 40-60 years (32,33) and is usually more noticeable on the alveolar ridge gingiva or on the tongue. (34)

The study by Aguirre-Urizar *et al.* following a systematic review of articles published in the last 5 years (2015-2020) showed that the percentage of malignant transformation of oral leukoplakia ranged from 1.1% to 40.8%. In addition, study findings showed that female sex, non-homogeneous clinical type of leukoplakia and presence of epithelial dysplasia were significantly related to malignant transformation. (35)

The same aspect related to the non-homogeneous clinical type of leukoplakia is presented by Pindborg *et al.* who show that speckled leukoplakia, characterized by the presence of the white nodular patches or white lesions, interspersed with erythematous areas, was often associated with epithelial dysplasia or carcinoma. (36) The reported leukoplakia malignant transformation rate is overall about 4,9% per year. (37)

In leukoplakia diagnosed patients the malignant transformation events can also occur in other tissue areas of oral cavity and upper aerodigestive tract, not only in the site of the primarily diagnosed lesion. Currently, leukoplakia is regarded as a great risk marker of the oral mucosa cancer. (38,39)

b) Proliferative verrucous leukoplakia is classified as a potentially malignant condition with a high rate of transformation into oral squamous cell carcinoma and verrucous carcinoma. (40) The incidence is

estimated at 3-9% of all oral mucosal cancers (43), and the rate of malignant transformation is 50%, averaging 57 months. (44)

It is a particular form of leukoplakia, multifocal, extensive, of unknown etiology, usually refractory to all therapeutic attempts and with frequent relapses. It occurs predominantly in older women, the most affected topographic areas being the gingiva, buccal mucosa, alveolar crest, while the tongue is less involved. (41)

Any leukoplakia lesion that becomes exophytic, extensive and multifocal after a period of time and has recurred after treatment should raise clinical suspicion of proliferative verrucous leukoplakia. (42)

c) Oral erythroplakia is defined as a premalignant lesion of the oral mucosa. (45,46) Traditionally it has been described by analogy with oral leukoplakia, as a red lesion that cannot be clinically or pathologically characterized as another well-defined lesion. (45)

Same as for leukoplakia, the main factors incriminated in this disease onset are represented by tobacco usage and alcohol intake. (47) According to several studies, oral erythroplakia has a high rate of malignant transformation, with variations between 14,3 to 50%. (48-50) Currently, p53 gene mutations are considered main actors in erythroplakia malignant transformations. These mutations were identified in exons 6 (33%), 5 (25%), 8 (25%) and 9 (17%) of p53 gene. (51)

This is one of the most important oral lesions because in 75-90% of cases it actually turns out to be carcinoma, carcinoma in situ or has severe dysplasia. (52)

d) Oral lichen planus (OLP) is considered by the World Health Organization as a possible precancerous condition. The premalignant potential of OLP is still a source of contention in the scientific literature. (53)

Fitzpatrick *et al.* following observational evaluation of 16 studies showed that in

individual studies the rate of malignant transformation of lichen lesions was up to 3.5%, and the most common topographic location of malignancy was the tongue. (54) Case report studies continuously highlight a possible significant risk of malignant transformation of OLP lesions to OSCC. (55-57)

Oral lichen planus (OLP) is defined as a chronic inflammatory condition of unknown etiology with characteristic periods of remissions and exacerbations. (58)

OLP is represented by a white, lacy, reticular pattern lesion, located on the buccal mucosa of the cheeks, usually on both sides of the internal cheeks. (59) Other forms include papular, atrophic or erosive lesions. (60) The erosive lesions may be quite painful and may develop multiple complications, such as secondary infections caused by *Candida* spp. (61)

Atrophic and erosive lesions of the tongue, especially if the patient is a smoker, have a higher risk of malignancy. A major challenge in the clinical follow-up of patients with OLP in terms of early diagnosis of malignant transformation of the disease relates to the difficulty in differentiating atrophic-erythematous lesions that frequently occur in OLP from erythroplakia lesions, which frequently occur as an expression of incipient carcinomas. (62)

e) Oral lichenoid lesions.

In the latest classification of OPMD (2020) (14) it was included in the group of potentially malignant conditions. In recent years, differentiation between oral lichen planus (OLP) and oral lichenoid lesions (OLL) has become important, as the latter may have a higher malignant potential. (63) Lichenoid reactions are classified into 3 types: (1) in relation to dental restorations, often amalgam, also called lichenoid contact lesions, (2) drug-related and (3) in association with chronic graft-versus-host disease (cGvHD). (43)

f) Oral submucous fibrosis are encountered particularly in population groups from Asia with specific lifestyle habits (the habit of chewing betel and areca nuts). (42)

g) Oral lupus erythematosus has been recognized by the WHO as a potentially malignant disease, but malignant transformation is known to be extremely rare. (64,65)

h) Oral epidermolysis bullosa in the latest classification of OPMD, due to lack of clear evidence, has been classified as a condition with limited evidence of malignant lesions. (14)

i) Actinic cheilitis is a potentially malignant lesion located on the lower lip caused by chronic exposure to ultraviolet radiation. The malignant transformation process to squamous cell carcinoma is significant (6-10%), with a high rate of metastasis (11% compared to 1% in other body areas). (66,67)

Risk factors for malignant transformation are considered to be people with excessive exposure to ultraviolet radiation, especially those living in tropical regions, with fair skin, males, older age and smokers. (68)

j) Oral manifestation of chronic graft-versus-host disease (GVHD) is a major complication of allogeneic hematopoietic stem cell transplantation and the leading cause of post-transplant morbidity and mortality.

Oral manifestations of GVHD include atrophic-ulcerative and keratotic lesions of the oral mucosa (similar to lichen planus), salivary gland dysfunction (similar to Sjögren's/Sicca syndrome), and limitation of mouth opening secondary to oral and perioral tissue sclerosis (similar to limitation in scleroderma). (69)

Therapeutic management may include both local and general long-term immunosuppressive therapy and/or corticosteroid therapy. The most common side effects of this therapy are the presence of local and general infections, hyperglycemia, renal dysfunction,

osteoporosis and possible development of secondary malignancies. Due to the increased risk of oral carcinoma in patients with oral GVHD, periodic evaluation is required, similar to other OPMD. (70)

Despite the accumulation of much knowledge in recent decades about early diagnosis and treatment of OSCC, patient survival rates have not improved, with overall survival at 5 years remaining around 50%. Late diagnosis, high relapse rate, local invasiveness and high risk of lymph node metastasis are among the reasons for the unchanged mortality rate over the last decade. The importance of early diagnosis and treatment at T1 or T2 stages is also known, with the percentage of relapse-free cases increasing to 84.6%. Deciphering the molecular mechanisms governing oral cancer is absolutely necessary for developing more effective diagnostic and therapeutic strategies. (71)

Therefore, the precise identification and characterization of certain molecular events with a key role in the malignant transformation of lesions with malignant potential is still a major research objective. PI3K/AKT/mTOR is considered one of the main, but discrete, players in malignant transformation. Consequently, it should be regarded as an important target in the studies focusing on the premalignant potential of oral lesions. (72-74)

2. PI3K/AKT/mTOR signaling pathway

Extracellular signaling is a strategy for body cells to interact and communicate with one another. This mechanism results with the production of elected molecules that will link to particular receptors on the surface of other cells. (75)

mTOR represents a mammalian target of rapamycin. This is a serine-threonine kinase that influences metabolism. (72) AKT, known as protein kinase B, is an evolutionarily conserved serine/ threonine kinase. In humans AKT has three isoforms, named AKT1, AKT2, AKT3. (73,76-79)

A cytokine molecule binding to a specific membrane receptor can induce phosphoinositide 3-kinase (PI3K) activation. PI3K is composed of two subunits, p85 and p110. p110 subunit enzymatic function activation, after cytokine receptor – PI3K interaction, triggers PIP2 phosphorylation to PIP3 in the membrane. (26,80-85) PIP2 phosphorylation to PIP3 represents a molecular event that triggers AKT recruitment to the membrane. PIP3 generation at the plasma membrane is controlled by the balance between PI3K activity and PTEN (phosphatase and tensin homolog), responsible for PIP3 dephosphorylation. (26,81-86) In this way PTEN inhibits AKT recruitment within the PI3K/AKT/mTOR signaling cascade. Activated AKT is involved in the downstream regulation cascade of mTORC1 and mTORC2 complexes, inducing in turn specific genes transcription and translation machinery proteins activation, in order to sustain cell growth and cell survival. (26,80-86) (Figure 1)

Insert Figure 1.

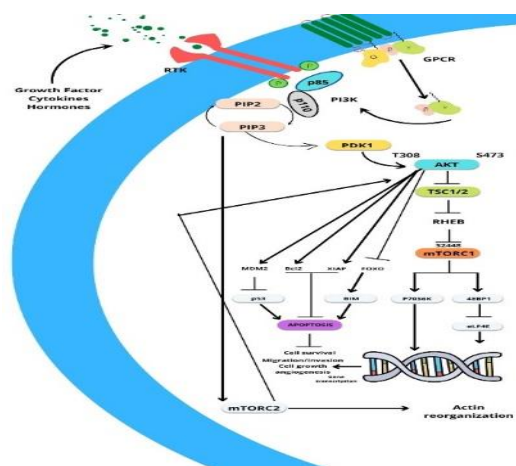


Figure 1. The AKT/mTOR signaling pathways and its role in various processes.

RTKs: a family of transmembrane proteins with inherent phosphotyrosine kinase activity which remain inactive in the plasma membrane until activated by ligand; GPCR: G-protein-coupled receptor; p85: a cytoplasmic domain through its regulatory subunit of PI3K (phosphoinositide 3-

kinase, also called phosphatidylinositol 3-kinases); p110: catalytic subunit of PI3K; PIP1 and PIP3: PtdIns(3,4,5) P3, phosphatidylinositol (3,4,5)-trisphosphate synthetic peptides; PDK1: 3-phosphoinositide-dependent protein kinase-1; AKT: Acute transforming retrovirus from AKR thymoma; TSC1/2: several genes of the pathway; RHEB: less understood complex; S2448: Phospho-mTOR; mTORC1: mammalian target of rapamycin complex 1; P70S6K: S6K, p70 ribosomal S6 kinase; 4EBP1: a multifactor regulated multifunctional protein; EIF4E: eukaryotic translation initiation factor 4E; MDM2: an E3 ubiquitin ligase that triggers p53 degradation; Bcl2: Antagonist of cell death protein; XIAP: X-linked inhibitor of apoptosis protein; FOXO: transcription factors; p53: cellular tumour antigen; Bim: Bcl-2-like protein 11, gene expression; mTORC2: an acutely rapamycin-insensitive protein complex formed by serine/threonine kinase mTOR that regulates cell proliferation and survival, cell migration and cytoskeletal remodeling.

AKT, as a central member of the PI3K/AKT/mTOR axis, plays key roles in controlling vital cellular signaling pathways, involved in metabolism and autophagy regulation and apoptosis suppression. AKT has also been assigned an important part in oncogenesis and angiogenesis. (87-89)

3. PI3K/AKT/mTOR signaling cascade and malignant transformation

It has been highlighted the involvement of the PI3K/AKT/mTOR signaling pathway in the molecular landscape of various human diseases, such as diabetes mellitus, cardiovascular disorders and neurological disorders. (73,74,90) Moreover, PI3K/AKT/mTOR cascade is regarded as one of the most deeply implicated signaling pathways in human cancer, including OSCC. (73) The important role played by this signaling pathway in oncogenesis and cancer evolution, oriented the researchers' attention toward studies of AKT and the PI3K/AKT/mTOR axis in the context of malignant transformation of oral

precancerous lesions. It must be emphasized the fact that many targeted therapies and chemotherapies in cancer are oriented towards controlling PI3K/AKT/mTOR signaling pathway, highlighting the main role played by AKT activation. (91) Miricescu *et al.* highlighted the important role played by an aberrant PI3K/AKT/mTOR cascade in oncogenesis and cancer evolution. (26) An upregulated level of expression of PI3K/AKT/mTOR axis key proteins has been reported in oral epithelial dysplasia. (92,93) Massarelli *et al.* showed that PI3K/AKT/mTOR signaling is directly involved in the progression of oral cancer and possibly in malignant transformation. A study conducted by Andrikopoulou *et al.* reported aberrant activation levels of AKT/mTOR signaling pathway in squamous intraepithelial lesions. The researchers tracked the expression of phosphorylated and activated versions of AKT in biopsy samples from cervical squamous intraepithelial lesions. Andrikopoulou results outlined the conclusion that the PI3K/AKT/mTOR signaling pathway played key roles in cervical cancer progression. Similar experiments should be conducted on biopsy samples collected from oral lesions with premalignant potential, in different stages of clinical evolution.

Many studies highlighted a tight molecular link between chronic inflammation and the PI3K/AKT/mTOR cascade. Sonis *et al.* results revealed abnormal levels of PI3K/AKT/mTOR pathway activity in OLP lesions. This study suggested the existence of a mechanism linking the activity of the signaling pathway in the context of chronic inflammation, and the premalignant potential of OLP lesions. Harsha *et al.* study highlighted aberrant activities of PI3K/AKT/mTOR signaling pathway in OSCC context. Gutkind *et al.* study revealed that in OSCC, metformin downregulated PI3K/AKT/mTOR axis activity, induced AMPK activation,

triggering the downregulation of the proliferative capacity of premalignant cells and encouraging their commitment to terminal differentiation. Gutkind *et al.* results led to the conclusion that metformin long treatment with metformin may trigger increased positive clinical response rates, because oral premalignant lesions needed time for the progressive remodeling process, despite the observation that the downregulation of cell proliferation and histological changes might have occurred more rapidly. According to Zhang *et al.*, the AKT/mTOR-autophagy relationship plays an important role in the local T cell-mediated immunoregulatory mechanism in OLP. The study results revealed how LC3B expression level might be a useful marker illustrating the progression direction of a premalignant lesions. (94) Ma *et al.* described how the aberrant IGF1-PI3K/AKT/MTOR signaling pathway may play a negative role in the immunoregulatory mechanism of OLP. (98)

CONCLUSIONS

The PI3K/AKT/mTOR signaling pathway plays one of the main roles in the molecular landscape of malignant transformation of oral potentially malignant disorders. It also keeps its important role during the evolution of the OSCC. Consequently, this signaling cascade should be regarded as a crucial subject for additional research, focusing not only on the early identification of malignant transformations of oral lesions, but also on opening new therapeutic ways in order to prevent malignant transformation and OSCC evolution. The experimental data obtained in the future studies targeting the PI3K/AKT/mTOR axis, in the context of malignant transformations, should pave more solid roads toward more suitable and specific treatment strategies of oral lesions with oral potentially malignant disorders.

Conflict of interest

The authors declare that they have no conflict of interest.

References

1. Ferreira AK, Carvalho SH, Granville-Garcia AF et al. Survival and prognostic factors in patients with oral squamous cell carcinoma. *Med Oral Patol Oral Cir Bucal*. 2021; 26(3):e387-e392.
2. Zittel S, Moratin J, Horn D et al. Clinical outcome and prognostic factors in recurrent oral squamous cell carcinoma after primary surgical treatment: a retrospective study. *Clin Oral Investig*. 2022; 26(2):2055-2064.
3. Ghantous Y, Nashef A, Sidransky D et al. Clinical and prognostic significance of the eighth edition oral cancer staging system. *Cancers (Basel)*. 2022; 14(19):4632.
4. Abati S, Bramati C, Bondi S et al. Oral cancer and precancer: A narrative review on the relevance of early diagnosis. *Int J Environ Res Public Health*. 2020; 17(24):9160.
5. Arduino PG, Magliano A, Gambino A et al. Risk of malignant transformation in 3173 subjects with histopathologically confirmed oral lichen planus: a 33-year cohort study in Northern Italy. *Cancers (Basel)*. 2021; 13(22):5740.
6. Auguste A, Deloumeaux J, Joachim C et al. Joint effect of tobacco, alcohol, and oral HPV infection on head and neck cancer risk in the French West Indies. *Cancer Med*. 2020; 9(18):6854-6863.
7. Aghiorghiesei O, Zanoaga O, Nutu A et al. The world of oral cancer and its risk factors viewed from the aspect of microrna expression patterns. *Genes (Basel)*. 2022; 13(4):594.
8. Jiang X, Wu J, Wang J et al. Tobacco and oral squamous cell carcinoma: A review of carcinogenic pathways. *Tob Induc Dis*. 2019; 17:29.
9. Johnson DE, Burtneess B, Leemans CR et al. Head and neck squamous cell carcinoma. *Nat Rev Dis Primers*. 2020; 6(1):92.
10. Irani S. New insights into oral cancer-risk factors and prevention: A review of literature. *Int J Prev Med*. 2020; 11:202.
11. Ilhan B, Epstein JB, Guneri P. Potentially premalignant disorder/lesion versus potentially premalignant patient: Relevance in clinical care. *Oral Oncol*. 2019; 92:57-58.

- 12.Crawford M, Johnson EH, Liu KYP et al. On the cutting edge of oral cancer prevention: finding risk-predictive markers in precancerous lesions by longitudinal studies. *Cells*. 2022; 11(6):1033.
- 13.Pires FR, Barreto ME, Nunes JG et al. Oral potentially malignant disorders: clinical-pathological study of 684 cases diagnosed in a Brazilian population. *Med Oral Patol Oral Cir Bucal*. 2020; 25(1):e84-e88.
- 14.Warnakulasuriya S, Kujan O, Aguirre-Urizar JM et al. Oral potentially malignant disorders: A consensus report from an international seminar on nomenclature and classification, convened by the WHO Collaborating Centre for Oral Cancer. *Oral Dis*. 2021; 27(8):1862-1880.
- 15.Kerr AR, Lodi G. Management of oral potentially malignant disorders. *Oral Dis*. 2021; 27(8):2008-2025.
- 16.Kumari P, Debta P, Dixit A. Oral potentially malignant disorders: etiology, pathogenesis, and transformation into oral cancer. *Front Pharmacol*. 2022; 13:825266.
- 17.Lorini L, Bescós Atín C, Thavaraj S et al. Overview of oral potentially malignant disorders: from risk factors to specific therapies. *Cancers (Basel)*. 2021; 13(15):3696.
- 18.Wetzel SL, Wollenberg J. Oral Potentially Malignant Disorders. *Dent Clin North Am*. 2020; 64(1):25-37.
- 19.Speight PM, Khurram SA, Kujan O. Oral potentially malignant disorders: risk of progression to malignancy. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2018; 125(6):612-627.
- 20.Curtius K, Wright NA, Graham TA. Evolution of premalignant disease. *Cold Spring Harb Perspect Med*. 2017; 7(12):a026542.
- 21.Bansal R, Nayak BB, Bhardwaj S et al. Cancer stem cells and field cancerization of head and neck cancer - An update. *J Family Med Prim Care*. 2020; 9(7):3178-3182.
- 22.Hayakawa Y, Nakagawa H, Rustgi AK et al. Stem cells and origins of cancer in the upper gastrointestinal tract. *Cell Stem Cell*. 2021; 28(8):1343-1361.
- 23.Poh CF, MacAulay CE, Laronde DM et al. Squamous cell carcinoma and precursor lesions: diagnosis and screening in a technical era. *Periodontol 2000*. 2011; 57(1):73-88.
- 24.de Bakker T, Journe F, Descamps G et al. Restoring p53 function in head and neck squamous cell carcinoma to improve treatments. *Front Oncol*. 2022; 11:799993.
- 25.Rascio F, Spadaccino F, Rocchetti MT et al. The pathogenic role of PI3K/AKT pathway in cancer onset and drug resistance: an updated review. *Cancers (Basel)*. 2021; 13(16):3949.
- 26.Miricescu D, Totan A, Stanescu-Spinu II et al. PI3K/AKT/mTOR signaling pathway in breast cancer: from molecular landscape to clinical aspects. *Int J Mol Sci*. 2020; 22(1):173.
- 27.González-Moles MÁ, Aguilar-Ruiz M, Ramos-García P. Challenges in the early diagnosis of oral cancer, evidence gaps and strategies for improvement: a scoping review of systematic reviews. *Cancers (Basel)*. 2022; 14(19):4967.
- 28.Mohammed F, Fairozekhan AT. Oral leukoplakia. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK442013>.
- 29.Warnakulasuriya S, Johnson NW, van der Waal I. Nomenclature and classification of potentially malignant disorders of the oral mucosa. *J Oral Pathol Med*. 2007; 36(10):575-80.
- 30.Hashim D, Genden E, Posner M et al. Head and neck cancer prevention: from primary prevention to impact of clinicians on reducing burden. *Ann Oncol*. 2019; 30(5):744-756.
- 31.Ganesh D, Sreenivasan P, Öhman J et al. Potentially malignant oral disorders and cancer transformation. *Anticancer Res*. 2018; 38(6):3223-3229.
- 32.Adhailiya N, Kalappanavar AN, Ali IM et al. Autophagy: A boon or bane in oral cancer. *Oral Oncol*. 2016; 61:120-6.
- 33.Ge S, Liu L, Zhou Q, Lou B, Zhou Z, Lou J, Fan Y. Prevalence of and related risk factors in oral mucosa diseases among residents in the Baoshan District of Shanghai, China. *PeerJ*. 2020; 8:e8644.
- 34.Alabdulaaly L, Almazyad A, Woo SB. Gingival Leukoplakia: Hyperkeratosis with Epithelial Atrophy Is A Frequent Histopathologic Finding. *Head Neck Pathol*. 2021; 15(4):1235-1245.
- 35.Aguirre-Urizar JM, Lafuente-Ibáñez de Mendoza I, Warnakulasuriya S. Malignant transformation of oral leukoplakia: Systematic review and meta-analysis of the last 5 years. *Oral Dis*. 2021;27(8):1881-1895.

36. Pindborg JJ, Renstrup G, Poulsen HE et al. Studies in oral leukoplakias. V. Clinical and histologic signs of malignancy. *Acta Odontol Scand.* 1963; 21:407-14.
37. Evren I, Brouns ER, Wils LJ et al. Annual malignant transformation rate of oral leukoplakia remains consistent: A long-term follow-up study. *Oral Oncol.* 2020; 110:105014.
38. van der Waal I. Oral leukoplakia, the ongoing discussion on definition and terminology. *Med Oral Patol Oral Cir Bucal.* 2015; 20(6):e685-92.
39. Irani S. Pre-cancerous lesions in the oral and maxillofacial region: a literature review with special focus on etiopathogenesis. *Iran J Pathol.* 2016; 11(4):303-322.
40. Akrish S, Eskander-Hashoul L, Rachmiel A et al. Clinicopathologic analysis of verrucous hyperplasia, verrucous carcinoma and squamous cell carcinoma as part of the clinicopathologic spectrum of oral proliferative verrucous leukoplakia: A literature review and analysis. *Pathol Res Pract.* 2019; 215(12):152670.
41. Capella DL, Gonçalves JM, Abrantes AAA et al. Proliferative verrucous leukoplakia: diagnosis, management and current advances. *Braz J Otorhinolaryngol.* 2017; 83(5):585-593.
42. Warnakulasuriya S. Clinical features and presentation of oral potentially malignant disorders. *Oral Surg Oral Med Oral Pathol Oral Radiol.* 2018; 125(6):582-590.
43. Țovaru S, Țovaru M, Costache M et al. *Medicină și patologie orală* QMed Publishing Press, Bucuresti, 2012.
44. Palaia G, Bellisario A, Pampena R et al. Oral proliferative verrucous leukoplakia: progression to malignancy and clinical implications. systematic review and meta-analysis. *Cancers (Basel).* 2021; 13(16):4085.
45. Holmstrup P. Oral erythroplakia-What is it? *Oral Dis.* 2018; 24(1-2): 138-143.
46. Yardimci G, Kutlubay Z, Engin B et al. Precancerous lesions of oral mucosa. *World J Clin Cases.* 2014; 2(12):866-72.
47. Mello FW, Melo G, Pasetto JJ et al. The synergistic effect of tobacco and alcohol consumption on oral squamous cell carcinoma: a systematic review and meta-analysis. *Clin Oral Investig.* 2019; 23(7):2849-2859.
48. Reichart PA, Philipsen HP. Oral erythroplakia-a review. *Oral Oncol.* 2005; 41(6):551-61.
49. Ferreira DM, Neves TJ, Lima LGCA et al. Prognostic implications of the phosphatidylinositol 3-kinase/Akt signaling pathway in oral squamous cell carcinoma: Overexpression of p-mTOR indicates an adverse prognosis. *Appl. Cancer Res.* 2017; 37:41.
50. Villa A, Villa C, Abati S. Oral cancer and oral erythroplakia: an update and implication for clinicians. *Aust Dent J.* 2011; 56(3):253-6.
51. Qin GZ, Park JY, Chen SY et al. A high prevalence of p53 mutations in pre-malignant oral erythroplakia. *Int J Cancer.* 1999; 80(3):345-8.
52. Bedi R, Scully C. Tropical Oral Health. *Manson's Tropical Infectious Diseases.* 2014:1073–1083.e2.
53. Shirasuna K. Oral lichen planus: Malignant potential and diagnosis. *Oral Sci. Int.* 2014; 11(1):1-7.
54. Fitzpatrick SG, Hirsch SA, Gordon SC. The malignant transformation of oral lichen planus and oral lichenoid lesions: a systematic review. *J Am Dent Assoc.* 2014; 145(1):45-56.
55. Roy NK, Monisha J, Padmavathi G et al. Isoform-specific role of Akt in oral squamous cell carcinoma. *Biomolecules.* 2019; 9(7):253.
56. Tampa M, Caruntu C, Mitran M et al. Markers of oral lichen planus malignant transformation. *Dis Markers.* 2018; 2018:1959506.
57. Arduino PG, Magliano A, Gambino A et al. Risk of malignant transformation in 3173 subjects with histopathologically confirmed oral lichen planus: a 33-year cohort study in Northern Italy. *Cancers (Basel).* 2021; 13(22):5740.
58. Cassol-Spanemberg J, Rodríguez-de Rivera-Campillo ME, Otero-Rey EM, Estrugo-Devesa A, Jané-Salas E, López-López J. Oral lichen planus and its relationship with systemic diseases. A review of evidence. *J Clin Exp Dent.* 2018; 10(9):e938-e944.
59. van der Meij EH, Schepman KP, Smeele LE et al. A review of the recent literature regarding malignant transformation of oral lichen planus. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 1999; 88(3):307-10.

60. Eisen D, Carrozzo M, Bagan Sebastian JV et al. Number V Oral lichen planus: clinical features and management. *Oral Dis.* 2005; 11(6):338-49.
61. Werneck JT, Costa Tde O, Stibich CA et al. Oral lichen planus: study of 21 cases. *An Bras Dermatol.* 2015; 90(3):321-6.
62. González-Moles MÁ, Aguilar-Ruiz M, Ramos-García P. Challenges in the early diagnosis of oral cancer, evidence gaps and strategies for improvement: a scoping review of systematic reviews. *Cancers (Basel).* 2022; 14(19):4967.
63. Cortés-Ramírez DA, Gainza-Cirauqui ML, Echebarria-Goikouria MA et al. Oral lichenoid disease as a premalignant condition: the controversies and the unknown. *Med Oral Patol Oral Cir Bucal.* 2009; 14(3):E118-22.
64. Melikoglu MA, Melikoglu M, Demirci E et al. Discoid lupus erythematosus-associated cutaneous squamous cell carcinoma in systemic lupus erythematosus. *Eurasian J Med.* 2022; 54(2):204-205.
65. Fernandes MS, Girisha BS, Viswanathan N et al. Discoid lupus erythematosus with squamous cell carcinoma: a case report and review of the literature in Indian patients. *Lupus.* 2015; 24(14):1562-6.
66. Muse ME, Crane JS. Actinic Cheilitis. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022.
67. Medeiros CK, Lopes ML, Silveira ÉJ et al. Actinic cheilitis: Proposal of a clinical index. *Med Oral Patol Oral Cir Bucal.* 2022; 27(4):e310-e318.
68. Dancyger A, Heard V, Huang B et al. Malignant transformation of actinic cheilitis: A systematic review of observational studies. *J Invest Clin Dent.* 2018; 9(4):e12343.
69. Fall-Dickson JM, Pavletic SZ, Mays JW et al. Oral Complications of chronic graft-versus-host disease. *J Natl Cancer Inst Monogr.* 2019; 2019(53):lgz007.
70. Dean D, Sroussi H. Oral Chronic Graft-Versus-Host Disease. *Front Oral Health.* 2022; 3:903154.
71. Mehterov N, Sacconi A, Pulito C et al. A novel panel of clinically relevant miRNAs signature accurately differentiates oral cancer from normal mucosa. *Front Oncol.* 2022; 12:1072579.
72. Harsha C, Banik K, Ang HL et al. Targeting AKT/mTOR in oral cancer: Mechanisms and advances in clinical trials. *Int J Mol Sci.* 2020; 21(9):3285.
73. Gutkind JS, Molinolo AA, Wu X et al. Inhibition of mTOR signaling and clinical activity of metformin in oral premalignant lesions. *JCI Insight.* 2021; 6(17):e147096.
74. Stanciu S, Ionita-Radu F, Stefani C et al. Targeting PI3K/AKT/mTOR signaling pathway in pancreatic cancer: From molecular to clinical aspects. *Int J Mol Sci.* 2022; 23(17):10132.
75. Müller P, Schier AF. Extracellular movement of signaling molecules. *Dev Cell.* 2011; 21(1):145-58.
76. Saxton RA, Sabatini DM. mTOR signaling in growth, metabolism, and disease. *Cell.* 2017; 168(6):960-976.
77. Xie J, Weiskirchen R. What Does the "AKT" Stand for in the Name "AKT Kinase"? Some Historical Comments. *Front Oncol.* 2020; 10:1329.
78. Wu M, Falasca M, Blough ER. Akt/protein kinase B in skeletal muscle physiology and pathology. *J Cell Physiol.* 2011; 226(1):29-36.
79. Manning BD, Toker A. AKT/PKB Signaling: Navigating the Network. *Cell.* 2017; 169(3):381-405.
80. Haddadi N, Lin Y, Travis G et al. PTEN/PTENP1: 'Regulating the regulator of RTK-dependent PI3K/Akt signalling', new targets for cancer therapy. *Mol Cancer.* 2018; 17(1):37.
81. He Y, Sun MM, Zhang GG et al. Targeting PI3K/Akt signal transduction for cancer therapy. *Signal Transduct Target Ther.* 2021; 6(1):425.
82. Mazloumi Gavgani F, Smith Arnesen V, Jacobsen RG et al. Class I phosphoinositide 3-kinase PIK3CA/p110 α and PIK3CB/p110 β isoforms in endometrial cancer. *Int J Mol Sci.* 2018; 19(12):3931.
83. Kotzampasi DM, Premeti K, Papafotika A et al. The orchestrated signaling by PI3K α and PTEN at the membrane interface. *Comput Struct Biotechnol J.* 2022; 20:5607-5621.
84. Singh P, Dar MS, Dar MJ. p110 α and p110 β isoforms of PI3K signaling: are they two sides of the same coin? *FEBS Lett.* 2016; 590(18):3071-82.
85. Brasseur K, Gévry N, Asselin E. Chemoresistance and targeted therapies in ovarian and endometrial cancers. *Oncotarget.* 2017; 8(3):4008-4042.

- 86.Haddadi N, Lin Y, Travis G et al. PTEN/PTENP1: 'Regulating the regulator of RTK-dependent PI3K/Akt signalling', new targets for cancer therapy. *Mol Cancer*. 2018; 17(1):37.
- 87.Wu YT, Tan HL, Huang Q et al. Activation of the PI3K-Akt-mTOR signaling pathway promotes necrotic cell death via suppression of autophagy. *Autophagy*. 2009; 5(6):824-34.
- 88.Yu JS, Cui W. Proliferation, survival and metabolism: the role of PI3K/AKT/mTOR signalling in pluripotency and cell fate determination. *Development*. 2016; 143(17):3050-60.
- 89.Coculescu BI, Manole G, Coculescu EC et al. Autophagy as a neuronal survival mechanism in ischemic stroke. *Rom J Leg Med*. 2018; 26(4):333-339.
- 90.Long HZ, Cheng Y, Zhou ZW et al. PI3K/AKT signal pathway: A target of natural products in the prevention and treatment of Alzheimer's disease and Parkinson's disease. *Front Pharmacol*. 2021; 12:648636.
- 91.Bouyahya A, El Allam A, Aboulaghras S et al. Targeting mTOR as a cancer therapy: recent advances in natural bioactive compounds and immunotherapy. *Cancers (Basel)*. 2022; 14(22):5520.
- 92.Matsuo FS, Andrade MF, Loyola AM et al. Pathologic significance of AKT, mTOR, and GSK3 β proteins in oral squamous cell carcinoma-affected patients. *Virchows Arch*. 2018; 472(6):983-997.
- 93.Martins F, de Sousa SC, Dos Santos E et al. PI3K-AKT-mTOR pathway proteins are differently expressed in oral carcinogenesis. *J Oral Pathol Med*. 2016; 45(10):746-752.
- 94.Massarelli E, Liu DD, Lee JJ et al. Akt activation correlates with adverse outcome in tongue cancer. *Cancer*. 2005; 104(11):2430-6.
- 95.Andrikopoulou M, Salakos N, Deligeoroglou E et al. The role of mTOR signaling pathway in premalignant and malignant cervical lesions. *Eur J Gynaecol Oncol*. 2015; 36(1):36-43.
- 96.Sonis ST, Amaral Mendes R. Could the PI3K canonical pathway be a common link between chronic inflammatory conditions and oral carcinogenesis? *J Oral Pathol Med*. 2016; 45(7):469-74.
- 97.Zhang N, Zhang J, Tan YQ et al. Activated Akt/mTOR-autophagy in local T cells of oral lichen planus. *Int Immunopharmacol*. 2017; 48:84–90.
- 98.Ma RJ, Tan YQ, Zhou G. Aberrant IGF1-PI3K/AKT/MTOR signaling pathway regulates the local immunity of oral lichen planus. *Immunobiology*. 2019; 224(3):455-461.