OSR
ORAL SCIENCES REPORTS

Review Article

Received: April 10, 2025
Revised: June 4, 2025
Accepted: June 6, 2025

Corresponding Author:
Bose Divya, Department of Oral Pathology & Microbiology, SRM Dental College, Bharati Salai, Ramapuram, Chennai-600089, Tamil Nadu, India.
E-mail: divyab.diffy@gmail.com

Oral Sci Rep. 2025; 46(3): 125-137

Novel Therapeutic Strategies for Oral Squamous Cell Carcinoma-An Overview

Mounika S, Bose Divya, Vasanthi V, Madhu Narayan, A. Ramesh Kumar, Rajkumar K

Department of Oral Pathology & Microbiology, SRM Dental College, Bharati Salai, Ramapurum, Tamil Nadu, India

Abstract

Surgery, radiotherapy, and chemotherapy have been the mainstay of oral squamous cell carcinoma (OSCC) management. However, these treatments often come with significant side effects and limitations, including systemic toxicity and drug resistance. Recent advancements in therapeutic strategies for oral cancer have focused on more targeted and less invasive approaches. Some of the novel therapeutic strategies for OSCC include immunotherapy, targeted therapy, photodynamic therapy, gene therapy, phytochemicals, nanotheranostics, and CRISPR/Cas technology. These novel strategies offer promising avenues for improving the outcomes and quality of life for patients with OSCC. Ongoing research and clinical trials are essential to further refine these approaches and make them widely available.

Keywords: oral cancer, recent, squamous cell carcinoma, treatment

Introduction

Oral cancer is a group of malignant diseases arising from the lips, gums, tongue, mouth, and palate and is the most fatal disease among other diseases in the world. Oral squamous cell carcinomas (OSCC), arising from surface epithelium, constitute more than 90% of all oral cancers, and they rank as the sixth most common neoplasm in the world.[1] There are more than 350,000 new cases of OSCC and 177,000 deaths every year, with considerable differences in geographic and environmental risk factors.[2] The incidence of OSCC has been decreasing in some parts of the world but the increase is observed in other countries with low socio-economic conditions. Despite the new management strategies, the 5-year survival rate is below 50%.[3] Low socioeconomic condition is in turn related to factors like nutrition, health care, living condition, lack of awareness, and risk behaviors, which contribute to the development of oral cancer. In many low-income and middle-income countries, including India, most of the population does not have access to a well-organized and well-regulated cancer care system. The high cost of novel therapies and treatments makes them inaccessible to many people in low-income regions. This economic barrier prevents patients from receiving the latest and most effective treatments. Addressing these challenges requires a comprehensive approach, including improving access to affordable therapies, enhancing healthcare infrastructure, and promoting early detection and screening.

Surgical resection is used as a primary treatment modality. Chemotherapy is used as an adjuvant to radiotherapy in patients who are diagnosed with stages 3 and 4, and in patients with local recurrence and metastasis. The continuous increase in oral cancer cases, the failure of conventional chemotherapies, and the excessive toxicity of chemotherapies demand alternative cancer treatments. The current review aimed to summarize the literature on novel therapeutic approaches available for oral cancer. Some of the novel therapeutic strategies for OSCC include immunotherapy, targeted therapy, Photodynamic Therapy, gene therapy, phytochemicals, Nanotheranostics, and CRISPR/Cas Technology. (Table 1)

Immunotherapy

Cancer immunotherapy involves stimulating specific components of the immune system and strengthening the immune system to counteract the signals that suppress them. Strengthening of the immune system is brought about by the cells of innate immunity, such as the neutrophils, macrophages, natural killer (NK) cells, dendritic cells, and eosinophils, and the cells of adaptive immunity, which include the B and T lymphocytes, commonly known as B and T cells. Neutrophils play a key role in cancer immunotherapy, acting as both tumor-promoting and tumor-suppressing agents. They contribute to immune evasion by releasing pro-inflammatory cytokines that support tumor growth, but they also enhance antitumor immunity by activating T cells and dendritic cells. tumor-associated neutrophils (TANs) can be polarized into either pro-tumor (N2) or anti-tumor (N1) phenotypes, influencing immunotherapy outcomes. B cells produce antibodies, and T cells generate CD4+ and CD8+ cells. CD4+ T cells, particularly T helper (Th) cells, support B cell activation and antibody production by releasing cytokines such as IL-4, IL-5, and IL-6. These cytokines enhance B cell proliferation and promote class switching, allowing B cells to produce tumor-targeting antibodies. CD8+ T cells, or cytotoxic T lymphocytes, directly attack cancer cells by recognizing tumor antigens presented via MHC class I molecules. Their activity is often enhanced by B-cell-derived antibodies, which facilitate antibody-dependent cellular cytotoxicity. B cells, beyond antibody production, contribute to antitumor immunity by forming tertiary lymphoid structures within tumors. These structures serve as immune hubs, fostering interactions between T cells and B cells to sustain long-term immune surveillance. The tumor cells escape the cells of the immune system by decreasing surface antigen expression and also alter the environment by synthesizing substances that suppress the immune system and thus increase the progression of the tumors.[4] Immune therapy is divided into two types, namely the active and passive types, where there is direct attack of the tumor cells. These are derived from the blood or tumor of the patient and cultured.

Active immunotherapy involves NK cells, dendritic cells, and cytotoxic T cells. In passive immunotherapy, there is targeting of cell surface receptors to form antibody-dependent cell-mediated immunity. Immunotherapy includes checkpoint inhibitors, targeted monoclonal antibodies, adoptive cell transfer, and cytokine immunotherapy.

Table 1: Comparison table highlighting key aspects of different novel therapeutic strategies.

Therapy	Mechanism	Advantages	Limitations	Response to therapy
Immunotherapy	Restore the ability of the immune system to detect and destroy cancer cells by overcoming the mechanisms by which tumors evade and suppress the immune response	Highly specific	Several factors influence tumor immunotherapy 1) Host immunity 2) Tumor cells 3) Environmental factors	1-year survival rate was ~19% higher with nivolumab (anti-programmed death 1 monoclonal antibody) than with standard therapy in HNSCC (head & neck squamous cell carcinoma).[44]
Targeted cancer stem cell therapy	Specifically targeting cancer stem cells	Selectivity, prevent recurrence	Drug resistance, challenging to identify, cancer stem cells	Preclinical study - CD44v6-specific CAR-NK cell therapy demonstrated a two-to-threefold increase in killing efficacy against various HNSCC cell lines compared to unmodified natural killer cells.[45]
Photodynamic Therapy	Uses light-sensitive drugs activated by specific wavelengths of light to destroy cancer cells	Minimally invasive, localized treatment	Limited penetration depth, requires specialized equipment	77% of patients with oral and oropharyngeal cancer achieved complete response and 42.3% of patients achieved local control.[46]
Gene Therapy	Modifies or replaces defective genes to inhibit cancer growth	Potential for long-term disease control	Ethical concerns, delivery challenges	Phase 1 trial - phIL-12 plasmid delivered via gene electrotransfer potentiates the immunostimulatory effects of local ablative therapies in basal cell carcinomas of the head and neck.[47]
Phytochemicals	Natural compounds with anticancer properties	Low toxicity, easily available	Variable efficacy	Phase I clinical trial- Histological improvements of precancerous lesions in oral cancer patients observed upon treatment with curcumin.[36]
Nanotheranostics	Uses nanoparticles	Enhanced drug delivery, real-time monitoring	Complex formulation, regulatory hurdles	Retrospective study - 77% of patients with metastatic cancer showed manageable tolerability and favorable response rates to nanoparticle albumin-bound paclitaxel (PacliALL™).[48]
CRISPR/Cas Technology	Gene-editing tool to correct mutations or disrupt cancer-promoting genes	High precision, potential for personalized treatment	Ethical concerns, risk of off-target effects	Preclinical study - CD147 gene knockout oral cancer cells, which were shown to decrease Cal27 tumor cell invasion and metastasis.[42]

Checkpoint inhibitor

Checkpoint inhibitors act against the pathways that suppress T cell activity, thus resulting in tumor regression. Evidence shows that immune checkpoints programmed cell death protein 1 (PD-1) promote the immune response against cancer.[5] Cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) creates new T cells and contributes to cancer regression. Apart from PD-1 and CTLA-4, other checkpoint inhibitor receptors such as lymphocyte-activation gene 3 (LAG3), mucin domain 3 (TIM-3), and T-cell immunoglobulin have demonstrated therapeutic effects in clinical trials in combination with PD-1 agents. Pembrolizumab was approved by the FDA for treating patients with recurrent head and neck squamous cell carcinoma (HNSCC).[6]

Targeted monoclonal antibodies

Monoclonal antibodies are either obtained from

human or murine antibody components. Epidermal growth factor receptor (EGFR) plays a major role in tumor progression by causing invasion, metastasis, angiogenic potential, and inhibition of apoptosis. Monoclonal antibodies such as cetuximab and panitumumab are EGFR-targeted therapies that are proven to be effective in oral cancer, either when used alone or along with radiotherapy.[7] P53 is the most commonly mutated gene in cancer, and antibodies against this gene have been useful in treating cases with nodal involvement.[8]

Adoptive cell transfer (ACT)

ACT is a procedure wherein T cells are obtained from the tumor sample or the patients and introduced with specific antigen receptors by genetic engineering and then reintroduced into the patients, therefore enhancing the ability to recognize the antigen. In the study conducted by Jiang *et al*, ACT has improved the survival rates in patients with HNSCC in comparison with the control group.[9] The side effects of ACT include fever, nausea, vomiting, rashes, and cytokine release syndrome, which occurs when the transferred T cells or other immune cells respond to the new T cells.

Cytokine immunotherapy

Cytokines are messengers that allow the cells of the immune system to work hand in hand to target the antigen (cancer cells). This method enhances the coordination of the tumor cells and the stromal cells. They are delivered either locally or systematically to elicit the anti-tumor response. Several cytokines are being explored for the treatment of HNSCC, including GM-CSF, IL-2, IFN-γ, IL-12, and an investigational multi-cytokine biologic known as IRX-2 (Primary cell-derived Biologic). Interleukin-2 enhances the activation of several types of leukocytes with antitumor activity, including natural killer cells, lymphokine-activated killer cells, antigen-specific T-helper cells, cytotoxic lymphocytes, macrophages, and B cells. IFN-γ and IL-12 are used in clinical trials that have been proven to be effective by increasing the number of natural killer cells.[10]

Immunotherapy has transformed cancer treatment, improving the outcome of cancers like melanoma, lung cancer, and renal cell carcinoma. Pembrolizumab resulted in improved progression-free survival of advanced melanoma patients at both 2 mg/kg and 10 mg/kg, with a 6-month progression-free survival of 34% when compared to 16% in the chemotherapy group.[11] When compared to standard chemotherapy, immunotherapy is expected to considerably enhance the overall and progression-free survival of patients with extensive-stage small-cell lung cancer. Overall survival was higher with anti-PD-L1 than with anti-PD-1 and anti-CTLA4.[12]

Patient selection plays a major role in the success of immunotherapy. Several side effects are associated with each approach. Checkpoint Inhibitors can cause immune-related adverse events, including colitis, pneumonitis, and endocrinopathies. Tumors can become resistant to treatment, and only a small subset of individuals respond well. Nivolumab and pembrolizumab have only been trialed for OSCC. The use of targeted monoclonal antibodies is restricted by their high cost, possible off-target effects, limited efficacy in advanced instances, and tumor-induced downregulation of target receptors, which lowers their efficacy. Cetuximab has been tested in combination with radiation therapy for oral cancer. Adoptive Cell Transfer requires complex genetic engineering, and solid tumors like oral cancer pose challenges for T-cell infiltration. Its drawbacks include high toxicity, cytokine release syndrome risk, and expensive production. For head and neck malignancies, CAR-T therapy that targets EGFR is being investigated. Cytokine Immunotherapy has a short half-life, requiring frequent dosing and systemic toxicity leading to severe inflammatory responses, including vascular leak syndrome. IL-12 gene electrotransfer has been tested for head and neck basal cell carcinoma.

Targeted cancer stem cell therapy

Oral cancer stem cells are a subpopulation of cells with tumor-initiating properties and several molecular similarities to embryonic and normal adult stem cells. Oral CSCs are also commonly resistant to conventional therapies targeting proliferating cells. Hence, they crucially contribute to metastasis and recurrence. It is important to increase knowledge of the molecular features and signaling pathways that are specific to the oral cancer stem cells to develop new targeted and efficient treatments for head and neck cancer. The self-renewal potential of cancer stem cells (CSCs) may be obtained through multiple endogenous signaling pathways such as the Wnt, Bmp, Pten, Notch, TGF-β, and Hedgehog. CSCs can be detected and isolated from the tumor mass by employing single or

combinations of numerous surface markers. Despite significant progress in identifying CSCs based on their specific surface markers like Aldehyde dehydrogenases (ALDHs), CD44, CD117, and CD133, the development of selective CSC therapies remains a challenge. ALDH1-positive cell subpopulations have been found to have higher tumorigenicity and are more resistant to chemotherapeutic agents than the negative population.

Adenosine triphosphate Phosphate Binding Cassette (ABC) transporters

ABC transporters are membrane transporters, which are capable of pumping different little molecules (e.g., anti-cancer drugs) out of the cells at the expense of ATP hydrolysis, and thus lead to decreased intracellular drug concentrations.[13] Cancer stem cells following chemo-radiotherapy become chemo-radio resistant or selectively improve the resistant cell population. Studies have found that overexpression of ABC transporters in cancer cells enhances their chemo-radio resistance. Suppressing ABC transporters elevates anti-cancer drug sensitivity in cancer.[14]

Histone demethylases

Epigenetic modifications like as DNA methylation, histone modification, and non-coding RNA regulation play a crucial role in oral cancer development, influencing gene expression, tumor progression, and therapeutic resistance. Hypermethylation of tumor suppressor genes leads to gene silencing, while hypomethylation of oncogenes results in genomic instability and increased tumor aggressiveness. Dysregulated miRNAs like miR-21, miR-200, and miR-34 contribute to tumor growth, metastasis, and drug resistance. Long non-coding RNAs influence epigenetic reprogramming, affecting the tumor microenvironment. Circular RNAs act as miRNA sponges, modulating cancer-related pathways. Histone Modifications include acetylation, phosphorylation, ubiquitination, and methylation. Histone methylation plays a key role in the regulation of gene expression and genetic stability, and dysregulation of this highly conserved process occurs in various cancers. There are two classes of enzymes involved in histone methylation: methyltransferases and demethylases. Histone demethylases are capable of removing methyl groups from histones and other proteins. The histone demethylases contribute to carcinogenesis, cell fate selection, and cell differentiation. The common histone demethylases are the JARID1, KDM4, LSD1, KDM6B, KDM6A, KMD3, KDM5. Jumonji domain–consisting of protein 6 (JMJD6) is a new molecular modulator of oral cancer stem cells.[15] JMJD6 overexpression increases both the CSC traits and the number of CSCs, which suggests that JMJD6 is a prominent modulator of the cancer stem cell phenotype and genesis in OSCC.

Calcium channels

Calcium is a global messenger in regulating several physiological processes and disruption of its homeostasis would be observed during carcinogenesis, which results in the deregulation of the rapid growth of the cells, emigration, and apoptosis inhibition. Studies have recently explored the role of calcium signaling in oral cancer.[16] Orai1, a calcium channel protein, enhances OSCC metastatic potentials, and suppression of Orai1 in the OSCC cell line resulted in suppressing CSC traits.[17]

Targeting these cancer stem cells would enhance efficacy and specificity for eradicating the tumors and reducing systemic toxicity. A better understanding of CSCs can provide unique opportunities to develop new therapeutic platforms for targeting CSCs in the treatment of cancers. They can significantly reduce side effects compared to traditional chemotherapy by specifically targeting CSCs. While targeted therapies can be highly effective, they may not work for all patients due to genetic variability. Additionally, cancer cells can develop resistance to these drugs over time.

Gene therapy

Gene therapy involves the transfer of a therapeutic gene into specific cells of an individual to repair a faulty gene. The objective of this method is to introduce new genetic material into target cells without causing any damage to the surrounding tissues. This alternate treatment option has been proven to increase the survival rates of OSCC patients.[18] The types of gene therapy include somatic and germ-line gene therapy. In somatic gene therapy, the therapeutic genes are introduced into the somatic cells, which restricts the effects of the individual and are not passed on to the next generation and in germ line gene therapy either the sperm or egg can be altered by introducing the therapeutic gene, which gets integrated into the genome.[19] The therapeutic genes are carried out

with the help of either viral or non-viral vectors.

The following are the techniques employed in gene therapy

Gene addition therapy

This approach adds a working copy of a gene into the cell. In this technique, the tumor growth is controlled by the introduction of tumor suppressor genes, which inactivate the carcinogenic cells. Genetic alterations in head and neck cancers include mutations of p53, the Retinoblastoma Gene, p16, and p21. In this method, tumor growth is controlled by the induction of tumor suppressor genes that inactivate the carcinogenic cells. P53 is the most commonly used gene with adenovirus as a viral vector. Studies are being carried out on adenovirus vector Ad5CMV-p53, which is first given by intramucosal injection, followed 2 h later by a mouthwash. From the next day, it is administered as a mouthwash twice daily for 2-5 days, and this treatment is repeated every 28 days. This method inhibits disease progression in precancerous lesions with no toxic effects.[20]

Gene excision therapy

In this technique, the defective oncogenes are removed, as a result of which, there is an inhibition in the growth of the tumor cells. The genes that control growth and cell cycle progression, including factors like TGF-α1, PDGF-α, and PTEN, are regulated by the expression of the transcription factor early growth response-1 (EGR-1). Thus, inhibiting this protein represents a good therapeutic approach for the tumor cells. Some studies demonstrated that inhibition of the protein kinase C reduces the expression of this gene, triggering higher sensitivity of the tumor to radiotherapy.[21]

Antisense RNA therapy

This method involves the introduction of the remedial gene that prevents the expression of a defective gene and is called "Antisense therapy." Gene expression can be inhibited by the RNA that is complementary to the strand of DNA expressing the gene. This technique can be directed towards carcinoma cells whose malignant phenotype is dependent upon the expression of particular oncogenes such as Myc, Fos, and Ras. Inhibition of the expression of these oncogenes may alter the phenotype, thus preventing the growth of the tumor.[22]

Patients with OSCC show defective function of several types of immune cells, which include natural killer cells, T-lymphocytes, and cytokines. The combined use of mIL-2 (murine interleukin 2) and mIL-12 (murine interleukin 12) gene therapy resulted in a significant reduction in the tumor due to increased activation of cytolytic T lymphocytes and natural killer cells. Radiosensitivity to γ radiation and chemosensitivity to 5-fluorouracil (5-FU) in oral squamous cell carcinoma can be enhanced after the suppression of NF-κB activity, which activates the anti-apoptotic proteins TNF, TRAF-1, TRAF-2, and cIAP-1.

The inhibition of NF-κB can decrease the expression of proinflammatory cytokines, e.g. IL-1α, IL-6, and IL-8, and of enzymes that degrade matrix metalloproteinase-9 (MMP-9). The progression and metastasis of OSCC can be prevented by inhibiting NF-κB activity, which may be a useful coadjuvant treatment in oral cancer therapy. Systemic administration of Anti-ICAM 2 induced the complete regression of OSCC. ICAM-2 is a glycosylated protein with surface adhesion that is expressed in endothelial cells and activated lymphocytes.[23]

Suicide gene therapy

It is also called gene-directed enzyme prodrug therapy. Suicide gene therapy introduces viral or bacterial genes into malignant cells that metabolize a non-toxic prodrug into a toxic compound. Suicide gene systems identified include the HSV-thymidine kinase gene (HSV-TK) with ganciclovir (GCV) and the cytosine deaminase gene (CD) with 5-fluorocytosine (5-FC). A Thymidine kinase gene of Herpes Simplex Virus (HSV) transforms ganciclovir into ganciclovir phosphate. Gene transfer of the HSVtk gene (Herpes simplex virus thymidine kinase gene) via adenovirus vector in combination with ganciclovir administration may be a good therapeutic option for OSCC. HSV-tk/GCV therapy in cultured oral squamous cancer cells has shown that tumor cell death occurs mainly by an apoptotic process, and the observed high cytotoxicity is due to the bystander effect, which is promoted by the diffusion of the toxic agent into neighboring cells via gap junctions.[24]

Both precise genetic changes and efficient delivery systems are necessary for targeted gene therapy to be effective. To overcome delivery-related challenges, several techniques have been developed, such as lipid nanoparti-

cles, exosomes, and viral vectors.

Gene therapy with the use of an oncolytic virus

In this method, a vector (virus) is genetically modified, which is replicated and causes lysis of the tumor cells. Adenovirus is the only vector to complete a phase III clinical trial study based on Herpes Simplex Virus thymidine kinase (HSV-tk) suicide gene therapy.[25] The methods of non-viral gene therapy include the injection of naked DNA, electroporation, the gene gun, and the use of oligonucleotides, dendrimers, and inorganic nanoparticles. HSV-tk/GCV system to achieve antitumor activity against oral cancer cells *in vitro* and *in vivo* using ligand-associated lipoplexes to enhance therapeutic delivery.[26]

Gene therapy is effective in cases of single-gene defects, but it also requires multiple visits by the patients, and the vectors can cause side effects. The use of insertional vectors to identify oncogenes by causing leukemia and solid tumors has raised concerns about insertional mutagenesis using the same vectors for gene therapy.

Photodynamic therapy

Photodynamic therapy (PDT) is used in the treatment of cancers due to its specificity and sensitivity to tumor cells. The antitumor effects of PDT may result directly from tumor cell death or indirectly from damage to tumor vasculature and activation of nonspecific and specific immune responses against the tumor cells. Due to its location and direct visibility, the oral cavity is an ideal model for conventional PDT.

Mechanism of action

PDT can activate the immune system in cancer treatment by inducing immunogenic cell death and releasing tumor-associated antigens, which in turn stimulate immune cells to target and eliminate cancer cells. A more detailed flowchart of the effect of PDT for immune activation in cancer is represented in Figure 1.

In PDT, a photosensitizer is administered, which accumulates in or around cancer cells. It is activated by exposure to a specific wavelength of light. The activated photosensitizer generates reactive oxygen species, causing damage to cancer cells and surrounding tissues. PDT-induced damage leads to cancer cell death, specifically immunogenic cell death. There is release of damage-associated molecular patterns (DAMPs), such as high-mobility group box 1 protein and heat shock proteins. DAMPs and tumor-associated antigens (TAAs) are activated and presented to antigen-presenting cells, such as dendritic cells (DCs). DCs present TAAs to T cells, leading to T cell activation and differentiation. Immune cells produce cytokines, such as TNF-α and IFN-γ, which further enhance the anti-tumor immune response to eliminate tumor cells. PDT can be combined with immunotherapy, such as checkpoint inhibitors, to further enhance the anti-tumor immune response.

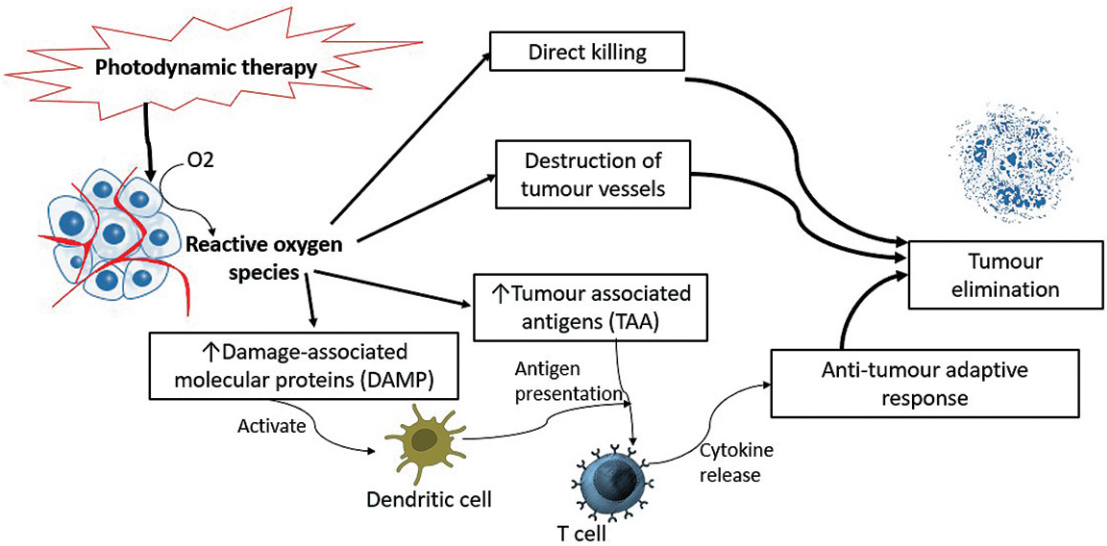

Figure 1: Mechanism of action of photodynamic therapy in tumour elimination.

Role in primary treatment

PDT is well-suited as a primary or alternative treatment modality for early oral cancer without nodal metastasis (i.e., T1 and T2 tumors) and is associated with significantly less morbidity compared to conventional therapy. Superficial cancers that are within the permeability range of the light source (i.e., 0.5-1 cm) show the best response according to studies. The advantages of PDT over conventional therapies, such as surgery, radiotherapy, and chemotherapy include minimal invasiveness, organ-sparing potential, excellent long-term functional and cosmetic results with improved quality of life, feasibility of repeating treatment at the same site for recurring lesions, minimal scarring after treatment, cost-effectiveness, and simplicity of technique.[27]

In the case of any relapse or development of a new primary tumor in the area previously treated by PDT, treatment can be repeated in the same area multiple times without cumulative toxicity, in contrast to ionizing radiation or surgery, where such retreatment results in extensive morbidity. In addition, the use of conventional therapies does not preclude the use of PDT, and the use of PDT does not compromise future surgical interventions or radiation therapy.

Role of PDT as an adjuvant or combination therapy

PDT has the advantage of being used as an adjuvant therapy for the treatment of surgical margins following resection of T3 and T4 head and neck cancers via superficial or interstitial light application. The use of PDT in combination with conventional therapies requires further investigation. PDT may be used to treat primary localized lesions alongside surgery and/or radiotherapy in cases that involve nodal metastasis. Recent *in vitro* and *in vivo* studies have utilized PDT in combination with other chemopreventive agents.[28] These combination therapies have shown enhanced anticancer effects, likely because a multifactorial disease such as cancer involves various pathological pathways. Therefore, a combination of treatment modalities can be used to target different disease processes, causing cell death via diverse mechanisms. Furthermore, the combination of different modalities can synergistically enhance selectivity and efficacy in comparison to either of the single therapies. This can eventually help to reduce the amount of cytotoxic drugs given to patients, resulting in reduced morbidity from side effects.[29]

Role of PDT in surveillance for cancer-free status

Numerous *in vitro* and *in vivo* studies have demonstrated the significant effect of PDT on the development of adaptive immunity. Dendritic cells (DCs) are considered the most important antigen-presenting cells and play a significant role in antitumor immune response by activation of CD8+ cytotoxic T-cells. DCs activated in response to PDT travel to tumor-draining lymph nodes, where they are known to stimulate T-cell activation. Saji *et al*.,[30] studied the combination of PDT with intratumoral injection of DCs and found synergistically enhanced tumor cure rates as compared to individual therapies. The immune response produced by the combination therapy was found to confer systemic antitumor effects that could induce the regression of distant untreated tumors

Recent developments have been made in PDT to ensure the specificity and efficacy of the method. It involves the following methods:

Targeted delivery

In the era of precision medicine, to increase specificity and minimize toxicity to normal tissues, photosensitizers are conjugated with targeting moieties. One such technique is to couple PSs to monoclonal antibodies that are directed against tumor-associated antigens (TAAs) specifically over-expressed in cancer cells. These TAAs include oncofetal antigens, growth factor receptors, and receptors for signal transduction pathways.

For cancers and precancers of the head and neck, EGFR overexpression has been frequently reported.[31] Thus, targeting EGFR with photoactive molecules linked to anti-EGFR antibodies may selectively destroy cancer cells whilst sparing adjacent normal cells expressing low levels of EGFR.

Vascular-targeted PDT

Vascular targeted PDT involves the use of PSs, such as TOOKAD (Steba Biotech, Luxembourg City, Luxembourg) and Visudyne (QLT Ophthalmics, Inc., Vancouver, BC, Canada), which are selectively retained in the neovasculature of targeted tumors, resulting in a preferential vascular response.[32] In comparison to conventional anti-cancer therapies directed against cancer cells, targeting tumor vasculature appears to be a more efficient

approach to killing cancer cells and has a lower probability of developing drug resistance.

Two-photon PDT

In two-photon (2-γ) PDT, ultrafast pulses of near-infrared light are used such that two photons of relatively low but identical energy are simultaneously absorbed by the PS. As each photon contributes to one-half of the excitation energy, a longer wavelength is needed for enough energy to produce singlet oxygen, allowing light to penetrate deeper into tissue due to lower scattering and absorption. Two-photon excitation has been utilized to target and selectively occlude blood vessels associated with neoplastic tissues while reducing the damage to adjacent normal tissues.

Clinical research suggests that PDT is a primary or alternate therapy option for early oral cancer without nodal metastases (i.e., T1 and T2 tumors) and is associated with lower morbidity than traditional treatment. In a study by Toratani S *et al*, superficial OSCC, when treated with Photofrin-mediated PDT, the wounds exhibited excellent functionality and aesthetic healing without any scar formation and with minimal side effects.[33]

PDT is minimally invasive and can be used as a primary treatment for early-stage cancers or in combination with other therapies. However, it is generally limited to treating superficial or localized tumors and may not be effective for larger or deeply infiltrating tumors. Light sensitivity and temporary side effects can also occur.

Phytochemicals

Natural foods, like cruciferous vegetables (e.g., cabbage and broccoli), alliums (e.g., garlic and onion), green tea, citrus fruits, soybeans, tomatoes, berries, and ginger, have chemopreventive activity.[34] Studies have suggested that lifestyle changes could prevent more than two-thirds of human cancers and that dietary factors contribute to approximately 35% of human cancer mortality.[35]

Mechanism of action:

• Free radicals are thought to be related to multistage carcinogenic processes. Peroxyl radicals and lipid peroxidation can independently cause DNA mutations, which are essential for the initiation of the carcinogenic process. Antioxidant phytochemicals can regulate the initiation of carcinogenic processes by protecting against DNA damage.

• Phytochemicals may exert their chemopreventive properties by blocking the critical events of tumor initiation and promotion, thereby reversing the premalignant stage.

• Phytochemicals can induce cancer cell death.

• Phytochemicals can also enhance innate immune surveillance and improve the elimination of transformed cells

Phytochemicals, such as flavonoids, phenolic acids, phytosterols, carotenoids, and stilbenes, have anticancer activity. Phytochemicals that have been studied for their effects on oral cancer include Curcumin, Green Tea Extract, Resveratrol, Isothiocyanates, Lycopene, and Genistein. Several *in vitro* and *in vivo* studies have concluded their anti-cancer effects. Preclinical and clinical studies are required to confirm their efficacy in oral cancer therapeutics. The cancer-preventive effect of curcumin has, however, been demonstrated in a Phase I clinical trial.[36] A Phase IIb clinical trial has shown the efficacy of curcumin (3.6 g for six months) in the treatment of oral leukoplakia.[37] It is also evident that curcumin is effective in delaying the onset and reducing the severity of radiation-induced oral mucositis in patients with head and neck cancer. Natural compounds are often hindered by low water solubility, low bioavailability, and deficient targeting; thus, numerous phytochemical delivery systems should be developed to compensate for these problems. More research is needed to fully understand their mechanisms and to develop effective delivery systems to enhance their bioavailability and therapeutic efficacy

Nanothernostics

Nanotherapeutics is the development of various nanomedicine strategies such as polymer conjugations, dendrimers, micelles, liposomes, metal and inorganic nanoparticles, and carbon nanotubes, nanoparticles of biodegradable polymers for sustained, controlled, and targeted co-delivery of diagnostic and therapeutic agents. This concept will have fewer side effects, which is essential for any therapy. Theranostic nanomedicine involves using colloidal nanoparticles in the range of 1 to 1000 nm (1 μm). They consist of absorbed, conjugated, entrapped macromolecular materials/polymers/carbon nanomaterials/metals and inorganic nanoparticles in which the diagnostic and therapeutic agents are absorbed.

A multifunctional nano-system for imaging-guided cancer treatment by Gu M *et al*, in which the terbium ion-doped hydroxyapatite nanoparticle was used as a luminescent probe to encapsulate both the near-infrared photothermal agent polydopamine (PDA) and anticancer doxorubicin. It promoted *in vitro* cell death through the overproduction of reactive oxygen species, cell cycle arrest, and increased cell apoptosis.[38]

In advanced theranostic nanomedicines, when conjugated with a targeting moiety will recognize specific targets, which then bind to specific receptors on the targeted cell membrane and be internalized by the diseased cells. It happens with most specific processes called-Receptor-Mediated Endocytosis.[39]

Erlotinib encapsulated in liposomal formulations showed significant anti-tumor effects against oral cancer when locally administered at the site of the tumor arising in the oral cavity.[40] The clinical trials that are completed or still ongoing in phase 1 or phase 2 are designed to evaluate the nanoparticles application in the treatment and diagnosis of head and neck cancer. None of the trials reached phase 3. It is observed that paclitaxel-albumin nanoparticles are widely studied for radiation therapy in most clinical trials. Cetuximab, cisplatin, and Hafnium Oxide nanoparticles are also assessed for their use in Head and neck cancer. The complexity of designing and manufacturing nanoparticles can be a limitation. There are also potential concerns about the long-term safety and toxicity of these materials. The major challenge is that the nanoparticles are rapidly eliminated or encountered by the immune system of a patient. Further studies are needed for clinical translation of nanotheranostics and their use in treating oral cancer.

CRISPR/Cas technology

CRISPR/Cas (molecular scissors) is a gene-editing tool that can modify or delete specific genes associated with cancer. By employing genome-wide CRISPR screens, vulnerabilities in oral cancer cells can be identified, revealing promising targets for therapeutic interventions.[41] Using the CRISPR/Cas9 gene-editing technology, Pan S *et al*, produced CD147 gene knockout oral cancer cells, which were shown to decrease Cal27 tumor cell invasion and metastasis *in vitro* and *in vivo*.[42] CRISPR/Cas9 technology has also been used to knock out p75NTR, which inhibited the proliferation, invasion, and migration of SCC-9 cells, suggesting that p75NTR is a viable target for tongue cancer therapy.[43]

CRISPR/Cas9 enables genome-wide screens to identify vulnerabilities in oral cancer cells, revealing promising targets for therapeutic interventions. CRISPR/Cas9 can also be used to perturb genes associated with drug resistance, thereby enhancing the efficacy of chemotherapy and other treatments. It can be combined with other therapeutic strategies, such as immunotherapy, to improve treatment outcomes.

While promising, CRISPR/Cas technology is still in its early stages and faces ethical and regulatory challenges. Off-target effects, where unintended genes are edited, are also a concern.

Limitations and future perspectives

Despite recent advancements in cancer diagnosis and treatment, the survival rate remains poor. Management of cancer should have a multidisciplinary approach and should include a team of health professionals such as surgeons, oncologists, radiologists, dental surgeons, nutritionists, and rehabilitation and reconstructive specialists. This is to support the overall oral and systemic health to improve the quality of life in survivors. A careful watch on patients is also necessary to prevent the risk of recurrences and the development of secondary tumors. Furthermore, each patient necessitates different treatment approaches depending on the type of cancer, stage of cancer, metastasis, lymph node involvement, age, and general systemic health. Early-stage cancers are mostly treated with a single treatment modality, like surgery or radiotherapy, while advanced cases are treated with a combination of two or more modalities. Patients with advanced stages of cancer often end up with extensive surgeries leading to facial disfigurement and often need facial prostheses with adjuvant therapies that assist with speech, mastication, saliva production, and psychological status. Survival following a diagnosis of OCs remains poor, with 50% of patients living up to and not beyond 5 years. Preventive measures like abstinence or avoidance of risk factors and screening at regular intervals must be enforced to control the disease in the early stages. Management of cancer has come a long way with the advent of new treatment modalities directed toward improving the survival rate. At the same time, efforts to improvise a disease-free interval and improve the quality of living of

cancer patients are of utmost importance.

Conclusion

• Immunotherapy offers a targeted and potentially less invasive option for treating head and neck cancer. Their use in oral cancer is still in pre-clinical trials.

• There are currently no clinically applied therapeutic trials to specifically target oral CSCs, and the different pluripotency-associated CSC surface markers are not specific to oral CSCs since they overlap with both normal somatic cells and their tissue-resident stem cells.

• Gene therapy has shown a positive effect in the treatment of oral cancer and prevention of invasion, metastasis, and recurrence in both *in vivo* and *in vitro* settings.

• Clinical studies have demonstrated the effectiveness of PDT as a primary treatment in early-stage oral cancers and as an adjuvant therapy for more advanced cases.

• Phytochemicals show great promise; further research and clinical trials are needed to fully understand their mechanisms of action and optimize their use in oral cancer therapy.

• Nanomaterials can be used as a drug carrier, the drug's action time of which is limited. This may cause toxicity. A targeted drug delivery system may help to improve the efficacy of treatment with nanoparticles.

• CRISPR/Cas9 holds great potential in oral cancer therapy; ongoing research is essential to address challenges such as off-target effects, efficient delivery mechanisms, and ethical considerations

Acknowledgments

None.

Funding

The authors declare that they received no funding for this work.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

1. Dhanuthai K, Rojanawatsirivej S, Thosaporn W, Kintarak S, Subarnbhesaj A, Darling M, *et al*. Oral cancer: a multicenter study. Med Oral Patol Oral Cir Bucal. 2018;23(1);e23-e29.
2. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, *et al*. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021;71(3): 209-49.
3. Du M, Nair R, Jamieson L, Liu Z, Bi P. Incidence trends of lip, oral cavity, and pharyngeal cancers: global burden of disease 1990-2017. J Dent Res. 2020;99(2):143-51.
4. Ljunggren HG, Jonsson R, Höglund P. Sem-inal immunologic discoveries with direct clinical implications: the 2018 nobel prize in physiology or medicine honours discoveries in cancer immunotherapy. Scand J Immunol 2018;88(6):e12731.
5. Messenheimer DJ, Jensen SM, Afentoulis ME, Wegmann KW, Feng Z, Friedman DJ, *et al*. Timing of PD-1 blockade is critical to effective combination immunotherapy with anti-OX40. Clin Cancer Res. 2017;23(20):6165-77.
6. Larkin J, Chiarion-Sileni V, Gonzalez R, Grob JJ, Cowey CL, Lao CD, *et al*. Combined nivolumab and ipilimumab or monotherapy in previously untreated melanoma. N Engl J Med. 2015;373(1):23-34.
7. Schlessinger J. Cell signaling by receptor tyrosine kinases. Cell. 2000;103(2):211-25.
8. Normanno N, De Luca A, Bianco C, Strizzi L, Mancino M, Maiello MR, *et al*. Epidermal growth factor receptor (EGFR) signaling in cancer. Gene. 2006;366(1):2-16.
9. Jiang P, Zhang Y, Archibald SJ, Wang H. Adoptive cell transfer after chemotherapy enhances survival in patients with resectable HNSCC. Int Immunopharmacol. 2015;28(1): 208-14.
10. Rapidis AD, Wolf GT. Immunotherapy of head and neck cancer: current and future considerations. J Oncol. 2009; 2009:346345. doi: 10.1155/2009/346345.
11. Knight A, Karapetyan L, Kirkwood JM. Immunotherapy in melanoma: recent advances and future directions. Cancers (Basel). 2023;15(4):1106.
12. Liu X, Xing H, Zhang H, Liu H, Chen J. Immunotherapy versus standard chemotherapy for treatment of extensive-stage small-cell lung cancer: a systematic review. Immunotherapy. 2021;13(12):989-1000.
13. Aadithi MG, Divya B, Nandhini G, Rajkumar K, Ramesh Kumar A, Sarangarajan R. Evaluation of ABCB5 immunostained epithelial stem cells in oral squamous cell carcinoma, inflammatory gingival hyperplasia and normal mucosa. Biotech Histochem. 2024;99(1):44-8.
14. Cui H, Zhang JA, Chen Chen, Liu JJ. ABC transporter inhibitors in reversing multidrug resistance to chemotherapy. Curr Drug Targets. 2015;16(12):1356-71.
15. Lee CR, Lee SH, Rigas NK, Kim RH, Kang MK, Park NH, *et al*. Elevated expression of JMJD6 is associated with oral carcinogenesis and maintains cancer stemness properties.

Carcinogenesis. 2016;37(2):119-28
16. Lee YF, Miller LD, Chan XB, Black MA, Pang B, Ong CW, *et al*. JMJD6 is a driver of cellular proliferation and motility and a marker of poor prognosis in breast cancer. Breast Cancer Res. 2012;14(3):R85. doi: 10.1186/bcr3200.
17. Baniebrahimi G, Mir F, Khanmohammadi R. Cancer stem cells and oral cancer: insights into molecular mechanisms and therapeutic approaches. Cancer Cell Int. 2020;20:113. doi: 10.1186/s12935-020-01192-0.
18. Ayllón Barbellido S, Campo Trapero J, Cano Sánchez J, Perea García MA, Escudero Castaño N, Bascones Martínez A. Gene therapy in the management of oral cancer: review of the literature. Med Oral Patol Oral Cir Bucal. 2008;13(1):E15-21.
19. Indu S, Ramesh V, Oza N, Prashad KV. Gene therapy: An overview. J Orofac Sci. 2013;5(2):83-7.
20. Saraswathi TR, Kavitha B, Vijayashree Priyadharsini J. Gene therapy for oral squamous cell carcinoma: an overview. Indian J Dent Res. 2007;18(3):120-3.
21. Madalli Vijaylaxmi B, Basavaraddi Shrinivas M, Sattur A, Burde K. Gene therapy for oral squamous cell carcinoma-a promising future. Int J Inf Res Rev. 2014;1:220-2.
22. Jacob KC, Yashoda R, Puranik MP. Gene therapy in the treatment and prevention of oral cancer: an overview. Int J Adv Health Sci. 2015;2(2):14-20.
23. Sunil PM, Joseph I, Susan Varghese S. Gene therapy in oral squamous cell carcinoma - a short review. Oral Maxillofac Pathol J. 2011;2(2):142-147.
24. Lakshmi LS, Radhika T, Jeddy N. Gene therapy in oral cancer–an update J Dr NTR Univ Health Sci. 2020;9(1):1-5.
25. Westphal M, Ylä-Herttuala S, Martin J, Warnke P, Menei P, Eckland D, *et al*. Adenovirus-mediated gene therapy with sitimagene ceradenovec followed by intravenous ganciclovir for patients with operable high-grade glioma (ASPECT): a randomised, open-label, phase 3 trial. Lancet Oncol. 2013;14(9):823-33.
26. Faneca H, Düzgüne¸s N, de Lima MP. Suicide gene therapy for oral squamous cell carcinoma. Methods Mol Biol. 2019;1895:43-55.
27. Tan IB, Dolivet G, Ceruse P, Vander Poorten V, Roest G, Rauschning W. Temoporfin-mediated photodynamic therapy in patients with advanced, incurable head and neck cancer: a multicenter study. Head Neck. 2010;32(12):1597-604.
28. Kim SG, Hong JW, Boo SH, Kim MG, Lee KD, Ahn JC, *et al*. Combination treatment of cetuximab and photodynamic therapy in SNU-1041 squamous cancer cell line. Oncol Rep. 2009;22(4):701-8.
29. Datta SN, Allman R, Loh C, Mason M, Matthews PN. Effect of photodynamic therapy in combination with mitomycin c on a mitomycin-resistant bladder cancer cell line. Br J Cancer. 1997;76(3):312-7.
30. Saji H, Song W, Furumoto K, Kato H, Engleman EG. Systemic antitumor effect of intratumoral injection of dendritic cells in combination with local photodynamic therapy. Clin. Cancer Res. 2006;12(8):2568-74.
31. Ke LD, Adler-Storthz K, Clayman GL, Yung AW, Chen Z. Differential expression of epidermal growth factor receptor in human head and neck cancers. Head Neck.1998;20(4): 320-7.
32. Chen B, Pogue BW, Hoopes PJ, Hasan T. Vascular and cellular targeting for photodynamic therapy. Crit Rev Eukaryot Gene Expr. 2006;16(4):279-305.
33. Toratani S, Tani R, Kanda T, Koizumi K, Yoshioka Y, Okamoto T. Photodynamic therapy using photofrin and excimer dye laser treatment for superficial oral squamous cell carcinomas with long-term follow up. Photodiagnosis Photodyn Ther. 2016;14:104-10.
34. Gullett NP, Ruhul Amin AR, Bayraktar S, Pezzuto JM, Shin DM, Khuri FR, *et al*. Cancer prevention with natural compounds. Semin Oncol. 2010;37(3):258-81.
35. Zhang YJ, Gan RY, Li S, Zhou Y, Li AN, Xu DP, *et al*. Antioxidant phytochemicals for the prevention and treatment of chronic diseases. Molecules. 2015;20(12):21138-56.
36. Cheng AL, Hsu CH, Lin JK, Hsu MM, Ho YF, Shen TS, *et al*. Phase I clinical trial of curcumin, a chemopreventive agent, in patients with high-risk or pre-malignant lesions. Anticancer Res. 2001;21(4B):2895-900.
37. Kuriakose MA, Ramdas K, Dey B, Iyer S, Rajan G, Elango KK, *et al*. A randomized double-blind placebo-controlled phase IIB trial of curcumin in oral leukoplakia. Cancer Prev Res (Phila). 2016;9(8):683-91.
38. Gu M, Jiang L, Hao L, Lu J, Liu Z, Lei Z, *et al*. A novel theranostic nanoplatform for imaging-guided chemo-photothermal therapy in oral squamous cell carcinoma. J Mater Chem B. 2021;9(30):6006-16.
39. Mahesh KP. Nanotheranostics-novel modality for integrating diagnosis and therapy for oral cancer. Acta Sci Dent Sci. 2018;2(9):57-8.
40. Hariharan K, Mehta T, Shah J, Dave H, Sami A, Omri A. Localized delivery of Erlotinib using liposomal gel formulations for the treatment of oral squamous cell carcinoma. Int J Pharm. 2023;642:123144.
41. Sowmya SV, Augustine D, Mushtaq S, Baeshen HA, Ashi H, Hassan RN, *et al*. Revitalizing oral cancer research: Crispr-Cas9 technology the promise of genetic editing. Front Oncol. 2024;14:1383062.
42. Pan S, Su Y, Sun B, Hao R, Gao X, Han B. Knockout of CD147 inhibits the proliferation, invasion, and drug resistance of human oral cancer CAL27 cells *in vitro* and *in vivo*. Int J Biol Macromol. 2021;181:378-89.
43. Huang P, Tong D, Sun J, Li Q, Zhang F. Generation and characterization of a human oral squamous carcinoma cell line SCC-9 with CRISPR/Cas9-mediated deletion of the p75 neurotrophin receptor. Arch Oral Bio. 2017;82:223-32.
44. Ferris RL, Blumenschein G Jr, Fayette J, Guigay J, Colevas AD, Licitra L, *et al*. Nivolumab for recurrent

squamous-cell carcinoma of the head and neck. N Engl J Med. 2016;375(19):1856-67.
45. Ciulean IS, Fischer J, Quaiser A, Bach C, Abken H, Tretbar US, *et al*. 250 Harnessing anti-CD44v6 CAR-NK cells for effective head and neck squamous cell carcinoma treatment. J Immunotherap Cancer. 2024;12(Suppl 2):288.
46. Lambert A, Nees L, Nuyts S, Clement P, Meulemans J, Delaere P, *et al*. Photodynamic therapy as an alternative therapeutic tool in functionally inoperable oral and oropharyngeal carcinoma: a single tertiary center retrospective cohort analysis. Front Oncol. 2021;11:626394.
47. Strojan P, Jesenko T, Omerzel M, Jamsek C, Groselj A, Tratar UL, *et al*. Phase I trial of phIL12 plasmid intratumoral gene electrotransfer in patients with basal cell carcinoma in head and neck region. Eur J Surg Oncol. 2025;51(5):109574.
48. Talreja V, Khetwani S, Nanadagopal E, Borkar NE, Khobragade K. Real-world study on the effect of nab-paclitaxel treatment on clinical outcomes and laboratory parameters in patients across metastatic tumor sites. IJMIO. 2024:9(2):46-52.