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REVIEW ARTICLE

3D BIOPRINTING FOR PRECISION TUMOR MODELS IN ORAL CANCER RESEARCH: A SYSTEMATIC REVIEW AND META-ANALYSIS

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Abstract

Background: The production of extremely precise and adaptable tissue models made possible by 3D printing has completely changed the field of biomedical research. With the use of this technology, especially 3D bioprinting with bioinks, it is now possible to replicate intricate tissue architectures for use in drug testing and regenerative medicine.
Aim: Evaluating developments in 3D bioprinting to produce accurate models of oral cancer tumors, emphasizing the contribution of bioinks to improving research efficacy, detection, and treatment.
Methodology: This systematic review examined the effects of 3D bioprinting and bioinks on the creation of accurate, customized tumor models for study on oral cancer, adhering to PRISMA principles. Upon doing an extensive search across PubMed, Scopus, Web of Science, Cochrane Library, and Google Scholar, 296 publications published between 2019 and 2024 were found. Three of the five chosen articles met the requirements for a meta-analysis when inclusion and exclusion were applied. The review evaluated the accuracy, therapeutic efficacy, and individualized treatment potentials of 3D bioprinted models versus conventional 2D cell cultures in investigations on oral cancer. Three reviewers extracted data and evaluated its quality to guarantee consensus and dependability. They used the Cochrane Risk of Bias Tool, ROBINS-I, and QUIN Tool for bias assessment.
Results: The systematic review emphasizes the precision with which 3D bioprinting can create tumor models and highlights its advancements for cancer research. Certain malignancies have been the focus of techniques like SLS, Inkjet 3DP, SLA, and FDM; high-quality studies have received scores of up to 11 on the QUIN Tool. Blinding and result details are two areas that require improvement, according to the quality assessment. 2 studies had low risk of bias according to the ROBINS-I assessment, but 1 study had a moderate level of concern because of confounding circumstances.
Conclusion: By producing more precise and useful tumor models, 3D bioprinting is transforming the field of cancer research. Drug testing and tailored medicine techniques are improved by these developments. For this technology to be properly utilized in clinical practice, future research should concentrate on standardizing bioprinting processes and addressing ethical issues.

Key-words: *Biomaterials; Bioprinting; Bioinks; Cell Culture Techniques; Inkjet 3D Printing; Tumor microenvironment*

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Introduction

Oral cancer continues to be a significant worldwide health problem, causing a substantial amount of illness and death. Siegel and colleagues found in 2020 that oral cancer remains difficult to diagnose and treat, as indicated by cancer statistics. The disease mainly affects the oral cavity and throat, frequently appearing in later stages because it has no symptoms in the beginning. The delayed discovery leads to relatively unfavorable prognosis and survival rates for patients. Additionally, treatment strategies are complicated by the intricate structure of the oral anatomy and the diverse nature of tumor cells. Developing models that accurately replicate the conditions of the tumor microenvironment (TME) is essential for studying the progression and therapeutic responses of oral cancers.[1,2]

Role of 3D printing in cancer research

The innovation of 3D printing technology has transformed cancer research by allowing the development of extremely accurate and customized tumor models. In contrast to conventional 2D cultures, these 3D printed models offer a more precise depiction of the tumor's structure and surrounding environment, especially advantageous for oral cancer studies. These models can mimic the intricate formations found in the mouth, leading to an enhanced comprehension of how tumors behave and react to treatment.[3-5] A particular type of this technology, known as 3D bioprinting, utilizes bioinks composed of living cells and biomaterials to create tissue-like structures that closely resemble tumor conditions found in the body.[6,7] This feature enables the creation of complex tumor models with different cell types and extracellular matrix elements, providing a thorough platform for researching cancer biology and testing new treatments. 3D bioprinting expands conventional 3D printing methods to the field of biology by allowing the development of intricate tissue and organ structures using bioinks deposited layer by layer. The bioinks consist of living cells and biocompatible substances, creating 3D constructs that imitate the structure and functions of natural tissues. Although 3D printing has been a mainstay in engineering and manufacturing since the 1980s, its potential for use in biomedicine only started to be investigated in the early 2000s. The medical profession first used bioprinting technology in 2003 when scientists began experimenting with printing living cells and biomaterials to build tissue-like structures. This marked a significant advancement in the area.

Steps in 3D bioprinting

- Structure designing: Using computer-aided design (CAD) software, the desired tissue structure is first digitally designed. To assure accuracy, this design is based on comprehensive imaging data from CT or MRI scans.
- Bioink preparation: Living cells are combined with biomaterials that offer nutrients and structural support to create bioinks. These bioinks need to be non-toxic, biocompatible, and able to encourage cell adhesion, differentiation, and proliferation.[7,8]
- Printing: In accordance with the digital design, the bioprinter applies the bioink layer by layer. It is possible to use a variety of printing methods, such as inkjet, laser-assisted, and extrusion-based printing. Every technique has benefits, and the best one is selected depending on the needs of the tissue to be printed.[6,7]
- Post-processing: Before being utilized in studies or therapeutic settings, the printed tissue might need to mature further in a bioreactor to acquire the required anatomical and functional characteristics.

Types of Bioinks

The successful generation of functional tissue constructions depends on the composition of bioinks, which are an indispensable part of the bioprinting process.

Typically made from synthetic or natural biomaterials, bioinks might be any of the following:

- **Hydrogels:** Bioinks are commonly available in the form of hydrogels. These are jelly-like, water-rich materials that offer a hydrated environment favorable to the survival and proliferation of cells. Collagen, gelatin, and alginate are a few such examples.
- **Synthetic polymers:** More control over mechanical characteristics and rates of degradation is possible with these materials. Polylactic acid (PLA) and polyethylene glycol (PEG) are two examples.
- **Decellularized extracellular matrix (dECM):** This kind of bioink comes from natural tissues that have undergone cell removal procedures, exposing a scaffold that is full with biochemical cues that promote tissue regeneration.[7,8]

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The benefits of 3D bioprinting in oral cancer research are manifold and can be enumerated as follows:

- **Precision and customization:** With the use of bioprinting, extremely accurate and personalized tissue models that closely resemble the in vivo tumor environment can be produced. This is essential for researching the intricate relationships found in the tumor microenvironment and for creating individualized treatment plans.[2-4]
- **Heterogeneity and biomimicry:** By combining various cell types and extracellular matrix components, advanced bioprinting techniques may create heterogeneous tissue architectures that closely resemble the chemical and physical characteristics of genuine tissues.[2,9] This is especially crucial for studies on oral cancer, since therapy response and cancer development are greatly influenced by the tumor microenvironment.[10,11]
- **Enhanced research and drug testing:** Compared to conventional 2D cell cultures, 3D bioprinted models offer a more precise platform for researching the biology of cancer and testing potential treatments. By increasing drug development speed and decreasing the need for animal testing, these models can more accurately forecast clinical outcomes.[5,12]

Though 3D bioprinting has great potential, there are several obstacles to overcome, such as the requirement for standardized materials and processes to guarantee consistency and repeatability in various applications and investigations. Another major obstacle is the creation of completely functional, vascularized tissues that can blend in perfectly with the body's inherent systems. Clinical translation is further complicated by the constantly changing regulatory environment surrounding bioprinted tissues and organs.

Personalized medicine and clinical applications

3D bioprinting shows potential for personalized medicine in addition to advancing our knowledge of cancer biology. Researchers can develop personalized tumor models that represent the distinct traits of a patient's cancer by utilizing cells taken from the patient. This method allows for more precise assessment of how treatments are working and the creation of personalized treatment plans.[13,14] Using 3D bioprinted models for drug screening and preclinical testing has the potential to bridge the gap between in vitro studies and clinical applications, improving the effectiveness of targeted cancer therapies. Moreover, these models can help in discovering new types of treatments and improving current therapies by offering a more accurate and dependable method for assessing the effectiveness and safety of treatments. Integrating 3D printing and bioprinting technologies into oral cancer research provides great benefits in developing accurate tumor models tailored to individual patients. These advancements play a crucial role in furthering our knowledge of oral cancer biology, enhancing diagnostic and treatment approaches, and ultimately improving patient results. The ongoing advancement and utilization of these technologies offer the potential to change the field of cancer research and treatment, possibly resulting in discoveries that could greatly enhance the survival rates and quality of life for individuals with oral cancer. The primary aim of this systematic review is to assess the progress in 3D bioprinting technology and its applications in producing precise and personalized tumor models for oral cancer research, emphasizing the growth and utilization of bioinks and their influence on cancer treatment and diagnosis.

Methodology

Research question: This systematic review's main research question is: *How have developments in 3D bioprinting technology specifically, the manufacturing of bioinks affected the production of precise and customized tumor models for the study and treatment of oral cancer?*

PICOS questions

- Population (P): studies involving tumor models for oral cancer
- Intervention (I): use of 3D bioprinting (bioinks) technology
- Comparison (C): traditional 2d cell culture models and other non-bioprinting methods
- Outcomes (O): accuracy of tumor models, therapeutic efficacy, patient-specific treatment strategies
- Study design (S): experimental studies, reviews,and clinical trials

Time frame: 2019 to 2024 (6 years)

Search strategy and keywords: The systematic review followed the PRISMA guidelines to ensure a thorough and unbiased identification of relevant studies. In accordance with PRISMA guidelines, a thorough search in various databases to collect relevant information on the application of 3D bioprinting and bioinks in studies related to oral cancer was performed. The databases that were searched were PubMed, Scopus, Web of Science, Cochrane Library, and Google Scholar. These databases were chosen because they cover a wide range of biomedical and clinical research, guaranteeing a comprehensive and diverse search. Different keywords and terms were used in the search to include all relevant studies. The key terms included "*3D bioprinting", "bioinks", "oral cancer", "tumor models", "cancer research", "personalized medicine", "bioprinting technology", and "tumor microenvironment", (or) "canter treatment", (AND) "cancer diagnosis", "3D culture"*. These terms were selected to cover different facets of the research subject, such as the technological, biological, and clinical aspects of 3D bioprinting for cancer research. This review aimed to comprehensively evaluate the current state and progress in 3D bioprinting technology for oral cancer research by adhering to PRISMA guidelines (Figure 1) and conducting an extensive search across various databases using specific keywords.

Inclusion criteria

- Studies published in peer-reviewed journal
- Research involving the use of 3D bioprinting for creating tumor models
- Studies focusing on oral cancer and especially experimental studies
- Articles written in English language
- Both experimental and review articles

Exclusion criteria

- Studies not related to oral cancer or cancer research
- Articles that do not involve 3D bioprinting technology
- Non-peer reviewed articles, abstracts, editorials, reports, and studies with no definitive concludingresults.
- Studies not written in English language

Data extraction, quality assessment and data synthesis: A total of 296 articles were searched through various databases from the time -period of 2019 till 2024. After meticulous review and analysis, keeping in mind all the inclusion and exclusion criteria, only 5 articles were included in the systematic review and out of them 3 were eligible for meta-analysis work further. The extraction of data and evaluation of quality were performed meticulously by two committed reviewers, known as VP and UM oversaw collecting information about the study, such as the study type, number of participants, and results that were reported. VP evaluated the studies' methodological quality using standardized assessment tools, ensuring a thorough assessment of the research's credibility. VP and UM both confirmed that the data extracted and conducted the quality assessments. Any inconsistenciesthat occurred during these procedures were addressed through in-depth conversation and agreement among the evaluators, guaranteeing dependability and uniformity in the evaluation. A narrative approach wasused to combine results from the studies for data synthesis purposes. Quantitative data were condensed using descriptive statistics when appropriate. The focus of the synthesis was to emphasize the progress in 3D bioprinting technology, the creation of bioinks, and their use in developing tumor models for oral cancer research. This thorough compilation sought to offer a unified comprehension of the present research and technology advancements in this area.

Risk of bias assessment: The Cochrane Risk of Bias Tool for randomized controlled trials and the ROBINS-I tool for non-randomized studies were used to evaluate the risk of bias in the included studies. Selection bias, performance bias, detection bias, attrition bias, and reporting bias were among the variables considered in the assessment. Based on these standards, each study was assigned a risk of bias rating: low, moderate, or high.

Figure 1. PRISMA Flowchart for the review

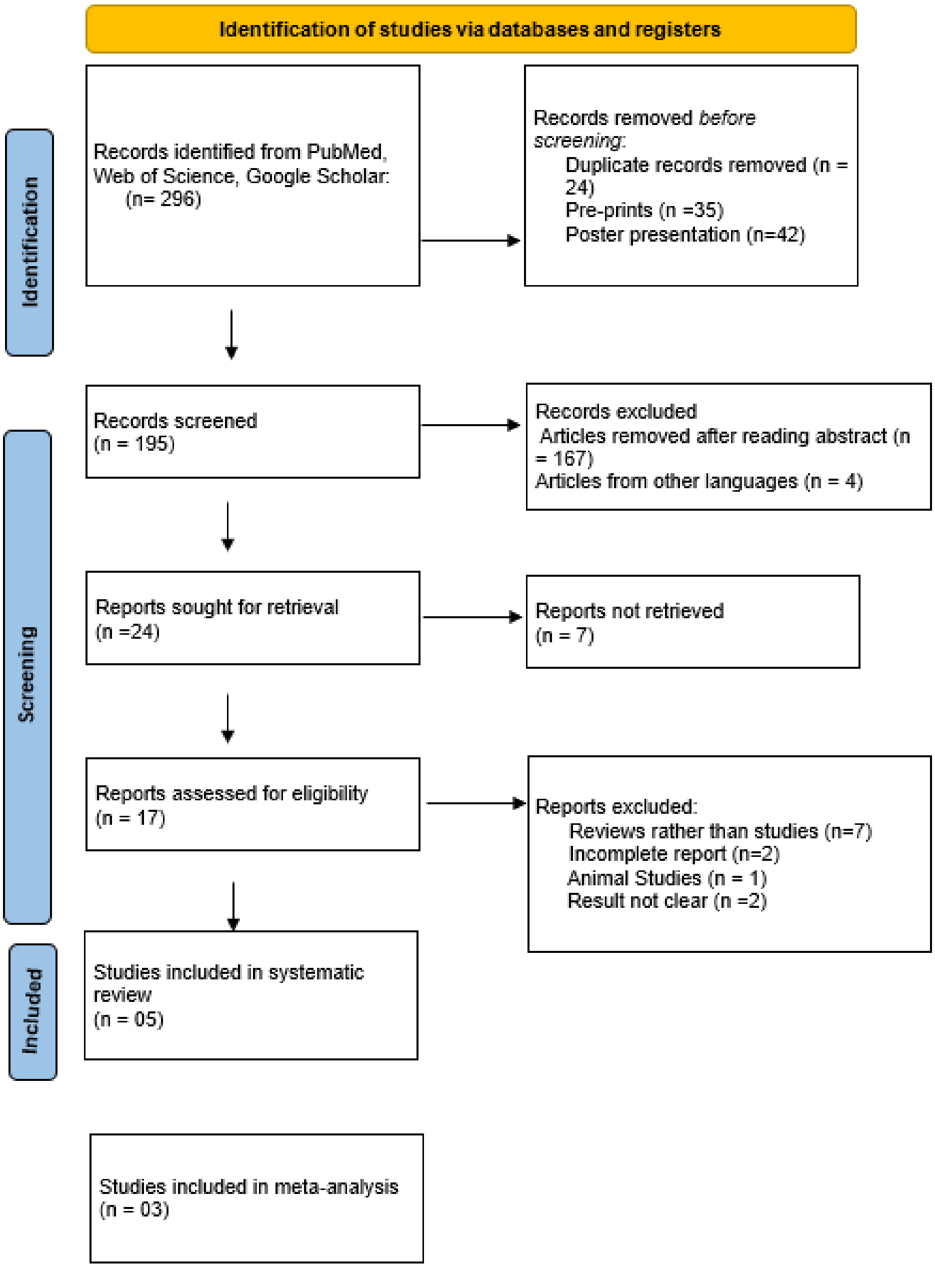

Table 1. Summary of experimental studies utilizing 3D Bioprinting in cancer research included in the systematic review

Author/year	Type of study/study design	Study objective	Methodology	Use of 3D bioprinting	Key findings or outcomes
Brambilla et al., (2021)[15]	Systematic review	To assess the frequency of characterisation tests for oral solid medication delivery systems and examine the use of 3D printing techniques in their development.	Examining and contrasting research on four 3D printing methods utilized in the production of oral solid formulations: stereolithography (SLA), selective laser sintering (SLS), inkjet 3D printing (Inkjet 3DP), and fused deposition modeling (FDM). The study looks at the frequency with which each technique has been examined using six distinct characterization tests: drug content, surface morphology, drug dissolution, hardness, friability, and weight uniformity.	This study assesses the viability of using 3D bioprinting methods to produce oral solid formulations, particularly for head and neck cancer treatment applications. The methods that are examined include Selective Laser Sintering (SLS) (4%), Inkjet 3D Printing (Inkjet 3DP) (13%), Stereolithography (SLA) (7%), and Fused Deposition Modeling (FDM) (76%). The emphasis is on how these technologies can be used to develop drug delivery systems that are tailored to each patient, which could increase treatment compliance and lower costs for patients with head and neck cancer.	An analysis of four different 3D printing processes' characterisation testing provides information about how well they work for treating head and neck cancer. Although SLA and SLS regularly investigated important criteria like drug content and solubility, SLS was found to be the most often tested method. But no single approach worked for everyone, highlighting the necessity of customizing decisions based on unique tumor characteristics and treatment needs in cases of head and neck cancer.
Derr et al., (2019)[16]	Experimental study	To create skin tissue equivalents through bioprinting that are both physiologically and morphologically identical to human skin in a reliable and repeatable manner. The model is intended to be used for disease modeling, toxicity screening, and chemical testing.	Creating a bioink for 3D printing using gelatin, collagen, and fibrin is part of the process. Skin tissue architectures are produced layer by layer using bioink. Histological examination, immunohistochemistry, MTT assays, and barrier function tests, such as electrical conductivity and penetration assays, are used to assess the printed tissues. For noninvasive imaging, optical coherence	Skin tissue equivalents are created via 3D bioprinting, which involves carefully applying layers of bioink that comprise cells and extracellular matrix elements. With the help of this technology, intricate tissue architectures with excellent spatial resolution can be created, guaranteeing structural integrity and homogeneity. This strategy enables the manipulation of tissue composition	The study produced skin tissues that are successfully bioprinted and resemble natural human skin in both structure and function. The bioprinted tissues showed low lateral contraction, acceptable cell viability, and efficient barrier function. Important indicators of tight junction development and epidermal differentiation were seen, and OCT offered a noninvasive

			tomography (OCT) is employed.	and size to conform to physiological parameters.	way to monitor quality in real time. The approach has potential uses in illness simulation and drug testing.
Langer et al., (2019)[17]	Experimental study	To create pancreatic cancer tissues using a 3D bioprinting technique, test it, and evaluate how the tissues react to drugs and external cues	A Novogen MMX Bioprinter was used to bioprint tissues from pancreatic cancer patients. To assess tissue structure, cell proliferation, and gene expression, histological examination, immunofluorescence, and quantitative RT-PCR were used. 3D viewing of intact tissues was made possible using CLARITY imaging.	Complex pancreatic cancer tissue models with exact control over cell distribution and spatial structure were made possible by 3D bioprinting.	The cellular shape and sensitivity to external cues were among the important characteristics of human tumors that the bioprinted pancreatic cancer tissues faithfully mimicked. They also showed dose-dependent responses to chemotherapy, indicating their potential as a platform for customized medicine and drug screening.
Leong et al., (2022)[18]	Systematic review	To appraise the therapeutic use of 3D bioprinted glioblastoma models for drug testing and the efficacy of bioinks in simulating the glioblastoma tumor microenvironment.	A thorough analysis of nineteen papers from the databases of PubMed, Medline Ovid, Web of Science, Scopus, and ScienceDirect was conducted. The capacity of several bioinks and bioprinting techniques to replicate the glioblastoma microenvironment was investigated.	Using various bioinks and printing processes, 3D bioprinting was used to create glioblastoma models that faithfully reflected the tumor microenvironment.	Alginate hydrogels showed great biocompatibility and were widely employed as bioinks. Models with increased drug resistance were produced by using multicomponent bioinks and advanced structural design. In comparison to 2D cultures, 3D bioprinted models demonstrated better drug response, indicating their potential for individualized treatment and high-throughput drug screening.
Mohammadrezaei et al., (2023)[19]	Experimental modeling	To create a CA model that can faithfully replicate the behavior of cells after printing in 3D bioprinted scaffolds. The study also sought to investigate the model's possible uses in tissue engineering and oncology, as well as to validate it using experimental data.	The fabrication of bioink, the 3D bioprinting of cell-hydrogel structures, and in-vitro tests to assess cellular behavior comprised the experimental technique. Based on experimental data, the computational methodology developed and calibrated a CA model	A key component of the study's experimental design was 3D bioprinting, which made it possible to create hydrogel structures packed with cells. This method made it possible to precisely regulate how the cells were arranged within the scaffold, which helped to validate the	The research effectively created a CA model that faithfully simulated in vitro cellular activity inside three-dimensional bioprinted scaffolds. The model accurately replicated the mobility, vitality, and proliferation of cells seen in experiments under different

			to simulate cellular dynamics within the printed scaffolds.	CA model and reveal information on the behavior of the cells after printing.	bioprinting circumstances. This method may lessen the requirement for lengthy in-vitro studies by forecasting cellular behavior in tissue engineering and cancer applications.

Results

The systematic review highlights the significant advancements and diverse applications of 3D bioprinting in cancer research, showcasing its potential to create precise and functional tumor models (Table 1). Brambilla et al., (2021)[15] conducted a comprehensive review of various 3D printing techniques for developing oral solid medication delivery systems aimed specifically at head and neck cancer treatments. They found that techniques such as Selective Laser Sintering (SLS), Inkjet 3D Printing (Inkjet 3DP), Stereolithography (SLA), and Fused Deposition Modeling (FDM) each offer unique benefits in drug content and solubility testing, with FDM being the most frequently tested. This study highlights the importance of tailoring 3D printing methods to specific tumor characteristics and treatment needs. Expanding on the application of 3D bioprinting, Derr et al., (2019)[16] successfully created skin tissue equivalents using a bioink composed of gelatin, collagen, and fibrin. The bioprinted skin models were evaluated through histological and functional assays, demonstrating structural integrity and homogeneity. These models closely resembled natural human skin in both physiology and morphology, making them valuable for disease modeling and drug testing.

Langer et al., (2019)[17] further showcased the capabilities of 3D bioprinting by creating complex pancreatic cancer tissues using the Novogen MMX Bioprinter. This technology allowed precise control over cell distribution and spatial structure within the bioprinted tissues. The resultant models accurately mimicked key characteristics of human tumors, including cellular morphology and response to chemotherapy, highlighting their potential for personalized medicine and drug screening. Similarly, Leong et al., (2022)[18] reviewed the therapeutic use of 3D bioprinted glioblastoma models, focusing on the efficacy of various bioinks such as alginate hydrogels in replicating the tumor microenvironment. Their analysis revealed that 3D bioprinted models provided more accurate drug response data compared to traditional 2D cultures, supporting their use in high-throughput drug screening and personalized treatment planning. Mohammadrezaei et al., (2023)[19] developed a computational cellular automata (CA) model validated with 3D bioprinted cell-hydrogel structures. This model accurately simulated cellular dynamics within the bioprinted scaffolds, offering insights into cellular behavior post-printing. Their findings suggest that 3D bioprinting enables precise control over cell arrangement, reducing the need for extensive in-vitro experiments and advancing applications in tissue engineering and oncology. Collectively, these studies highlight the transformative potential of 3D bioprinting in cancer research, particularly in creating accurate and functional tumor models. These models enhance our understanding of cancer biology and improve drug testing and personalized treatment strategies.

Quality assessment for the invitro studies were done using the QUIN Tool (Quality assessment tool for invitro studies) was developed by Sheth VH, (2024).[20] This tool contains 12 domains and the studies were assessed under these conditions and the study of Mohammadrezaei et al., (2023)[19] were found to be of highest quality with 11 points and the least quality was found by the Deer K et al (2019).[16] This study failed to address the blinding and outcome details of the study (Table 2). The QUIN Tool quality assessment of in vitro research is shown in Table 2. Despite lacking information on operator specifics, technique, and result assessor blinding, the authors received a score of 9 out of 12. Using simply blinding, Langer et al., (2019)[17] received a score of 10. The group that scored the highest, Mohammadrezaei et al. (2023)[19], only missed blinding. The studies all demonstrated strong sampling strategies, methods, and statistical analyses; nevertheless, the lack of blinding in operator details and outcome evaluation draws attention to areas in which study design transparency must be improved.[16,17] Figures 2 and 3 Risk of bias assessment for the invitro studies were done using the ROBINS – I. Among the 3 studies, 2 studies were found to be low risk of bias[17,19] and other study were found to be of moderate concern.[16] In overall risk of bias assessment among Invitro studies high risk was found due to confounding and low risk was found due to missing data. Table 3 depicts the studies excluded from the present systematic review.

Figure 2. Risk of Bias assessment done using the ROBINS – I

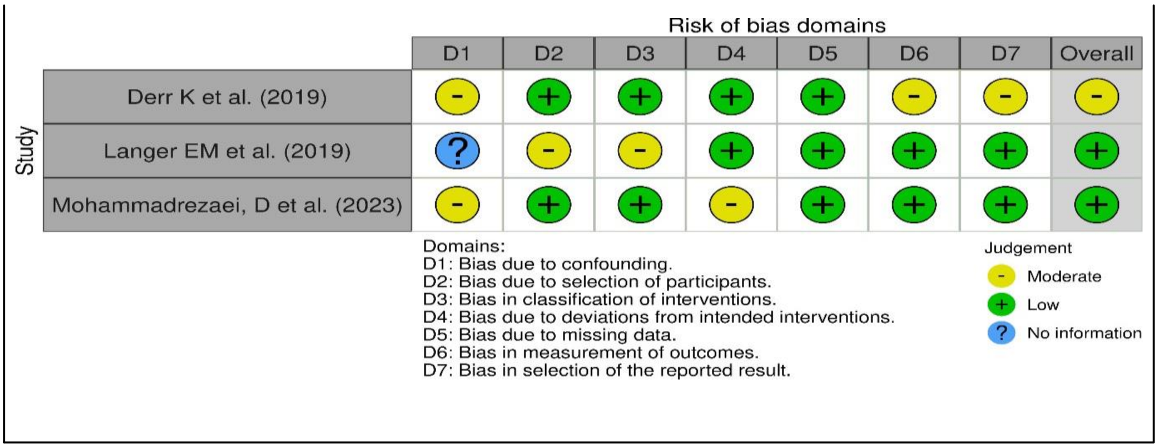

Figure 3: Risk of Bias assessment done using the ROBINS – I Overall

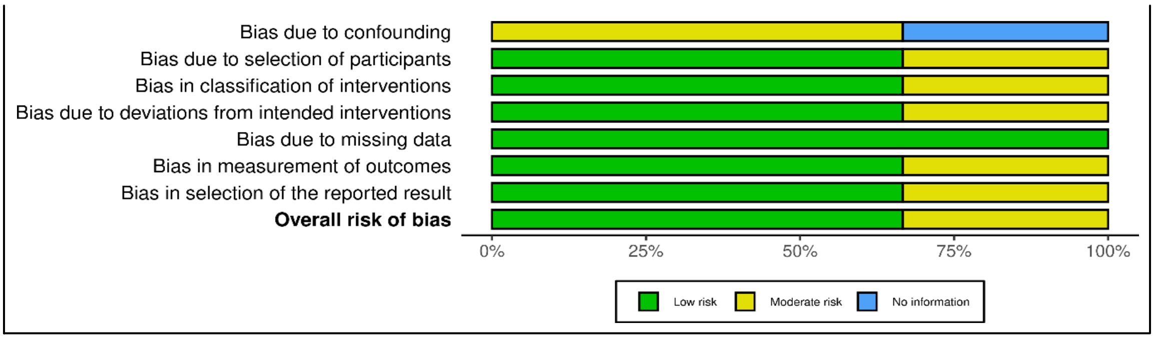

Table 2. Quality assessment done using the QUIN Tool (Quality assessment tool for in-vitro studies)

S.No	Author/ year	Aim / Objec-tive	Sample Size	Samp-ling tech-nique	Compa-rison Group	Metho-dology	Opera-tor details	Rando-mization	Out-come	Out-come Asses-sor	Blin-ding	Statis-tical Ana-lysis	Presen-tation of results	Total
1	Derr et al., (2019)[16]	*	*	*	*	*	-	*	*	-	-	*	*	9
2	Langer et al., (2019)[17]	*	*	*	*	*	*	*	*	-	*	*	*	10
3	Mohammadrezaei et al., (2023)[19]	*	*	*	*	*	*	*	*	*	-	*	*	11

Table 3. Studies excluded from the systematic review

S. No	Author & year	Title of the Study	Reason for exclusion
1	Zaid et al., (2019)[21]	Creating customized oral stents for head and neck radiotherapy using 3D scanning and printing	Out of scope of the study
2	Cornejo et al., (2022)[22]	Anatomical Engineering and 3D Printing for Surgery and Medical Devices: International Review and Future Exponential Innovations.	Study design - Review study
3	Shi et al., (2019)[23]	Drop-On-Powder 3D Printing of Tablets with an Anti-Cancer Drug,5-Fluorouracil.	Out of scope of the study
4	Sharafeldin et al., (2021)[24]	Detecting cancer metastasis and accompanying protein biomarkers at single cell levels using a 3D-printed microfluidic immunoarray.	Out of scope of the study

Discussion

In cancer research, 3D bioprinting has become a game-changer, providing previously unheard-of capacity to produce accurate and useful tumor models. These developments have greatly improved our knowledge of the biology of cancer, paving the way for more precise medication testing and individualized treatment plans. This review explores the many facets of 3D bioprinting, as demonstrated by current research, and looks at the unique ways in which these technologies might be applied to cancer treatment and research.

New approaches to modeling TME and studying cancer biology in unprecedented depth have been made possible by recent developments in 3D bioprinting technology, which has had a significant impact on cancer research. The novel technology under consideration tackles the ongoing obstacles brought to light by Siegel et al., 2020, underscoring the vital requirement for inventive methods to improve therapeutic approaches in the treatment of cancer.[25] According to Klemm & Joyce, (2015),[1] conventional 2D cell cultures frequently fall short of accurately capturing the intricate relationships that exist between tumors and their surrounding microenvironment. These interactions are essential for comprehending drug resistance mechanisms and therapeutic responses. 3D bioprinted models, on the other hand, have become a reliable substitute that can more accurately replicate the complex cellular makeup and architecture of tumors. There is great potential in being able to use 3D bioprinting to replicate tumor models unique to individual patients. With this method, researchers may quickly discover innovative medicines and facilitate customized medicine methods by creating experimental circumstances that closely mimic the *in vivo* tumor environment. The advances in 3D bioprinting to target the TME enhance treatment outcomes were explored which allowed for detailed studies of tumor growth, metastasis, and treatment response by including a variety of cell types and extracellular matrix components.[2]

The complexity and utility of these models are improved by the incorporation of multi-material and multi-cellular 3D bioprinting processes, as investigated by Khorsandi D, (2024)[26] and Zhang Y (2023).[27]

This ability is essential for researching the dynamic behaviors and interactions of cells within the tumor position, helping to understand the mechanisms driving the progression of cancer and accelerating the development of novel therapeutic approaches.

Beyond the study of tumor biology, 3D bioprinting has been applied to diagnostic purposes. Recent advancements in 4D printing technology as they relate to cancer therapies are highlighted by Chinnakorn et al., (2023)[28], who showed how they can be used to develop customized treatment plans and responsive platforms for drug delivery. These developments highlight how 3D bioprinting might be used to further oncology research and therapeutic applications. Moreover, compared to conventional approaches, the application of high-content screening in 3D bioprinted tumor models enables complete evaluation of medication efficacy and toxicity profiles in a more physiologically appropriate context.[29,30] This method expedites the transition of potential medicines from bench to bedside and improves the predictive value of preclinical trials. In cancer research, 3D bioprinting is a paradigm shift that offers advanced capabilities to model intricate tumor biology and therapeutic responses. Through the utilization of these technical developments, scientists may now better understand the pathophysiology of cancer and expedite the creation of tailored treatment plans, which will ultimately lead to better oncology patient outcomes.

In 3D bioprinting, materials called bioinks materials made of living cells and biocompatible matrices are deposited layer by layer to construct intricate tissue architectures. To simulate the variety of tumor tissues, this procedure can include different cell types, extracellular matrix elements, and biochemical signals. For example, Chowdhury T, (2023)[31] emphasized single-molecule and single-cell characterisation while discussing the use of 3D bioprinting in cancer diagnostics and prognostics. An essential component of 3D bioprinting is bioink. According to Zhang Y et al., (2024)[32] and Wang J et al., (2024)[33], these bioinks are made to maintain tissue integrity while offering the mechanical characteristics required for cell survival, proliferation, and differentiation. Mandrycky et al., (2016)[34] described how improvements in bioink formulations have made it possible to create more intricate and useful tissue structures, thereby expanding the possibilities for cancer simulation.

3D bioprinting has various important implications for cancer research. According to Roma-Rodrigues et al., (2019)[2], focusing on the TME can enhance the effectiveness of cancer treatments. Researchers can identify new treatment targets by improving their understanding of the interactions between cancer cells and the surrounding stroma through the creation of more precise tumor models. The creation and testing of drugs have benefited greatly from the use of 3D bioprinted models. Compared to conventional 2D cultures, Baker & Chen, (2012)[3] found that three-dimensional culture systems can modify cellular signals, leading to more physiologically appropriate drug responses. This development is especially important for anticancer drug screening since it makes it possible to identify molecules that traditional methods could have missed. 3D bioprinting makes it possible to create tumor models that are unique to each patient in customized medicine. These models can be used to examine individual reactions to different treatments, enabling more individualized and successful therapy techniques.[8] By improving the ability to forecast drug toxicity and efficacy, this method not only increases the accuracy of cancer treatment but also lowers the possibility of side effects. 3D bioprinting has various important implications for cancer research. High-content screening could screen better cancer treatment leads, as evidenced by Kochanek et al., (2019)[30] who used it to describe multicellular tumor spheroid cultures of head and neck squamous cell carcinoma. Yan et al., (2013)[35] demonstrated the use of 3D bioprinted models in investigating cancer heterogeneity by identifying CD166 as a novel marker for cancer stem-like cells in head and neck squamous cell carcinoma using plasma membrane proteomics. Yang et al., (2013)[36] demonstrated the use of 3D bioprinted models to research cancer cell behavior by demonstrating how suppression of bone morphogenetic protein can reduce mesenchymal migration of head and neck cancer cells. Colley et al., (2011)[37] improved the utility of 3D bioprinting in simulating cancer progression by creating tissue-engineered models of early invasive oral squamous cell carcinoma and oral dysplasia. They developed three-dimensional co-culture models derived from distinct phases of human tongue carcinogenesis, offering valuable perspectives on the advancement of neoplastic processes, and offering prospects for customized therapeutic applications. Majumder et al., (2015)[38] demonstrated how 3D bioprinting can enable tailored treatment approaches by creating an ex vivo platform that captures tumor heterogeneity and predicts clinical response to anticancer medications. Recent studies show promising outcomes when combining different therapeutic techniques with advanced technologies such as 3D bioprinting to tackle difficult medical conditions. Shah SU et al. (2023)[39] found that using triamcinolone, pentoxifylline, and vitamin E together was effective in improving mouth opening and symptoms severity in patients with oral submucous fibrosis. Another research emphasized the importance of increased inclusion of oral pathologists in histopathology labs to enhance the accuracy of diagnosing oral lesions, as general pathologists often fail to refer challenging cases to oral pathologists despite acknowledging their expertise.[40]

Therefore, the amalgamation of cutting-edge technology, especially 3D bioprinting, is transforming the field of cancer research by offering more precise models for analyzing the behavior of tumors and assessing the effectiveness of treatments. The complexity of cancer can now be better understood due to the 3D bioprinted models, which also enables to create individualized treatment plans that more precisely resemble the human body reaction to various treatments.[24,29] Alongwith these advances in 3D bioprinting, nanotechnology also has improved medicine delivery and minimized the side effects, especially when treating illnesses like oral mucositis caused by radiation therapy.[41] Furthermore, by using 3D bioprinted models, the investigation of novel therapeutic drugs, such as calotropin, which targets important cancer pathways, could be further improved and their effectiveness in treating oral squamous cell carcinoma precisely evaluated.[42] The investigation of biomarkers such as RFC3, which is markedly elevated in HNSCC and linked to important carcinogenic pathways, is made easier by this accurate modeling, thus improving the treatment approaches and patient outcomes.[43] When taken as a whole, these developments are opening the door to more precise and successful cancer treatments.

The incorporation of these approaches into cancer research is a breakthrough that opens new possibilities for understanding tumor biology and creating individualized treatments. Through more accurate replication of the intricate tumor microenvironment than is possible with conventional techniques, 3D bioprinting advances our knowledge of cancer progression and treatment outcomes. This technology has enormous potential to advance customized medicine and improve the results of cancer treatments as it develops. The revolutionary potential of 3D bioprinting in furthering cancer research is highlighted by this systematic study, especially in the creation of precise disease models and creative drug delivery systems. The evaluation assesses the effectiveness of different 3D printing processes, including Stereolithography (SLA), Fused Deposition Modeling (FDM), Selective Laser Sintering (SLS), and Inkjet 3D Printing (Inkjet 3DP), to demonstrate how well they can create accurate and useful models that are customized to meet certain treatment requirements.

The results of Brambilla et al., (2021)[15] highlight the importance of choosing the right bioprinting techniques, with FDM being found to be the best option for drug content and solubility testing in the administration of medications for head and neck cancer. Other researches showed that 3D bioprinting can accurately mimic complex tissue settings, including glioblastoma models and skin and pancreatic cancer tissues.[16-19] These developments help to improve drug screening procedures and customized treatment strategies in addition to deepening our understanding of cancer biology. The QUIN and ROBINS-I tools, when used in a systematic manner to evaluate study quality and bias, offer important insights into the methodological rigor and transparency of the discipline.

Identifying studies with low to moderate risks of bias is critical to confirming the validity and use of bioprinted models in oncology research. The development of more precise and useful tumor models made possible by 3D bioprinting technology has completely changed the field of cancer research. Research has indicated that several 3D printing methods are effective in simulating intricate tissue architectures and medication delivery systems. The necessity for increased transparency in study design, especially regarding blinding and operator details, is underscored by the quality and danger of bias assessments. All things considered, 3D bioprinting has enormous potential to advance our knowledge of cancer biology, enhance medication testing, and create individualized treatment plans. To fully exploit the potential of these technologies in cancer therapy and diagnostics, future research should carry out more exploration and refinement.

Conclusion

This comprehensive analysis emphasizes how important 3D bioprinting is to the advancement of dental and cancer research. The evaluated studies demonstrated how the technology may be used to build complex tissue models that closely resemble human physiology, which can help with accurate drug testing and customized treatment plans for oral malignancies. Benefits include improved treatment relevance and model accuracy, however there are also drawbacks, such as inconsistent printing materials and difficult tumor microenvironment replication. However, the study's conclusions highlight important advancements in bioprinting's ability to improve therapeutic outcomes and diagnostic skills, pointing to a time when individualized therapies based on a patient's unique tumor biology will be the norm. Future research recommendations include standardizing bioprinting procedures, addressing ethical concerns, and expanding collaborative efforts across interdisciplinary areas to maximize the technology's potential influence in clinical practice. Ultimately, by paving the way for more customized medical care and effective cancer treatments in the future, 3D bioprinting holds the potential to completely transform the domains of oncology and dentistry.

Declarations

Conflicts of interest and financial disclosures

The author declares that he has no conflict percent and there was no external source of funding for the research in question.

Ethical approval

The study was approved by the University ethics committee and was conducted in accordance with the Declaration of the World Medical Association.

Informed consent

Informed consent was obtained from all individual participants included in the study.

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REFERENCES

1. Klemm F, Joyce JA. Microenvironmental regulation of therapeutic response in cancer. *Trends Cell Biol*. 2015;25(4):198-213. doi:10.1016/j.tcb.2014.11.006
2. Roma-Rodrigues C, Mendes R, Baptista PV, Fernandes AR. Targeting Tumor Microenvironment for Cancer Therapy. *Int J Mol Sci*. 2019;20(4):840. doi:10.3390/ijms20040840
3. Baker BM, Chen CS. Deconstructing the third dimension: how 3D culture microenvironments alter cellular cues. *J Cell Sci*. 2012;125(13):3015-24. doi:10.1242/jcs.079509
4. Zhang YS, Duchamp M, Oklu R, Ellisen LW, Langer R, Khademhosseini A. Bioprinting the Cancer Microenvironment. ACS Biomater Sci Eng.2016;2(10):1710-1721. doi:10.1021/acsbiomaterials.6b00246
5. Melissaridou S, Wiechec E, Magan M, et al. The effect of 2D and 3D cell cultures on treatment response, EMT profile and stem cell features in head and neck cancer. *Cancer Cell Int*. 2019;19:16. doi:10.1186/s12935-019-0733-1
6. Ashammakhi N, Ahadian S, Xu C, et al. Bioinks and bioprinting technologies to make heterogeneous and biomimetic tissue constructs. *Mater Today Bio*. 2019;1:100008. doi:10.1016/j.mtbio.2019.100008
7. Hospodiuk M, Dey M, Sosnoski D, Ozbolat IT. The bioink: A comprehensive review on bioprintable materials. *Biotechnol Adv*. 2017;35(2):217-239. doi:10.1016/j.biotechadv.2016.12.006
8. Bhuskute H, Shende P, Prabhakar B. 3D Printed Personalized Medicine for Cancer: Applications for Betterment of Diagnosis, Prognosis and Treatment. *AAPS PharmSciTech*. 2021;23(1):8. doi:10.1208/s12249-021-02153-0
9. Almela T, Tayebi L, Moharamzadeh K. 3D Bioprinting for In Vitro Models of Oral Cancer: Toward Development and Validation. *Bioprinting*. 2021;22:e00132. doi:10.1016/j.bprint.2021.e00132
10. Augustine R, Kalva SN, Ahmad R, et al. 3D Bioprinted cancer models: Revolutionizing personalized cancer therapy. *Transl Oncol*. 2021;14(4):101015. doi:10.1016/j.tranon.2021.101015
11. Datta P, Dey M, Ataie Z, Unutmaz D, Ozbolat IT. 3D bioprinting for reconstituting the cancer microenvironment. *NPJ Precis Oncol*. 2020;4:18. doi:10.1038/s41698-020-0121-2
12. van't Veer LJ, Bernards R. Enabling personalized cancer medicine through analysis of gene-expression patterns. *Nature*. 2008;452(7187):564-70.

doi:10.1038/nature06915

13. Li J, Liang D, Chen X, Sun W, Shen X. Applications of 3D printing in tumor treatment. *Biomedical Technology*. 2024;5:1-3

14. Safhi AY. Three-Dimensional (3D) Printing in Cancer Therapy and Diagnostics: Current Status and Future Perspectives. *Pharmaceuticals (Basel)*. 2022;15(6):678. doi:10.3390/ph15060678

15. Brambilla CRM, Okafor-Muo OL, Hassanin H, ElShaer A. 3DP Printing of Oral Solid Formulations: A Systematic Review. *Pharmaceutics*. 2021;13(3):358. doi:10.3390/pharmaceutics13030358

16. Derr K, Zou J, Luo K, et al. Fully Three-Dimensional Bioprinted Skin Equivalent Constructs with Validated Morphology and Barrier Function. *Tissue Eng Part C Methods*. 2019;25(6):334-343. doi:10.1089/ten.TEC.2018.0318

17. Langer EM, Allen-Petersen BL, King SM, et al. Modeling Tumor Phenotypes In Vitro with Three-Dimensional Bioprinting. *Cell Rep*. 2019;26(3):608-623.e6. doi:10.1016/j.celrep.2018.12.090

18. Leong SW, Tan SC, Norhayati MN, Monif M, Lee SY. Effectiveness of Bioinks and the Clinical Value of 3D Bioprinted Glioblastoma Models: A Systematic Review. *Cancers (Basel)*. 2022;14(9):2149. doi:10.3390/cancers14092149

19. Mohammadrezaei D, Moghimi N, Vandvajdi S, Powathil G, Hamis S, Kohandel M. Predicting and elucidating the post-printing behavior of 3D printed cancer cells in hydrogel structures by integrating in-vitro and in-silico experiments. *Scientific Reports*. 2023;13(1):1211

20. Sheth VH, Shah NP, Jain R, Bhanushali N, Bhatnagar V. Development and validation of a risk-of-bias tool for assessing in vitro studies conducted in dentistry: *The QUIN. J Prosthet Dent*. 2024;131(6):1038-1042. doi:10.1016/j.prosdent.2022.05.019

21. Zaid M, Bajaj N, Burrows H, et al. Creating customized oral stents for head and neck radiotherapy using 3D scanning and printing. *Radiat Oncol*. 2019;14(1):148. doi:10.1186/s13014-019-1357-2

22. Cornejo J, Cornejo-Aguilar JA, Vargas M, et al. Anatomical Engineering and 3D Printing for Surgery and Medical Devices: International Review and Future Exponential Innovations. *Biomed Res Int*. 2022;2022:6797745. doi:10.1155/2022/6797745

23. Shi K, Tan DK, Nokhodchi A, Maniruzzaman M. Drop-On-Powder 3D Printing of Tablets with an Anti-Cancer Drug, 5-Fluorouracil. *Pharmaceutics*. 2019;11(4):150. doi:10.3390/pharmaceutics11040150

24. Sharafeldin M, Chen T, Ozkaya GU, et al. Detecting cancer metastasis and accompanying protein biomarkers at single cell levels using a 3D-printed microfluidic immunoarray. *Biosens Bioelectron*. 2021;171:112681. doi:10.1016/j.bios.2020.112681

25. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2020. *CA Cancer J Clin*. 2020;70(1):7-30. doi:10.3322/caac.21590

26. Khorsandi D, Rezayat D, Sezen S, et al. Application of 3D/4D/5D and 6D Bioprinting in Cancer Research: How Does the Future Look Like? *Journal of Materials Chemistry B*. 2024

27. Zhang Y. 3D Printing for Cancer Diagnosis: What Unique Advantages Are Gained? *ACS Mater Au*. 2023;3(6):620-635. doi:10.1021/acsmaterialsau.3c00046

28. Chinnakorn A, Nuansing W, Bodag Brambilla hi M, Rolfe B, Zolfagharian A. Recent progress of 4D printing in cancer therapeutics studies. *SLAS Technol*. 2023;28(3):127-141. doi:10.1016/j.slast.2023.02.002

29. Shan F, Close DA, Camarco DP, Johnston PA. High-Content Screening Comparison of Cancer Drug Accumulation and Distribution in Two-Dimensional and Three-Dimensional Culture Models of Head and Neck Cancer. *Assay Drug Dev Technol*. 2018;16(1):27-50. doi:10.1089/adt.2017.812

30. Kochanek SJ, Close DA, Johnston PA. High

Content Screening Characterization of Head and Neck Squamous Cell Carcinoma Multicellular Tumor Spheroid Cultures Generated in 384-Well Ultra-Low Attachment Plates to Screen for Better Cancer Drug Leads. *Assay Drug Dev Technol*. 2019;17(1):17-36. doi:10.1089/adt.2018.896

31. Chowdhury T, Cressiot B, Parisi C, et al. Circulating Tumor Cells in Cancer Diagnostics and Prognostics by Single-Molecule and Single-Cell Characterization. *ACS Sens*. 2023;8(2):406-426. doi:10.1021/acssensors.2c02308

32. Zhang Y, Yang Q, Wan H, et al. 3D bioprinting in otorhinolaryngology: from bench to clinical application. *International Journal of Bioprinting.* 2024:3006

33. Wang J, Zhao Z, Yang K, Bai Y. Research progress in cell therapy for oral diseases: focus on cell sources and strategies to optimize cell function. *Frontiers in Bioengineering and Biotechnology*. 2024;12:1340728

34. Mandrycky C, Wang Z, Kim K, Kim DH. 3D bioprinting for engineering complex tissues. *Biotechnol Adv*. 2016;34(4):422-434. doi:10.1016/j.biotechadv.2015.12.011

35. Yan M, Yang X, Wang L, et al. Plasma membrane proteomics of tumor spheres identify CD166 as a novel marker for cancer stem-like cells in head and neck squamous cell carcinoma. *Mol Cell Proteomics*. 2013;12(11):3271-84. doi:10.1074/mcp.M112.025460

36. Yang WH, Lan HY, Tai SK, Yang MH. Repression of bone morphogenetic protein 4 by let-7i attenuates mesenchymal migration of head and neck cancer cells. *Biochemical and biophysical research communications*. 2013;433(1):24-30

37. Colley HE, Hearnden V, Jones AV, et al. Development of tissue-engineered models of oral dysplasia and early invasive oral squamous cell carcinoma. *Br J Cancer*. 2011;105(10):1582-92. doi:10.1038/bjc.2011.403

38. Majumder B, Baraneedharan U, Thiyagarajan S, et al. Predicting clinical response to anticancer drugs using an ex vivo platform that captures tumour heterogeneity. *Nat Commun*. 2015;6:6169. doi:10.1038/ncomms7169

39. Shah SU, Nigar S, Yousofi R, et al. Comparison of triamcinolone with pentoxifylline and vitamin E efficacy in the treatment of stage 2 and 3 oral submucous fibrosis: A randomized clinical trial. *SAGE Open Med*. 2023;11:20503121231200757. doi:10.1177/20503121231200757

40. Maqsood A, Faheem S, Mirza D, et al. An insight into perceptions of general pathologists about the need for oral pathology services: An observational study. *SAGE Open Med*. 2023;11:20503121231200758. doi:10.1177/20503121231200758

41. Dharman S, Maragathavalli G, Shanmugam R, Shanmugasundaram K. Current perspectives of nanotherapies in the prevention and treatment of radiotherapy/chemotherapy-induced oral mucositis in head and neck cancer—A narrative review. Journal of International Oral Health. 2023 Nov 1;15(6):491-9.

42. Selvaraj FM, Joseph AP, Pillai VR, Ramani P, Pazhani J, Mony V. Significance of tumour budding and invasive characteristics in grading of oral squamous cell carcinoma. J Oral Maxillofac Pathol. 2023 Oct-Dec;27(4):642-648. doi: 10.4103/jomfp.jomfp_410_23. Epub 2023 Dec 20. PMID: 38304506; PMCID: PMC10829472.

43. Avs KR, Pandi C, Kannan B, Pandi A, Jayaseelan VP, Arumugam P. RFC3 serves as a novel prognostic biomarker and target for head and neck squamous cell carcinoma. Clin Oral Investig. 2023 Nov;27(11):6961-6969. doi: 10.1007/s00784-023-05316-4. Epub 2023 Oct 20. PMID: 37861747