Aundhia et al., *BioImpacts.* 2026;16:33650
doi:10.34172/bi.33650
https://bi.tbzmed.ac.ir/

Hydrogel-based smart bioinks for 3D bioprinting: design strategies and applications in brain tissue engineering and tumor microenvironment modelling

Chintan Aundhia[1*], Jagruti Desai[2], Swayamprakash Patel[2], Chitrali Talele[1], Bhupendra G. Prajapati[3*]

[1]Department of Pharmacy, Sumandeep Vidyapeeth Deemed to be University, Piparia, Vadodara, Gujarat 391760, India
[2]Department of Pharmaceutical Technology, Ramanbhai Patel College of Pharmacy, Charotar University of Science and Technology (CHARUSAT), CHARUSAT Campus, Changa 388421, India
[3]Department of Pharmaceutics, Parul Institute of Pharmacy, Faculty of Pharmacy and Research & Development Cell, Parul University, Waghodia, Vadodara, Gujarat 391760, India

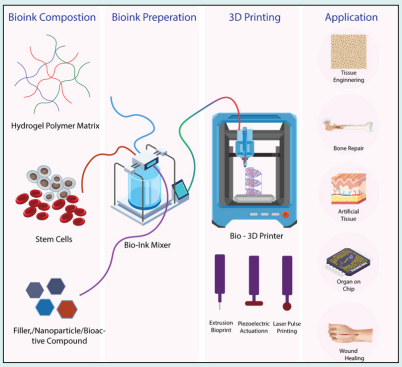
Article Info

Article Type:
Review

Article History:
Received: 10 Mar. 2026
Revised: 9 Jun. 2026
Accepted: 27 Jun. 2026
ePublished: 16 Aug. 2026

Keywords:
Hydrogel bioinks
Smart bioinks
3D bioprinting
Brain tissue engineering
Tumor microenvironment
Cancer-on-a-chip
Biomimetic scaffolds
Biofabrication

Abstract



Smart bioinks based on hydrogel have emerged a revolutionary platform in three-dimensional bioprinting that facilitates the development of biomimetic tissue constructs with a higher level of structural and biological precision. These advanced bioinks exhibit dynamic responsiveness to physicochemical and biological stimuli, allowing precise modulation of cell matrix interactions and microenvironmental conditions. The rational design of smart bioinks has achieved considerable advances in recent years by means of controlled optimization of rheological properties, crosslinking processes, and incorporation of bioactive agents. This review gives an in-depth account of hydrogel-based smart bioinks, their design principles, physicochemical and biological properties, and printability aspects. Their use in brain tissue engineering and tumor microenvironment modelling is of special interest as their capability to replicate complex cellular structures, biochemical gradients and dynamic signalling plays a critical role. Additionally, latest trends such as recent developments in nanocomposite bioinks, co-culture, and AI-assisted bio fabrication are also discussed. Although significant advances have been made, vascularization issues, long-term functionality, and scalability issues continue to pose major barriers to clinical translation. Altogether, hydrogel-based smart bioinks may be viewed as a promising approach to the development of tissue engineering, disease modelling, and precision medicine applications.

Introduction

Three dimensional (3D) bioprinting has already become among the most disruptive technologies in the field of regenerative medicine and tissue engineering, providing a capability to build complex, biomimetic tissue scaffolds with an unmatched precision of their spatial arrangement. In contrast to traditional scaffold fabrication technology, where a cell line often cannot exert fine control over geometry and cellular deposition are not readily accessible, through 3D bioprinting, cells, biomaterials and signalling molecules can be deposited in layer-by-layer which enables the formation of functional tissue analogs. This significantly impacts the development of personalized medicine, modelling and drug screening platforms.

The primary feature of successful three-dimensional bioprinting is the choice of a suitable bioink which is a collection of cells in a biomaterial formulation that regulates the structural integrity, biological functionality and biological relevance of printed structures.[1] The selection of bioink is a critical factor in supporting the cell viability throughout the fabrication process, as well as, allowing the formation of the tissue architectures that closely resemble the native biological systems.

Amongst the different types of bioinks, hydrogels have become the most commonly used due to their inherent

*Corresponding authors: Chintan Aundhia, Email: aundhia@gmail.com; Bhupendra G. Prajapati, Email: bhupen27@gmail.com

Study Highlights

- Hydrogel-based smart bioinks enable biomimetic 3D bioprinting by integrating tunable physicochemical properties, stimuli-responsiveness, and bioactivity, with promising applications in brain tissue engineering and tumor microenvironment modelling. Emerging approaches incorporating nanocomposites, co-culture, organoids, vascularization, and AI-assisted design further expand their potential for advanced biofabrication and precision medicine

compatibility with the living cells and close resemblance with the extracellular matrix of the living cells. These properties make hydrogel-based systems especially advantageous in bio fabrication of biologically relevant three-dimensional constructs in a variety of tissue engineering and disease-modelling applications.[2]

To provide conceptual clarity, "smart bioinks" can be defined as advanced hydrogel-based biomaterial systems that are capable of dynamically responding to internal or external stimuli, including biochemical signals, mechanical forces, or environmental changes such as pH, temperature, and oxidative stress. Unlike conventional hydrogel bioinks, which primarily serve as passive structural matrices, smart bioinks exhibit adaptive and feedback-driven behaviour, enabling spatiotemporal modulation of cellular responses, matrix properties, and biological functionality. This dynamic responsiveness allows smart bioinks to more closely replicate the continuously evolving microenvironment of native tissues, particularly in complex systems such as neural tissues and tumor microenvironments. In this context, the key distinction between conventional and smart bioinks lies not only in stimuli-responsiveness, but also in their ability to actively regulate cell–matrix interactions, enable controlled release of bioactive factors, and adapt to disease-specific microenvironmental cues. The design of smart bioinks can be broadly understood through three interconnected dimensions which are dynamic physicochemical responsiveness, cell-instructive bioactivity and printability with adaptive structural stability.

Notably, neural tissue and brain models have shown all bioink types to be highly effective, with the microenvironment softness and dynamic complexity of the native extracellular matrix (ECM) requiring biomaterials that meet biophysiological aspects as closely as possible.[3]

Compared to other tissues, brain tissue engineering poses unique challenges due to the extreme softness, viscoelastic nature, and biochemical specialization of neural extracellular matrices.[4] The ability to recreate this microenvironment in a highly regulated form along with maintaining neuronal viability, differentiation, and network formation continues to pose fundamental challenge in the development of physiologically relevant neural constructs.

In addition to regenerative applications, bioinks using hydrogels have also been investigated to model the tumor microenvironment (TME), specifically in brain cancers including glioblastoma.[5] Conventional two-dimensional (2D) cell or tissue culture models fail to elucidate the complexity of tumor growth, invasion and drug resistance in large part because they lack the 3D structure, cellular heterogeneity and dynamic cell-matrix interactions of native tumors.[6] Likewise, animal models offer more physiologically relevant models, but these models usually have interspecies discrepancies and reduced translatability to human disease. In comparison, 3D bioprinted tumor models enable researchers to replicate the structural, mechanical, and biochemical aspects of the TME that enable further accurate study of tumor progression, metastasis and responsiveness to therapy. Hydrogels have the ability to encapsulate multiple cell types and integrate biochemical gradients and are thus mimic these microenvironments more efficiently, thus providing new knowledge on cancer biology and unique treatment approach.[7]

Despite significant advances in hydrogel-based bioinks, there are still unresolved issues regarding the formation of functional brain tissues and physiologically relevant tumor models, related to construct maturation, long-term viability, and the integration of vascular-like networks. It is important to overcome these limitations in order to translate bioprinted systems from experimental platforms to clinically and biologically relevant models. Neural and tumor microenvironments are highly dynamic and their biochemical and mechanical fluctuations undergo a continuous set of changes that affect cellular behaviour and the progression of disease. This driven growing interest in smart, stimuli-responsive bioinks which have the ability to respond to local stimuli to support more biologically relevant tissue constructs and disease models.

The potential applications of the hydrogel-based smart bioinks are further emphasized by their use in drug discovery and evaluation. These systems have the ability to significantly reduce the use of animal models, enhance predictive power of preclinical research and expedite the discovery of therapeutic candidates by providing clinically relevant physiological models of tissues of the brain and brain tumours in 3D. This technology makes use of patent specific bioinks which in turn are derived from autologous cells. This helps in creating individualized disease models or therapeutic implants and hence holds immense advantage in the field of precision medicine.

Methodology of literature search

A comprehensive literature search was conducted using electronic databases including PubMed, Web of Science, and Scopus. Relevant keywords and their combinations included "hydrogel bioinks," "smart bioinks," "3D bioprinting," "brain tissue engineering," and "tumor microenvironment modelling." The search was restricted to peer-reviewed articles published in the English

language. Selection criteria prioritized studies and reviews focusing on the design, functionalization, and application of hydrogel-based smart bioinks for neural and cancer-related bioprinting applications. Editorials, conference abstracts, and articles not directly aligned with the scope of this review were excluded.

Hydrogel-based smart bioinks: properties and design considerations

Physicochemical properties

The functionality of the bioink is based mostly on the physicochemical properties of hydrogels. Soft tissues such as brain have elastic modulus values between 0.1-1 kPa, which require hydrogels that accurately mimic the mechanical properties environments to avoid adverse cellular reactions.[8] Hydrogel-based systems based on gelatin methacrylate (GelMA), hyaluronic acid (HA), and polyethylene glycol (PEG) allow fine adjustment of the stiffness by modifying the crosslinking density or polymer concentration. Rheological behaviour, shear-thinning behaviour and viscoelastic recovery are also important as they directly affect extrusion-based bioprinting.[9]

In addition to the mechanical properties, hydrogel swelling and degradation are crucial factors for governing the nutrient flow, elimination of waste, and the temporal stability of the printed components. To model tumor microenvironment, hydrogels must demonstrate degradability to facilitate cell invasion and their angiogenesis, but in the case of neural tissue engineering regulated degradation is essential for sustained tissue modelling. The integration of dynamic functionalities, such as pH, temperature, or reactive oxygen species (ROS) responsiveness, has also enabled traditional hydrogels to become smart bioinks that can respond to pathological signals. These controllable physicochemical characteristics allow hydrogels to mimic sophisticated tissue conditions and increase their application in biomimicking biofabrication.[10]

Biological compatibility

The success of hydrogel-based bioinks in tissue engineering applications depends on the biological performance of the material. Cytocompatibility is one of the fundamental parameters in this context that facilitate the survival and viability of encapsulated or seeded cells during and after printing. Hydrogels meant for brain tissue engineering often include biomimetic compounds such as laminin-derived peptides, RGD motifs, or HA backbones.[11] These help to facilitate neuronal adhesion, axonal growth, and synaptic connection. Similarly, in the case of tumor models, the hydrogels need to provide biochemical information that mimics the heterogeneity of the tumour microenvironment such as oxygen, nutrient and growth factor gradients.[12]

Bioactive molecules can also be stored in hydrogel as reservoirs. As an example, growth factors may be incorporated in bioinks and released over an extended time span, inducing neurogenesis or angiogenesis in printed constructs, e.g. brain-derived neurotrophic factor (BDNF) or vascular endothelial growth factor (VEGF). Increasingly, however, immunomodulatory characteristics of bioinks are also essential, especially in tumor modelling, where immune cell infiltration and cytokine signalling represent important facets of disease pathophysiology. Biofunctional design is important as robust immunomodulatory capabilities could be achieved, and sustained stability of scaffolds despite the presence of host immune responses may be ensured.

Another fundamental component of hydrogel bioink biology are the cell-matrix interactions. Unlike the synthetic scaffolds, natural hydrogels possess an inherent ability to maintain integrin binding and cell signalling. However, synthetic hydrogels may be functionalized to deliver comparable bioactivity, with added control of degradation rate, and mechanical properties. Therefore, hybrid systems that integrate natural and synthetic polymers are quite common due to their ability to combine biological components with finely adjustable physicochemical characteristics.[13]

Printability and structural fidelity

Besides material composition and biological compatibility, the success of hydrogel-based smart bioinks has also been linked to printability as well as their ability to maintain structural fidelity upon assembly. Printability is a measure of how readily a bioink can be extruded or deposited in fine-scale patterns and maintain cell viability. The desired characteristic of ideal hydrogels is their ability to undergo shear thinning under the influence of stress, allowing drop viscosity to decrease during delay and regaining stability with great speed. This property reduces the mechanical forces on the cells and results in high-resolution constructs.[14]

The structural fidelity is also crucial in specific applications, especially in brain tissue constructs when microarchitectural organization affects the connectivity of neurons, or in tumor models with spatial heterogeneity to promote disease progression. To accomplish this, crosslinking techniques of either photo-crosslinking, ionic bonding, or enzymatic reactions needs to be optimally adjusted. As an example, GelMA bioinks have a broad application because of the capacity to photo-crosslink and can be used to stabilize the printed construct rapidly and with customizable stiffness. Similarly, hydrogels derived by using alginate also employ ionic crosslinking but are usually mixed with other polymers to enhance their mechanical and biological properties.[15]

The viscosity and gelation dynamics of hydrogel are also related to the scalability of printing. Gelation that is too slow may cause structural collapse but excessively fast gelation can block nozzles and ruin resolution. A predominant technique now used is to integrate the pre crosslinking with post print stabilization, commonly referred to as hybrid approach. The ability of hydrogels to form highly complex functional tissue constructs can be achieved by the incorporation of sacrificial bioinks,

microfluidic printing and 4D bioprinting approaches.[16]

Design and development of smart bioinks

The rational engineering of smart hydrogel-based bioinks is a significant new development in three-dimensional bioprinting that allows printing constructs that are not only structurally compliant but also biologically reactive to complex tissue microenvironment.[17] In contrast to traditional bioinks, smart bioinks are designed to respond dynamically to biochemical and biophysical stimuli with printability and cytocompatibility, which is especially challenging when performing neural and tumour models. This balance is dependent on precise regulation over chemical functionalisation, cross linking designs and incorporation of bioactive factors all of which determine mechanical fidelity, cellular behaviour and long-term construct functionality.

Strategies for hydrogel functionalization

To convert traditional hydrogels into smart bioinks capable of guiding cell behavior and tissue-specific outcomes various functionalization techniques are essential. Natural polymers can be modified chemically by the introduction of reactive functional groups which can help regulate the spatiotemporal control of bioink behaviour.[18] Typical examples in this case are GelMA or the thiolation of hyaluronic acid. These modifications facilitate the inclusion of bioactive ligands, adhesion receptors as well as degradation-sensitive motifs, which enhance cell attachment, migration, and hydrogel matrix remodelling.[19]

The addition of stimuli-responsive groups, including pH, redox, or other stimulus responsible groups imparts bioinks with adaptive capabilities which is especially beneficial in in tumour modelling where microenvironmental variability is frequent.[20] More sophisticated conjugation methods also allow the coupling of signalling molecules or extracellular matrix derived peptides to the hydrogel scaffold, allowing specific control of any cell fate choices essential for neuronal development or tumor stroma interactions.

Crosslinking mechanisms: physical, chemical, and enzymatic

Crosslinking serves as a crucial mechanism for stabilizing hydrogel-based bioinks in the printing process, and post-printing phase which has a direct effect on construct fidelity, mechanical robustness and cellular response. Physical crosslinking methods including ionic interactions and hydrogen bonds provide mild and reversible gelation environments that are highly advantageous to cell survival often exhibit insufficient long-term stability.[21] The use of chemical crosslinking methods, especially photo-crosslinking using methacrylate functional groups, provide precise spatiotemporal regulation of gelation, and enables the creation of complex structures at high resolution level, but with the drawback of cytotoxic effects of photoinitiators and UV radiation. Enzyme crosslinking, which utilizes the actions of an enzyme, like the transglutaminase or horseradish peroxidase has evolved into a sustainable alternative that enables fast gelation under physiological conditions without the need of harsh reagents.[22]

Hybrid approaches are being developed to take advantage of the benefits of each of the formulation techniques which include physical, chemical and enzymatic crosslinking. These approaches help in optimizing various variables such as mechanical stiffness and degradation rates. This helps in developing hydrogels resulting in hydrogels that concurrently maintain printability, mechanical integrity, and cellular remodelling. Tumor models often require stiffer and more diverse networks to replicate the pathological microenvironment. This is in contrast to brain tissue engineering which requires softer crosslinked matrices that mimic native tissue elasticity.

Role of nanomaterials and growth factors in smart bioinks

The inclusion of nanomaterials and growth factors in the hydrogel-based bioinks has emerged as an effective strategy of improving mechanical behaviour, bio-functionality and of spatiotemporal regulation of cellular behaviour. Graphene oxide, nanoclays, silica nanoparticles and conductive polymers nanomaterials may be embedded into hydrogel matrices to increase the mechanical strength, shear-thinning behaviour and electrical conductivity, which is particularly important in neural tissue constructs where signal transmission and structural integrity is important. Nanoscale fillers can also be used to regulate the rheology of a hydrogel, to attain enhanced print fidelity but without excessive cellular damage caused by shear during the extrusion of printed objects.[23,24]

In addition to structural roles, nanomaterials are widely used to deliver bioactive molecules which can be localized and sustained in printed constructs using delivery of growth factors, cytokines or therapeutic agents. Growth factors like BDNF, VEGF and transforming growth factor-β (TGF-β) can be encapsulated in nanoparticle reservoirs or nanocomposite hydrogels to enable controlled release patterns, which resemble physiological signalling dynamics. This can be of particular benefit to brain tissue engineering where long term neurotrophic support is necessary to enhance neuronal survival, synaptic maturation and network stability, and tumour models where angiogenic and paracrine signalling are primary contributors to disease pathogenesis.

However, nanomaterials and growth factors integration also have critical design limitations that should be addressed. High loading of nanoparticles may negatively influence hydrogel porosity, diffusivity, cytocompatibility and unregulated release of growth factors can cause non-physiological signalling, loss of spatial gradients, or rapid depletion.[25] To overcome these limitations, strategies including the surface functionalisation of nanoparticles, tethering of growth factors using cleavable linkers, or encapsulation with degradable nanocarriers

have been developed, resulting in the ability to deliver bioactive cues with precise spatiotemporal control.

Within the framework of tumour microenvironment modelling, further insights into the mechanisms of therapeutic response and resistance can be examined using nanomaterial supported smart bioinks. A nanocomposite hydrogel can also be designed to potentially include chemotherapeutic agents or immunomodulators and thus modelling of tumour progression and treatment response may be effectively performed in a three-dimensional, physiologically relevant hydrogel can also be achieved. These systems provide a distinct advantage compared to conventional models of culture through the ability to allow localized exposure of drugs, gradient formation, and dynamic cell matrix interactions that influence treatment efficacy.

Printability, rheology, and shape fidelity considerations

The practical effectiveness of smart hydrogel-based bioinks is determined not solely by their biological functionality but also by their printability and structural integrity during and post-bioprinting. Rheological behaviour ranks as one of these factors that is highly significant for deciding whether a bioink can be accurately deposited while maintaining cells viability. Shear-thinning behaviour, which involves a decrease in viscosity on application of stress, is an important requirement for extrusion-based bioprinting as it permits smooth extrusion through the nozzle. Once the material is deposited, the rapid recovery of the viscosity is essential to maintain the printed structure. This causes a reduction in cellular damage caused by shear while enabling high-resolution production. This behavior directly influences the fabrication of well-defined filament architectures and grid-like constructs with high spatial fidelity, as shown in Fig. 1. The printed structures exhibit uniform filament deposition with consistent strut diameter (~432 ± 27 μm) and inter-filament spacing (~1000 μm), closely matching the designed geometry. Additionally, minimal filament spreading and effective shape retention enable stable multilayer stacking and reproducible large-area constructs. The successful formation of grid and multi-well architectures highlights the excellent printability, dimensional accuracy, and self-supporting capability of the optimized hydrogel bioink.[26]

The kinetics of gelation are equally significant as the flow behaviour when considering shape fidelity. If the gelation process is too slow, it can cause the filament to spread out and the structure to fall apart whereas excessively rapid process can clog the nozzles and reduce precision. To resolve this, multi-stage crosslinking strategies are increasingly being adopted that combine preprint viscosity tuning with post print stabilization.[27] Despite these improvements, it is challenging to find a balance between biological and mechanical requirements. Soft hydrogels are ideal for neural and tumor models due to their biomimetic properties, but they are difficult to print with high structural accuracy. Thus, rheology modifiers such as nanoclays and temporary physical networks are incorporated to improve the viscoelastic recovery without permanently increasing the stiffness.[28]

Additionally, the rates of degradation must be carefully synchronized with the cellular activity. Rapid degradation can compromise structural integrity prior to tissue maturation, whereas highly stable matrices can prevent cell migration and extracellular matrix remodelling. Therefore, smart bioinks must be designed to enable gradual transition from material-supported to cell-driven tissue architecture. In general, printability and shape fidelity shouldn't be considered of as separate factors. Instead, they should be considered of as part of a dynamic interaction between the properties of the material, the conditions of printing, and biological functionality. Optimizing these interdependent factors is essential for achieving reproducible and high-fidelity bioprinted constructs.[29] The overall design framework of hydrogel-based smart bioinks for 3D bioprinting, highlighting the relationship between material selection, bioink design strategies, resulting properties, and biomedical applications, is summarized in Fig. 2.

Hydrogel-based bioinks for brain tissue engineering

The human brain can be described as one of the most complex organs known to science, as it is characterized by an exceptional cellular heterogeneity, a highly organized architecture, and a strictly controlled biochemical environment.

In contrast to peripheral tissues, the central nervous system has a limited regenerative power, which can result in permanent functional deficits after injuries or

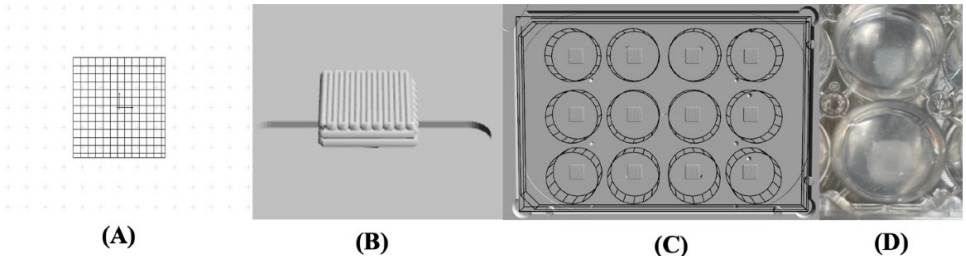


Fig. 1. Bioprinting process and structural fidelity of hydrogel constructs. (A) Computer-aided design (CAD) of the scaffold generated using BioCAD™ software. (B) 3D-rendered model illustrating the designed architecture. (C) Printed construct within a multi-well plate demonstrating spatial accuracy and reproducibility. (D) Cell-laden hydrogel printed within an agarose support bath, maintaining structural integrity during deposition. The results highlight precise filament deposition and faithful replication of the digital design using extrusion-based bioprinting. Adapted from Sousa et al licensed under CC BY 4.0.[26]

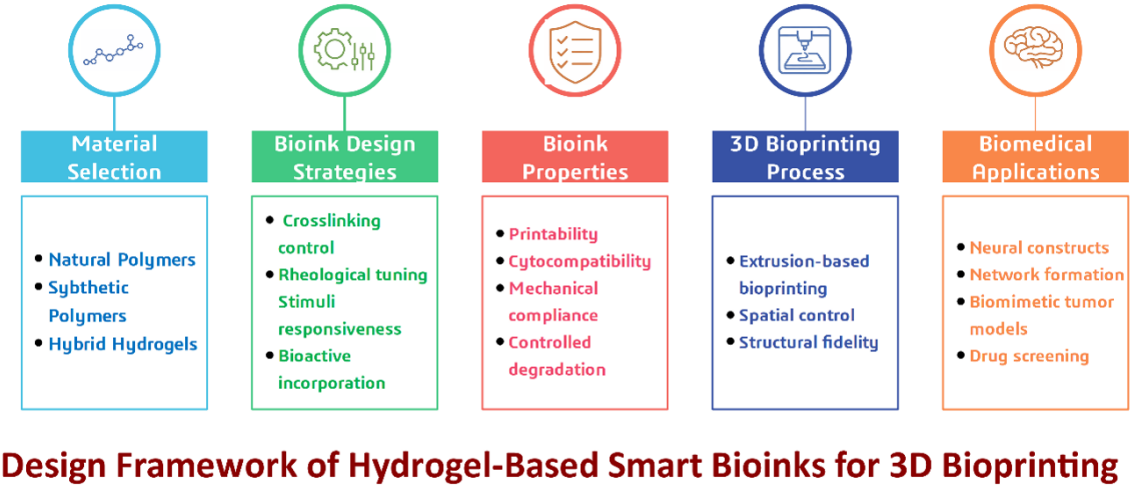


Fig. 2. Schematic representation of the design framework for hydrogel-based smart bioinks used in 3D bioprinting, illustrating the relationship between material selection, bioink design strategies, resulting physicochemical properties, bioprinting process, and biomedical applications in brain tissue engineering and tumor microenvironment modelling.

neurodegenerative disorders.[30] Therefore, brain tissue engineering has turned out to be a viable strategy to restore damaged neural structures and to promote functional recovery. Hydrogel based bioinks provide an appealing platform, given that they possess the capacity to replicate the soft, hydrated and bioactive conditions of native brain tissue. Bioinks that are capable of maintaining neuronal integrity, driving axonal growth and reconstituting the complexity of cytoarchitecture of cerebral networks can be developed by integrating structural versatility of hydrogels with advanced 3D bioprinting technologies.[31]

Challenges in brain tissue regeneration

Brain tissue regeneration presents fundamental challenges that distinguish it from regenerative strategies applied to other organs. Neurons in the central nervous system exhibit a limited intrinsic capacity for self-repair, and damage is frequently accompanied by the formation of inhibitory glial scars that restrict axonal regeneration and disrupt network reorganization. These biological constraints significantly limit functional recovery following injury or neurodegenerative processes.[32]

Besides the lack of regenerative capacity, there are other barriers to tissue engineering approaches generated by the specific mechanical and immunological properties of the brain. Neural tissues are highly viscoelastic and low in stiffness and implanted or engineered materials have to be very similar in these characteristics, otherwise mechanical incompatibility, inflammation and foreign body reactions can occur. Minor mechanical deviation in compliance can negatively impact on neuronal survival, differentiation and integration with host tissue.

The regeneration of brain tissue requires effective biomaterial platforms thus necessitating the availability of biomaterials to support the tissue in addition to the fine tuning of mechanical compatibility, immune tolerance and long-term integration of functions. These obstacles continue to be among the major focus of the development of the hydrogel-based bioinks to be used in neural repair and in modelling.

Biomimetic hydrogel scaffolds for neuronal growth

Biomimetic hydrogel scaffolds serve as an ideal platform for brain tissue engineering due to their ability to replicate hydration, compliant and viscoelastic characteristics of the neural extra cellular matrix.[33] Other than providing passive structural support, these scaffolds are also involved in regulating neuronal adhesion, migration and network assembly through controlled mechanical and biochemical signals that govern cell-matrix interactions in the central nervous system.[34]

The stiffness of the matrices is a critically important parameter of neuronal behaviour in-gel-scaffolds. This needs to be limited to a small physiological range to enable neurite proliferation and synaptic activity. Excessively stiff matrices inhibit axonal growth and inhibit synaptic transmission, whereas excessively flexible hydrogels do not provide sufficient mechanical for cell alignment and network stability.[35] Apart from elastic modulus, viscoelastic properties such as stress relaxation and time-dependent deformation also play an important role to stimulate neuronal survival and enhance axonal regeneration.[36] Hydrogels that are designed to support viscoelasticity of the brain have shown enhancement of neurite outgrowth and synaptic connectivity in comparison to purely elastic systems.[37]

To further enhance scaffold biomimicry, extracellular matrix-derived biochemical motifs are incorporated to provide cell-adhesive and signalling signals required by neurons to cells during their maturation. Laminin-mimetic peptides and integrin-binding ligands mediate neuronal adhesion, neurite direction, and strengthen synapse formation and hyaluronic acid-based backbones help maintain native neural phenotypes.[38] The presentation of these biochemical signals must be integrated with suitable mechanical properties to facilitate synergistic regulation of neuronal behaviour.

Microarchitectural structure is another mechanism of control in biomimetic hydrogel scaffolds for neural applications. Recent advances in three-dimensional bioprinting can now enable the fabrication of spatially patterned constructs that incorporate aligned fibers, microchannels or layered geometries, which in turn directs neuronal orientation, and connectivity. Such architectures are particularly useful in replicating the anisotropic structure of nervous tissue, and facilitating long range axon guidance. Aligned anisotropic hydrogels can be fabricated using an aspiration–ejection technique, which induces directional organization of collagen fibrils. The perfusable microchannels also enable the diffusion of nutrients and elimination of waste, which contributes to long term neuronal viability and functional maturation.

Overall, biomimetic hydrogel scaffolds can be effectively used for neuronal growth, though their success depends on the combination of mechanical compliance, viscoelastic behavior, biochemical signalling and architectural guidance on a single platform. By systematically maintaining these parameters, hydrogel-based bioinks can sustain the formation of structurally organized and functionally active neural networks thereby contributing to the advancement of physiologically relevant brain-tissue models and regenerative therapies.

The key design elements and biological outcomes associated with smart hydrogel bioinks for brain tissue engineering are summarized in Fig. 3.

Incorporation of neural stem cells and signalling cues

The incorporation of viable cellular components into bioinks in hydrogel form is a key requirement to the creation of functional brain tissue constructs, and neural stem cells (NSCs) have become a target cellular source. NSCs have the property of self-renewal, as well as the potential of multilineage differentiation.[39] These abilities give rise to neurons, astrocytes and oligodendrocytes thus facilitating the recreation of cellular heterogeneity that can be found in normal neural tissue. The encapsulation of these cells hydrogel mediums not only provides mechanical protection to the bioprinting procedure, but also provides a permissive environment that supports cell survival and early differentiation.[40]

Besides basic encapsulation, developmental pathway and functionalisation of NSCs is highly controlled by the spatial-temporal configuration of biochemical signalling signals incorporated into the hydrogel scaffold. The neurotrophic factors, including BDNF and nerve growth factor (NGF), are relevant to the survival of neurons, axonal proliferation and synaptic regeneration, whereas angiogenic mediators enhance construct survival indirectly by enhancing vascular integration.[41]

Similar significance is given to biophysical signals included in the hydrogel meshwork, including stiffness gradient, viscoelasticity and topographical characteristics. Minor variations in matrix mechanics have been shown to influence the development of NSC. Softer microdomains preferentially induce neuronal phenotypes, whereas stiffer microenvironments promote astrocytic lineage differentiation. This mechanosensitive reaction emphasizes the need for spatially diverse hydrogel structures that more accurately simulate the mechanical environment of native brain tissue.[42]

The simultaneous integration of biochemical and biophysical stimuli into a single hydrogel substrate enables precise control of neuronal development pathways and tissue structure. Advanced bioinks can promote the formation of organized neural networks with region specific

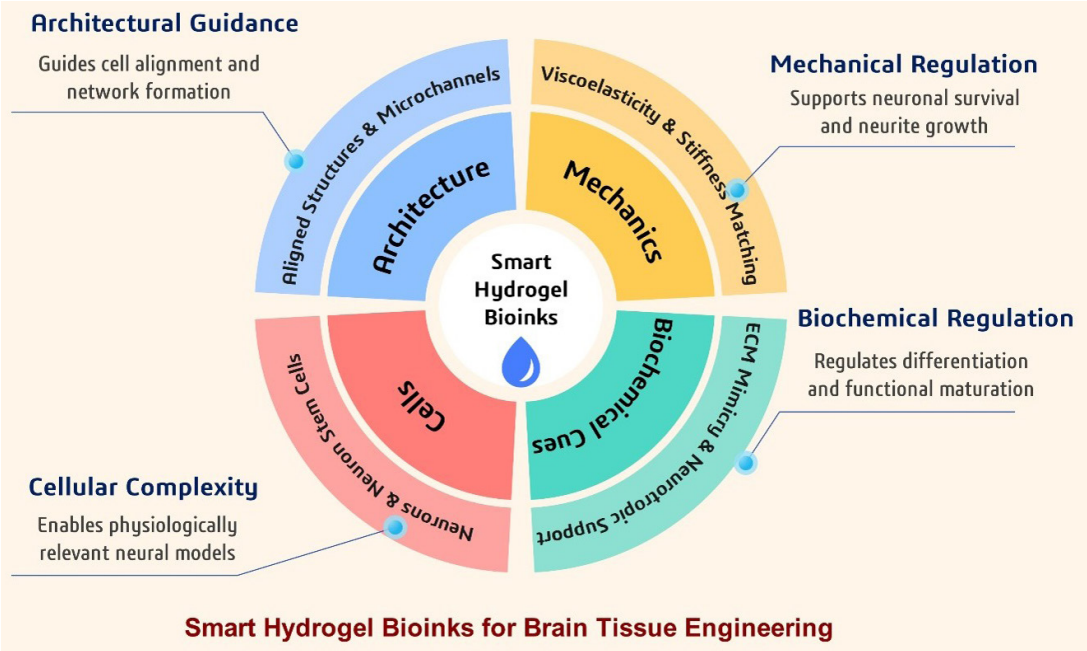


Fig. 3. Two-layer circular schematic illustrating the key design domains of smart hydrogel-based bioinks for brain tissue engineering. The inner ring represents primary bioink design cues, while the outer ring highlights essential attributes related to mechanical regulation, biochemical signalling, cellular composition, and structural architecture that collectively support functional neural outcomes.

cellular composition and functional interconnectivity.[43] This may be achieved by synchronizing growth factor distribution and mechanical patterning. The methodologies are of particular importance to the recapitulation of neurodevelopmental dynamics, neurodegenerative pathology, and injury-related remodelling, in which highly controlled emulation of cell-cell and cell-matrix crosstalk is essential.

Together, hydrogel based bioinks, which includes NSCs in addition to precisely calibrated signalling cues, comprise a versatile platform for neural tissue-engineered tissues with enhanced biological fidelity. Continued optimization of the cell-instructive hydrogel design is further expected to enhance maturation, stability, and translational viability of bioprinted brain tissue constructs.[44] Representative studies highlighting the application of hydrogel-based bioinks in brain tissue engineering, including both in vitro and in vivo models, are summarized in Table 1.

Hydrogel-based bioinks for tumor microenvironment

The tumor microenvironment is an important determinant of oncogenesis, disease progression, and resistance to therapy due to its ability to mediate complex interactions between neoplastic cells, stromal fibroblasts, immune components, and vascular networks. Typical characteristics such as dysregulated extracellular matrix remodelling, hypoxia, acidosis and immune suppression are integrated to regulate tumor biology and determine therapeutic responses. Thus, accurate replication of the dynamic and heterogeneous environments is essential for developing physiologically relevant cancer models.

Tumor heterogeneity and limitations of 2D models

Intratumoral heterogeneity poses a significant challenge in oncology as malignant lesions comprise of heterogeneous units of cells which exhibit discrete genetic signatures, metabolic phenotype and react to therapy differently. These malignant cells coexist with stromal and immune components of a complex interacting environment that coordinates tumor development and treatment responses.[49]

Conventional two-dimensional culture models are not sufficient for replicating this complexity because tumor cells are grown on rigid surfaces under homogeneous nutrient and oxygen supplies that inadequately resemble the in-situ tumour conditions. These artificial conditions disrupt cellular morphology, polarity and transcriptomic phenotypes, reducing their predictive ability of pharmacodynamic effects and resistance mechanisms. Consequently, many drugs demonstrating effectiveness in planar models do not successfully transition to clinical applications.[50] A schematic comparison highlighting the limitations of conventional 2D models and the enhanced biomimetic capabilities of 3D hydrogel-based tumor models is presented in Fig. 4.

Hydrogels for replicating ECM stiffness, hypoxia, and nutrient gradients

The extracellular matrix of solid tumours plays an active and dynamic role in oncogenic development which goes beyond mere passive structural support. Tumour related ECM is often characterized by increased rigidity which can be explained by excessive collagen deposition, enzyme crosslinking and aberrant matrix remodelling that has far-reaching consequences on cellular behaviour.[51] Increased matrix rigidity facilitates integrin-based signalling cascades, cytoskeletal reorganisation and enhances epithelial -mesenchymal transition, thus contributing to

Table 1. Studies using hydrogel-based bioinks in brain tissue engineering and tumor modelling

Study	Hydrogel composition	Cell type/ species	Model	Experimental level	Key outcomes	Reference
Injectable conductive silk-fibre/MXene hydrogel for neural stem cells mediated brain repair	Silk fibroin + MXene nanosheets hydrogel with neural stem cells	Rat neural stem cells; traumatic brain injury rats	Brain injury model	In vitro + in vivo	Enhanced axonal growth, reduced glial scar formation, increased neuronal differentiation, improved motor recovery and lesion repair	45
Capillary alginate hydrogel seeded with neural stem/ progenitor cells for spinal cord repair	Ca^{2+} cross-linked alginate hydrogel with capillary like channels	Rat embryonic day 14 NSPCs; adult rats (T8 transection SCI)	Spinal cord injury model	In vitro + in vivo	38.3 % cell survival; 40.7 % neurons, 26.6 % astrocytes; axonal extension across lesion; restored electrophysiological conduction and locomotor function	46
Conductive composite hydrogel scaffold for spinal cord injury repair	A novel conductive polymer within gelatin/PEG matrix	Neural stem cells; rat spinal cord injury model	Spinal cord injury model	In vitro + in vivo	Promoted neurite elongation and synaptic contact; reduced glial scarring; improved neuronal differentiation and functional recovery	47
Architected GelMA/ PEGDA hydrogels with microchannels for vascularization	GelMA + PEGDA printed with 0.5–1 mm channels	Murine subcutaneous implantation	Vascularization model	In vivo	1 mm channels showed optimal blood-vessel infiltration and perfusion; demonstrated importance of microarchitecture for viability	48

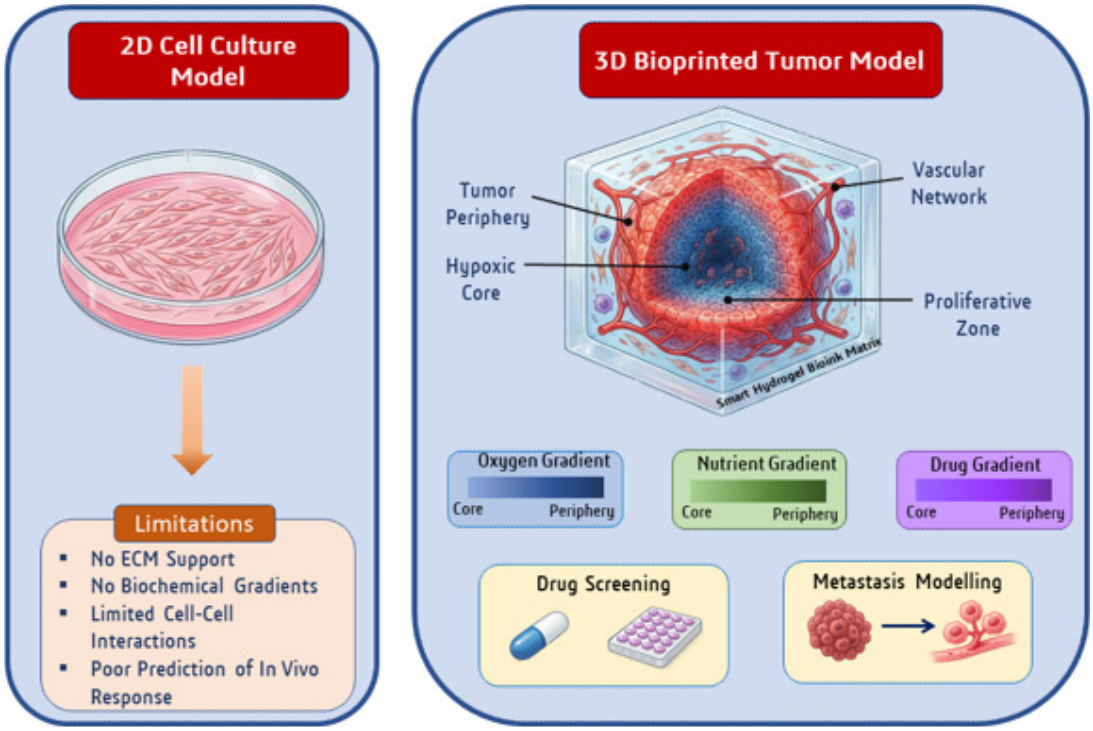


Fig. 4. Comparison of 2D monolayer culture and 3D bioprinted tumor models. The 3D smart hydrogel bioink-based system replicates tumor structure, cellular interactions, and oxygen, nutrient, and drug gradients, enabling more physiologically relevant drug screening and metastasis modelling compared to conventional 2D systems

invasive and metastatic phenotypes.[52]

Hydrogel-based bioinks provide a flexible medium for simulating pathological mechanical signals by precisely adjusting polymer concentration, cross-linking density, and composite formulation. Bioprinted tumor models may be designed to simulate various malignancies, by adjusting matrix stiffness within physiologically relevant ranges. These systems enable systematic examination of mechano transduction signalling and its role in tumour cell proliferation, migration and chemotherapeutic resistance with insights that are difficult to achieve in conventional culture counterparts.[53]

Hypoxia is another defining characteristic of the tumour microenvironment, which accompanies modified ECM mechanics, which is caused by abnormal vascular structure and inadequate perfusion. Oxygen deprivation stabilises hypoxia-influencing factors, thus initiating transcriptional programmes to stimulate angiogenesis, metabolic restructuring, immune evasion, and therapeutic resistance. Modulable hydrogel systems are able to simulate hypoxic gradients by regulating diffusional characteristics, construct thickness, or by incorporating oxygen-absorbing components. They thus help in creating spatially heterogeneous microenvironments that are more accurately reflect the tumour physiology.[54]

Establishment of regulated concentrations of nutrients and metabolic by products also contributes to the increase in physiological importance of hydrogel-based tumour models. Nutrient deprived neoplastic cells in these constructs exhibit adaptive metabolic changes, which resemble those in invasive tumours such as enhanced glycolysis and modified mitochondrial activity. Drug penetration and efficacy is also modulated by these gradients which makes it possible to estimate treatment responses within clinically relevant transport constraints.

In addition to mechanical and metabolic signals, hydrogels may be functionalised with proteolytically degradable signals as well as adhesive ligands that promote tumour cell invasion and stromal interaction. These kinds of dynamic matrices enable cancer cells to reorganize the surrounding environment and facilitates group migration and interaction with endothelial and immune components. By combining stiffness modulation, hypoxia, and biochemical signals on a single platform, cell hydrogel-based bioinks provide a potent tool in simulating the complex and dynamic tumour microenvironment.[55]

In addition to mechanical and metabolic cues, the tumor microenvironment is critically governed by dynamic biochemical crosstalk mediated through paracrine signaling. Tumor cells continuously interact with surrounding stromal components, including cancer-associated fibroblasts, immune cells, and endothelial cells, through the secretion of cytokines, growth factors, and extracellular vesicles. These signalling pathways regulate key processes such as tumor proliferation, invasion, angiogenesis, and therapeutic resistance.

Smart hydrogel-based bioinks offer a unique advantage in replicating this dynamic communication by enabling co-culture systems and controlled presentation of bioactive molecules within three-dimensional constructs. By incorporating multiple cell types and programmable release of signaling factors, these platforms can more accurately mimic the evolving tumor microenvironment compared to models that rely solely on static physical

parameters. However, achieving precise spatial and temporal control of these signaling gradients remains a significant challenge and represents an important direction for future research. Therefore, integrating both physical and biochemical signaling mechanisms is essential for developing physiologically relevant tumor models.

3D Bioprinting of cancer-on-a-chip models

Recent developments in three-dimensional bioprinting have significantly expanded the functional range of hydrogel-based bioinks that can be used to assemble tumour models that exhibit a highly organised spatial structure with cellular heterogeneity. The diverse fabrication strategies employed in hydrogel-based 3D bioprinting play a pivotal role in defining the structural complexity, mechanical integrity, and biological functionality of engineered tumor models. As illustrated in Fig. 5, multiple advanced bioprinting approaches have been developed to achieve precise spatial organization and microenvironmental mimicry. Embedded bioprinting techniques (Fig. 5a) enable the fabrication of channel-integrated tumor constructs within a supportive matrix, allowing the incorporation of perfusable features that closely resemble tumor–vascular interfaces. Such architectures facilitate the establishment of physiologically relevant gradients of oxygen, nutrients, and signaling molecules, which are critical determinants of tumor progression and therapeutic resistance.[56]

Support-bath-based bioprinting strategies (Fig. 5b) further enhance structural fidelity by enabling the deposition of low-viscosity or soft hydrogel bioinks within a stabilizing medium. This approach minimizes deformation during printing and allows the generation

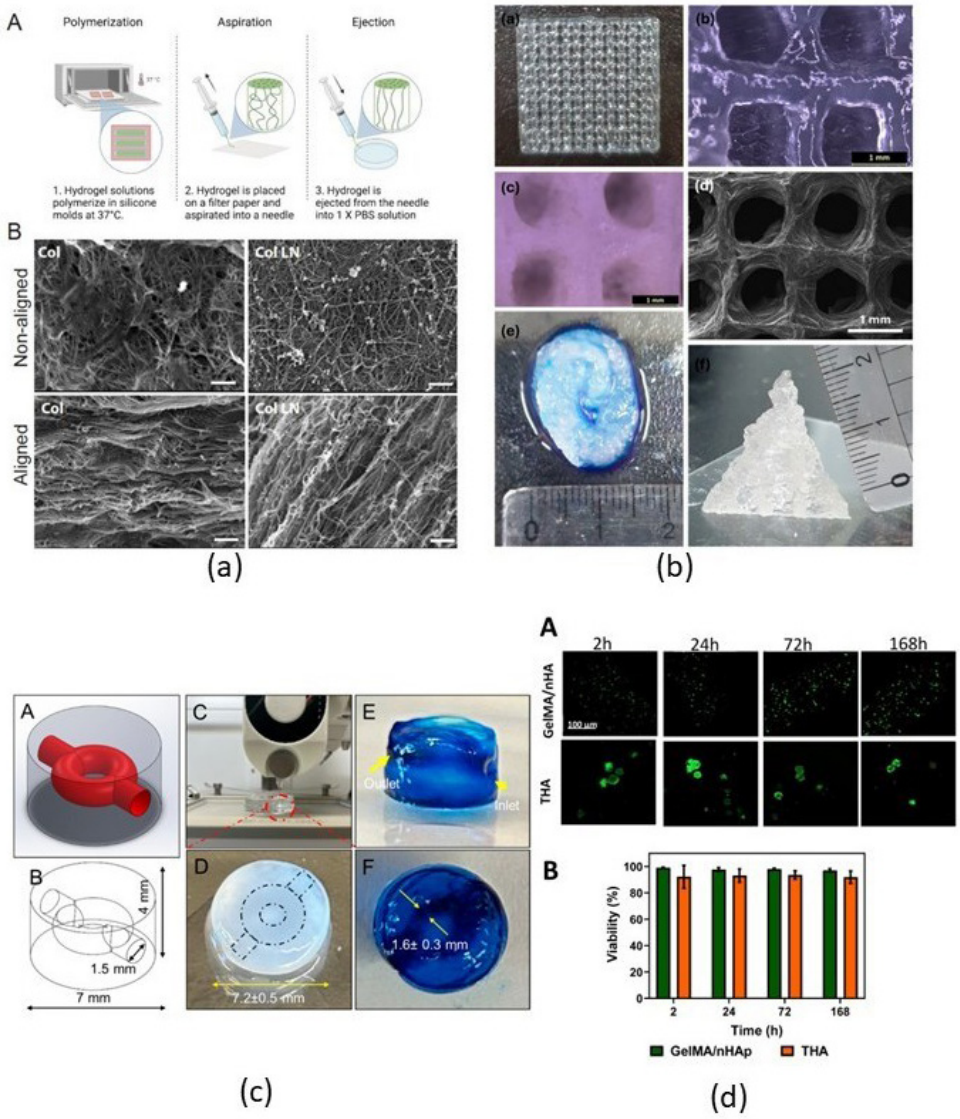


Fig. 5. Representative examples of extrusion-based bioprinting patterns demonstrating printability and structural fidelity of hydrogel bioinks. (a) Grid-based filament deposition and aligned microstructure illustrating controlled anisotropic architecture formation; (b) layer-by-layer extrusion showing continuous filament alignment and structural fidelity of self-healing hydrogel constructs; (c) multi-well printing layout demonstrating precise spatial control in bioprinted glioblastoma models; (d) printed hydrogel constructs in well-plate format with sustained structural integrity and cell viability. These images highlight the ability of optimized bioinks to maintain geometric precision, reproducibility, and biological functionality during fabrication. Adapted from Severs et al, Nagaraja et al, Schroyer et al and Jahangir et al; licensed under the Creative Commons Attribution (CC BY 4.0) license.[56-59]

of complex, free-form geometries that would otherwise collapse under gravitational forces.[57] The ability to maintain geometric precision during fabrication is particularly important for recreating heterogeneous tumor architectures, including necrotic cores and proliferative outer regions. In parallel, extrusion-based bioprinting approaches (Fig. 5c) demonstrate the formation of high-resolution constructs with controlled filament deposition and layer-by-layer assembly. The fidelity of these structures is strongly governed by bioink rheological properties, including shear-thinning behaviour and rapid post-printing recovery, as well as crosslinking kinetics that stabilize the printed architecture. These features enable reproducible fabrication of scaffolds with defined porosity and interconnectivity, which are essential for cell infiltration, nutrient transport, and waste removal.[58] Importantly, the functional relevance of these fabrication strategies is validated by their ability to support sustained cellular viability over extended culture durations (Fig. 5d). Live/dead staining analysis demonstrates consistently high viability across multiple time points, indicating that the hydrogel matrices provide a protective microenvironment that mitigates shear-induced cellular damage during printing while supporting long-term cellular activity. This sustained viability is crucial for modeling dynamic tumor behaviours, including proliferation, invasion, and drug response.[59]

Collectively, these integrated fabrication and validation approaches highlight the versatility of hydrogel-based bioinks in generating structurally and biologically relevant tumor models. By combining architectural precision with cytocompatibility and functional stability, these systems provide a robust platform for replicating key features of the tumor microenvironment and improving the predictive accuracy of in vitro cancer models.

Unlike the conventional three-dimensional cell culture models, bioprinting helps to engineer the precise location of malignant cells, stromal fibroblasts, endothelial cells, and immune components in architecturally defined structures. This spatial accuracy is extremely necessary to replicate the tumour-stroma and tumour-vasculature interaction which regulates invasion, angiogenesis and therapeutic responsiveness.[60]

Cancer-on-a-chip designs combine bioprinted hydrogel models with microfluidic systems to create dynamic tumour micro environments that more accurately replicate in vivo conditions. Microfluidic perfusion has the benefit of providing continuous supply of nutrients and oxygen and, at the same time, eliminates the metabolic waste, hence maintaining the long-term survival of tumour tissues. Additionally, the flow of fluids in these systems creates physiologically relevant shear stress and creates concentrations gradients of soluble factors and pharmacological agents, which makes it possible to explore transport-limited penetration and heterogeneous treatment effects.[61] Cancer-on-a-chip systems based on hydrogels have been used to mimic major properties of solid tumours, such as invasive growth factors, hypoxia-associated signalling, and abnormal vascularisation. Invasion patterns and resistance mechanisms replicated in bioprinted hydrogels with perfusable microchannels that mimic orthotopic tumours have been shown in glioblastoma models.[62] Similarly, it has been shown by breast and colorectal cancer-on chip assays that extracellular matrix rigidity and interstitial flow affect chemoresistance and metastatic capacity. The great flexibility of cancer-on-a-chip platforms is a significant benefit in personalized medicine. Bio printable hydrogel matrices can be loaded with patient-derived tumor cells to form personalized disease models that are genetically heterogeneous and have clinically relevant phenotypes. These constructs which are patient-specific allow comparative analysis of therapeutic regimens, prediction of drug sensitivity and investigation of resistance encountered in a controlled, physiologically-relevant setting.[63]

Moreover, the integration of immune cells into cellular multi-culture on a chip cancer models has opened up new opportunities to study tumour-immune interactions and provide immunotherapeutic solutions. These models provide the capability to create a predictive system to measure immune infiltration, cytokine signalling and responsiveness to immunomodulators by facilitating the co-culture of tumour with immune components in a spatially resolved drive-to-fate format with dynamical flow conditions. Together, three-dimensional bioprinted cancer-on-a-chip platforms constitute a paradigm shift in the sphere of translational oncology, thus filling the gap between simple in vitro tests and the complexity of in vivo experiments.[64] Key studies demonstrating the use of hydrogel-based bioinks for tumor microenvironment modelling are summarized in Table 2.

Advanced applications and integrations

Hydrogel-based smart bioinks have evolved beyond their initial role as structural scaffolds to become multifunctional platforms capable of replicating complex physiological and pathological processes. These advanced systems integrate biological responsiveness, spatial control, and dynamic functionality, enabling more accurate modelling of tissue behavior and disease progression. However, their successful application depends on the ability to coordinate multiple parameters, including cellular interactions, vascularization, and computational design.

Co-culture and vascularization strategies

One of the primary limitations of engineered tissue constructs is the lack of functional vascular networks, which restricts oxygen and nutrient diffusion in dense hydrogel systems. Smart bioinks address this challenge by enabling co-culture strategies that incorporate endothelial cells alongside parenchymal and stromal cells to promote angiogenic signaling and microvascular organization.

Recent advances in bioprinting have enabled the fabrication of perfusable microchannels using sacrificial bioinks and microfluidic-assisted printing techniques.

Table 2. Representative studies using hydrogel-based bioinks in tumor microenvironment modelling

Study	Hydrogel composition	Cell type	Model system	Experimental level	Key outcomes	Reference
3D bioprinted perfusable hydrogel model of glioblastoma	Fibrin-based bioink with endothelial-lined perfusable channels	Patient-derived glioblastoma cells, astrocytes, microglia, HUVECs	Microfluidic tumor model (cancer-on-a-chip)	In Vitro (microfluidic chip)	Reproduced tumour invasion fronts, hypoxia gradients and drug responses resembling orthotopic GBM; improved drug-penetration modelling	[65]
GelMA/hyaluronic-acid hydrogel for biomimetic GBM culture	GelMA + hyaluronic acid composite	Glioblastoma stem-like cells	3D tumor culture model	In Vitro	Supported proliferation, maintained stemness, and modulated phenotype depending on stiffness; enhanced physiological relevance vs 2D culture	[66]
GSC-laden 3D bioprinted hydrogel co-cultured with HUVECs	Gelatin/alginate/fibrinogen hydrogel with endothelial co-culture	Glioma stem cells + HUVECs; xenograft mice	Tumor angiogenesis model	In Vitro + In Vivo	Enhanced angiogenesis, $CD105^{+}$ endothelial infiltration, and vascular lumen formation; xenografts resembled human glioma vasculature	[67]

These structures facilitate dynamic perfusion, thereby improving long-term cell viability and mimicking physiological transport conditions.

Despite these advances, achieving stable and fully functional vascular integration remains a major challenge. Issues such as incomplete endothelial maturation, limited perfusion efficiency, and poor integration with host vasculature continue to hinder clinical translation. Future strategies should focus on combining bioactive signaling cues with optimized architectural design to achieve robust and scalable vascular networks.[68]

Integration with organoid technologies

Organoid systems have emerged as powerful tools for modeling human development and disease; however, their inherent lack of structural control often results in variability in size, organization, and functional maturity. The integration of hydrogel-based smart bioinks provides a means to overcome these limitations by offering defined extracellular matrix analogues that support spatial organization and mechanical stability.

Bioprinted hydrogel scaffolds can guide the positioning and growth of organoid structures, enabling improved reproducibility and controlled tissue architecture. In neural applications, this approach enhances network formation and functional connectivity, while in tumor models it preserves cellular heterogeneity and microenvironmental interactions.

Nevertheless, challenges remain in achieving full maturation and long-term stability of organoid–bioink hybrid systems. Standardization of bioink composition and optimization of cell–matrix interactions are essential to ensure reproducibility and translational relevance.[69]

AI-assisted design and simulation in hydrogel bioprinting

The design of smart bioinks involves multiple interdependent parameters, making conventional trial-and-error approaches inefficient and resource-intensive. Artificial intelligence (AI) and machine learning have emerged as promising tools to address this complexity by enabling data-driven optimization of bioink formulations and bioprinting parameters.

Recent studies have begun to demonstrate the practical application of artificial intelligence in bioink design and optimization. For instance, machine learning models have been employed to predict relationships between hydrogel composition, crosslinking density, and rheological properties, enabling identification of formulations with optimal printability and cell viability. Additionally, data-driven approaches have been used to optimize extrusion parameters such as pressure, nozzle diameter, and printing speed to minimize shear-induced cell damage while maintaining structural accuracy.[70,71] Furthermore, supervised learning algorithms have been applied to integrate large datasets of material properties and biological responses, allowing predictive modeling of bioink performance under varying conditions. Such approaches improve reproducibility and facilitate rational design of application-specific bioinks.[72]

Despite these promising developments, the application of AI in bioink design remains at an early stage. Limitations such as the lack of standardized datasets, variability in experimental protocols, and challenges in model generalization must be addressed. Future research should focus on integrating AI with high-throughput experimentation to develop predictive and scalable biofabrication platforms.

Drug screening and personalized medicine using hydrogel-based models

Hydrogel-based smart bioinks have emerged as advanced platforms for drug screening due to their ability to recreate physiologically relevant three-dimensional microenvironments. These systems allow evaluation of drug efficacy, toxicity, and resistance under conditions that more closely resemble in vivo tissue architecture compared to conventional two-dimensional models.

The incorporation of patient-derived cells into bioink

matrices enables the development of personalized disease models that capture individual genetic and microenvironmental variability. Such platforms facilitate comparative assessment of therapeutic regimens and support precision medicine approaches.

However, several challenges limit their widespread adoption, including scalability, reproducibility, and integration with high-throughput screening systems. Additionally, regulatory considerations and standardization of model systems remain critical barriers. Addressing these issues will be essential for translating hydrogel-based platforms from research tools to clinically relevant drug development systems.[73]

Challenges, Limitations, and Future Perspectives

As much as significant progress has been made in hydrogel-based bioinks in brain tissue engineering and tumor microenvironment modelling, there are still various challenges that limit the potential towards translation. One of the main technical constraints is the creation of high resolution bioprinting with reproducibility across a variety of platforms.[74] Although extrusion based bioprinting has found extensive use in hydrogel deposition because of its flexibility, it often compromises resolution and cell survival because of shear stress.[75] On the other hand, the resolution of techniques like digital light processing and inkjet bioprinting is much superior, but is limited by the range of hydrogels that these techniques can operate with, especially those with the increased viscosities needed to recapitulate the complex brain extracellular matrix. In addition, the reproducibility of mechanical properties, degradation rates and bioactivity of hydrogel formulations across batches is a significant issue because a slight deviation may lead to deviations in cellular responses.[76]

The success of hydrogel bioinks is also hindered by biological constraints. The key issue in maintaining long-term cell viability in printed constructs is that cells demand a very finely tuned balance of nutrients, oxygen and waste clearance, which is hard to maintain in large hydrogel scaffolds. Lack of an efficient vascularization of engineered brain tissue of tumor models is one of the most serious bottlenecks. In the absence of functional networks of blood vessels, cells located in the inner parts of constructs develop hypoxia and nutrient deprivation, leading to necrosis and dysfunctional tissue functioning.[77] Moreover, although bioinks can be modified to re-create the extracellular matrix, recapitulating brain or tumor microenvironment biochemical and biophysical complexity is not yet complimented by the current technological advancement. There is the added complexity of immune response, where implanted hydrogels can induce inflammation, fibrosis, or rejection to limit long-term integration and performance in vivo.[78]

In addition to technical and biological limitations, economic, manufacturing, regulatory, and ethical challenges represent significant barriers to the clinical and industrial translation of hydrogel-based smart bioinks. The incorporation of bioactive components such as growth factors, peptides, nanoparticles, and stimuli-responsive elements substantially increases production costs, limiting their feasibility for large-scale applications. Furthermore, the complexity of multi-component bioink formulations poses challenges in achieving batch-to-batch reproducibility, quality control, and long-term storage stability.[79]

Bioengineered tissues and bioprinted constructs have yet to receive widespread regulatory approval, as current evaluation frameworks are not fully equipped to assess highly complex, patient-specific living systems. Issues related to long-term safety, reproducibility, and quality assurance further hinder clinical translation. From a manufacturing perspective, scaling up hydrogel-based bioinks from laboratory-scale prototypes to industrial production remains a critical challenge. Factors such as sterilization compatibility, process standardization, and preservation of biological functionality during large-scale processing must be carefully addressed.

Ethical considerations also play a crucial role, particularly in brain tissue models where the potential for higher-order neural activity raises philosophical and regulatory concerns. Similarly, patient-derived tumor models require careful handling of issues related to consent, privacy, and data security. Addressing these interconnected challenges will be essential to enable the successful translation of smart bioinks from experimental platforms to clinically and commercially viable technologies.[80]

Looking forward, future studies in hydrogel based bioink technology are likely to overcome most of these challenges. Multi-material bioprinting is one such option that can facilitate simultaneous deposition of a variety of bioinks with different compositions, thus making it possible to form a heterogeneous tissue construct that is better able to represent native brain and tumor microenvironment. Hydrogel based hybrid scaffolds that incorporate synthetic polymers or bioactive nanomaterials are also an option as an approach to strike a balance between mechanical stability and biological functionality. A revolutionary trend is in-situ bioprinting, where constructs are printed directly into a living organism or onto damaged tissue, which has the potential to repair brain injuries or recapitulate tumor microenvironment in real time. New methods of vascularization, such as the use of endothelial cells, angiogenic growth factors, and sacrificial bioinks to create perfusable channels, are be expected to significantly enhance tissue viability and function.[81]

Furthermore, the integration of computer modelling with artificial intelligence can help to overcome the existing limitations. Such a system can help to predict optimum bioink composition along with printing specifications, and scaffold structures. This can help in minimizing trial-and-error experimentation and expediting development schedules. Diffusion of nutrients, mechanical stresses and cellular responses in constructs can also be simulated using simulation platforms, which are difficult to measure experimentally.

Conclusion

Hydrogel-based smart bioinks have emerged as a transformative platform in three-dimensional bioprinting, enabling the recreation of complex biochemical, mechanical, and structural features of native tissues. Their ability to integrate stimuli-responsiveness, bioactivity, and tunable physicochemical properties makes them particularly valuable for applications in brain tissue engineering and tumor microenvironment modelling. These systems have demonstrated significant potential in improving the physiological relevance of in vitro models, advancing regenerative strategies, and enabling more predictive drug screening platforms. Despite these advances, several critical challenges remain that limit their translational impact. Among these, the most pressing issues include achieving robust vascularization, ensuring long-term functional stability of engineered tissues, and maintaining reproducibility across bioink formulations and printing processes. In addition, the integration of dynamic biochemical signalling—particularly in complex systems such as tumor microenvironments—remains an area requiring further development. Economic and manufacturing constraints, including scalability and cost of functionalized bioinks, further complicate their transition from laboratory research to clinical and industrial applications.

Looking forward, future research should prioritize the development of multi-functional and adaptive bioink systems capable of simultaneously addressing biological, mechanical, and manufacturing requirements. The integration of advanced strategies such as multi-material bioprinting, organoid–bioink hybrid systems, and AI-assisted design frameworks is expected to accelerate the rational development of next-generation bioinks. In particular, the convergence of data-driven modeling with experimental biofabrication holds promise for improving reproducibility and reducing development timelines. Ultimately, the successful translation of smart bioinks will depend not only on technological innovation but also on the ability to integrate interdisciplinary approaches spanning biomaterials science, biology, engineering, and regulatory science. By addressing these challenges in a coordinated manner, hydrogel-based smart bioinks have the potential to bridge the gap between experimental models and clinically relevant biomedical applications, thereby advancing the fields of precision medicine and translational bioengineering.

Acknowledgments

The authors sincerely acknowledge all colleagues and peers who provided informal scientific discussions and constructive feedback during the preparation of this manuscript. The authors also acknowledge assistance received in language refinement and proofreading during manuscript development.

Authors' Contribution

Conceptualization: Chintan Aundhia.
Data Curation: Jagruti Desai, Swayamprakash Patel.
Formal Analysis: Chintan Aundhia, Chitrali Talele.
Investigation: Jagruti Desai, Swayamprakash Patel.
Methodology: Jagruti Desai, Swayamprakash Patel.
Supervision: Chintan Aundhia, Bhupendra G. Prajapati.
Visualization: Chitrali Talele.
Writing–Original Draft: Chintan Aundhia.
Writing–Review & Editing: Chintan Aundhia, Jagruti Desai, Chitrali Talele, Bhupendra G. Prajapati.

Competing Interests

The authors declare no conflicts of interest.

Data Availability Statement

Not applicable.

Ethical Approval

Not applicable. This article does not report any studies involving human participants or animals performed by any of the authors.

Funding

None.

References

1. Qiao T, He C, Xia P, Liu G, Sun Y, Sun M, et al. Bioink design for organ-scale projection-based 3D bioprinting. *Nat Protoc* **2026**; 21: 894-923. doi:10.1038/s41596-025-01221-0
2. Fatimi A, Okoro OV, Podstawczyk D, Siminska-Stanny J, Shavandi A. Natural hydrogel-based bio-inks for 3D bioprinting in tissue engineering: a review. *Gels* **2022**; 8: 189. doi:10.3390/gels8030179
3. Choi T, Park J, Lee S, Jeon HJ, Kim BH, Kim HO, et al. 3D bioprinted neural tissues: emerging strategies for regeneration and disease modeling. *Pharmaceutics* **2025**; 17: 1176. doi:10.3390/pharmaceutics17091176
4. Mangani S, Vetoulas M, Mineschou K, Spanopoulos K, Vivanco MD, Piperigkou Z, et al. Design and applications of extracellular matrix scaffolds in tissue engineering and regeneration. *Cells* **2025**; 14: 1076. doi:10.3390/cells14141076
5. Ozbek II, Saybasili H, Ulgen KO. Applications of 3D bioprinting technology to brain cells and brain tumor models: special emphasis to glioblastoma. *ACS Biomater Sci Eng* **2024**; 10: 2616-35. doi:10.1021/acsbiomaterials.3c01569
6. Rauner G, Gupta PB, Kuperwasser C. From 2D to 3D and beyond: the evolution and impact of in vitro tumor models in cancer research. *Nat Methods* **2025**; 22: 1776-87. doi:10.1038/s41592-025-02769-1
7. Wu Y, Xiao Y, Yin B, Wong SHD. Dynamic hydrogels: adaptive biomaterials for engineering tumor microenvironment and cancer treatment. *Int J Mol Sci* **2025**; 26: 9502. doi:10.3390/ijms26199502
8. Boucq P, Ucakar B, Debuisson F, Riva R, des Rieux A, Jérôme C, et al. High internal phase emulsion-templated hydrophilic polyphosphoester scaffolds: tailoring the porosity and degradation for soft tissue engineering. *Biomacromolecules* **2025**; 26: 1935-47. doi:10.1021/acs.biomac.4c01740
9. Azmir MS, Moni MN, Gobetti A, Ramorino G, Dey K. Advances in modulating mechanical properties of gelatin-based hydrogel in tissue engineering. *Int J Polym Mater Polym Biomater* **2025**; 74: 215-50. doi:10.1080/00914037.2024.2330420
10. Pu M, Cao H, Zhang H, Wang T, Li Y, Xiao S, et al. ROS-responsive hydrogels: from design and additive manufacturing to biomedical applications. *Mater Horiz* **2024**; 11: 3721-46. doi:10.1039/d4mh00289j
11. Khodve G, Banerjee S, Kumari M, Ravichandiran V, Banerjee S, Roy S. Exploration of biomaterial and stem cell-based strategies for promoting neuronal regeneration and creating engineered 3D in-vitro disease models. *J Transl Med* **2025**; 23: 1197. doi:10.1186/s12967-025-07266-9
12. Karakaya M, Karataş RB, Ayaz F, Aydemir E. Hydrogel applications in tumor microenvironment modeling. *Discov Biotechnol* **2025**; 2: 21. doi:10.1007/s44340-025-00029-8
13. Lou J, Mooney DJ. Chemical strategies to engineer hydrogels for cell culture. *Nat Rev Chem* **2022**; 6: 726-44. doi:10.1038/s41570-022-00420-7
14. Hinton TJ, Jallerat Q, Palchesko RN, Park JH, Grodzicki MS, Shue HJ, et al. Three-dimensional printing of complex biological structures by freeform reversible embedding of suspended

hydrogels. *Sci Adv* **2015**; 1: e1500758. doi:10.1126/sciadv.1500758
15. Xiang L, Cui W. Biomedical application of photo-crosslinked gelatin hydrogels. *J Leather Sci Eng* **2021**; 3: 3. doi:10.1186/s42825-020-00043-y
16. Arias-Peregrino VM, Tenorio-Barajas AY, Mendoza-Barrera CO, Román-Doval J, Lavariega-Sumano EF, Torres-Arellanes SP, et al. 3D printing for tissue engineering: printing techniques, biomaterials, challenges, and the emerging role of 4D bioprinting. *Bioengineering (Basel)* **2025**; 12: 936. doi:10.3390/bioengineering12090936
17. Yarali E, Mirzaali MJ, Ghalayaniesfahani A, Accardo A, Diaz-Payno PJ, Zadpoor AA. 4D printing for biomedical applications. *Adv Mater* **2024**; 36: e2402301. doi:10.1002/adma.202402301
18. Jiang T, Yang Y, Lin Z, Hong Y, Luo Z. Modified polysaccharides: potential biomaterials for bioprinting. *J Funct Biomater* **2025**; 16: 338. doi:10.3390/jfb16090338
19. Xiao X, Yang Y, Lai Y, Huang Z, Li C, Yang S, et al. Customization of an ultrafast thiol-norbornene photo-cross-linkable hyaluronic acid-gelatin bioink for extrusion-based 3D bioprinting. *Biomacromolecules* **2023**; 24: 5414-27. doi:10.1021/acs.biomac.3c00887
20. Sadraei A, Naghib SM. 4D printing of physical stimuli-responsive hydrogels for localized drug delivery and tissue engineering. *Polym Rev (Phila Pa)* **2025**; 65: 104-68. doi:10.1080/15583724.2024.2427184
21. Thareja P, Swarupa S, Ahmad S, Jinugu ME. Hydrogel-based inks for extrusion 3D printing: a rheological viewpoint. *Curr Opin Colloid Interface Sci* **2025**; 77: 101918. doi:10.1016/j.cocis.2025.101918
22. Li Z, Lu F, Liu Y. A review of the mechanism, properties, and applications of hydrogels prepared by enzymatic cross-linking. *J Agric Food Chem* **2023**; 71: 10238-49. doi:10.1021/acs.jafc.3c01162
23. Tipa C, Cidade MT, Borges JP, Costa LC, Silva JC, Soares PIP. Clay-based nanocomposite hydrogels for biomedical applications: a review. *Nanomaterials (Basel)* **2022**; 12: 3308. doi:10.3390/nano12193308
24. Unterman S, Charles LF, Strecker SE, Kramarenko D, Pivovarchik D, Edelman ER, et al. Hydrogel nanocomposites with independently tunable rheology and mechanics. *ACS Nano* **2017**; 11: 2598-610. doi:10.1021/acsnano.6b06730
25. Shan BH, Wu FG. Hydrogel-based growth factor delivery platforms: strategies and recent advances. *Adv Mater* **2024**; 36: e2210707. doi:10.1002/adma.202210707
26. Sousa AC, McDermott G, Shields F, Alvites R, Lopes B, Sousa P, et al. Innovative ink-based 3D hydrogel bioprinted formulations for tissue engineering applications. *Gels* **2024**; 10: 831. doi:10.3390/gels10120831
27. Patrocinio D, Galván-Chacón V, Gómez-Blanco JC, Miguel SP, Loureiro J, Ribeiro MP, et al. Biopolymers for tissue engineering: crosslinking, printing techniques, and applications. *Gels* **2023**; 9: 890. doi:10.3390/gels9110890
28. Chandra DK, Reis RL, Kundu SC, Kumar A, Mahapatra C. Nanomaterials-based hybrid bioink platforms in advancing 3D bioprinting technologies for regenerative medicine. *ACS Biomater Sci Eng* **2024**; 10: 4145-74. doi:10.1021/acsbiomaterials.4c00166
29. Schwab A, Levato R, D'Este M, Piluso S, Eglin D, Malda J. Printability and shape fidelity of bioinks in 3D bioprinting. *Chem Rev* **2020**; 120: 11028-55. doi:10.1021/acs.chemrev.0c00084
30. Rocha LA, Silva D, Barata-Antunes S, Cavaleiro H, Gomes ED, Silva NA, et al. Cell and tissue instructive materials for central nervous system repair. *Adv Funct Mater* **2020**; 30: 1909083. doi:10.1002/adfm.201909083
31. Bae M, Hwang DW, Ko MK, Jin Y, Shin WJ, Park W, et al. Neural stem cell delivery using brain-derived tissue-specific bioink for recovering from traumatic brain injury. *Biofabrication* **2021**; 13: 044110. doi:10.1088/1758-5090/ac293f
32. Patel JC, Shukla M, Shukla M. Cellular and molecular interactions in CNS injury: the role of immune cells and inflammatory responses in damage and repair. *Cells* **2025**; 14: 918. doi:10.3390/cells14120918
33. Thakur N, Sharma V, Singh B. Designing biomimetic hydrogels for neuro-therapeutic delivery devices for brain soft tissue injury: integrating antioxidant, cell viability and tissue adhesion properties to enhance neural regeneration via a synergistic approach. *React Chem Eng* **2026**; 11: 196-218. doi:10.1039/d5re00316d
34. Samanta S, Ylä-Outinen L, Rangasami VK, Narkilahti S, Oommen OP. Bidirectional cell-matrix interaction dictates neuronal network formation in a brain-mimetic 3D scaffold. *Acta Biomater* **2022**; 140: 314-23. doi:10.1016/j.actbio.2021.12.010
35. Han R, Luo L, Wei C, Qiao Y, Xie J, Pan X, et al. Stiffness-tunable biomaterials provide a good extracellular matrix environment for axon growth and regeneration. *Neural Regen Res* **2025**; 20: 1364-76. doi:10.4103/nrr.Nrr-d-23-01874
36. Dai X, Wu D, Xu K, Ming P, Cao S, Yu L. Viscoelastic mechanics: from pathology and cell fate to tissue regeneration biomaterial development. *ACS Appl Mater Interfaces* **2025**; 17: 8751-70. doi:10.1021/acsami.4c18174
37. Roth JG, Huang MS, Navarro RS, Akram JT, LeSavage BL, Heilshorn SC. Tunable hydrogel viscoelasticity modulates human neural maturation. *Sci Adv* **2023**; 9: eadh8313. doi:10.1126/sciadv.adh8313
38. Zhang W, Zhang Y, Ma H, Duan L, Zhang W, Ding L, et al. Biomaterial and hydrogel strategies for regenerative microenvironment reconstruction in peripheral nerve conduits. *Gels* **2025**; 11: 898. doi:10.3390/gels11110898
39. Llorente V, Velarde P, Desco M, Gómez-Gaviro MV. Current understanding of the neural stem cell niches. *Cells* **2022**; 11: 3002. doi:10.3390/cells11193002
40. Sullivan MA, Lane S, Volkerling A, Engel M, Werry EL, Kassiou M. Three-dimensional bioprinting of stem cell-derived central nervous system cells enables astrocyte growth, vasculogenesis, and enhances neural differentiation/function. *Biotechnol Bioeng* **2023**; 120: 3079-91. doi:10.1002/bit.28470
41. Ciszowski K, Gomółka E, Gawlikowski T, Szpak D, Potoczek A, Boba M. Brain-derived neurotrophic factor (BDNF) and nerve growth factor (NGF) blood levels in patients with acute carbon monoxide poisoning - a preliminary observations. *Przegl Lek* **2016**; 73: 552-9.
42. Wang J, Zhang Y, Lei Z, Wang J, Zhao Y, Sun T, et al. Hydrogels with brain tissue-like mechanical properties in complex environments. *Mater Des* **2023**; 234: 112338. doi:10.1016/j.matdes.2023.112338
43. Yan Y, Li X, Gao Y, Mathivanan S, Kong L, Tao Y, et al. 3D bioprinting of human neural tissues with functional connectivity. *Cell Stem Cell* **2024**; 31: 260-74.e7. doi:10.1016/j.stem.2023.12.009
44. Mancuso S, Bhalerao A, Cucullo L. Advances and challenges of bioassembly strategies in neurovascular in vitro modeling: an overview of current technologies with a focus on three-dimensional bioprinting. *Int J Mol Sci* **2024**; 25: 11000. doi:10.3390/ijms252011000
45. Yang Z, You Y, Liu X, Wan Q, Xu Z, Shuai Y, et al. Injectable *Bombyx mori* (*B. mori*) silk fibroin/MXene conductive hydrogel for electrically stimulating neural stem cells into neurons for treating brain damage. *J Nanobiotechnology* **2024**; 22: 111. doi:10.1186/s12951-024-02359-x
46. Zhou J, Wu Y, Tang Z, Zou K, Chen J, Lei Z, et al. Alginate hydrogel cross-linked by Ca2 + to promote spinal cord neural stem/progenitor cell differentiation and functional recovery after a spinal cord injuryhh. *Regen Biomater* **2022**; 9: rbac057. doi:10.1093/rb/rbac057
47. Song S, Li Y, Huang J, Cheng S, Zhang Z. Inhibited astrocytic differentiation in neural stem cell-laden 3D bioprinted conductive composite hydrogel scaffolds for repair of spinal cord injury. *Biomater Adv* **2023**; 148: 213385. doi:10.1016/j.bioadv.2023.213385
48. Fowler M, Moreno Lozano A, Krause J, Bednarz P, Pandey S, Ghayour M, et al. Guiding vascular infiltration through architected GelMA/PEGDA hydrogels: an in vivo study of channel diameter, length, and complexity. *Biomater Sci* **2025**; 13: 2951-60. doi:10.1039/d5bm00193e
49. Ni Y, Zhou X, Yang J, Shi H, Li H, Zhao X, et al. The role of tumor-stroma interactions in drug resistance within tumor microenvironment. *Front Cell Dev Biol* **2021**; 9: 637675. doi:10.3389/fcell.2021.637675
50. Loewa A, Feng JJ, Hedtrich S. Human disease models in drug development. *Nat Rev Bioeng* **2023**; 1: 545-59. doi:10.1038/s44222-023-00063-3
51. Zhang M, Zhang B. Extracellular matrix stiffness: mechanisms

in tumor progression and therapeutic potential in cancer. *Exp Hematol Oncol* **2025**; 14: 54. doi:10.1186/s40164-025-00647-2
52. Wu T, Xiong S, Chen M, Tam BT, Chen W, Dong K, et al. Matrix stiffening facilitates the collective invasion of breast cancer through the periostin-integrin mechanotransduction pathway. *Matrix Biol* **2023**; 121: 22-40. doi:10.1016/j.matbio.2023.05.006
53. Lee SH, Lee J, Choi MS, Ahn M, Yoon S, Lee D, et al. Mechanical stimuli-induced manipulation of malignant behavior in bioprinted cancer microtissues via PI3K/NF-κB activation. *Adv Sci (Weinh)* **2026**; 13: e18295. doi:10.1002/advs.202518295
54. Zhang Y, Fu Q, Sun W, Yue Q, He P, Niu D, et al. Mechanical forces in the tumor microenvironment: roles, pathways, and therapeutic approaches. *J Transl Med* **2025**; 23: 313. doi:10.1186/s12967-025-06306-8
55. Jacobs M, Gllareva V, Moser L, Pravato S, Mora Pimentel E, Leeners B, et al. Three-dimensional ovary model to improve and study murine follicle growth. *Adv Healthc Mater* **2026**; 15: e04643. doi:10.1002/adhm.202504643
56. Severs LJ, Katta A, Cates LN, Dewees DM, Hoagland RT, Horner PJ, et al. Biomimetic 3D hydrogels with aligned topography for neural tissue engineering. *Polymers (Basel)* **2024**; 16: 3556. doi:10.3390/polym16243556
57. Nagaraja K, Bhattacharyya A, Jung M, Kim D, Khatun MR, Noh I. 3D bioprintable self-healing hyaluronic acid hydrogel with cysteamine grafting for tissue engineering. *Gels* **2024**; 10: 789. doi:10.3390/gels10120780
58. Schroyer KA, Schmitz KM, Raheja G, Su B, Lathia JD, Ning L. Bioprinting of GelMA-based hydrogels to aid in creation of biomimetic 3D models for glioblastoma. *Micromachines (Basel)* **2025**; 16: 654. doi:10.3390/mi16060654
59. Jahangir S, Vecstaudza J, Augurio A, Canciani E, Stipniece L, Locs J, et al. Cell-laden 3D printed GelMA/HAp and THA hydrogel bioinks: development of osteochondral tissue-like bioinks. *Materials (Basel)* **2023**; 16: 7214. doi:10.3390/ma16227214
60. Staros R, Michalak A, Rusinek K, Mucha K, Pojda Z, Zagożdżon R. Perspectives for 3D-bioprinting in modeling of tumor immune evasion. *Cancers (Basel)* **2022**; 14: 3126. doi:10.3390/cancers14133126
61. Grebenyuk S, Abdel Fattah AR, Kumar M, Toprakhisar B, Rustandi G, Vananroye A, et al. Large-scale perfused tissues via synthetic 3D soft microfluidics. *Nat Commun* **2023**; 14: 193. doi:10.1038/s41467-022-35619-1
62. Tang M, Rich JN, Chen S. Biomaterials and 3D bioprinting strategies to model glioblastoma and the blood-brain barrier. *Adv Mater* **2021**; 33: e2004776. doi:10.1002/adma.202004776
63. Liu S, Jin P. Advances and challenges in 3D bioprinted cancer models: opportunities for personalized medicine and tissue engineering. *Polymers (Basel)* **2025**; 17: 948. doi:10.3390/polym17070948
64. Manduca N, Maccafeo E, De Maria R, Sistigu A, Musella M. 3D cancer models: one step closer to in vitro human studies. *Front Immunol* **2023**; 14: 1175503. doi:10.3389/fimmu.2023.1175503
65. Neufeld L, Yeini E, Reisman N, Shtilerman Y, Ben-Shushan D, Pozzi S, et al. Microengineered perfusable 3D-bioprinted glioblastoma model for in vivo mimicry of tumor microenvironment. *Sci Adv* **2021**; 7: eabi9119. doi:10.1126/sciadv.abi9119
66. Tang M, Xie Q, Gimple RC, Zhong Z, Tam T, Tian J, et al. Three-dimensional bioprinted glioblastoma microenvironments model cellular dependencies and immune interactions. *Cell Res.* 2020;30:833–853. doi:10.1038/s41422-020-0338-1.
67. Wang X, Xu T, Niu C. Vascularization ability of glioma stem cells in different three-dimensional microenvironments. *Regen Biomater* **2024**; 11: rbad094. doi:10.1093/rb/rbad094
68. Kong Z, Wang X. Bioprinting technologies and bioinks for vascular model establishment. *Int J Mol Sci* **2023**; 24: 891. doi:10.3390/ijms24010891
69. Mierke CT. Bioprinting of cells, organoids and organs-on-a-chip together with hydrogels improves structural and mechanical cues. *Cells* **2024**; 13: 1638. doi:10.3390/cells13191638
70. Freeman S, Calabro S, Williams R, Jin S, Ye K. Bioink formulation and machine learning-empowered bioprinting optimization. *Front Bioeng Biotechnol* **2022**; 10: 913579. doi:10.3389/fbioe.2022.913579
71. Rafieyan S, Ansari E, Vasheghani-Farahani E. A practical machine learning approach for predicting the quality of 3D (bio)printed scaffolds. *Biofabrication* **2024**; 16: 045014. doi:10.1088/1758-5090/ad6374
72. Zhang Z, Tao ZZ, Du R, Huo R, Zheng X. Artificial intelligence informed hydrogel biomaterials in additive manufacturing. *Gels* **2025**; 11: 981. doi:10.3390/gels11120981
73. Mallya D, Gadre MA, Varadharajan S, Vasanthan KS. 3D bioprinting for the construction of drug testing models-development strategies and regulatory concerns. *Front Bioeng Biotechnol* **2025**; 13: 1457872. doi:10.3389/fbioe.2025.1457872
74. Zandrini T, Florczak S, Levato R, Ovsianikov A. Breaking the resolution limits of 3D bioprinting: future opportunities and present challenges. *Trends Biotechnol* **2023**; 41: 604-14. doi:10.1016/j.tibtech.2022.10.009
75. Askari M, Afzali Naniz M, Kouhi M, Saberi A, Zolfagharian A, Bodaghi M. Recent progress in extrusion 3D bioprinting of hydrogel biomaterials for tissue regeneration: a comprehensive review with focus on advanced fabrication techniques. *Biomater Sci* **2021**; 9: 535-73. doi:10.1039/d0bm00973c
76. Mo X, Ouyang L, Xiong Z, Zhang T. Advances in digital light processing of hydrogels. *Biomed Mater* **2022**; 17: 042002. doi:10.1088/1748-605X/ac6b04
77. Raju RR, AlSawaftah NM, Husseini GA. Modeling of brain tumors using in vitro, in vivo, and microfluidic models: a review of the current developments. *Heliyon* **2024**; 10: e31402. doi:10.1016/j.heliyon.2024.e31402
78. Barthes J, Lagarrigue P, Riabov V, Lutzweiler G, Kirsch J, Muller C, et al. Biofunctionalization of 3D-printed silicone implants with immunomodulatory hydrogels for controlling the innate immune response: an in vivo model of tracheal defect repair. *Biomaterials* **2021**; 268: 120549. doi:10.1016/j.biomaterials.2020.120549
79. Perin F, Ouyang L, Lim KS, Motta A, Maniglio D, Moroni L, et al. Bioprinted constructs in the regulatory landscape: current state and future perspectives. *Adv Mater* **2026**; 38: e04037. doi:10.1002/adma.202504037
80. Hou X, Du C, Lu L, Yuan S, Zhan M, You P, et al. Opportunities and challenges of patient-derived models in cancer research: patient-derived xenografts, patient-derived organoid and patient-derived cells. *World J Surg Oncol* **2022**; 20: 37. doi:10.1186/s12957-022-02510-8
81. An J, Leong KF. Multi-material and multi-dimensional 3D printing for biomedical materials and devices. *Biomed Mater Devices* **2023**; 1: 38-48. doi:10.1007/s44174-022-00038-9