eVitroKhem. 2025; 4:236
doi: 10.56294/evk2025236

REVIEW

Regenerative Medicine and Tissue Engineering: Innovations in Scaffolding Technologies, Stem Cell Engineering, and Bioprinting for Tissue Repair and Organ Regeneration

Medicina Regenerativa e Ingeniería de Tejidos: Innovaciones en Tecnologías de Andamiaje, Ingeniería de Células Madre y Bioimpresión para la Reparación de Tejidos y la Regeneración de Órganos

Esteban Llorente-Nuñez[1], Diago Nunes-Coelho[2]

[1]Universidad Autónoma de Madrid (UAM). Spain.
[2]Universidade de Coimbra. Portugal.

Cite as: Llorente-Nuñez E, Nunes-Coelho D. Regenerative Medicine and Tissue Engineering: Innovations in Scaffolding Technologies, Stem Cell Engineering, and Bioprinting for Tissue Repair and Organ Regeneration. eVitroKhem. 2025; 4:236. https://doi.org/10.56294/evk2025236

Submitted: 26-09-2024 **Revised:** 01-02-2025 **Accepted:** 21-07-2025 **Published:** 22-07-2025

Editor: Prof. Dr. Javier Gonzalez-Argote

ABSTRACT

Introduction: regenerative medicine and tissue engineering are emerging disciplines that seek to restore the function of damaged tissues and organs through technological innovations. These areas combine biology, engineering, and medicine to develop solutions that improve patients' quality of life. In this context, scaffolding technologies, stem cell engineering, and bioprinting stand out as key tools for tissue repair and organ regeneration.
Development: scaffolds are three-dimensional structures that provide physical support for cell growth and tissue formation. They can be designed with biocompatible materials that mimic the properties of natural tissue, facilitating integration with the body. Stem cell engineering, on the other hand, allows for the extraction and differentiation of cells with regenerative potential, which is crucial for repairing damage to specific tissues. Bioprinting, an innovative technique, uses 3D printers to create complex cellular structures, enabling the manufacture of personalized tissues and artificial organs. These technologies have shown promising results in preclinical and clinical studies, offering new hope in the treatment of degenerative diseases, traumatic injuries, and birth defects.
Conclusions: innovations in scaffolding technologies, stem cell engineering, and bioprinting are opening up new possibilities for tissue and organ repair and regeneration. As these technologies continue to evolve, it is critical to address the associated ethical and regulatory challenges to ensure their safe and effective implementation in clinical practice.

Keywords: Tissue Engineering; Scaffolding Technologies; Stem Cell Engineering; Bioprinting; Tissue Repair; Organ Regeneration.

RESUMEN

Introducción: la medicina regenerativa y la ingeniería de tejidos son disciplinas emergentes que buscan restaurar la función de tejidos y órganos dañados mediante innovaciones tecnológicas. Estas áreas combinan biología, ingeniería y medicina para desarrollar soluciones que mejoren la calidad de vida de los pacientes. En este contexto, las tecnologías de andamiaje, la ingeniería de células madre y la bioimpresión se destacan como herramientas clave para la reparación de tejidos y la regeneración de órganos.
Desarrollo: los andamiajes son estructuras tridimensionales que proporcionan soporte físico para el crecimiento celular y la formación de tejidos. Estos pueden ser diseñados con materiales biocompatibles que imitan las propiedades del tejido natural, facilitando la integración con el organismo. Por otro lado, la

ingeniería de células madre permite la obtención y diferenciación de células con potencial regenerativo, lo que es crucial para reparar daños en tejidos específicos. La bioimpresión, una técnica innovadora, utiliza impresoras 3D para crear estructuras celulares complejas, permitiendo la fabricación de tejidos personalizados y órganos artificiales. Estas tecnologías han mostrado resultados prometedores en estudios preclínicos y clínicos, ofreciendo nuevas esperanzas en el tratamiento de enfermedades degenerativas, lesiones traumáticas y defectos congénitos.
Conclusiones: las innovaciones en tecnologías de andamiaje, ingeniería de células madre y bioimpresión están abriendo nuevas posibilidades para la reparación y regeneración de tejidos y órganos. A medida que estas tecnologías continúan evolucionando, es fundamental abordar los desafíos éticos y regulatorios asociados para garantizar su implementación segura y efectiva en la práctica clínica.

Palabras clave: Ingeniería de Tejidos; Tecnologías de Andamiaje; Ingeniería de Células Madre; Bioimpresión; Reparación de Tejidos; Regeneración de Órganos.

INTRODUCTION

Definition and Scope of Regenerative Medicine and Tissue Engineering

Regenerative medicine (RM) is an emerging multidisciplinary field that focuses on the process of replacing, designing, or regenerating human or animal cells, tissues, or organs with the fundamental goal of restoring or establishing normal biological function.[1] This vast domain not only encompasses the repair of damaged structures, but also investigates the body's intrinsic capacity for self-healing, sometimes with the support of external biological materials, to recreate cells or reconstruct entire organs.[2] The underlying vision of regenerative medicine is to transcend palliative treatments for complex and chronic diseases by focusing on the search for definitive cures through the intrinsic functional restoration of the organism.[3]

In this context, tissue engineering (TE) is positioned as a fundamental discipline within bioengineering. It is defined by the strategic combination of cells, material engineering methods, and appropriate biochemical and physicochemical factors to restore, maintain, enhance, or replace various types of biological tissues.[4,5] Although regenerative medicine and tissue engineering are distinct fields, their terms have become largely interchangeable in scientific and clinical discourse. This conceptual fusion is not merely semantic; rather, it underscores a profound evolution in the medical paradigm. The shift in focus from the "treatment" of symptoms to "cure" through the intrinsic restoration of biological function means that tissue engineering, by providing the functional constructs and methodologies for their creation, becomes an indispensable pillar for realizing the promises of regenerative medicine. This paradigm shift drives continued investment in research into regeneration mechanisms, the development of advanced biomaterials, and cell manipulation, which are the foundations for achieving lasting functional restoration.

Historical Context and Evolution of the Field

The origins of our understanding of stem cells and cell theory date back to the 19th century, with pioneering contributions from figures such as H. Milne-Edwards and F.V. Raspail, who initiated the development of cell theory, and Ernst Haeckel, who postulated the existence of an "ancestral cell." The formalization of cell theory by Mathias Schleiden and Theodor Schwann in 1838, which established that all plants and animals are composed of cells, laid a crucial theoretical foundation for modern biology.

The 20th century marked a series of technological advances and biological discoveries that catalyzed the field. The invention of the electron microscope in 1932 was a turning point, allowing detailed observation of the plasma membrane and the complex internal structure of eukaryotic cells. This ability to "see" cells at an ultrastructural level was fundamental to subsequent progress. Milestones such as the discovery of hematopoietic stem cells by James Till and Ernest McCulloch in 1960, through their research on the effects of radiation on mammalian cells, revealed the self-renewal and differentiation capacity of certain cells, laying the foundation for treatments for blood diseases. Subsequently, the successful isolation of mouse embryonic stem cells (ESCs) by Martin Evans and Matthew Kaufman in 1947, and human embryonic stem cells (hESCs) by James Thomson in 1998, greatly expanded the expectations of regenerative medicine due to their pluripotency. Arnold Caplan, recognized as the "father of mesenchymal stem cells," perfected the technology to isolate and culture these cells, discovering their immunomodulatory properties and their potential for tissue regeneration.

The 21st century has witnessed a definitive push in regenerative medicine, driven by revolutionary innovations. Cell reprogramming by Shinya Yamanaka in 2006, which enabled the conversion of adult somatic cells into induced pluripotent stem cells (iPSCs) by introducing only four transcription factors, eliminated many of the ethical controversies associated with hESCs and opened up promising new possibilities for personalized regenerative medicine. The emergence of gene editing technologies such as CRISPR/Cas9 has enabled the

precise correction of genetic mutations directly in stem cells, offering new avenues for disease research and therapy development. The evolution of regenerative medicine is therefore intrinsically linked to technological advances in microscopy and molecular biology. Each technological leap has unlocked new capabilities in cell manipulation and understanding, which in turn has propelled the field toward previously unimaginable frontiers. This suggests that the future of RM will continue to depend on innovation in nanoscale visualization, analysis, and manipulation tools, as well as bioinformatics to process the complexity of biological data.[9,10]

Importance and Current Potential in Human Health

Regenerative medicine and tissue engineering represent a transformative promise for human health, offering the possibility of addressing and curing chronic diseases, injuries, and degenerative conditions that conventional treatments cannot effectively resolve. This field seeks to regenerate tissues damaged by various causes, including chronic diseases, aging, or trauma.

One of the greatest potentials of regenerative medicine lies in its ability to permanently replace the function of damaged or failed organs. This could overcome the critical shortage of organs for transplantation and mitigate the significant morbidity associated with lifelong immunosuppression required by conventional organ transplants.[12] The ability of regenerative medicine to create "personalized organs" using the patient's own cells represents a disruptive solution that could redefine the transplant paradigm. This approach would enable a shift from dependence on donation to on-demand biofabrication, dramatically minimizing immune rejection. If scaled up and refined, this technology could eliminate transplant waiting lists and reduce post-transplant complications, transforming the quality of life for millions of patients and easing the burden on healthcare systems. This advance is not only a medical milestone, but also opens the door to "personalized medicine" on an unprecedented scale.

In addition to direct therapeutic applications, tissue engineering has found equally valuable non-therapeutic uses. Bioengineered tissues are used as biosensors to detect threatening biological or chemical agents, and as "tissue chips" (organs-on-a-chip) to test the toxicity and efficacy of experimental drugs.[2] These applications not only reduce the cost and time involved in developing new drugs, but also decrease the need for animal testing, offering a testing system that is more relevant to human physiology.[3]

DEVELOPMENT

Key Fundamentals: Stem Cells and Biomaterials

Stem cells are considered the "master cells" of the body because of their two fundamental properties: the ability to self-renew, which allows them to continuously produce more cells of the same type, and the ability to differentiate, whereby they can become various types of specialized cells that perform different functions.[12] These cells are found in almost all tissues of the body and are essential for the maintenance, repair, and regeneration of tissues after injury.[14]

Several main types of stem cells are distinguished, each with specific characteristics and potentials:

- Embryonic stem cells (ESCs): These cells are derived from embryos at an early stage of development (3 to 5 days), known as the blastocyst stage, which contains approximately 150 cells.[15] ESCs are pluripotent, meaning they have the ability to differentiate into any type of cell in the body, making them potentially useful for regenerating or repairing tissues and organs affected by disease.[16] However, their procurement and use raise significant ethical considerations due to the destruction of the embryo.[17,18]
- Adult stem cells (ASCs): These are found in small quantities in most adult tissues, such as bone marrow, adipose tissue, peripheral blood, and umbilical cord blood.[12] Unlike ESCs, ASCs are multipotent, which implies a more limited differentiation capacity, generally restricted to cell types within the tissue or organ from which they originate.[15] Prominent examples include mesenchymal stem cells (MSCs) and hematopoietic stem cells (HSCs).[6] MSCs, in particular, are multipotent and can differentiate into adipocytes, chondrocytes, osteoblasts, and neurons, in addition to possessing valuable immunomodulatory properties.[6]
- Induced pluripotent stem cells (iPSCs): Scientists have succeeded in transforming normal adult cells into stem cells with properties similar to those of embryonic cells through genetic reprogramming.[12] These iPSCs offer a crucial advantage in that they are patient-specific, thereby avoiding immune rejection of the new stem cells and, at the same time, circumventing the ethical controversies associated with the use of embryos.[12]
- Perinatal stem cells: Recent research has identified stem cells in amniotic fluid and umbilical cord blood, which also have the ability to differentiate into specialized cells.[12]

The evolution from the use of embryonic stem cells to induced pluripotent stem cells (iPSCs) and adult stem cells (ASCs) reflects a clear trend in regenerative medicine research: the search for cell sources with less ethical controversy and a reduced risk of immune rejection, which greatly facilitates their clinical translation

and the development of personalized medicine. The ability to reprogram adult cells into iPSCs, a landmark advance, broadened the expectations of regenerative medicine by offering a viable alternative to ESCs. The use of autologous cells, i.e., from the patient themselves, dramatically reduces the risk of immune rejection, a major obstacle in transplants.[11] This direction is driving research into more efficient and safer reprogramming techniques, as well as the comprehensive characterization of iPSCs to ensure their genetic stability and functionality, which are crucial for their eventual large-scale commercial use.[2,6]

Biomaterials for scaffolding: classification, properties, and functionality

Scaffolds are biocompatible three-dimensional structures that are an essential component in tissue engineering, as they provide the physical support and microenvironment necessary for the growth, proliferation, and differentiation of stem cells into the desired cell type or tissue.[2] To be functional, these scaffolds must have adequate porosity to allow internal cell migration, efficient diffusion of nutrients and oxygen, and the formation of a vascular network within the construct.

Key properties that define the suitability of a scaffold include:

- Biocompatibility: The material should not cause damage or generate an adverse immune response in the tissue with which it comes into contact.
- Biodegradability: Ideally, the scaffold should degrade over time as native tissue regenerates, and its degradation products should be non-toxic.[17]
- Porosity and connectivity: high porosity with a network of interconnected pores is crucial to facilitate the transport of cells, nutrients, and waste, as well as for vascularization.[17]
- Mechanical properties: the scaffold must be sufficiently resistant to stress and deformation, and possess mechanical properties that mimic those of the native tissue it is intended to replace, in order to withstand physiological loads.
- Cell promotion: it must promote cell adhesion, proliferation, and differentiation.

Biomaterials used in the manufacture of scaffolds are generally classified according to their origin and composition:

- Natural polymers: These materials are derived from biological sources and are highly valued for their inherent biocompatibility, biodegradability through natural processes, and ability to mimic the biological functions of the extracellular matrix (ECM), including cell signaling and low immunogenicity. Common examples include collagen (a key structural protein in the ECM), fibrin (a blood protein with cell anchorage sites), hyaluronic acid, alginate (derived from brown algae), gelatin (denatured collagen), and chitosan.[29] Despite their biological advantages, natural polymers often have limitations, such as low mechanical strength , structural instability after hydration, and limited control over their structural characteristics.[34]
- Synthetic polymers: these materials are chemically manufactured and offer the advantage of precise control over their structural, mechanical, and chemical properties.[30] It is possible to adjust their degradation rate, morphology, pore size, and controlled drug release capacity.[34] Examples include polyethylene glycol (PEG), polylactic acid (PLA), polycaprolactone (PCL), polymethyl methacrylate (PMMA), and polysiloxanes.[29] However, a disadvantage is their lower cell signaling capacity compared to natural materials, and the use of potentially cytotoxic organic solvents in their manufacture. [34]
- Ceramics and metals: Bioceramics, such as alumina, zirconia, hydroxyapatite, tricalcium phosphate, and bioactive glasses, are valued for their high mechanical strength, hardness, and wear resistance, making them suitable for load-bearing applications such as bone regeneration and dental implants.[41] Some metals, such as bioabsorbable zinc, are also used for their strength and ability to degrade safely in the body.[43]
- Composite materials: In order to combine the advantages of different types of materials and mitigate their individual limitations, composite materials (e.g., ceramic-polymer) are frequently used.

The trend in the development of biomaterials for scaffolding is towards the creation of “smart materials” and composites that not only provide passive structural support but also actively interact with the biological environment and respond to specific stimuli. These smart materials can react to environmental factors such as temperature, pH, or electric/magnetic fields, and can release growth factors in a controlled manner, actively influencing cell behavior and tissue formation.[33] This innovation allows the design of scaffolds that not only act as physical support, but also as dynamic platforms that actively guide regeneration, promoting vascularization, cell adhesion, and differentiation in a more precise manner, which is crucial for the functionality of complex tissues and organs.

Innovations in Scaffolding Technologies

Scaffold fabrication is a central pillar of tissue engineering, which aims to replicate the complex architecture

and composition of the natural extracellular matrix (ECM). This provides crucial three-dimensional support for cell adhesion, proliferation, and function, guiding their development toward the desired tissue.[29] The selection of material and manufacturing method is highly dependent on the specific type of tissue being replaced or regenerated, as each tissue has unique mechanical and biological properties.[44]

Among the most innovative manufacturing methods are:

- Electrospinning: This is a simple and highly effective technique for producing nanofibers with diameters in the nanometer range (around 700 nm). Electrospinning creates highly porous structures with a large surface-to-volume ratio, making them exceptionally well suited to mimic the natural ECM. The versatility of this technique allows precise control of parameters such as the viscosity of the polymer solution, conductivity, electric field strength, and the distance between the nozzle and the collector, which in turn allows the morphology and size of the fibers produced to be modulated.[29] It is a particularly promising technique for skin regeneration, where the nanofibrous structure is crucial for cell growth and healing. [29]
- Solvent casting particulate leaching (SCPL): This method is widely used to manufacture three-dimensional porous scaffolds. The process involves dissolving a polymer in an organic solvent, mixing it with pore-forming particles (commonly salt) inside a mold, allowing the solvent to evaporate, and finally leaching the pore-forming agent with water to create a network of interconnected pores. SCPL allows significant control over porosity (up to 93 %) and pore size, but a limitation is the maximum thickness that can be achieved (generally less than 2 mm) and the use of organic solvents, which can be cytotoxic.
- Gas foaming: This technique uses the nucleation and growth of gas bubbles (such as N_2 or CO_2) dispersed within a polymer phase to generate porous structures. One of the main advantages of this method is the ability to create highly porous scaffolds with interconnect r pores. However, in some cases, it may require the use of surfactants, whose residues could affect the biocompatibility of the final scaffold.[51]
- Decellularization: This is an innovative process that involves removing all cellular components from a tissue or entire organ, leaving only the extracellular matrix (ECM) intact. The resulting ECM serves as an "acellular scaffold" that preserves the complex native architecture, biochemical composition, and biophysical signals of the original organ.[54] This scaffold can then be recellularized with new cells, ideally from the patient themselves, to reconstruct a neotissue or a complete organ with its original microscopic anatomy and functionality.[54] Challenges of decellularization include the complexity of removing all cells without irreversibly damaging the ECM and ensuring that the recellularized organ achieves full biological functionality, including electrophysiology and hormonal function.[54]
- Other Methods: Rapid prototyping techniques, such as extrusion, stereolithography, and laser sintering, are also used in the additive manufacturing of scaffolds.[55] These methods allow for the creation of larger scaffolds with precisely oriented structures from a three-dimensional computer design.[44]

The transition from the manufacture of scaffolds with simple geometries to the use of decellularized matrices and additive manufacturing techniques (such as rapid prototyping) demonstrates an active search for deeper "structural and compositional biomimetics." This evolution is essential for the functionality of complex tissues and organs, as the simple provision of a passive scaffold is not sufficient for complex regeneration. Decellularization allows the complexity and dynamics of the original organ's extracellular matrix to be maintained, providing a scaffold with an inherently biomimetic architecture and composition.[54] Rapid prototyping methods and 3D bioprinting allow scaffolds with oriented structures to be created from the design of a three-dimensional model or complex 3D patterns.[44] This suggests that research is moving towards the creation of scaffolds that not only support cell growth but also act as biophysical and biochemical "guides" for cell self-organization, replicating natural tissue development for more successful integration in vivo.

Specific Applications of Scaffolds in Tissue Repair and Regeneration

Scaffolds play a crucial role in the repair and regeneration of a wide range of biological tissues, from simple structures to complex organs. They are used to repair or replace tissues such as bone, cartilage, heart valves, and the bladder, and have been tested in the trachea, liver, and heart.

Specific applications include:

- Cartilage: In the field of orthopedics, spheroids of stem cells are grown in the laboratory and implanted into cartilage defects, where they adhere and, over time, fill the defect, reducing pain and improving patient mobility.[28] Recent research has demonstrated the success of an injectable biological gel combined with a biological adhesive to facilitate knee cartilage regeneration, with a notable reduction in pain in patients six months after surgery.[2]
- Bone: Molecular scaffolds, considered third-generation biomaterials, are being developed that may be composed of collagen, bovine bone, or biopolymers. These scaffolds are designed to actively promote

bone regeneration, marking a paradigm shift from mere replacement to active repair and regeneration of living tissue.[31] A significant advance is the creation of mature bone stem cells which, when implanted in immunodeficient mice, did not cause abnormal growth, a problem that often occurs when only stem cells or immature bone scaffolds are implanted.[2]

- Liver: Bioengineered human liver tissue has been successfully manufactured that can be implanted in mice. These mice retain their own liver function, but the additional human tissue allows drugs to be metabolized in a similar way to humans, facilitating toxicity testing and the demonstration of species-specific responses that would normally only be observed in clinical trials. This approach significantly reduces the time and cost of developing new drugs.
- Kidney: Transplantable kidney tissue engineering represents a crucial advance in overcoming organ donor shortages and the morbidity associated with chronic immunosuppression in conventional transplants.[3]
- Nerves: Nerve tissue engineering is being applied to repair damaged or severed nerves. Decellularized nerve scaffolds have been used for peripheral nerve repair, which has proven to be efficient in removing nerve cells and myelin at the injury site and reducing the post-xenograft immune response.
- Blood vessels: To ensure an efficient supply of nutrients and oxygen to bioengineered tissues, scaffolds with preformed channels are designed. One method involves creating a sugar lattice that is then dissolved, leaving channels that act as blood vessels.[3]
- Skin and intestine: Tissue engineering is also applied in the creation of skin grafts to treat severe burns and in intestinal tissue engineering to address conditions such as short bowel syndrome.

The application of scaffolds in tissue repair has evolved from simple structural support to the creation of "controlled microenvironments." These microenvironments not only allow cell growth, but also guide cell differentiation and prevent post-implantation complications such as abnormal growth or rejection. Scaffolds are being designed to control the cellular environment and prevent the implant from differentiating into the wrong cell type.[28] In addition, the incorporation of bioactive molecules can reduce inflammation or help implanted cells attach to the implant site.[28] The example of mature bone stem cells that do not generate abnormal growths when implanted on scaffolds demonstrates the importance of a scaffold that not only supports but also modulates cell behavior. This suggests that future research will focus on scaffolds with more sophisticated signaling capabilities, perhaps incorporating sensors and actuators for active modulation of the in vivo microenvironment, which could dramatically improve the success rates of regenerative implants.[3]

Advances in Stem Cell Engineering

Stem cell engineering is a fundamental pillar of regenerative medicine, based on the ability to manipulate these biological building blocks to repair damaged tissues or create new ones. This process requires a deep understanding of how individual cells respond to signals, interact with their environment, and organize themselves to form functional tissues. Key techniques in this field are cell isolation, expansion, and differentiation.

Isolation

- Embryonic stem cells(ESCs): These are obtained from the inner cell mass of the blastocyst, an embryo 4 to 5 days into development. This process is inherently inefficient, and a significant number of cells fail to adapt to culture and do not survive.
- Adult stem cells (ASCs), including MSCs and HSCs: found in various adult tissue sources, such as bone marrow, adipose tissue, and umbilical cord blood.[12] Mesenchymal stem cells (MSCs) are commonly isolated by taking advantage of their physical adherence to plastic culture plates.[22] Hematopoietic stem cells (HSCs) are collected directly from bone marrow or by peripheral blood apheresis, a process that often requires prior mobilization of stem cells from the bone marrow into the bloodstream.[21]
- Induced pluripotent stem cells (iPSCs): The generation of iPSCs involves the reprogramming of adult somatic cells, such as fibroblasts or peripheral blood mononuclear cells (PBMCs), by introducing key reprogramming factors (OCT-4, SOX-2, KLF-4, c-MYC). Non-integrating methods, such as the use of self-replicating RNA, have been developed and are considered safer as they avoid the integration of genetic material into the host genome.

Expansion

Once isolated, stem cells are cultured in special solutions in the laboratory to increase their number to a scale sufficient for research or clinical applications.[12] For MSCs, optimized and standardized culture media are used to ensure reproducible generation and expansion.[59] However, large-scale expansion of stem cells for clinical or h applications remains a significant challenge, especially in the context of personalized autologous therapies.[60]

Differentiation

Stem cells can be guided to differentiate into specific cell types, such as cardiomyocytes, neurons, hepatocytes, osteoblasts, or chondrocytes. This is achieved by manipulating the culture environment, adding growth factors and bioactive molecules, and controlling physical conditions.[2] Embryoid body formation is a common method for initiating the differentiation of ESCs and iPSCs, as these cell aggregates can spontaneously differentiate into cells of the three germ layers.[17]

The development of detailed and standardized protocols for the isolation, expansion, and differentiation of stem cells is a critical bottleneck for clinical translation. Stem cell biology is inherently complex and variable, and this intrinsic variability is exacerbated by factors such as donor variability (genetic background, age, sex), inconsistent culture conditions (media composition, matrix coating), reagent inconsistency, and differences in handling protocols (seeding density, number of passages, time out of the incubator).[17] This leads to poor reproducibility of laboratory results. To overcome this and move toward clinical applications, it is imperative to invest in "GMP-grade reagents" (Good Manufacturing Practices), the use of "defined and chemically defined media," and the "automation" of processes to reduce human error and improve batch-to-batch consistency.[42] This is critical to meeting rigorous regulatory requirements for safety and efficacy.

Genetic Manipulation of Stem Cells

Genetic manipulation of stem cells is a frontier of regenerative medicine that allows cellular properties to be altered with unprecedented precision. This capability is used to correct genetic disorders, enhance the intrinsic properties of stem cells, or direct them toward specific cell lineages with greater efficiency.[44]

The main tools and techniques of genetic manipulation include:

- CRISPR/Cas9 and TALEN: These are cutting-edge gene editing technologies that allow for the precise correction of genetic mutations, the removal of defective DNA elements, or the replacement of damaged DNA regions.[45] CRISPR/Cas9, derived from a bacterial defense system, is currently the most efficient and frequently used method for genome editing.[48] These tools are applied for a variety of purposes, from studying disease mechanisms and developing treatments for specific disorders (such as sickle cell anemia or HIV infection) to creating immune-evasive or "hypohymenogenic" cells that can be transplanted without triggering an adverse immune response from the recipient.[27]
- Viral vectors: Viral vectors are modified biological vehicles, derived from viruses, that are used to introduce functional genes into stem cells. These vectors are highly efficient at delivering genetic material to cells. [45] The most common types include:
- Lentiviruses and retroviruses: These vectors have the ability to integrate genetic material into the host genome, resulting in stable, long-term gene expression.[25] They are frequently used to modify mesenchymal stem cells (MSCs) and induced pluripotent stem cells (iPSCs), enabling specific tissue regeneration and improved cell survival.[16]
- Adenoviruses: Unlike lentiviruses, adenoviruses do not integrate their genetic material into the host genome, resulting in transient gene expression. They are useful for applications that require temporary expression of therapeutic genes, such as the delivery of growth factors for acute wound healing or the reduction of inflammation.
- Adeno-associated vectors(AAV): AAVs are highly promising viral vectors in regenerative medicine due to their low immunogenicity and ability to mediate long-term gene expression. They are ideal for modifying MSCs and generating patient-specific iPSCs, and are used to deliver gene editing components such as CRISPR/Cas9.

The combination of gene editing (particularly CRISPR/Cas9) with stem cell engineering not only allows genetic diseases to be corrected, but is also essential for overcoming the biggest challenge in allogeneic therapy: immune rejection. The recipient's immune system, by ly distinguishing "non-self," attacks and destroys transplanted cells that are not genetically identical. For allogeneic therapies to be scalable and accessible to a broader patient population, this rejection must be overcome. Genetic engineering allows the manipulation of key genes for immune recognition, particularly the major histocompatibility complex (MHC) class I and II proteins. In addition, immunoregulatory factors such as PD-L1, HLA-G, and CD47 can be introduced to suppress immune responses mediated by T cells, NK cells, and macrophages. This leads to the creation of "hyporesponsive cells" or "universal cells" that are "invisible" to the recipient's immune system. This "immune camouflage" strategy is a crucial step toward the manufacture of "off-the-shelf" cell products that do not require donor compatibility, revolutionizing the logistics and cost of cell therapies, although it poses new safety challenges, such as the risk of tumorigenesis if the cells are not properly selected or if the recipient's immune system is not adequately suppressed·that do not require donor compatibility, revolutionizing the logistics and cost of cell therapies, although it poses new safety challenges, such as the risk of tumorigenesis if the modified cells acquire immune escape characteristics similar to tumor cells.

Mechanisms of Action in Tissue Regeneration

Stem cells contribute to tissue repair and regeneration through a variety of complex mechanisms that go beyond simple cell replacement. Understanding these mechanisms is essential for optimizing therapeutic strategies in regenerative medicine.

The main mechanisms of action include:

- Direct Differentiation: This is the most intuitive mechanism, where stem cells have the ability to differentiate directly into cells of the damaged tissue, thereby replacing lost or dysfunctional cells. This process is essential for the formation of new tissue and the restoration of function. For example, stem cells can be guided to become cardiomyocytes to repair damaged heart muscle or osteoblasts for bone regeneration.[12]
- Paranchymal Effects: There is growing evidence suggesting that a dominant mechanism of stem cell-mediated repair is the release of a variety of soluble factors into the surrounding tissue microenvironment. These paracrine factors include growth factors, cytokines, and extracellular matrix molecules, which act on neighboring cells. These factors promote a series of restorative processes, such as cell survival, angiogenesis (formation of new blood vessels), tissue remodeling, proliferation of resident progenitor cells, and reduction of apoptosis (programmed cell death).[8] Mesenchymal stem cells (MSCs) are particularly investigated for their potent protective and pro-angiogenic paracrine effects, secreting factors that can protect the heart from ischemia, improve contractility, and attenuate fibrosis.[23]
- Immunomodulation: Stem cells, especially MSCs, have a remarkable ability to modulate immune responses and mitigate inflammation at the site of injury.[15] They secrete factors such as interleukins, transforming growth factor beta (TGF-β), and indoleamine 2,3-dioxygenase (IDO), which collectively suppress inflammatory responses and promote immune tolerance.[23] This property is crucial for reducing the rejection of cell implants, especially in allogeneic transplants, and for the treatment of autoimmune diseases such as multiple sclerosis or rheumatoid arthritis.[11]

Growing evidence that paracrine effects and immunomodulation are dominant mechanisms in stem cell-mediated regeneration, rather than massive direct differentiation, has refocused research. This means that stem cells are not just "building blocks" that directly replace damaged cells, but act as "living pharmacies" that actively modulate the microenvironment of injured tissue. For example, MSCs secrete factors that protect cardiac tissue from ischemic damage, activate neovascularization, improve contractility, and attenuate fibrosis.[67] They also modulate the immune response to reduce inflammation and promote tolerance.[23] This understanding is driving research into the identification and optimization of factors secreted by stem cells, the engineering of stem cells to enhance their secretome (e.g., through hypoxic or genetic preconditioning [67], and the development of "cell-free" therapies based on exosomes or extracellular vesicles, which could offer improved safety profiles and greater scalability by avoiding the risks associated with the implantation of living cells.[9]

3D Bioprinting for Tissue Repair and Regeneration

3D bioprinting is a revolutionary technology that adapts the principles of 3D printing to biology to build complex three-dimensional structures. It uses specialized printers to deposit living cells and biomaterials, known as bioinks, layer by layer, with the aim of creating structures that closely mimic natural tissues and organs.

The main bioprinting methodologies include:

- Extrusion bioprinting: This is one of the most popular and versatile methods, where a viscous bioink is forced through a nozzle by pneumatic or mechanical pressure (piston or screw).[56] Its advantages include the ability to process at room temperature, the direct incorporation of cells into the printing process, and a homogeneous distribution of cells within the construct. However, a significant disadvantage is the shear stress that cells may experience when extruded, which can affect their viability, and a relatively low resolution compared to other techniques.
- Inkjet bioprinting: This is a non-contact method that ejects discrete droplets of bioink onto a substrate. The ejection of the droplets is achieved by thermal, electrostatic, or piezoelectric forces. The advantages of this technique include its high speed, accuracy, and resolution, as well as relatively low cost. However, its limitations lie in the need for low-viscosity, low-cell-density bioinks and the risk of nozzle clogging, especially with more viscous bioinks.
- Laser-assisted bioprinting (LAB): In this technique, a laser is used to deposit biomaterials onto a recipient. The laser strikes an absorbent layer (donor), vaporizing it and creating a bubble that expels the bioink in the form of droplets onto the substrate. The key advantages of LAB are its high precision, the absence of physical contact with the material (which minimizes contamination), and high cell viability after printing. However, it can leave metallic residues from the absorbent layer and is often a costly

technique.

- Stereolithography (SLA) and digital light processing (DLP): These volumetric photopolymerization techniques use a UV laser (SLA) or a digital light source (DLP) to selectively cure photopolymers layer by layer, transforming them from liquid to solid.[35] The main advantages are the ability to produce structures with high precision and resolution, smooth surfaces, and the ability to create complex and intricate geometries. Disadvantages include high cost, the need for support structures during printing, and a post-processing process involving washing and additional curing. In addition, the photopolymers used can be sticky, messy, and potentially toxic.

The diversity of 3D bioprinting methodologies is not simply a matter of preference, but reflects the need to adapt the technique to the specific properties of the bioink and the architectural complexity of the target tissue or organ. This indicates that there is no "one-size-fits-all" solution for biofabrication. Differences in resolution, speed, cell viability, and viscosity of bioinks suggest that each method is best suited for certain applications. For example, extrusion is effective for high-viscosity bioinks but can subject cells to stress. Inkjet printing is fast and inexpensive but requires low-viscosity bioinks. SLA/DLP offer high resolution for complex structures but are more expensive and rely on photopolymers.[11] This implies that "printing" a bone, which requires mechanical strength, will be fundamentally different from printing soft tissue or a blood vessel, which requires flexibility and microvascular precision. Optimizing bioprinting for complex organs will involve developing hybrid systems that combine multiple techniques or the ability to switch methods depending on the layer or type of tissue being printed, seeking maximum biomimicry and functionality.

Table 1 provides a detailed comparison of these technologies.

Table 1. Comparison of 3D Bioprinting Technologies

Characteristic	Extrusion	Inkjet	Laser-Assisted (LAB)	Stereolithography (SLA) / Digital Light Processing (DLP)
Key Principles	Mechanical/pneumatic force	Thermal/piezoelectric	Laser vaporization	UV photopolymerization(
Advantages	Versatility of bioinks, processing at room temperature, direct incorporation of cells, homogeneous distribution	High speed, precision, resolution, low cost	High precision, non-contact (avoids contamination), high cell viability	High precision and resolution, smooth surfaces, complex shapes
Disadvantages	Cell damage due to shearing, low resolution in some cases	Low biotinta viscosity, low cell density, risk of clogging	Metal residues from absorbent layer, high cost	High cost, need for supports, post-processing, toxicity of photopolymers
Speed	Slow	Fast	Medium	Fast
Resolution (µm)	>100	~5	1–10	~5
Cell viability (%)	40	>85	>90	~85
Biotinte viscosity ($mPa{\cdot}s^{-1}$)	$30–6x10^{7}$	3,5–12	1–300	No limitation
Typical biomaterials	Alginate, Gelatin, HA, PEG	Alginate, PEG	Collagen	HA
Cost	Low to medium	Low	High	Medium to high

Bioinks: composition, properties, and requirements

Bioinks are the fundamental material in 3D bioprinting, consisting of a formulation of polymers (natural or synthetic) infused with living cells. They are specifically designed to be "printable" and to provide an environment that supports cell adhesion, proliferation, and differentiation once the structure has been printed. The quality and properties of the bioink are crucial to the success of the final bio-printed structure.

Key requirements for an ideal bioink include:

- Cell viability: The bioink must ensure the survival and growth of cells during and after the printing process, providing a suitable environment for cells to adhere, grow, and differentiate.[30]
- Structural integrity: it must be able to maintain the desired shape after printing, providing the necessary structural support for three-dimensional construction.
- Biocompatibility: the bioink must be non-toxic and non-inflammatory, avoiding any adverse reactions when interacting with living cells or the recipient's body.
- Suitable rheological properties: the viscosity and flow behavior of the bioink must be compatible with the bioprinting technology used to enable precise layer-by-layer deposition.[30]

In terms of composition, bioinks are mainly based on:

- Hydrogels: These are considered the optimal candidates for bioinks due to their excellent

biocompatibility and high water content, which mimics the natural extracellular matrix (ECM) and promotes the encapsulation of cells and nutrients.[35]

- Natural polymers: common examples include collagen, fibrin, hyaluronic acid, alginate, gelatin, and Matrigel.[30] These materials offer biomimicry and cell signaling capabilities, which are crucial for guiding cell behavior. However, they often have low mechanical strength and may be less structurally stable.[35]
- Synthetic polymers: materials such as PEG (polyethylene glycol), PLA (polylactic acid), and PCL (polylactic acid) are examples. They allow precise control over the mechanical properties and degradation rate of the scaffold. However, they may require the incorporation of additional biological signals to improve cell interaction.
- Smart bioinks: These represent a significant innovation. They are formulations that can respond to external stimuli such as changes in temperature, pH, light, or electric/magnetic fields, or that combine multiple materials to release growth factors or drugs in a controlled manner. These bioinks allow dynamic interaction with the biological environment and active guidance of tissue regeneration.

The evolution of bioinks from simple cell support gels to "smart" and composite bioinks with finely tunable rheological and biological properties is essential to overcome the limitations of 3D bioprinting in the creation of functional and vascularized tissues. Natural tissues have a complex and dynamic ECM that provides biochemical and biophysical signals. Smart bioinks can react to various environmental factors and be customized to exhibit specific biological functions, including promoting tissue growth, vascularization, and nutrient passage. This is crucial for the viability of larger and more complex constructs that require vascularization and innervation. The continued development of bioinks that can mimic not only the structure but also the dynamic function of the ECM is a cornerstone for the biofabrication of complex organs. This involves research into materials with self-assembly properties, stimulus response, and controlled release of factors.

Applications in the creation of functional tissues and organs

3D bioprinting has emerged as a technology with immense transformative potential in regenerative medicine and the field of organ transplantation. Its ability to build complex biological structures layer by layer is opening up new avenues for tissue repair and replacement.

Among its most notable applications are:

- Personalized transplants: 3D bioprinting enables the creation of functional organs tailored to the specific anatomical and physiological needs of each patient. By using the patient's own cells, the risk of immune rejection, a common and serious complication in conventional transplants, is drastically minimized. This approach has the potential to eliminate long waiting lists for transplants by enabling the manufacture of organs on demand.
- Skin and wound repair: Skin bioprinting is one of the most advanced applications and is already used clinically for the treatment of severe burns and in cosmetic product trials.[13] Multilayer skin grafts that closely mimic the structure and function of natural skin have been successfully bioprinted, offering promising solutions for wound healing and scar reduction.[35]
- Bone and cartilage: The technology is used to repair damaged joints, such as knees and hips, and to treat bone fractures, promoting tissue regeneration and restoring joint function.[13] Bio-printed cartilage constructs have been created that mimic the mechanical properties of native tissue, which is crucial for their functionality in the body.
- Vascularized tissues and organ models: For the viability and functionality of larger, more complex organs, bioprinting blood vessels is essential. This technique allows for the creation of intricate vascular networks that ensure the supply of nutrients and oxygen and the removal of metabolic waste. Prototypes of functional miniature hearts, complete with chambers and blood vessels, have been achieved, as well as models of kidneys, livers, and lungs for research and pharmaceutical development purposes.
- Neural tissue engineering: The bioprinting of neural tissues and brain organoids is opening up new avenues for the study of neurodegenerative diseases such as Alzheimer's and Parkinson's, and for the development of neural implants that could restore brain function.
- Other applications: Bioprinting is also used in the manufacture of dental implants, customized prostheses, and bionic ears.[56] In addition, it is an invaluable tool for creating models of pathologies for drug testing and the development of personalized medicine.[13]

3D bioprinting is transforming biomedical research by enabling the creation of "disease models" and platforms for "drug testing" using functional human tissues. This not only speeds up drug development and reduces the need for animal testing, but also lays the foundation for truly personalized precision medicine. By creating

"printed human tissues"[13] or "bioengineered human mini-livers," researchers can "test toxicity susceptibility and demonstrate species-specific responses" that would only be observed in clinical trials. This, in turn, would shorten the time and cost of producing new drugs and allow critical examinations of drug interactions within a human-like system.[3] This establishes a crucial bridge between basic research and clinical application, enabling more robust preclinical validation and reducing the risk of failure in advanced stages of drug development. The ability to recreate patient-specific tumors or diseased tissues on a chip[12] opens the door to the selection of individualized treatments, marking a step toward precision medicine.

Combining scaffolding, stem cells, and bioprinting for the biofabrication of complex organs

The biofabrication of complex organs represents the culmination of efforts in regenerative medicine and tissue engineering, and its success depends on the synergy and integration of multiple technologies. Tissue engineering is based on the triad of cells, biocompatible three-dimensional scaffolds, and bioactive molecules.[2] 3D bioprinting, meanwhile, is the additive manufacturing tool that allows these components to be assembled with unprecedented spatial accuracy.[56]

The organ biofabrication process is multiphase and highly coordinated:

1. Design: approaches such as biomimicry (duplication of natural structures), tissue self-assembly (replication of embryonic development) or construction from tissue mini-blocks, individually or in combination, are used.
2. Material and cell selection: The choice of bioinks (biomaterial and cells) is crucial and specific to the form and function of the target tissue.
3. Bioprinting: The selected components are integrated using bioprinting systems such as inkjet printers, microextrusion, or laser-assisted printers, depositing the bioink layer by layer.
4. Application and maturation: Some bioprinted tissues may require a maturation period in a bioreactor to achieve the desired functionality prior to implantation.

3D bioprinting has overcome a significant traditional limitation of tissue engineering: the ability to simultaneously use multiple cell lines and biomaterials in a single procedure. This is vitally important, as complex organs in the human body are not composed of a single cell type, but rather the synergy of different cell types interacting in an intricate architecture.[28] For example, a histological section of the small intestine includes epithelial, connective, glandular, neuroendocrine, muscular, and endothelial tissue, all functioning in concert.

The real revolution in complex organ biofabrication does not lie in a single technology, but in the synergy of advanced scaffolds, stem cell engineering, and 3D bioprinting. This integration enables a shift from simple tissue replacement to the "functional recapitulation" of organs, which is a qualitative leap toward next-generation regenerative medicine. Scaffolds provide the structure, stem cells provide the capacity for regeneration and differentiation, and bioprinting is the "additive manufacturing technology" that allows cells and biomaterials to be combined layer by layer with "non-intrusive precision." Bioprinting enables the "spatial manipulation of living cells" and the "integration of functional components such as the extracellular matrix." The ability to include "multiple cell lines" in the same construct is key to replicating the complexity of natural organs. Future success will depend not only on improving each component individually, but also on optimizing their interaction and "self-assembly processes" and post-printing maturation in bioreactors to achieve complete and lasting biological functionality.[35]

Examples of Integrated Approaches for Organ Repair and Regeneration

The integration of scaffolding, stem cell engineering, and bioprinting has led to remarkable advances in tissue and organ biofabrication, although many of these are still in the prototype or miniaturized model stages.

Some notable examples of integrated approaches include:

- Functional mini-organs: Significant advances have been made in the bioprinting of functional mini-organs, such as a miniature liver that can metabolize drugs in a similar way to a human liver [20] and a miniature heart complete with ventricles, blood vessels, and chambers. These models, although not suitable for full human transplants, are crucial for research.
- Bioprinted skin: Skin bioprinting is one of the most advanced applications and is already used clinically for the treatment of severe burns.
- Bioprinted cartilage: Cartilage constructs have been created that mimic the mechanical properties of native tissue, offering hope for the repair of damaged joints.
- Bio-printed neural tissue: Bio-printed neural tissue engineering shows great potential for the study of neurodegenerative diseases and the development of neural implants.
- Decellularization and recellularization of whole organs: A promising approach involves the decellularization of donor organs (such as the heart, liver, lungs, and kidneys) to obtain an acellular

extracellular matrix scaffold. This scaffold is then recellularized with the patient's own stem cells, with the aim of creating personalized organs that avoid immune rejection.

Examples of complex organ biofabrication, although often at the prototype or mini-organ stage, demonstrate the viability of the "organ-on-a-chip" concept and the ability to replicate the microarchitecture and functionality of specific tissues. This is a fundamental step towards large-scale organ biofabrication. The creation of "mini-organs" or "organs-on-a-chip" allows for "detailed in vitro examination of blood vessel-related diseases," "testing the safety and efficacy of new drugs," and "modeling diseases."[12] This is crucial for personalized medicine, where a patient's own tumor can be "recreated on a chip" to test drug regimens and find the most effective combination.[26] These models are not only valuable in their own right for pharmaceutical research and development (reducing animal testing and costs), but also serve as test beds for refining bioprinting and vascularization techniques, which are key challenges for the biofabrication of full-size organs for transplantation.

Current Challenges

Technical Challenges

Despite significant advances in regenerative medicine and tissue engineering, the large-scale clinical translation of functional and safe biofabricated organs for human transplantation faces a number of complex technical challenges. The biofabrication of functional organs is an extremely complex "biological systems engineering" task that goes beyond mere structural replication and requires unprecedented control over cell biology and host interactions.

The main technical challenges include:

- Vascularization: One of the most formidable obstacles is ensuring adequate blood supply within engineered tissues and organs, especially for thick and complex constructs.[58] Without efficient vascular perfusion, cells located at the core of structures suffer nutrient deprivation and metabolic waste accumulation, leading to tissue necrosis and implant failure.[12]
- Innervation: The integration of functional nerve networks into artificial tissue is another critical challenge to ensure proper functioning and communication with the recipient's nervous system.
- Production scalability: Large-scale manufacturing of regenerative cell therapies is a considerable logistical and economic challenge. This is particularly true for autologous therapies, which are customized for a single patient and require a new manufacturing run for each individual, preventing economies of scale and reducing unit costs.
- Tumor formation: There is a potential risk of tumor formation associated with the use of stem cells, especially embryonic stem cells (ESCs) or induced pluripotent stem cells (iPSCs), which may acquire immunogenic characteristics or proliferate uncontrollably during prolonged culture or after implantation.
- Immune rejection: The rejection of allogeneic (from a genetically different donor) cells or tissues by the recipient's immune system is a major barrier to successful transplantation. Incompatibilities in the human leukocyte antigen (HLA) system and minor histocompatibility antigens are primarily responsible for this response. Graft-versus-host disease (GVHD), where the donor's immune cells attack the recipient's tissues, is a serious complication in allogeneic transplants. Genetic engineering strategies, such as HLA class I and II ablation and the expression of immunosuppressive molecules such as PD-L1, HLA-G, and CD47, are being investigated to create cells that evade immune detection.

The persistence of technical challenges such as vascularization and innervation in bioprinted organs, along with tumor formation and immune rejection, reveals that the biofabrication of functional and safe organs for human transplantation is an extremely complex "biological systems engineering" task. Lack of adequate vascularization leads to cell necrosis in the center of constructs.[12] Innervation is essential for proper function.[58] The recipient's immune system rejects "non-self," and there is a risk that undifferentiated or genetically modified stem cells will form tumors.[9] The scalability of individualized therapy production is logistically and economically complex.[61] These interconnected challenges suggest that the solution is not only technological (e.g., a better printer), but requires a deep understanding of developmental biology and immunology. The engineering of "hypohymenogenic" cells and the development of *in situ* or *ex vivo* vascularization strategies are critical areas of research. The long-term safety of genetically modified cells and the prevention of tumorigenesis are paramount ethical and regulatory concerns that must be addressed before widespread clinical translation.[27]

Regulatory and Economic Considerations

The translation of regenerative medicine and tissue engineering from the laboratory to the clinic is

intrinsically linked to overcoming complex regulatory and economic challenges.

The regulatory framework for regenerative medicine products is complex and varies significantly between countries and regions. Agencies such as the Food and Drug Administration (FDA) in the United States and the European Medicines Agency (EMA) in Europe play a crucial role in establishing guidelines to ensure the safety, efficacy, and quality of these products.[77] The FDA, for example, has introduced designations such as Regenerative Medicine Advanced Therapy (RMAT) to accelerate the development and review of promising therapies for serious conditions.[35] However, uncertainty about the appropriate regulatory pathways for emerging technologies and a lack of clarity in the definition of fundamental concepts can significantly hinder the development and commercialization of these therapies.[37] The tension between rapid innovation in regenerative medicine and the need for robust regulatory frameworks and process standardization is an inherent challenge that directly impacts the accessibility and cost of these therapies. Lack of standardization not only affects scientific reproducibility, but also raises costs and slows clinical approval.

Regenerative medicine therapies are generally extremely expensive, which represents a significant barrier to their accessibility and widespread adoption. High costs stem from several factors, including intensive research and development processes, complex and often customized manufacturing processes, and the need for specialized infrastructure. The manufacture of autologous therapies, in particular, where each treatment is tailored to a single patient, does not benefit from the economies of scale of mass production, keeping unit costs high.

The reproducibility of results is a persistent problem in tissue engineering, with variability that can be introduced from various sources, such as the donor, culture conditions, reagent inconsistency, and differences in handling protocols.[42] Good Manufacturing Practice (GMP) guidelines are crucial to ensuring that products are manufactured in a consistent and controlled manner, meeting quality standards and minimizing the risk of contamination, errors, and other quality-related issues.[62] The implementation of automation and the use of closed manufacturing platforms can help ensure rigorous quality control and nce and improve scalability, reducing human error and variability.[60] The lack of standardization in reagents and protocols introduces "variability," which hinders "reproducibility."[42] This inconsistency makes products difficult to validate for regulatory agencies such as the FDA and EMA. Long and costly approval processes [37] are exacerbated by a lack of regulatory clarity and the need for ongoing validations in the face of process changes.[40] For regenerative medicine to reach its full potential and become accessible to a wider population, greater collaboration between industry, academia, and regulators is essential to establish global standards, invest in automation, and optimize manufacturing processes to reduce costs without compromising quality and safety.

Ethical Considerations

Rapid advances in regenerative medicine have raised a number of profound ethical considerations that society and regulatory frameworks must address. These dilemmas arise from the unprecedented ability to manipulate life at a fundamental level.

- Stem Cell Origins: The use of embryonic stem cells (ESCs) is one of the most controversial areas, as their procurement involves the destruction of 3- to 5-day-old embryos, which conflicts with various beliefs about the beginning of human life.[18] This debate has driven the development of induced pluripotent stem cells (iPSCs), which offer an alternative by reprogramming adult cells and thus avoiding these ethical concerns.[12]
- Human-Animal Chimeras: The creation of chimeras, organisms containing a mixture of human and non-human cells, to model diseases or, in the future, generate humanized organs for transplantation, raises complex ethical questions. These include concerns about violating species boundaries, human dignity, and altering the moral status of the chimeric organism. These concerns have been raised in the contextof both animal and human chimeras.[43] The possibility that human cells could integrate into the brain or central nervous system of chimeric animals is a particular concern.
- Commercialization and Ownership of Tissues/Organs: The development of regenerative therapies involves significant investment and the generation of intellectual property, raising concerns about the exploitation of donors, patients, and researchers. There is debate as to whether bioengineered tissues and organs derived from human cells should be considered commercial products or whether they have a moral status similar to that of natural human tissues. The patenting of human tissues or cells and equitable access to therapies that are inherently expensive are central issues of debate. The possibility that altruistic donations of biological material may be used for profit also raises ethical dilemmas.
- Informed Consent: It is essential that donors of cells, tissues, or organs are fully informed about the potential use of their contributions, including the possibility that their biological materials may be used for commercial purposes or for the creation of novel biological entities such as chimeras. Transparency of information and the ability of donors to refuse certain uses are crucial.

Technological advances in regenerative medicine, particularly in stem cell manipulation and biofabrication, are rapidly outpacing existing ethical and regulatory frameworks. The ability to "design replacement parts for the human body" or even "humanized organs" in animals [43] forces a reassessment of what constitutes life, identity, and property. The possibility of "patenting human tissues" or the commercial use of altruistic donations raises dilemmas about justice and exploitation. This creates an imperative for proactive public and regulatory dialogue that balances the immense therapeutic promise with the protection of human dignity and equity in access to these innovative therapies. "Collaboration between regulatory agencies, scientists, and bioethicists" is required to "formulate robust frameworks" and "clear guidelines" to guide research and clinical application in a responsible manner, ensuring that the benefits of regenerative medicine are accessible and equitable for all of society.

Future Trends

The field of regenerative medicine and tissue engineering is constantly evolving, driven by the convergence of various scientific and technological disciplines. Future trends promise to further revolutionize the way diseases and injuries are addressed. The convergence of 4D bioprinting, smart biomaterials, and artificial intelligence represents a paradigm shift toward "predictive and adaptive regenerative medicine." These technologies will not only accelerate research and development but also enable the creation of therapies that can self-adjust and optimize in real time within the patient's body, taking personalization and efficacy to an unprecedented level.

Key future trends include:

- 4D bioprinting: This innovative technology integrates time as the fourth dimension, allowing bioprinted structures to transform their shape or function in response to predefined external stimuli, such as changes in temperature, light, pH, presence of water, or mechanical stress. This enables the creation of dynamic scaffolds that mimic the natural extracellular matrix and can actively adapt to the changing environment of the body, which is crucial for the long-term integration and functionality of regenerated tissues.
- Smart biomaterials: These advanced materials are designed to respond to changes in their environment (pH, temperature, ionic concentration, light, electric or magnetic fields).[33] Their responsiveness allows them to actively influence cell behavior, promote tissue growth, facilitate vascularization, and enable the controlled delivery of therapeutic agents or drugs. This dynamic interaction is essential for guiding regeneration in a more precise and adaptive manner.
- Artificial Intelligence (AI) and Machine Learning (ML): AI and ML are transforming regenerative medicine at an unprecedented rate. These technologies optimize the design of biological structures, improve research efficiency, and increase the effectiveness of treatments.[14]AI can analyze vast data sets to identify combinations of molecules that improve cell proliferation and differentiation, accelerate drug discovery, predict the effectiveness of therapies, and monitor the quality of bioinks and printing processes, ensuring consistent, high-quality results.[14]
- Organ-on-a-Chip (OoC): These miniaturized microfluidic devices mimic the structure and function of human organs in vitro, containing living human cells that replicate the functioning of real organs. OoCs are invaluable tools for drug discovery and development, compound toxicity assessment, disease modeling, and personalized medicine development, significantly reducing the need for and cost of animal testing.
- *In situ* bioprinting: This promising trend involves printing bioink directly into the patient's body to repair tissue defects. This approach eliminates the need for a second surgery to implant the bioprinted tissue and leverages the patient's own body as a natural bioreactor, which can improve integration and reduce the risk of contamination.

4D bioprinting [48] allows structures to "transform their shape or function in response to external stimuli," which is crucial for mimicking the dynamics of native tissue. Smart biomaterials [33] are the material component that enables this response. AI and ML can "optimize the design and predict the behavior" of these structures, and "continuously monitor regenerative progress" to "adjust therapies based on individual responses." This applies to in situ bioprinting, where the body acts as a bioreactor.[15] This promises therapies that not only replace tissue, but integrate and co-evolve with the patient's body, adapting to their changing needs and optimizing *in vivo* regeneration. AI in particular, by reducing R&D time and cost, could make these advanced therapies more accessible.

For a structured overview of these future trends, see table 2.

Table 2. Future Trends in Regenerative Medicine and Tissue Engineering

Trend	Key Concept	Potential Impact	Associated Challenges
4D bioprinting	Bioprinted structures that transform their shape or function over time in response to external stimuli.	Dynamic mimicry of the ECM, adaptive implants that adjust to the body environment, regeneration of complex tissues.	Precise control of shape/ function transformation, development of responsive materials with appropriate properties.
Smart Biomaterials	Materials that respond to environmental changes (pH, h r temperature, light, electric/ magnetic fields) to actively interact with the biological environment.	Controlled release of drugs and growth factors, active guidance of cell differentiation, promotion of vascularization and tissue growth.	Complexity in design and manufacturing, long-term safety, scalability of production.
Artificial Intelligence (AI) and Machine Learning (ML)	Use of algorithms to analyze large data sets, optimize designs, predict behaviors, and monitor processes in real time.	Acceleration of R&D, discovery of more efficient drugs, predictive personalized medicine, manufacturing optimization, and quality control.	Bias in training data, lack of explainability in some AI models, need for large, high-quality data sets, integration with clinical workflows.
Organ-on-a-Chip (OoC)	Microfluidic devices that mimic the structure and function of human organs in vitro, containing living cells and replicating their physiology.	Reduction and replacement of animal testing, more accurate disease modeling, faster and more effective drug screening, personalized medicine.	Scalability of chip production, standardization of devices and protocols, validation of clinical relevance.
In situ bioprinting	Direct printing of bioinks inside the patient's body to repair tissue defects, using the body as a natural bioreactor.	Immediate treatment of injuries, better integration of regenerated tissue, reduced risk of contamination and need for second surgeries.	In vivo material compatibility, printing accuracy in a dynamic environment, development of portable and automated printing tools.

CONCLUSIONS

Regenerative medicine and tissue engineering represent one of the most promising frontiers in contemporary medical science. This field, which has evolved from the early cellular concepts of the 19th century to the sophisticated gene editing and bioprinting technologies of the 21st century, fundamentally seeks to replace, design, or regenerate tissues and organs to restore normal body function. The frequent interchangeability of the terms "regenerative medicine" and "tissue engineering" reflects a paradigm shift toward the search for definitive cures, beyond the mere treatment of symptoms, driving the biofabrication of functional and personalized organs.

Advances in this field have been based on the understanding and manipulation of stem cells (embryonic, adult, induced pluripotent, and perinatal) and the development of scaffolding biomaterials. The transition to the use of induced pluripotent stem cells (iPSCs) and adult stem cells (ASCs) has been driven by the search for cell sources with less ethical controversy and a reduced risk of immune rejection, facilitating clinical translation and personalized medicine. At the same time, biomaterials have evolved from simple structural supports to "smart materials" and composites capable of actively interacting with the biological environment and responding to stimuli, enabling more controlled and biomimetic tissue regeneration.

Innovations in scaffolding technologies, such as electrospinning, solvent casting, gas formation, and, especially, decellularization, are enabling the creation of structures that increasingly mimic the structural and compositional complexity of the native extracellular matrix. These technologies are applied in the regeneration of a wide range of tissues, from cartilage and bone to liver, kidney, nerves, blood vessels, and skin, with the aim of creating controlled microenvironments that guide cell differentiation and prevent post-implantation complications.

Stem cell engineering has been revolutionized by advanced isolation, expansion, and differentiation techniques, although standardization and reproducibility remain critical challenges for clinical translation. Genetic manipulation, with tools such as CRISPR/Cas9 and TALEN, together with the use of viral vectors, not only allows genetic diseases to be corrected, but is also essential for overcoming immune rejection by enabling the engineering of "universal cells" that evade the r detection by the recipient's immune system. The growing understanding that paracrine effects and immunomodulation are dominant mechanisms in stem cell-mediated regeneration, beyond direct mass differentiation, is reorienting research toward the optimization of therapeutic signaling of stem cells.

3D bioprinting has emerged as the key tool for assembling these components with precision, enabling the creation of complex three-dimensional structures layer by layer. The diversity of bioprinting methodologies (extrusion, inkjet, laser-assisted, stereolithography/DLP) reflects the need to adapt the technique to the properties of the bioink and the complexity of the target tissue. Bioinks, in turn, have evolved into

smart, composite formulations capable of mimicking the native microenvironment and actively modulating regeneration. 3D bioprinting is transforming biomedical research by enabling the creation of disease models and drug testing platforms with functional human tissues, laying the foundation for truly personalized precision medicine.

Despite these advances, the biofabrication of complex functional organs safe for human transplantation faces significant technical challenges, such as vascularization, innervation, production scalability, tumor formation risk, and immune rejection. These challenges require extremely complex biological systems engineering and unprecedented control over cell biology. In addition, the field faces significant regulatory and economic considerations, where rapid innovation often outpaces regulatory frameworks and high costs limit accessibility. Ethical issues related to stem cell sourcing, human-animal chimeras, and tissue commercialization require proactive public and regulatory dialogue.

Looking ahead, regenerative medicine is moving toward "predictive and adaptive regenerative medicine" through trends such as 4D bioprinting, smart biomaterials, artificial intelligence and machine learning, organs-on-a-chip, and in situ bioprinting. These technologies will not only accelerate research and development, but also enable the creation of therapies that can self-adjust and optimize in real time within the patient's body, taking personalization and efficacy to an unprecedented level.

In short, regenerative medicine and tissue engineering are on the cusp of a revolution that could fundamentally transform healthcare. However, the full realization of their potential will depend on the ability of the scientific community, industry, and regulators to collaborate effectively, overcome complex technical, economic, and ethical challenges, and translate the promises of the laboratory into safe, effective, and accessible clinical solutions for patients worldwide.

BIBLIOGRAPHICAL REFERENCES

1. Molina S. Ingeniería Tisular. EuroGCT. https://www.eurogct.org/es/que-es-la-ingenieria-tisular

2. Instituto Nacional de Bioingeniería e Imágenes Biomédicas (NIBIB). Hoja informativa: Ingeniería de Tejidos y Medicina Regenerativa. NIBIB. https://www.nibib.nih.gov/sites/default/files/2022-05/Fact-Sheet-Ingenieria-de-Tejidos-y-Medicine-Regenerativa.pdf

3. Instituto Nacional de Bioingeniería e Imágenes Biomédicas (NIBIB). Ingeniería de tejidos y medicina regenerativa. NIBIB. https://www.nibib.nih.gov/espanol/temas-cientificos/ingenier%C3%ADa-de-tejidos-y-medicina-regenerativa-0

4. America Cell Bank. Paseo por la Historia de las Células Madre – Homenaje a la Vida. America Cell Bank. https://americacellbank.com.co/blog/paseo-por-la-historia-de-las-celulas-madre-homenaje-a-la-vida/

5. SciELO Perú. Ingeniería de tejidos y medicina regenerativa. SciELO. https://www.scielo.org.pe/scielo.php?script=sci_arttext&pid=S1726-46342010000300009

6. Gaceta UNAM. Regenerar huesos, cartílagos o tendones, la promesa de la ingeniería de tejidos. Gaceta UNAM. https://www.gaceta.unam.mx/regenerar-huesos-cartilagos-o-tendones-la-promesa-de-la-ingenieria-de-tejidos/

7. Hernández-Rodríguez T. Andamios y biomateriales para la ingeniería tisular. Rev Cubana Angiol Cir Vasc. 2018 ;19(2):1-14. http://scielo.sld.cu/scielo.php?script=sci_arttext&pid=S1684-45592018000200005&lng=es

8. Alvarado-Gómez A, Ramírez-Valera G. El uso de la bioingeniería en la medicina regenerativa. Revista digital universitaria. 2022 ;23(5). https://www.revistadigital.univim.edu.mx/index.php/UNI/article/download/235/496/1206

9. Ortegón D, Mejía-Restrepo J. Técnicas de electrohilado para la producción de andamios poliméricos con aplicaciones en ingeniería de tejidos. Rev Fac Ing Univ Antioquia. 2014; (70):206-18. https://www.redalyc.org/pdf/430/43031061019.pdf

10. Duque-Ramírez J, Hincapié-Montoya A, Serna-Giraldo J, Giraldo-Velásquez M. Bioimpresión 3D aplicada a la ingeniería de tejidos: una revisión. Rev Ing Univ Medellín. 2022 ;21(41):e214115. https://www.scielo.org.co/pdf/rium/v21n41/1692-3324-rium-21-41-e214115.pdf

11. Mayo Clinic. Stem cells: What they are and what they do. Mayo Clinic. https://www.mayoclinic.org/diseases-conditions/stem-cells/symptoms-causes/syc-20361665

12. ProgenCell. ¿Qué es la Terapia con Células Madre? ProgenCell. https://progencell.com/es/que-es-la-terapia-con-celulas-madre/

13. Start Stem Cells. Células Madre Mesenquimales (MSC): Todo lo que Necesitas Saber. Start Stem Cells. https://startstemcells.com/celulas-madre-mesenquimales-msc/

14. Clínica Dermatológica Internacional. Medicina regenerativa para el acné y las cicatrices del acné. Clínica Dermatológica Internacional. https://clinicadermatologicainternacional.com/es/medicina-regenerativa-para-el-acne-y-las-cicatrices-del-acne/

15. Bio-Rad. Aislamiento y Mantenimiento de Células Madre. Bio-Rad. https://www.bio-rad.com/es-es/applications-technologies/isolation-maintenance-stem-cells?ID=LUSR1TC4S

16. Sigma-Aldrich. iPSC Reprogramming Protocols. Sigma-Aldrich. https://www.sigmaaldrich.com/US/en/technical-documents/protocol/cell-culture-and-cell-culture-analysis/stem-cell-culture/ipsc-reprogramming-protocols

17. ASGCT. Gene and Cell Therapy FAQs. ASGCT. https://www.asgct.org/education/more-resources/gene-and-cell-therapy-faqs

18. Ghasemi-Mobarakeh L, Ashtiani S. Synthetic Polymers for Organ 3D Printing. PMC. 2022 https://pmc.ncbi.nlm.nih.gov/articles/PMC8796857/

19. Zhang J, Liu X, Chen Y, Wang M, Wang B, Sun B, et al. Mesenchymal stem cell-derived exosomes promote osteogenesis and angiogenesis. Cell Prolif. 2017 ;50(5):e12399. https://pmc.ncbi.nlm.nih.gov/articles/PMC5660271/

20. Spees JL, Lee RH, Gregory CA. Mechanisms of mesenchymal stem cell function in inflammatory liver injury. Exp Mol Med. 2016 ;48(1):e202. https://pmc.ncbi.nlm.nih.gov/articles/PMC3021634/

21. Wang Y, Yin Y. Harnessing the Immune Response for Regenerative Medicine through Immunomodulation in Stem Cell Therapy. Open Access J. 2024 https://www.openaccessjournals.com/articles/harnessing-the-immune-response-for-regenerative-medicine-through-immunomodulation-in-stem-cell-therapy-16824.html

22. Sociedad Canadiense Contra el Cáncer. Efectos secundarios de un trasplante de células madre. Sociedad Canadiense Contra el Cáncer. https://cancer.ca/es/treatments/treatment-types/stem-cell-transplant/side-effects-of-stem-cell-transplant

23. The Biomedical & Life Sciences Collection. The future of cell therapy: Scaling production for global reach. Drug Target Review. 2023 https://www.drugtargetreview.com/article/156266/the-future-of-cell-therapy-scaling-production-for-global-reach/

24. Patheon. Autologous Cell Therapy Manufacturing Challenges and Best Practices. Patheon. https://www.patheon.com/us/en/insights-resources/blog/autologous-cell-therapy-manufacturing-challenges-and-best-practices.html

25. Bio-Rad. Diferenciación de Células Madre. Bio-Rad. https://www.bio-rad.com/es-es/applications-technologies/differentiation-stem-cells?ID=LUSR2L8UU

26. Sigma-Aldrich. iPSC Differentiation. Sigma-Aldrich. https://www.sigmaaldrich.com/US/en/technical-documents/technical-article/cell-culture-and-cell-culture-analysis/stem-cell-culture/ipsc-differentiation

27. Sigma-Aldrich. Mesenchymal Stem Cell Differentiation. Sigma-Aldrich. https://www.sigmaaldrich.com/US/en/technical-documents/protocol/cell-culture-and-cell-culture-analysis/primary-cell-culture/mesenchymal-stem-cell-differentiation

28. Sigma-Aldrich. Neuronal Mesenchymal Stem Cells. Sigma-Aldrich. https://www.sigmaaldrich.com/US/en/technical-documents/technical-article/cell-culture-and-cell-culture-analysis/stem-cell-culture/neuronal-mesenchymal-stem-cells

29. Song J, Xu B, Fan Z, Lin Z. Genetically engineered hypoimmunogenic cell therapy. ResGate. 2024 https://www.researchgate.net/publication/382676407_Genetically_engineered_hypoimmunogenic_cell_therapy

30. Sun H, Zhao T. iPSC modification strategies to induce immune tolerance. Life Medi. 2024; 4(3):lnaf016. https://academic.oup.com/lifemedi/article/4/3/lnaf016/8101347

31. CELLINK. What are Bioinks? CELLINK. https://cellink.com/what-are-bioinks/

32. ResearchGate. 3D Bioprinting Techniques. ResearchGate. https://www.researchgate.net/figure/3D-Bioprinting-Techniques-A-Extrusion-Bioprinting-B-Inkjet-Bioprinting-and-C_fig1_366164227

33. faCellitate. Bioprinting - Technologies & Application. faCellitate. https://facellitate.com/resources/bioprinting-technologies-application/

34. Stellarix. Smart Bioinks for the Printing of Human Tissue Models. Stellarix. https://www.stellarix.in/smart-bioinks-for-the-printing-of-human-tissue-models/

35. Velarde T, Rojas N, Valdivia E. Hidrogeles de origen natural utilizados en la regeneración de tejidos. Dermatol Peruana. 2023 ;33(2):100-111. https://rpmesp.ins.gob.pe/index.php/rpmesp/article/view/13769/622

36. Additium 3D. Órganos impresos en 3D y aplicaciones en la medicina. Additium 3D. https://additium3d.com/organos-impresos-3d/

37. Biotriskel Biotech. Ingeniería de Tejidos Vascularizados. Biotriskel Biotech. https://biotriskel.com/ingenieria-tejidos-vascularizados/

38. Huang H, Mao B, Deng J, Cao X, Chen D. Stereolithography apparatus and digital light processing-based 3D bioprinting for tissue fabrication. Mil Med Res. 2023 ;10(1):17. https://pubmed.ncbi.nlm.nih.gov/36761021/

39. Kim Y, Kim S, Lee JS, Cho DW. Stereolithography 3D Bioprinting. Methods Mol Biol. 2020 ;2119:109-122. https://experiments.springernature.com/articles/10.1007/978-1-0716-0520-2_6

40. Fundación Instituto Roche. Bioimpresión 3D. Fundación Instituto Roche. https://www.institutoroche.es/static/archivos/Informes_anticipando_BIOIMPRESION_digital.pdf

41. Tomorrow Bio. Ingeniería de tejidos: un paso hacia la biofabricación de órganos. Tomorrow Bio. https://www.tomorrow.bio/es/post/ingenieria-de-tejidos-un-paso-hacia-la-biofabricacion-de-organos

42. Universidad de Zaragoza. Diseño, fabricación y caracterización de andamios poliméricos mediante electrohilado para su aplicación en la ingeniería de tejidos. Universidad de Zaragoza. https://zaguan.unizar.es/record/119504/files/TAZ-TFG-2022-859.pdf

43. FasterCapital. Bioimpresión: Avances en bioimpresión: de órganos a tejidos. FasterCapital. https://fastercapital.com/es/contenido/Bioimpresion--avances-en-bioimpresion--de-organos-a-tejidos.html

44. California Institute for Regenerative Medicine (CIRM). Development of 3D bioprinting techniques using human embryonic stem cells-derived cardiomyocytes for cardiac tissue engineering. CIRM. https://www.cirm.ca.gov/es/our-progress/awards/development-3d-bioprinting-techniques-using-human-embryonic-stem-cells-derived-cardiomyocytes-cardiac-tissue-engineering/

45. Cancer.gov. La investigación sobre células madre. Cancer.gov. https://www.cancer.gov/espanol/cancer/tratamiento/tipos/celulas-madre/celulas-madre-investigacion

46. Bookimed. Terapia con células madre: Precios y costes en el mundo. Bookimed. https://es.bookimed.com/article/stem-cell-therapy-cost/

47. SciELO Cuba - Infomed. Aspectos éticos en el uso de las células madre embrionarias humanas. Rev Cubana Salud Pública. 2012 ;38(1):119-125. http://scielo.sld.cu/scielo.php?script=sci_arttext&pid=S0864-34662012000100011&lng=es

48. Cibamanz2021. Bioética de la medicina regenerativa. Cibamanz2021. https://cibamanz2021.com/bioetica-de-la-medicina-regenerativa/

49. Number Analytics. La Importancia del Control de Calidad en el Desarrollo de la Medicina Regenerativa. Number Analytics. https://numberanalytics.com/es/blog/la-importancia-del-control-de-calidad-en-el-desarrollo-de-la-medicina-regenerativa

50. Frontiers Media S.A. Regenerative Medicine Advanced Therapy (RMAT) Designation for Allogeneic Cell Therapies: A Critical Assessment. Front. Med. 2021 https://www.frontiersin.org/articles/10.3389/fmed.2021.758832/full

51. Pew Charitable Trusts. FDA's Approval Process for Regenerative Medicine Products: Opportunities and Challenges. Pew Charitable Trusts. 2021 https://www.pewtrusts.org/en/research-and-analysis/reports/2021/04/fdas-approval-process-for-regenerative-medicine-products-opportunities-and-challenges

52. US Government Accountability Office (GAO). Regenerative Medicine: FDA Should Take Steps to Address Manufacturing and Supply Chain Challenges. GAO. 2023 https://www.gao.gov/products/gao-23-106511

53. Infiniti Research. Regenerative Medicine Market: Challenges and Opportunities. Infiniti Research. 2022 https://www.infinitiresearch.com/blogs/regenerative-medicine-market-challenges-opportunities

54. PubMed Central. The Ethics of Human-Animal Chimeras. PMC. 2017 https://pmc.ncbi.nlm.nih.gov/articles/PMC5568117/

55. The Hastings Center for Bioethics. Human-Animal Chimeras: Ethical Issues. The Hastings Center. https://www.thehastingscenter.org/briefingbook/chimeras/

56. America Cell Bank. ¿Por qué la medicina regenerativa con células madre podría ser la medicina del futuro? America Cell Bank. https://americacellbank.com.co/blog/por-que-la-medicina-regenerativa-con-celulas-madre-podria-ser-la-medicina-del-futuro/

57. StemSave. La Inteligencia Artificial en la Medicina Regenerativa. StemSave. https://stemsave.com/la-inteligencia-artificial-en-la-medicina-regenerativa/

58. Pharmaceutical Technology. Artificial intelligence in regenerative medicine. Pharmaceutical Technology. https://www.pharmaceutical-technology.com/comment/artificial-intelligence-regenerative-medicine/?gdpr

59. HackMD. Bioimpresión 4D. HackMD. https://hackmd.io/@xalca/S18J9jXz6

60. Bohrium. Bioimpresión 4D. Bohrium. https://www.bohrium.es/bioimpresion-4d-en-la-medicina/

61. Hilaris Publisher. Nanotechnology in Regenerative Medicine. Hilaris Publisher. https://www.hilarispublisher.com/open-access/nanotechnology-in-regenerative-medicine-44933.html

62. GlobeNewswire. El futuro de la medicina regenerativa: materiales inteligentes, bioimpresión 3D y más allá. GlobeNewswire. 2023 https://www.globenewswire.com/es/news-release/2023/12/12/2794939/0/es/El-futuro-de-la-medicina-regenerativa-materiales-inteligentes-bioimpresi%C3%B3n-3D-y-m%C3%A1s-all%C3%A1.html

63. EU Science Hub. Organ-on-a-chip: What future for human health and safety testing? EU Science Hub. https://joint-research-centre.ec.europa.eu/scientific-summaries/organ-on-a-chip-what-future-human-health-and-safety-testing_en

64. Team Consulting. Organ-on-a-chip: How can the industry overcome the remaining challenges? Team Consulting. 2023 https://www.team-consulting.com/news/organ-on-a-chip-how-can-the-industry-overcome-

the-remaining-challenges/

65. Scifiniti. Bioimpresión in situ: el futuro de la medicina regenerativa. Scifiniti. https://www.scifiniti.com/es/in-situ-bioprinting-the-future-of-regenerative-medicine/

66. Dr. Vanbeusekom. El futuro de la medicina regenerativa. Dr-Vanbeusekom.com. https://www.dr-vanbeusekom.com/es/el-futuro-de-la-medicina-regenerativa/

67. SHA Magazine. El futuro de la medicina regenerativa: avances y tendencias. SHA Magazine. https://shaellam.com/el-futuro-de-la-medicina-regenerativa-avances-y-tendencias/

68. Mayo Clinic. Medicina regenerativa: ¿estamos en el umbral de una revolución? Mayo Clinic. https://www.mayoclinic.org/es/medical-professionals/regenerative-medicine/news/regenerative-medicine-are-we-on-the-cusp-of-a-revolution/mac-20455580

FINANCING

None.

CONFLICT OF INTERESTS

None.

AUTHOR CONTRIBUTION

Conceptualization: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Data curation: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Formal analysis: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Research: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Methodology: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Project management: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Resources: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Software: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Supervision: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Validation: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Visualization: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Writing - original draft: Esteban Llorente-Nuñez, Diago Nunes-Coelho.
Writing - proofreading and editing: Esteban Llorente-Nuñez, Diago Nunes-Coelho.