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3D BIOPRINTING: AN OVERVIEW OF TECHNIQUES, MATERIALS AND APPLICATIONS

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ABSTRACT

3D bioprinting is an innovative technology that creates three-dimensional biological structures layer by layer using living cells, supportive biomaterials, and bioactive molecules. It helps create tissues that work better by placing cells in a controlled and organized way, which solves some of the problems seen in older methods that use pre-made scaffolds. 3D bioprinting has transformative promise in biomedical research, with applications in organ transplantation, tissue engineering, and regenerative medicine. 3D bioprinting has opened new possibilities in medicine, such as tissue repair, drug testing, organ transplantation, and regenerative therapies. It replicates authentic tissues through a layer-by-layer deposition of bio-inks. This review provides a comprehensive overview of the main 3D bioprinting techniques, types of bioinks and materials used, and current applications in medicine. It also discusses the advantages, challenges, and future directions of 3D bioprinting, showing how it could transform personalized medicine, reduce the need for animal testing, and advancing healthcare outcomes.

KEYWORDS: 3D Bioprinting, Bioinks, Bioprinting Techniques, Tissue Engineering, Regenerative Medicine.

HISTORY

The idea of 3D bioprinting came in the 1980, But its applications began in the early 2000s. When scientist started the way print to living cells. This new field became to known as 3D bioprinting. In 2003, Scientist Thomas Bend and his team modified an inkjet printer to print living cells instead of ink. This was a key moment in bioprinting history.[1] Between 2008 – 2010, researchers successfully printed multi-layered tissue structures such as blood vessels and skin-like constructs. These breakthroughs showed that bioprinting could produce structures similar to real tissues.[2] In 2013, a biotech company Organovo printed first the first functional human liver tissue for drug testing, it showed that how bioprinting played role in pharmaceutical research and reduced animal testing.[3] In 2019, a scientist 3D printed a small-scale heart with cells and blood vessels using a patient's own cells and biomaterials. While it was not yet suitable for clinical use or transplantation, but it was a major step towards personalized organ printing. Today, bioprinting is used to create the skin, cartilage, blood vessels, and mini-organs (organoids) for research and testing.[4]

INTRODUCTION

3D bioprinting is an advanced technology that enables the fabrication of complex three-dimensional biological structures using viable cells, biomaterials, and biological molecules, offering immense potential for tissue engineering and regenerative medicine.[5] This technology uses bio-inks made of living cells, growth factors, and biocompatible materials to make exact copies of tissues and organs. This has changed the way biomedical research is done, drug testing is done, and transplants are done.[6] 3D bioprinting has great promise for tissue engineering and regenerative medicine by making complex three-dimensional biological structures using living cells, biological materials, and biological molecules.[7]

Figure 1: 3D bioprinter.

Pre-bioprinting

Pre-bioprinting is the stage of preparation in which imaging techniques like MRI and CT scans are used to create digital models of tissues or organs. After that, a lab cultivates cells, typically from a biopsy, and mixes them with bio-ink, a gel-like substance that promotes cell growth and survival.[5]

Bioprinting or 3D Bioprinting

The actual printing process, known as "bioprinting," uses bio-ink to produce intricate biological structures. Printers utilize the digital model as a reference to determine the exact location of cells. This approach, which mimics natural tissues, may use laser-assisted printing, extrusion, and inkjet techniques.[7]

Post-bioprinting

Post-bioprinting is the process that occurs after the actual bioprinting step. It ensures that the printed tissue or organ becomes functional and viable once the bio-ink has been printed into the desired 3D structure. During this phase, the printed structure is maintained in a bioreactor, which provides a controlled environment that supports the cell growth, interaction, and development into functional tissues. This phase is critical for the tissue's development and the overall success of the bio printed structure.[8]

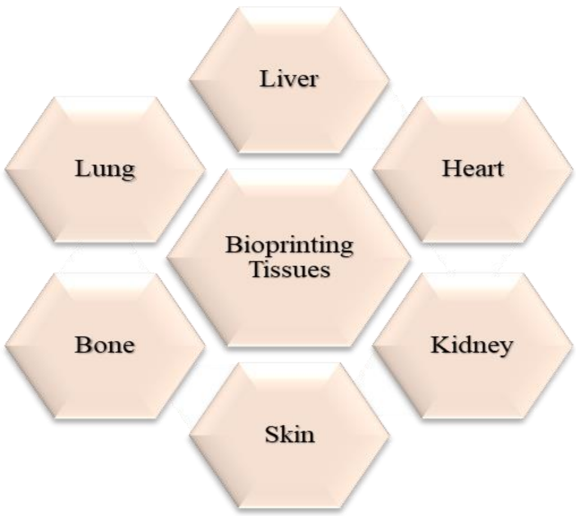

Figure 2: Bioprinting Tissues.

3D BIOPRINTING TECHNIQUES

There are four main types of 3D bioprinting techniques as shown in Figure 3.

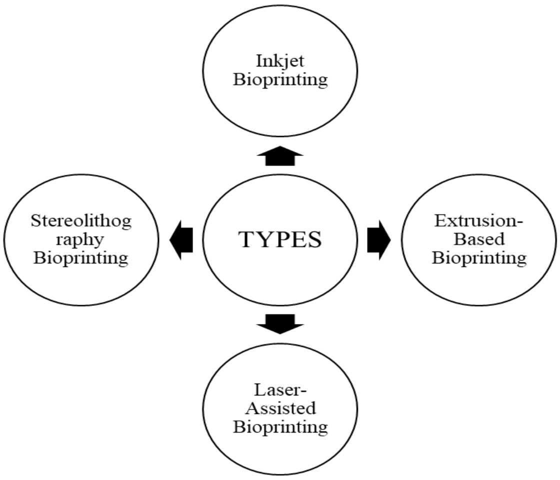

Figure 3: Types of 3D Bioprinting Techniques.

Inkjet Bioprinting

Inkjet bioprinting is a droplet-based technique that deposits bio inks layer-by-layer to fabricate complex biological structures with high precision.[9] Inkjet bioprinting uses droplets of the bioink containing living cells instead of ink, similar to a standard inkjet printer. People carefully place these droplets on a surface using heat or sound waves. This technique has multiple benefits, including low cost, excellent designs, and skin-like tissues. It is used to test the effect of drugs on cells. However, it may damage some cells due to the heat or pressure involved. It is only compatible with thin bio ink.[10]

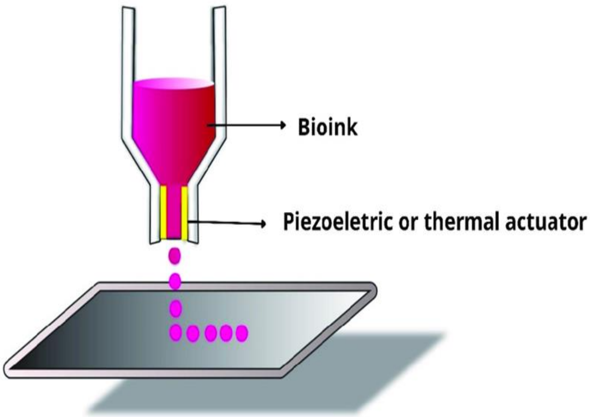

Figure 4: ***Schematic representation of a droplet-based inkjet bioprinting system.*** **Adapted from*: Lima TD, Canelas CA, Concha VO, Costa FA, Passos MF.* J Funct Biomater, *2022; 13(4): 214.*[11]**

Extrusion based Bioprinting

Extrusion-based bioprinting is a technique that uses mechanical or pneumatic forces to continuously dispense bio inks through a nozzle to create 3D biological structures.[12]

Extrusion-based bioprinting works similarly to cake icing. Thick bio inks are forced through a nozzle to operate it. It is one of the most common 3D bioprinting techniques, as it can print thick materials and create large, substantial structures.[13] It can also be used to produce blood vessels, bone, and cartilage. A downside is that it is a little slower and lacks fine details. Further, the force required to push the bio ink out leads to sometimes damaging the cells.[5]

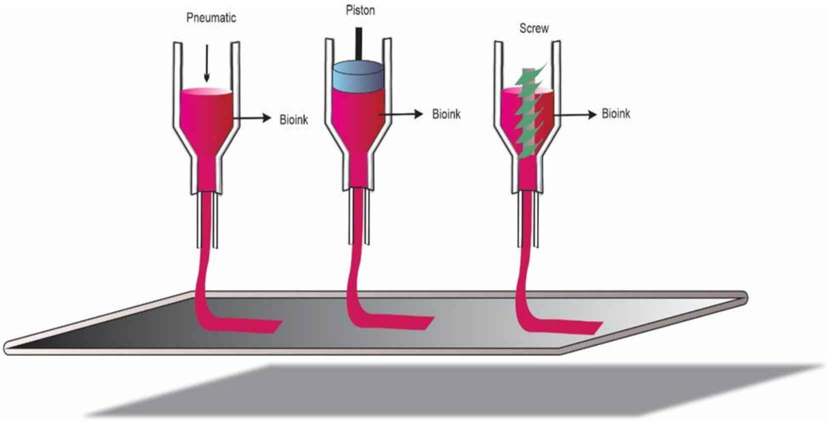

Figure 5: *Schematic representation* of the extrusion-based bioprinting system. Adapted from*: Lima TD, Canelas CA, Concha VO, Costa FA, Passos MF*. J Funct Biomater. *2022; 13(4): 214.*[13]

Laser Assisted Bioprinting

Laser-assisted bioprinting is a nozzle-free technique that uses laser pulses to precisely deposit bioinks on to a substrate.[14] It distributes small amounts of bio ink on to a surface using short laser pulses. This method does not use a nozzle, which means there is no risk of clogging.[15] It maintains the health and vitality of the cells because it is very precise and gentle. For this reason, it is used in the construction of sensitive tissues such as neurons or small blood vessels. The main disadvantages are that it's expensive and complicated to set up and run.[16]

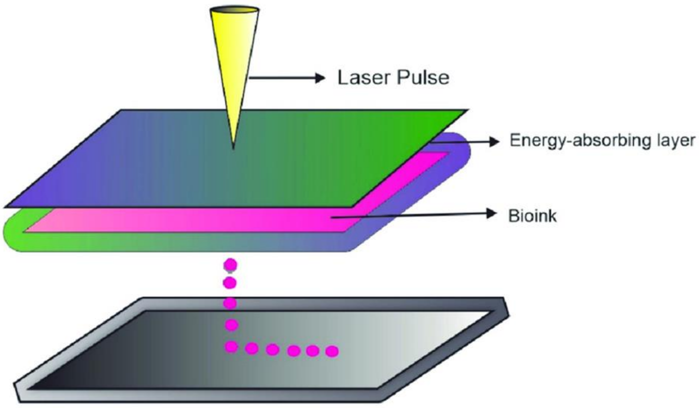

Figure 6: Schematic *representation* of the laser-assisted bioprinting technique. Adapted from*: Lima TD, Canelas CA, Concha VO, Costa FA, Passos MF*. J Funct Biomater. *2022; 13(4): 214.*[13]

Stereolithography Bioprinting

Stereolithography bioprinting is a technique that builds 3D structures layer by layer using UV or visible light. It works by shining light in a unique liquid bioink that hardens in the presence of light. It creates strong structures from the bottom up by repeatedly.[17] The technique is good for printing soft tissues or blood vessel networks since it is highly precise. It can create small, complex parts. One big advantage is that it is gentle on cells and allows high precision. But the bioinks used for bioprinting must be light-sensitive, and some materials or light exposure can damage the cells if not carefully managed.[18]

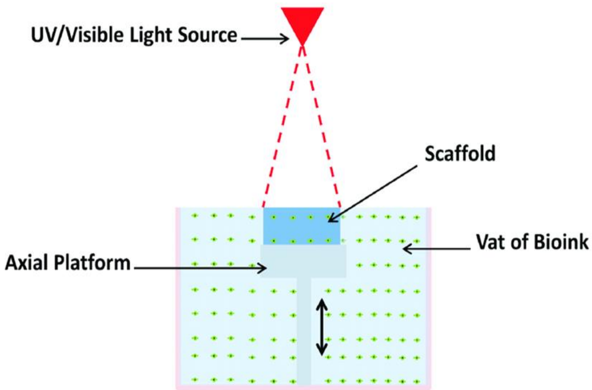

Figure 7: Schematic representation of stereolithography-based bioprinting, where a UV/visible light source polymerizes a photosensitive bioink layer-by-layer to form a scaffold on an axial platform. ***Adapted from: Kačarević ŽP, Rider PM, Alkildani S, Retnasingh S, Smeets R, Jung O, Ivanišević Z, Barbeck M. An introduction to 3D bioprinting: possibilities, challenges and future aspects. Materials. 2018 Nov 6; 11(11): 2199. p.9.*[17]**

BIOINKS FOR 3D BIOPRINTING

Bioinks are soft gel like materials used in 3D Bioprinting to create living tissues. They support cell growth. Common bioinks include alginate, gelatin, and collagen. On the bases of application, there are two types of bioinks are used. Natural bioink and Synthetic bioinks.[19]

Natural bioinks

Natural bioinks are obtained from plants and animal tissues. (e.g., alginate, gelatin, collagen). These are rich in biological cues that support cell growth and mimic the natural environment of tissues. They have lack of mechanical strength.[20]

Synthetic bioinks

These bioinks made by polymers such as PEG (polyethylene glycol) or PCL (polycaprolactone). These provides excellent control over properties like stiffness and degradation rate. However, they may need surface modification to support cell attachment and enhance bioactivity.[20]

Hydrogels

Hydrogels are water-rich, jelly-like materials commonly used as bioinks in bioprinting. They provide a cell-friendly environment that supports cell viability and function. Common hydrogel materials include alginate, gelatin, and GelMA. All these materials are well-suited for cell encapsulation and 3D printing applications.[21]

Decellularized Extracellular Matrices (dECM)

Decellularized extracellular matrices (dECM) are natural materials made by removing cells from tissues. They help in cell growth and build tissues in 3D bioprinting, making dECM bioinks ideal for tissue-specific applications like heart, liver, or skin bioprinting.[22]

Composite Bioinks

Composite bioinks are made by mixing of natural and synthetic polymers to improve printing and support cell growth. They create strong and functional tissues. For example, mixing alginate with GelMA, or PCL with collagen, creates a more stable structure while maintaining biocompatibility and mechanical strength.[23]

Table 1: Types of Boinks and their sources.

Bioink Type	Examples	Source	Reference
Natural Polymers	Alginate, Gelatin,	Plant- or animal-derived	7

	Collagen, Fibrin	extracellular matrices	
Synthetic Polymers	PEG, PCL, PLGA	Chemically synthesized	10
Composite Bioinks	Alginate + Gelatin + GelMA; GelMA + Nanoclay	Combination of natural and synthetic	24
Decellularized extracellular matrices (dECM)	Heart dECM, Liver dECM	Extracted from Tissues/Organs	25

Table 2: Advantages and limitations of different Bioinks.

Bioink Type	Advantages	Limitations	References
Natural	Biocompatible	Poor mechanical strength	7
Synthetic	Tunable	Limited bioactivity	10
Composite	Combine strength of both type	Complex formulations	24
dECM	Mimics native tissue	Source variability	25

Table 3: Common polymers used in Bioinks.

Polymer	Type	Properties	Applications	References
Alginate	Natural	Biocompatible	Vascular tissue	26,7
Gelatin	Natural	Cell adhesion	Skin, Bone, Cardiac tissue	5,7
Collagen	Natural	ECM mimic	Soft tissue regeneration	27,7
PEG	Synthetic	Tunable	Hydrogels, soft tissue	28,7
PCL	Synthetic	High mechanical strength	Bone, Cartilage	29,7
PLGA	Synthetic	Biodegradable	Bone scaffolds, Drug delivery	29,7

ADVANTAGES

3D bioprinting provides possible to create customized organs and tissues. It provides a promising solution for the global organ donor shortage. Bio printed tissues can be used for accurate drug testing and development.[5] The technology enables rapid prototyping of biological models. 3D bioprinting can produce complex and precise tissue structures.[30] In the long term, 3D bioprinting may reduce overall healthcare costs. It also enhances surgical planning and medical training.[31] Furthermore, 3D bioprinting supports advancements in regenerative medicine. It minimizes transplant rejection. It provides valuable tools for education and research.[32]

DISADVANTAGES

There are various disadvantages such as high cost of equipment and bio inks. The process of bioprinting is highly complex, requiring precise control to ensure the success.[5] Incomplete organ functionality. There are ethical concerns surrounding 3D bioprinting.[33] Regulatory Hurdles – Which can delay or prevent the approval processes for clinical use.[34] Maintaining the cell viability and functionality during and after the printing process is difficult.[35] Limited Scalability – Difficult to scale up from lab models to clinical use. There is a limited variety of bio inks and biomaterials available.[10] The time required to develop, test, and approve bio printed tissues is very long. Bio printed tissues and organs may carry unforeseen risks, including infections, structural failure, or rejection.[36]

APPLICATION OF 3D BIOPRINTING

3D bioprinting has several applications in tissues engineering, organ printing, drug testing, disease modeling and education model. It is used to create 3D structures using biomaterials. There are various applications as shown in figure 8.

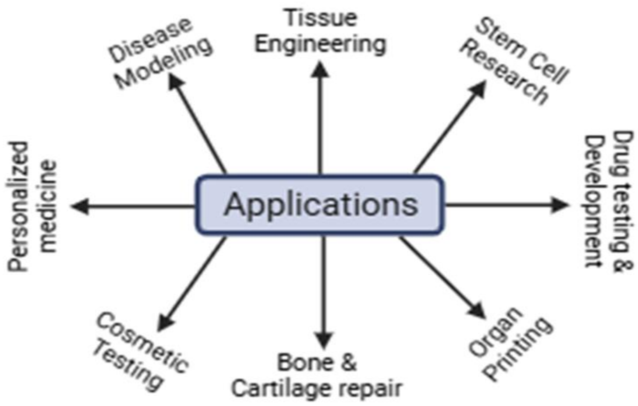

Figure 8: Key applications of 3D Bioprinting

1. Tissue Engineering and Regenerative Medicine

In tissue engineering, researchers use 3D technique to make parts of the body—like pieces of bone, cartilage, or blood vessels—that can replace damaged ones. It's kind of like repairing or rebuilding body parts using a printer that prints with cells. In regenerative medicine, 3D bioprinting helps the body heal by creating living tissues or organs that can be implanted. These printed tissues support healing and reduce the need for surgeries or the risk of transplant rejection.[37]

2. Organ Printing

Organ printing is very helpful for people who need organ transplants. Many patients wait a long time for donor organs, and sadly, some don't get them in time. If doctors could print organs using the patient's own cells, it would help solve the problem of not having enough donors. Right now, we can't print full organs for transplants yet, but scientists have already printed small parts of organs and are getting closer every year. 3D bioprinting could give patients the organs they need and reduce the need for donated ones.[38]

3. Cancer research and Drug testing

Scientists uses 3D bioprinting for creates the small, realistic models of human tumors. These models are made using real human cells, which allows scientists to study how cancer grows and test new drugs more safely and accurately before trying them on real people.[39]

4. Bone and Cartilage repair

3D bioprinting is a powerful tool in the field of bone and cartilage repair. This technology creates the custom-shaped grafts that perfectly fit a patient's bone defect. In terms of cartilage repair, 3D bioprinting makes it possible to create the structure that mimics the unique layered structure of cartilage. This is especially beneficial in repairing the damage from injuries or conditions like osteoarthritis.[40]

5. Personalized medicine

3D bioprinting is playing an important role in personalized medicine by enabling thecreation of patient-specific medical solutions. With the use of 3D bioprinting, medical professionals and researchers can create personalized implants, artificial limbs, implants for teeth, implants for breasts, surgical guides, and other surgical models that are tailored to each patient's particular anatomy. Better results arise from this, and surgeons are able to design procedures more precisely. 3D bioprinting is used to explored for developing personalized drug delivery systems and printing tissues that can be tailored to an individual's biological and genetic profile.[41]

6. Disease Modeling

3D bioprinting is also used to create models of human tissues and organs to study diseases. Instead of testing on animals or people, scientists can use these printed models to see how a disease grows and how different medicines might work. This makes research faster, safer, and often more accurate.[42]

7. Stem cell research

In stem cell research, 3D bioprinting is used to create tissues and small organ like structures. Scientists uses stem cells, which can turn in to variety of cells types and print them layer by layer to build tissues. This helps researchers' studies into cell growth, drug testing, and the development of future living organs.[43]

8. Cosmetic testing

3D bioprinting is used to create the skin models that mimic the human skins. These models allow companies to test cosmetics and skincare products without using animals. Scientists can evaluate how items affect real skin, like if they induce irritation or allergic reactions, by printing layers of human cells to generate a realistic skin structure. This approach is more ethical and can provide results that are more relevant to humans.[44]

CHALLENGES AND LIMITATIONS

There are various challenges and limitations of 3D Bioprinting such as Technical challenges, Cell viability & vascular problem and Regulatory & Ethical considerations as shown in (Figure 9).[45,46]

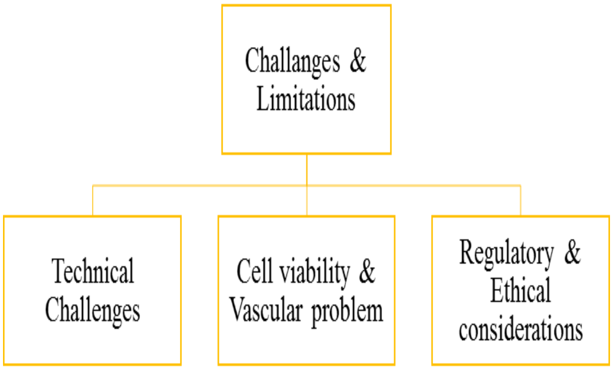

Figure 9: Challenges of 3D Bioprinting.

FUTURE PERSPECTIVES AND INNOVATIONS

There are various future prospective and innovations of 3D Bioprinting such as AI and machine learning integration, Emerging bioprinting technologies, Bioprinting of complex tissues and organs and Commercialization and clinical translation as shown in (Figure 10).[5]

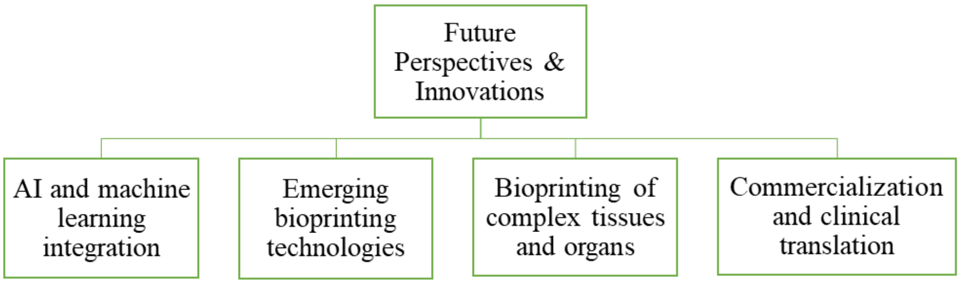

Figure 10: Future Perspectives & Innovations of 3D Bioprinting.

CONCLUSION

In this review, we discussed 3D bioprinting technology, focusing on its different types, advantages, disadvantages, limitations, and applications. 3D bioprinting is a modern technology that creates body parts like skin, tissues, and tiny organs using living cells and special materials. It offers a new hope for personalized treatments and reduces the need for animal testing and organ donors. In the future, this technology may enable the creation of fully functional organs, transforming personalized medicine and transplantation.

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