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REVIEW: INTRODUCTION TO 3D BIOPRINTING

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ABSTRACT

3D bioprinting is an advanced additive manufacturing approach evolved from traditional an printing to fulfill specific requirements for the applications of issue engineering and during the past decade, bioprinting has progressed regenerative medicine substantially showing its extraordinary potential for applications in different biomedical fields, however, a huge gap still exists for these Large scale industrialization and commercialization of this technology. In this review article first discus successful bioprinting companies. and their current Affairs effects bi commercialization of bioprinting technology. thin future challenges on the that restrict the application. of bioprinting will be discussed, finally, we will conclude several. factors, which Key Inhibit the the translational development of bioprinting, while should be addressed soon.

KEYWORDS: 3d bioprinting, methods of 3d bioprinting, history, need of the bioprinting, classification, types of bioprinting techniques, bioprinters, bioinks.

INTRODUCTION

In the 21st century, bioprinting is undoubtfully the most exciting area of additive manufacturing. technology this tec follows the lay-by layer manufacturing process, while utilizingnot only biomaterials but live cells, extracellular mater (ECM), growth factions, etc. for creating bio engineered constructed in the feildoftissueengineering and pharmaceutical development. for the bioprinting approaches, they can be separated into four major Categories.

1. Extrusion based
2. Droplet-based
3. SIA (stereolithography) based
4. Sex assisted bioprinting.[1,2]

Bioprinting is a of additive manufacturing (AN) also Subcateogary of known d's three-dimensional (3D) printing. It is defined as the printing of structures using viable calls, biomaterials and biological molecules.[3,4] Bioprinting Must produce Scaffolds with a Suitable microarchiteclute to provide Mechanical stability and promote cellingrowth. While also considering the manufacture on chemical all viability for instance chemical cytotoxicity the impactcaused by the use of solvents of pressure -induced opoptoticeffect produced during the extrusion of material.

The advantage of Homogeniouslydistributed Cell-Laden Scaffolds has been demonstrated faster integration with the host tissues, lower risk of rejection and most importantly uniform tissue growth in vivo.[5,6] Conventional cell seeding techniques are either static or dynamic and while the latter one results into the scaffolds. It is known as known affect cell morphology.[7] Bioprinting. techniques have been employed to fabricate micro-vascular like structure and have thepotential to position endothelical cells within the 30 startures as privascularization step s prior to implantation.[8] Bioprinting can be applied in a clinical of setting, where it can be used to create ingenerative scaffolds to suit patient specific requirements.[9] The process of Applying bioprinting Clinical sitting is depicted in fig.1

HISTORY

- In 1984 Charles Hull invented stereolithgraphy for printing 3D objects from digital data, symbolizing the birth of 3D printing. Bioprinting was first demonstrated in 1998 while Klebs using a stand a standard Hewlott – Packard inkjet printer to deposit cells by Cytoscrbing Technology.[10]
- In 1996 Forgacs and co-workers draw a conclusion that apparent tissue surface tension was the microscopic manifestation of molecular adhesion between cells and provide a quantitative measure for tissue cohesion.[11]
- In 1999 Odde and Renn first utilized laser assisted bioprinting to deposit living cells for developing analogs with complex anatomy.[12]

- In 2001direct printing of a Scaffold in the shape of bladder and seeding of human cells took place.[13]
- In 2002 the first extrusion based bioprinting technology was reported by Landers et al, which was later commercialized as " 3D – Bioplotter ".[14]
- Wilson and Bolond developed the first inkjet Bioprinter in 2003 by modifying on hp standard inkjet printer.[15]
- In 2019, Noor et al succeeded in manufacturing a perfusable scale -down heart.[16]

Classification

The process of Bio- Printing can be classified into main 4 steps:

1. Data Acquisition
2. Material Selection
3. Bio – Printing
4. Functionalization

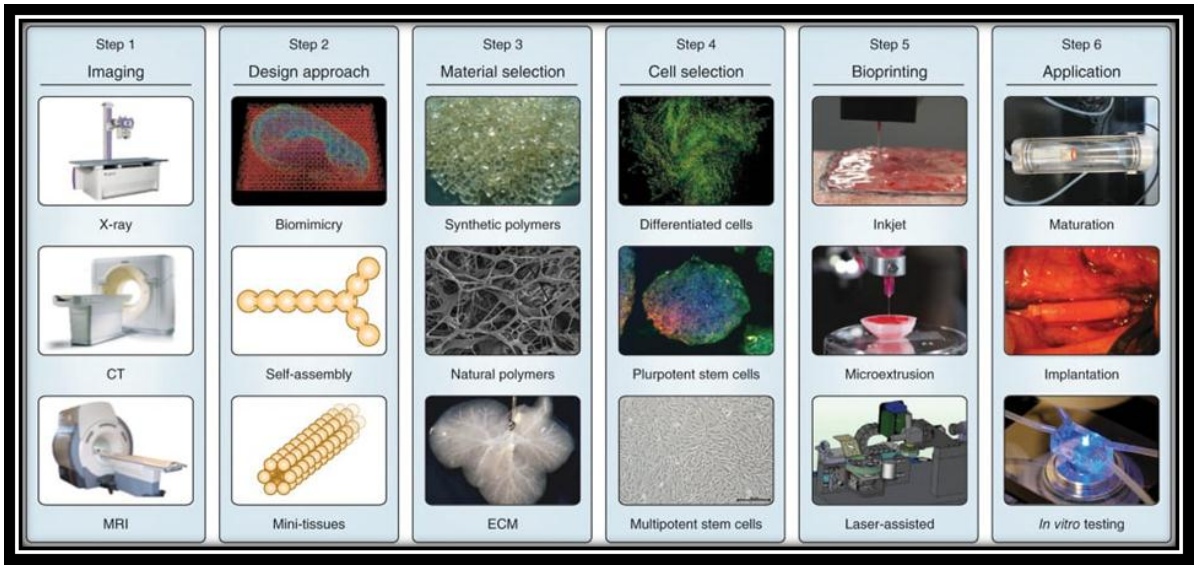


Figure No:- 1.

1: Data Acquisition

3D models can be obtained by using X- ray. computed tomography, magnetic resonance imaging [MRI] etc techniques to scan and reconstruct or directly using computer aided design [CAD] software to establish. 3D models would then be divided into 2D horizontal Slicus with customizable size and orientation by specific software. These data would be processed into particles or filaments according to different bioprinting approaches.

2: Material Selection

Materials including cells growth factor hydrogels etc should be chosen carefully accordingto requirements of printed structures and approaches. Strictly speaking the combination of these biomaterials is called bioinks while they could also be simply regarded as cell-laden. Hydrogels in most cases. The selection of bioinks is crucial to guranteebicompatibility printability and mechanical property, which would be further discussed in the last part of this review.

3: Bio- Printing

Before bioprinting appropriate configuration of printing parameters needs to be confirmed and observation during printing process necessary to make adjustment when uncounters any problems.

4: Functionalization

After printing to make dispersed cells forming connections and generating some functions of natural tissues organ through physical and chemical stimulation is the target.

Methods used for 3d bioprinting

Accordingto different prototyping principles and printing material, 3D bioprinting is mainly based on 3 central approaches:

1: Extrusion based bioprinting

2: Droplet based bioprinting

3: Photocuring based bioprinting

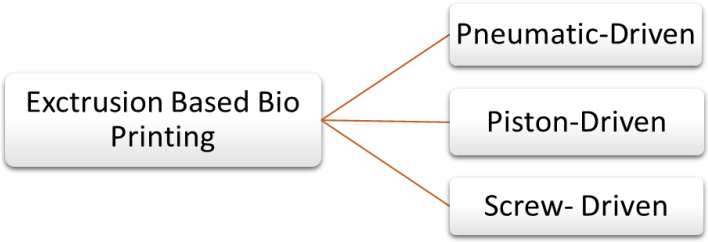


Figure No:- 2.

1: Extrusion – based bioprinting

It is also called Direct - Ink Writing which is derived from inkjet printing. It is the most widely approach of 3D bioprinting because of its versality and affordability. instead of single droplet extrusion based bioprinting produces ongoing filaments through continuous extrusion force this approach can be used and different concentrations of cells.[14] For this reason

researchers prefer extrusion based bioprinting to build tissue structures with sufficient mechanical property.[22]

Principle

Theortically extrusion based bioprinting extrudes bioink usually from a syringe through a nozzle by means of mechanical or pneumatic driven to form continuous micro filaments which are subsequently deposited on them receiving substrate and finally stacked into desired structures.

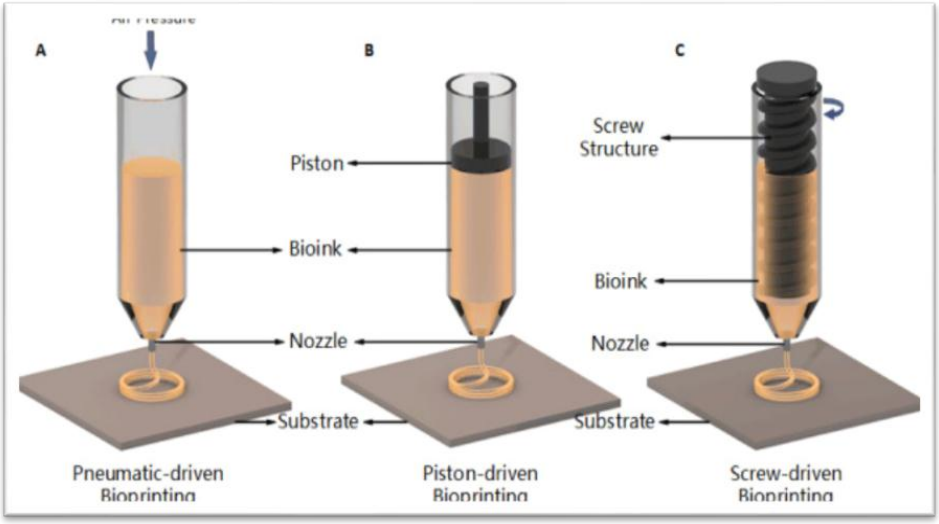


Figure No:- 3.

Extrusion based Bioprinting

1: Pneumatic Driven Extrusion

2: Piston Driven Extrusion

3: Screw Based Extrusion

1: Pneumatic Driven Extrusion

This technology extrudes materials using compressed air to achieve liquid dispensing. Typically, it consists of a piper and an air pump that are coupled to a syringe that is attached to a Bioink-filled air pump. Because they maintain their filament condition after extrusion, hydrogels with shear-thinning properties function well with pneumatically powered systems.

b: Piston Driven Extrusion

It is generally accepted that mechanically driven liquid dispensing systems are better suited for the extrusion of high viscocity biomaterials, such as artificial or naturally occurring high

molecular polymers. One of them, piston-driven extension, is highly popular, and related products like micro infusion pumps are widely available.

c: Screw Based Extrusion

Screw-driven devices are another type of mechanically driven liquid dispensing system that offer more volumetric control and support the notion of extruding biomaterials with higher viscosities. Similar to a piston-driven drive, a screw-driven drive uses a motor to directly drive a screw that is used for extrusion rather than a piston.

Bioprinters

There are several commercial bioprinters based on the extrusion method available on the market since it is the most practical, accessible, and widespread method for bioprinting.

We believe that printing scaffolds using FDM and then transplanting cells is not truly bioprinting technology. As a result, the 3D Bioplotter®, the world's first commercial 3D bioprinter, could be said to be capable of producing cells-rich biomaterials.[23] It was created by a University of Freiburg research team and quickly put into use by EnvisionTEC (which was established in 1999 and reorganised into EnvisionTEC GmbH[24] in 2002). It can also print rigid polymers, inorganic ceramic materials like PCL, hydroxyapatite (HA), and tricalcium phosphate (TCP) particles in addition to cell-laden hydrogels like gelatin, fibrin, alginate, agarose, etc. construct scaffolds that are not biodegradable.[25] NovoGen MMXBioprinterTM, developed by Organovo (established in Delaware, USA, in 2007), was another famous bioprinter.[26]

Multi- material bioprinting

Multiple material 3D bioprinting, which uses two or more types of materials to jointly construct a building, is currently becoming more and more popular. For example, Lee et al. used various bioinks encapsulating keratinocytes, fibroblasts, and collagen to print different parts of skin tissue[27]; Daly et al. adopted no-cell-laden PCL polymer with cell-laden bioink to print bone tissue precursor in turn; or it could be to use different material properties to form composite bioink to get around the limitations of various printing technologies and post tissue culture. Droplet-based and photocuring-based multi-material bioprinting have also been regularly reported, despite the fact that extrusion-based multi-material bioprinting has been widely employed.

In 2014 Levato et al. suggested a technique fusing microcarrier technology with bioprinting in 2014. The benefits of a hybrid bioink dubbed GelMA-GG MC-MSCs, which contained GelMA, gellan, and mesenchymal stromal cell (MSC)-loaded polylactic acid microcarriers, were demonstrated after comparing six distinct bioink compositions. Researchers used this bioink to create a bilayered cartilage structure, demonstrating the method's potential for building bone and osteochondral structures via microcarrier-based bioprinting.[28]

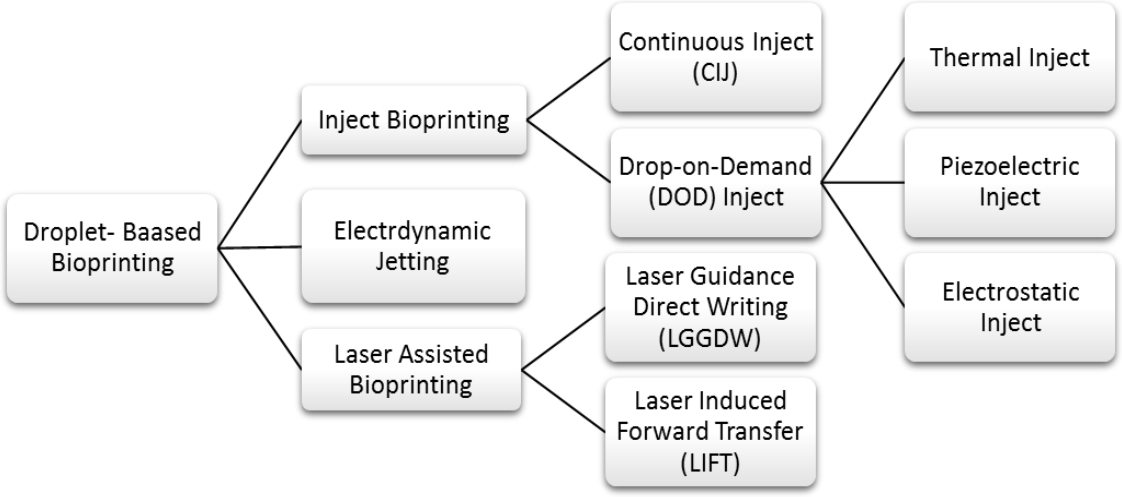
2: Droplet Based Bioprinting

In contrast to extrusion-based bioprinting, which uses continuous filaments as its basic unit, droplet-based bioprinting treats independent and discrete droplets as its fundamental unit. This method produces prints with a higher resolution than extrusion-based bioprinting. mainly because of its clarity and precision in handling biologics, particularly cells. Droplet-based bioprinting has various uses in tissue engineering, regenerative medicine, transplantation, clinical pharmacy, high throughput screening, and cancer research. Growth factors, medications, biomaterials, etc.

Principle

Droplet-based bioprinting can be split into three categories: inkjet bioprinting, electrodynamic jetting (EHDJ), and laser assisted bioprinting (LAB). Additionally, continuous inkjet (GIJ) printing and drop-on-demand (DOD) inkjet bioprinting are subcategories of inkjet bioprinting, while laser aided bioprinting is subcategorised as laser guidance. both laser-induced forward transfer (LIFT) and direct writing (LGDW).

Types of droplet – based bioprinting



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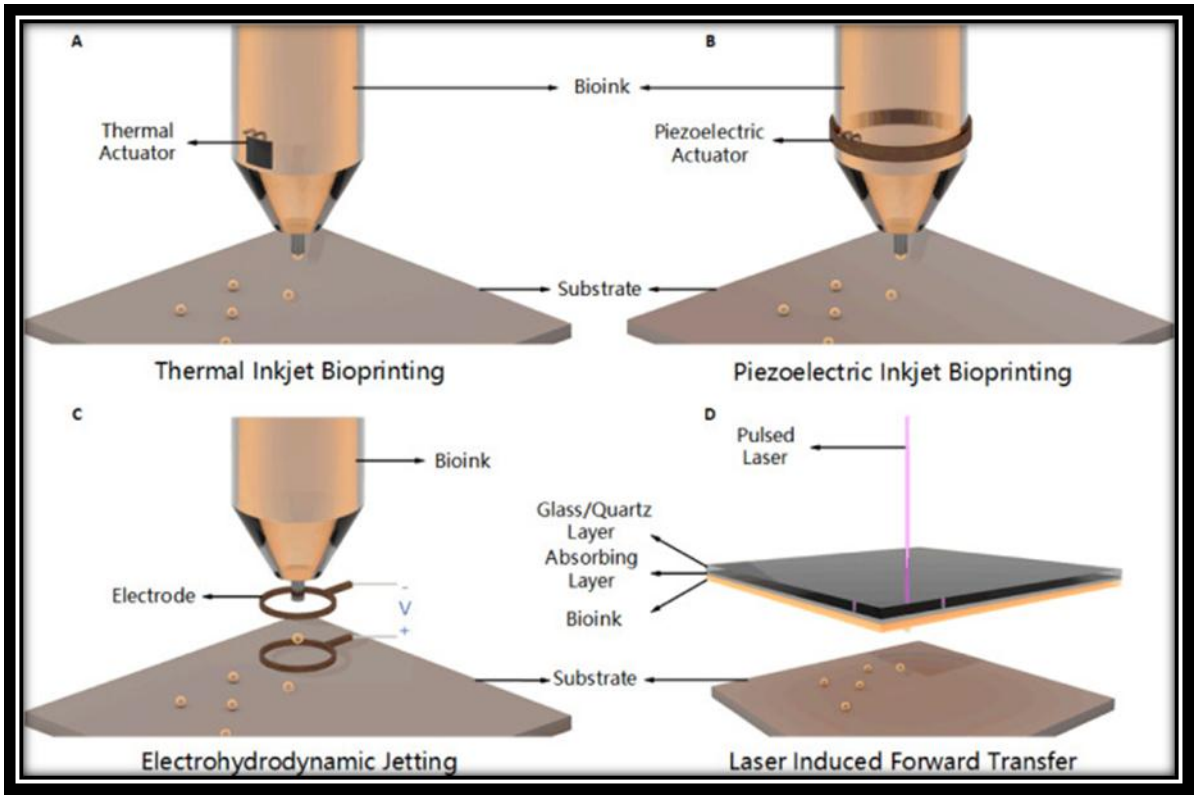

Figure No:- 05.

Inkjet bioprinting

Since Elmqvist of Siemens patented the first usable inkjet system in 1951, inkjet bioprinting is regarded as the earliest bioprinting technique.[29] Stanford University professor Sweet created the CIJ printing method later in the 1960s. The DOD inkjet printing system was created in the 1970s by Zoltan, Kyser, and Sears and licenced for use in the Siemens PT-80, the first commercial printer, in 1977.[30] In 1988, Klebe applied inkjet technology to bioprinting for the first time, utilising a Hewlett-Packard [HP] thermal DOD inkjet printer to deposit a collagen and fibronectin bioink solution.[6] Inkjet printing can be broken down into two steps: forming distinct droplets that are guided to specific areas of the substrate; and interacting with the substrate. As we mentioned above, there are two methods to form droplets: CIJ leverages a natural phenomenon called Rayleigh-plateau instability, which exhibits the natural tendency for a stream of liquid to undergo a morphological transformation to a train of discrete drops.

Electrohydrodynamic jetting

By forcing bioink via nozzles, inkjet bioprinting creates droplets. In this instance, a small diameter nozzle is used under a dramatic pressure that could occasionally impair the viability of the cells. EHDJ, on the other hand, makes use of an electric field, which would certainly

prevent bioink from being put under too much strain.[31] Figure 12C depicts the EHDJ's operating system. Surface tension causes the metallic nozzle to first be filled with bioink, which causes a spherical meniscus to form at the tip. Next, a high voltage is applied between the nozzle and substrate to create an electric field, which causes mobile ions to collect at the meniscus. Electrostatic repulsions between the ions cause the meniscus to become a Taylor cone as a result. Studies have demonstrated that elements used in EHDJ, such as electric field strength, cell concentrations, bioink composition, etc., would have an impact on the long-term survival of cells after printing.[32]

Laser- assisted bioprinting

LAB, which includes LGDW, LIFT, AFA-LIFT, biological laser processing (BioLP), matrix-assisted pulsed laser evaporation direct writing (MAPLE-DW), etc., is a category of non-contacting, nozzle-free printing processes to precisely deposit bio materials onto a substrate. AFA-LIFT, BioLP, and MAPLE-DW technologies are a few of them that are tailored from LIFT for various application scenarios.

Known as laser guiding direct writing, Odde and Renn employed laser-induced optical pressures to deposit living cells into a 2D pattern in 1999.[33] The basic idea behind LGDW is that a weakly focused laser beam, like an 800 nm tunable diode laser beam[34], is aimed towards a suspension of cells, optically catches them, and then directs them onto a substrate. When cells interact with light, a gradient force is created that pulls the cells to the centre of the light and directs their transfer along the axial direction onto a substrate (such as a coverslip). The varied refractive indices of the cells and surrounding fluid are the main component of this method.

Bioprinters

There are not as many droplet-based bioprinters on the market as there are compared to the development of commercial extrusion-based bioprinters.

By modifying an HP ordinary inkjet printer, Thomas Boland of Clemson University created the first inkjet bioprinter in 2003.[35] Organovo in the United States received related patents but has not yet made any inkjet bioprinters available for purchase.[36] The majority of inkjet-based bioprinters are lab-made or specially modified conventional inkjet printers. For instance, Nishiyama et al. from the University of Toyama constructed a hollow cylinder structure in their electrostatic inkjet bioprinter in 2009 by combining stepper motors and

EPSON's Sea-JetTM nozzles.[37] Other than the Sea-JetTM nozzles, reports have also been made about the DMP-2800 from Fujifilm Dimatrix, Xaar-126 piezoelectric inkjet print heads, etc. a range of cells and other biologics using bioprinting.[38] Founded in 1997, LabJet-Bio is a very accurate piezoelectric inkjet dispensing technology by Microjet in Japan. It can be used for bioprinting proteins, antibodies, enzymes, cells, and reagents, making biochips and biosensors, designing circuits with nanometal ink, screening drugs, and evaluating cell sheet production.

Applications

Skin

In 2012, On the basis of EHDJ, Sofokleous et al. created a portable, handheld multi-needle device in 2012. This apparatus may spray PLGA or poly(methylsilsesquioxane) (PMSQ) solution into piezoelectric inkjet bioprinting adult rt retinal ganglion cells (RGC) and glial cells under the control of a high voltage electric field and according to reference angle (RA).[39]

Photocuring-based bioprinting

Literally, photocuring-based bioprinting is a method of bioprinting that makes use of the photopolymerization property of photosensitive polymers under carefully regulated lighting. It often significantly improves printing resolution and printing speed when compared to other bioprinting methods. Additionally, it has the inherent benefit of not requiring concern over nozzle plugging or shear stress influencing cell viability**.**

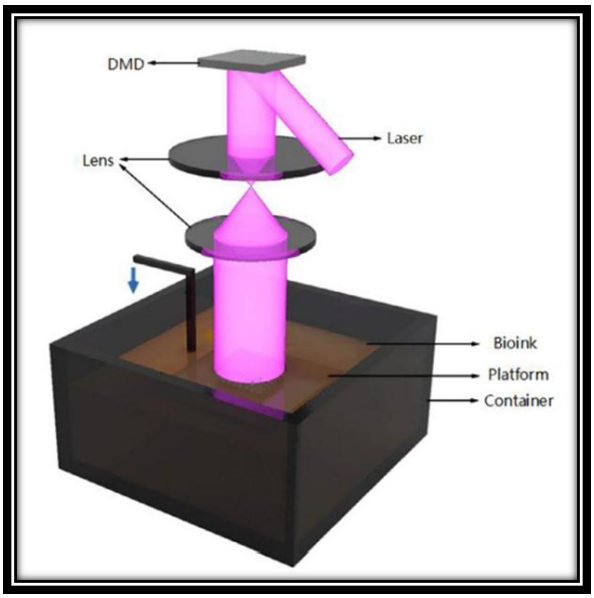

Figure No:-06.

Principle

The most popular use of photocuring-based bioprinting is the creation of cell-free scaffolds for post-printing cell seeding. However, recent reports have also mentioned cell-filled photocuring-based bioprinting. Photocuring-based bioprinting can be further divided into stereolithography (SLA) and digital light processing (DLP) based on various light scanning modes.

Types of photocuring- based bioprinting

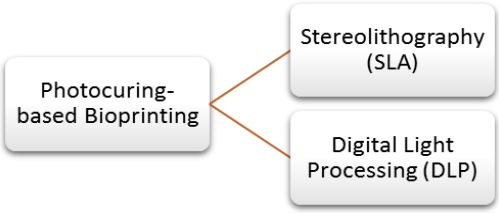

Figure No:-07.

Sterelithography

Layering software is used to slice the 3D digital model according to a certain Charles W. Hull created the first 3D printing method that was used for commercial purposes in 1984 with SLA. By selectively applying UV light to a vat of light-sensitive material, he was able to cure it layer by layer and produce 3D structures. Due to its high resolution, SLA is frequently used in the field of bioprinting to produce precise tissue scaffolds with adjustable geometry and porosity structure. It wasn't until 2004 that Boland's team at Clemson University used a commercial SLA printer (SLA-250, 3D Systems, Valencia, CA) to achieve cell-loaded SLA printing.[40] Following that, numerous research teams continued to refine SLA technique, which prompted further growth in the field of bioprinting. However, compared to cell-loaded bioprinting, scaffold printing has currently seen more application of SLA technology. Bioink is loaded in a tank with a moving platform for a typical SLA bioprinter. The usage of a dynamic mask during printing allows for photocuring one entire layer at a time. To put it simply, the mask contains a design pattern that light can travel through and sends to the receiving substrate.

The freshly cured ink solution is firmly bonded to the prior layer by submerging the platform in the liquid and pushing it up/down by a distance equal to the layer height. This procedure is repeated layer after layer until the last 3D construct is finished.[41] The majority of the time, extra bioink needs to be cleaned, and more photo-curing is needed after printing. The

precision of SLA will depend on elements like laser power, scanning speed, exposure duration, laser spot size, laser wavelength, etc.[42]

Digital light processing

In contrast to SLA, which solidifies each point individually, DLP solidifies the entire layer at once. A typical bottom-to-top DLP bioprinter is depicted in Fig. 16, which indicates that the bottom layer is printed first and that subsequent layers are printed on top of one another. A container filled with bioink, such as photocurable hydrogel or photosensitive resin that can solidify when exposed to a laser of a particular wavelength (typically UV light), photoinitiator, cells, etc.; a lifting platform that ensures lowering to a certain height (equal to the thickness of one layer) after one layer is finished being exposed, making it a new layer for photocuring; and an imaging system above the container make up this device.

Bioprinters

Most photocuring-based research teams throughout the world use a combination of biocompatible resins as bioink to convert the majority of commercial photocuring-based printers (SLA or DLP) into bioprinters. It is important to note that the dynamic optical projection stereolithography (DOPSL) technique has long been used in research by Professor Shaochen Chen's group at the University of California, San Diego. Due to its broad use, it has drawn significant interest from academia and is a representative application of DLP technology in the bioprinting field.

A photocurable bioprinter that uses photocuring has been marketed by our team.

Levels of optimization, the BP8600 is specifically made for printing hydrogels with great precision, particularly GelMA hydrogels. Several functional modules, such as the computer-free module, the material supplementary module, the multi-material, multi-cell bioprinting module etc.

Need OF THE PRESENT WORK

3d – bioprinting for health - sector

A chronic metabolic disease that is becoming more common worldwide is diabetes mellitus (DM).[45] Persistent hyperglycemia linked to insulin [type 1] production deficit is a hallmark of DM. Long-term DN resistance [type 2 DM].[46] causes the physiological function of organ systems to decrease. Serious health issues are caused by factors such the integumentary, neurological, and immune systems.[47] One of the most frequent problems in diabetes patients

is diabetic foot ulceration (DEU). Roughly 15–25% of diabetic people will experience a DFU at some point in their lifetime that necessitates intensive wound care.[48] Despite this High Major Public Health Concern, developing long-term healthcare systems for diabetic individuals is expensive. Therefore, it is crucial to enhance present therapies and create fresh therapeutic techniques to handle DFUs. Recent advancements in bioprinting technology have hastened the creation of individualised prosthetics and other healthcare fields. manufacture of tissue and implants.[49]

The Human Skin Structure

The skin is a unique organ that acts as a barrier between the body and the outside world. The body's appendages and skin. The integumentary system, which includes hair follicles, eccrine and glands, is important in immunological defence, thermoregulation, protection from ultraviolet light, and vitamin D synthesis.[50] The three main skin layers are.

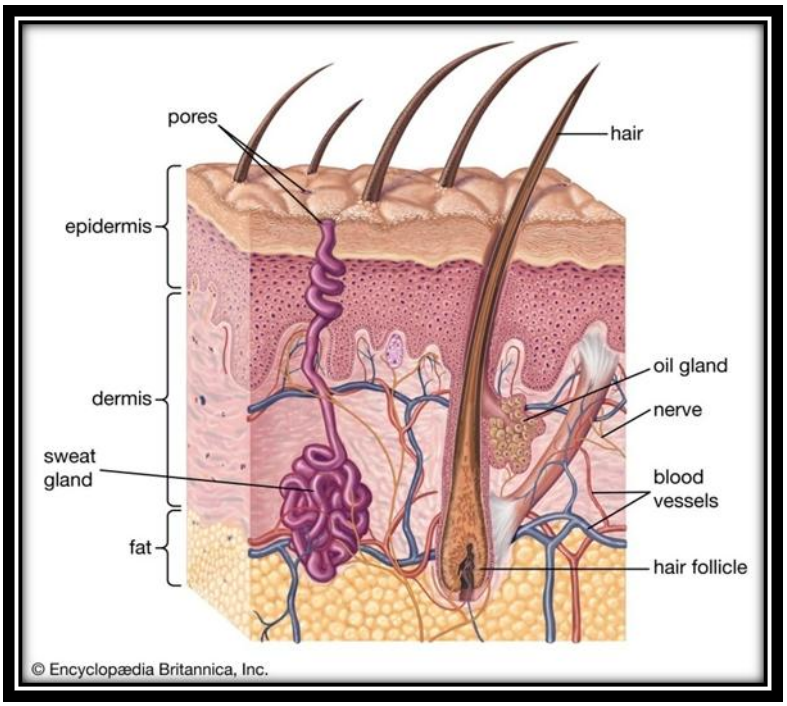

Figure No:- 08.

1: The skin.

2: The dermis

3: Hypodermal.

In order to prevent excessive water loss from the body and to defend against chemical assaults, the skin's outermost layer comprises successive sublayers that function as

permeability barriers.[51] The dermis is, too. The third primary layer of skin, the subcutaneous fat layer [hypodermis], is made up of two sublayers, the papillary layer and the reticular layer. prevents physical harm to the body and serves as an insulator for the the skin's supporting tissues.[52]

CONCLUSION

The potential disruption of the current design paradigm by 3D printing and bioprinting could be the biggest one yet. Throughout this century, including the delivery of healthcare and research. A paradigm shift in the use of 3D printing for surgery is expected to result from the inclusion of human cells and biocompatible materials, which will allow for the 3D printing of living tissue and organs. Patients may be able to obtain customised treatments at every stage of their healthcare journey because to the promise of 3D printing de novo body parts, eliminating the necessity for organ transplantation, and replacing the function of animals in the development and testing of novel pharmaceuticals.

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