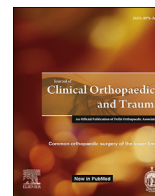




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Original article

3D printing applications in bone tissue engineering

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ABSTRACT

Purpose: 3D printing technology provides an excellent capability to manufacture customised implants for patients. Now, its applications are also successful in bone tissue engineering. This paper tries to provide a review of the applications of 3D printing in bone tissue engineering.

Methods: Searching by keywords, from the Scopus database, to identify relevant latest research articles on 3D printing in bone tissue engineering, through “3D printing” “bone tissue engineering”. This study makes a bibliometric analysis of the identified research articles and identified major applications and steps.

Results: 3D printing technology creates innovative development in bone tissue engineering. It involves the manufacturing of a scaffold with the combination of cells and materials. We identified a total number of 257 research articles through bibliometric analysis by searching through keywords “3D printing” “bone tissue engineering”. This paper studies 3D printing technology and its significant contributions, benefits and steps used for bone tissue engineering. Result discusses the essential elements of bone tissue engineering and identifies its five significant advancements when 3D printing is used. Finally, ten useful applications of 3D printing in bone tissue engineering are identified and studied with a brief description.

Conclusion: In orthopaedics, bone defects create a high impact on the quality of life of the patient. It leads to a higher demand for bone substitutes for replacement of bone defect. Bone tissue engineering can help to replace a critical defect bone. 3D printing is a useful technology for the fabrication of scaffolds critical in bone tissue engineering. There are different binders which can create bone scaffolds with requisite mechanical strength. These binders are used to create excellent osteoconductive, bioactive scaffolds. Computed tomography (CT) and Magnetic resonance imaging (MRI) help to provide images of specific defects of an individual patient, and these images can further be used for 3D printing the defective object. A bone defect caused by specific disease is sorted out by transplantation in clinical practice. Now a day bone tissue engineering opens a new option for this treatment of bone defects with the manufacturing of porous bone scaffold using 3D printing technology.

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1. Introduction

Bone tissue engineering is an interdisciplinary approach which

utilizes cells and combination of biomaterials to restore required functions of tissues. Scaffolds are an essential part of bone tissue engineering. They are 3D structures which provide a template for cell attachment in bone tissue formation.¹ There are various critical parameters to define the performance of scaffolds; these include, pore-volume, pore size, chemical properties and mechanical strength. It is made as per the understanding of bone mechanics, bone structure, and tissue formation for new functional bone tissues. There are varieties of methods extensively used for making porous bone scaffolds. These methods are chemical foaming, freeze-drying, solvent casting and foam-gel.^{2,3}

3D printing is first introduced to create a prototype for the

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development of products of specific industries. This technology is part of additive manufacturing (AM) which is used to create parts, layer by layer from the 3D Computer-Aided Design (CAD) model. In printing scaffold, using Polyjet printing, parameters are optimised to obtain appropriate quality, these include powder flowability, binder drop volume, powder packing density and layer thickness. There are different types of 3D printing technology, each having its capability to print parts and are materials specific. These technologies use, material such as gypsum powder, plastics, resins, aluminate, ceramics, sand, metal, Polyether ether ketone (PEEK), graphene and other materials to create the required part. Inkjet 3D printing technology provides disruptive innovation for bone tissue engineering. It facilitates the requisite distribution of cells in the scaffold. It also uses bioink for efficient regeneration and part reconstruction.^{4,5}

3D printing technology creates advancement in the field of bone regeneration medicine by fabricating biological tissues or organs. 3D bones tissue are easily created using bioink to overcome the challenges. This bioink is in the form of hydrogels and viscous fluid. Bioink is used to manufacture cell-laden 3D structures. Thus, 3D printing and bioink are successfully used to manufacture tissue of bone and considers different parameters for developing specific bioink such as printability, mechanical integrity etc.^{6,7}

This emerging field aims for the treatment of bone disease. There is a vital role in cell adhesion, differentiation, proliferation, migration, and mineralized bone tissue formation. Bone consists of cancellous, while cortical bone has two different structures. Cancellous is the inner part of the bone, and cortical is the outer part of the bone. These two collectively have capabilities of self-healing to restore normal operations. Also, there are options of autograft and allograft, by which bone tissue engineering provides the capability to restore and improve its functions. This review based article provides a study on 3D printing as adopted in bone tissue engineering and addresses the following issues:

Issue 1: to study the 3D printing technology, its significant contributions and significant benefits for bone tissue engineering;

Issue 2: to briefly describe the concepts of bone tissue engineering with its basic elements;

Issue 3: to identify steps of 3D printing in bone tissue engineering;

Issue 4: to study different advancement in bone tissue engineering using 3D printing;

Issue 5: to study and analyse the research status of 3D printing in bone tissue engineering;

Issue 6: to identify significant applications of 3D printing in bone tissue engineering;

Issue 7: to study the miRNA therapy integration with 3D printing technology.

2. 3D printing

In 1980, 3D printing technology was introduced, and today it is providing extensive benefits over traditional manufacturing technologies. It can manufacture different parts with different and unconventional materials. The applications of 3D printing are growing, and now it has uses in the field of bone tissue engineering. Here, this technology provides excellent capabilities to print porous scaffolds with required shape and size using layer by layer technique. 3D printing is becoming popular due to its capability manufacturing of the customised parts at lesser cost and time. For printing any part, first, step is the importing of the 3D digital CAD file and converting to Standard Triangulate Language (STL) file

format, for facilitating 3D printing of the part. After that setup files for printing are provided with proper orientation of the part in 3D Print software.⁸ Finally, the part is printed with selected material. Post-processing is done as per the requirements of quality and strength of part.^{9,10}

AM also plays a vital role in product development and provides a modified concept in the industry. The design and development of the medical implants, tool, instrument and devices are easily possible by using this technology. It increases the efficiency and quality of the product, but there is a reduction in development cycle time and cost of the product. 3D printing provides incremental innovation in medical, dentistry, cardiology, orthopaedics, engineering, research, food, agriculture, automobile, architecture, aerospace and various other fields.^{11–15}

3. Bone tissue engineering

Bone tissue engineering is an essential technique, where a scaffold for the cell is manufactured using a biomaterial having suitable biochemical and physicochemical properties. It is used to replace or improve bone with surrounding tissues. It is used to create advancement in the orthopaedics field. Thus, bone tissue engineering, with its extensive capability of formation of bone structure provides significant progress.¹⁶

3D printing provides an excellent opportunity by creating scaffolds for bone tissue engineering. It enhances bone formation for cell culture to optimize osteogenesis. Bone tissue engineering is now becoming feasible in today medical field. This technique is used successfully for grafting of bone, by which a surgeon can repair the bone of a patient. Scientists and researcher use scaffolds for bone tissue. It resembles bone and has required porosity and composition. A scaffold is created according to the desired area, which has to be repaired using cells. Two methods are used to produce bone tissues, either these tissues are taken from existing bone or stem cells. These are used successfully for bone in the scaffold.^{17,18}

4. Essential elements of bone tissue engineering

The important part of tissue engineering is to repair a bone defect, may be caused by fracture or trauma. In conventional techniques, there is a change in structure and function by biological substitutes when used for repairing of damaged tissue.^{19,20} Thus, to sort out bone defects, 3D printing is used to construct artificial bone with required shape, size, properties and mechanical strength. This technology is more suitable as compared to other traditional manufacturing technique and consists of the following elements:

- Bone Scaffolds
- Cells
- Growth factors

4.1. Bone scaffold

Scaffolds are mainly applied to make a physical and mechanical stable bone-like structure. In clinical medicine, it has an excellent capability to rebuilt organs with normal function. It allows attachment and migration of the cell to retain biochemical and other required biological properties. Scaffolds help all cells to integrate and fulfil the requirements of bone tissue treatment. This scaffold is created by using AM (3D printing) technologies as per the required shape and size of the individual customer. It helps to treat the disease/fracture successfully, by fulfilling the challenge to regenerate and growth of new tissue for medical applications,

thereby improving patient survival.²¹

4.2. Cells

In living beings, cells are the basic building blocks and are specific to the body part. These cells provide structure to the body and help carry out specialized functions. Cells perform different functions and provide shape to the body part. 3D printing is introduced to assist localized cell growth as per the specifications.^{22,23}

4.3. Growth factors

There are different growth factors like fibroblast growth factor, vascular endothelial growth factor and bone morphogenic proteins also are required in bone tissue engineering. 3D printed scaffold minimizes the side effect.²⁴

5. Major contributions of 3D printing

3D printing is used for the development in different fields and provides better flexibilities to achieve improved accuracy. This technology explores the manufacturing of complex products as per the customer's requirements. There are different capabilities, advantages and limitations of this technology during its practical implementation. Its applications areas include medical, dentistry, engineering, food, agriculture and related fields. There are different types of 3D printing, with different material, processes and part property. These technologies now provide excellent surface resolution, surface finish, colour 3D parts, recycling of waste materials, lightweight parts, lesser maintenance cost, environmentally friendly production and capability to make parts with different types of material.^{25,26} Coming subsections provide major areas wise contributions of 3D printing.

5.1. Academia

3D printing technology helps students, researchers and teachers to study facts in detail with lesser risks and with the quicker fabrication of prototype before starting commercial production. It contributes to the innovative application in education and training for students to develop concepts. For medical students, it is the best tool by which they can plan and perform mock surgery before the actual surgery. Complex medical cases are easily understandable by manufacturing a patient-specific replica.

5.2. Industry

It plays a vital role to enhance the design and development ability of a conceived product in different interdisciplinary fields. It can quickly help identify the mistakes in assembly. In industries, it provides better ideas for product design, development, and manufacturing. This technology speeds up the first stages of product development with quicker production of prototypes. It helps creative ideation for the manufacturing of challenging to make a product. This technology provides complex geometries/lightweight parts at a lower cost.

5.3. Research and development

This technology shortens research & development (R&D) time for new products/applications. The design and development of any product can be done effectively as per the requirements such as ergonomics, aesthetics and aerodynamics. It helps to do quicker modification in R&D of industrial products. In the medical industry,

it has a significant contribution because every patient data is not the same. Thus, it is used for the development of implants, tool, instruments and various other devices as per the requirements of the patient.

6. Steps used in 3D printing for bone tissue engineering

Bone tissue engineering with the help of additive manufacturing provides a promising solution for the treatment of orthopaedics patients. It provides scaffolds as per the required shape and size and uses a process similar to natural bone regeneration. Fig. 1 provides different steps as required for bone tissue engineering in conjunction with additive manufacturing.

In bone tissue engineering, the first step is to capture the data of defective bone of individual patient using CT/MRI. Different software like 3D slicer, 3D doctors, magics, mimics etc. are used to convert this data in 3D CAD form and create a precise design of the scaffold. During manufacturing of scaffold, it requires the exact shape, size, mechanical and biological properties to improve the reliability of the patient outcome after the surgery. Then digital CAD data is exported in 3D printing technology in STL format and is printed with the required material. After printing of the scaffold, cells and tissues are planted on the scaffold.⁷ Finally, it is implanted in the body of the patient through a successful surgery.

7. Material properties required 3D printed scaffold

Scaffolds are produced by using varieties of biomaterials which are helpful to regenerate the organs and tissue in the body.^{27,28}

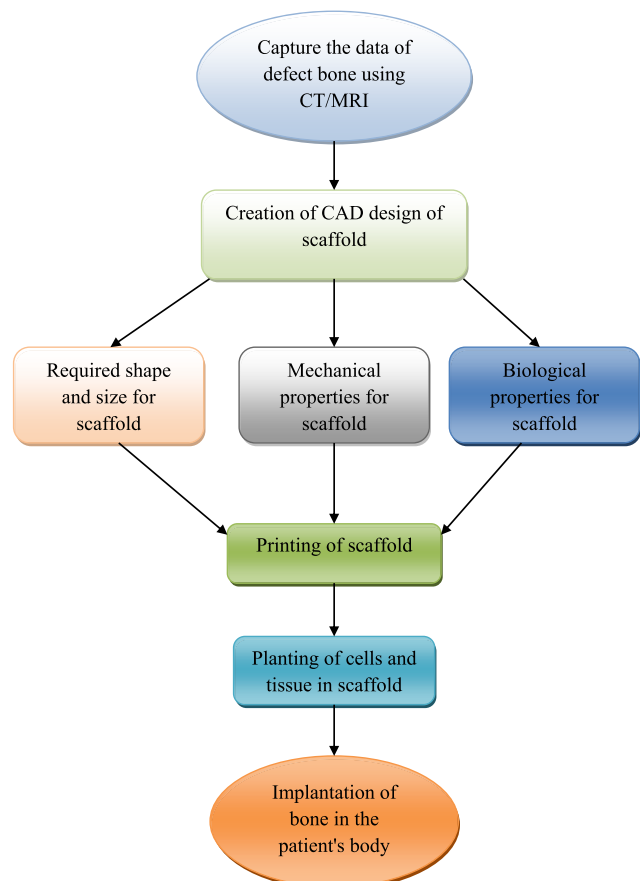


Fig. 1. Steps used by Additive Manufacturing for bone tissue engineering.

There different properties of material required for 3D printed scaffold used for bone tissue engineering are:

7.1. Mechanical properties

Scaffold required specific mechanical properties to be implanted successfully during a practical perspective. It must be strong so that it can be handled easily during the surgical situation/during implantation. It has to be appropriate for bone or cartilage.

7.2. Chemical properties

It consists of bioceramics, polymers and hybrid materials which supports the growth of cells in scaffolds. These provide the ability to create complex shape and improve attachment of cells. Scaffold with excellent chemical properties overcome different complications in bone tissue engineering.

7.3. Biological properties

Scaffold requires biological and nontoxicity properties so that cells can grow and reproduce a new matrix. It produces perfect and complete regeneration of bone tissue. The perfect biological properties significantly create an interaction of scaffolds with organs and tissue. It promotes proper cellular interaction, differentiation/migration and incorporates cells in scaffolds.

7.4. Biodegradability

It should be biodegradable, which helps to construct tissue from their body cells. A scaffold is not a permanent implant as it allows to produce own extracellular matrix from cells. It plays a prominent role in clinical practice and research areas.

7.5. Biocompatible material

A biocompatible material is required to create scaffolds helps cells to perform routine functions. These materials reduce reaction in the body and increase efficiency after implantation.

7.6. Precise shape and size of scaffold

3D printing is applicable to manufacture scaffolds for bone

tissue engineering as per required shape and size. CT/MRI is used to capture the data of the patient. This technique is suitable for manufacturing of patient-specific scaffolds. It is to be cost-effective than a customised scaffold which is used generally in bone tissue engineering.

7.7. Porous structure

Scaffolds should be highly porous and should have an interconnected pore structure to ensure cellular penetration. It creates an extracellular matrix from the cells and exit from the body without any interference of surrounding tissue and organs.

8. Advancement of 3D printing for bone tissue engineering

Accurate designing of scaffold is essential for proper implantation in the patient body. It has to support the vascular growth to meet the requirement of bone regeneration mechanical requirement. There are highly dynamic and diverse tissues in bone with both structurally and functionality. Table 1 discusses the significant advancement of 3D printing for bone tissue engineering.

3D printing creates various advancements by the printing of smart biomaterials which can perform desired functions. There are different Hydrogels used to create innovations in bone tissue engineering filed. Bioceramic scaffolds are made with different biocompatible materials. This technology creates advancements by printing living tissues which have broader applications in the medical field.

9. Research status on 3D printing in bone tissue engineering

Research on 3D printing in bone tissue engineering is rapidly increasing. By searching keywords as “3D printing” bone tissue engineering”, a total of 257 research articles are identified up to September 2019. In the year 2004, the first article on this specific area was published and this year, only one research article was published. In 2005, three articles and in 2006, again no article was published. One research article published in year 2007 and 2008. In 2009, two, in 2010, one and in 2011 no research article published. In 2012 (1), 2013 (7), 2014 (13), 2015 (29), 2016 (21), 2017 (54), 2018 (60), and in year 2019, total 64 research articles are published so far up to September.

There are different journals and sources publishing research

Table 1
Advancement of 3D printing for bone tissue engineering.

SNo	Advancement	Description	Reference/s
1	Biomaterials	<ul style="list-style-type: none"> This technology provides an ability to print smart biomaterial to form a bone These materials have specific biological, physical and chemical properties and have appropriate functionality Having the capability to build hybrid biomaterials to cater to required functionalities Create a new class of bone scaffolding applications using different composite 	Butscher et al., 2011 ²⁹ ; Javaid and Haleem, 2018 ³⁰
2	Hydrogels	<ul style="list-style-type: none"> Due to its unique characteristics, it is used for tissue engineering to promote tissue regeneration Having capability for self-assembling to form scaffolds Represent a new class of bone tissue engineering applications 	Bose et al., 2013 ³¹ ; Liu et al., 2017 ³²
3	Bioceramic scaffolds	<ul style="list-style-type: none"> AM aims for personalized treatment of bone tissue engineering with the precise, fast and controllable fabrication process It provides a different composition for bioceramic scaffolds to enhance the biological properties The applications of 3D printing are also helpful for tumour therapy and other specific diseases 	Castilho et al., 2015 ³³ ; Ma et al., 2018 ³⁴
4	Calcium Phosphate Ceramics	<ul style="list-style-type: none"> Calcium Phosphate Ceramics used as input materials for bone regeneration by 3D printing The performance and reliability of the final product gets improved by using this material It creates a proper combination of growth, cell populations and other osteoinductive elements for bone regeneration 	Trombetta et al., 2016 ³⁵ ; Jammalamadaka and Tappa, 2018 ³⁶
5	Living tissue	<ul style="list-style-type: none"> 3D printing is used to construct vascularised tissue for organ engineering with a wide variety of cells It is the best approach for survival and maturation of cells with desired growth This technology creates a new version of organs to create development in the bone tissue engineering field 	Holmes, 2016 ³⁷ ; Egan, 2019 ³⁸ ; Tamay et al., 2019 ³⁹

articles on 3D printing in bone tissue engineering. Out of these, we have reported the top five journals. These are: “Materials Science and Engineering C” which has published a maximum number of research articles (14). Whereas, “ActaBiomaterialia” and “Materials” each published nine research articles. Journal “ACS Applied Materials and Interfaces” and “Biofabrication” again published an equal number of publications (8). Other sources and journals also have provided an excellent contribution to the research on 3D printing in bone tissue engineering.

In area wise contribution to this valuable area, “Materials Science” field has a maximum contribution of 27%. In contrast, other fields provide engineering (25%), Biochemistry, Genetics and Molecular Biology (13%), Chemical Engineering (10%), Medicine (6%), Physics and Astronomy (6%), Chemistry (5%) and 8% of contribution, these include Computer Science, Pharmacology, Toxicology and Pharmaceuticals, Dentistry, Mathematics, Agricultural and Biological Sciences, Environmental Science, Immunology and Microbiology, Energy, Health Professions, Multidisciplinary, Neuroscience, Social Sciences and Veterinary fields.

We have analysed from the Scopus databases that the applications of 3D printing are increasing in bone tissue engineering. Researchers from the different fields are providing a significant contribution to this innovative field. In the upcoming days, more research will be carried out in this field to create advancement.

10. 3D printing applications in bone tissue engineering

3D printing technology gains attraction in the field of bone tissue engineering due to its extensive capability of higher precision, ease of use and cost-effectiveness for customised requirements. The results show that this technology allows the reproducing of the required structure. Table 2 discusses different applications of 3D printing in bone tissue engineering.

3D printing provides extensive applications towards repairing of bone defects and is helpful to perform precise surgery. It is helpful in the planting of cells which are used to perform required biological functions.^{63,64} This technology can respond to different types of input materials for enhancing the mechanical properties of the scaffold. It is used for the various innovative applications aimed at better treatment of the patient.

11. miRNA therapy integration with 3D printing technology

In recent times, there has been a surge in integrating miRNA therapy with 3D printing technology. miRNAs are small non-coding RNAs that can regulate gene expression at the post-transcriptional level by binding to complementary sites on target messenger RNAs. miRNA cannot easily cross the cell membrane; hence, vectors are required to protect and deliver miRNAs into cells. Recently, three-dimensional printed hybrid scaffolds were engineered with ex-situ miR-148b expressing delivery system for osteogenic induction of ‘rat bone marrow stem cells; (rBMSCs) in vitro, and also in vivo in critical-sized rat calvarial defects which improved calvarial bone regeneration in rats. Scaffolds are designed as highly porous structures and then loaded into collagen gel that was subsequently used to infill a 3D printed polymeric frame which enabled the loading of a high number of cells laden in collagen compared to conventional 3D printed scaffolds, where cells can be only loaded on the surface of filaments. This collagen-infilled 3D printed hybrid scaffolds, expressing significant levels of osteogenic markers, improved bone regeneration.^{65–67} In another study, a versatile material poly-lactide was used as a 3D printing filament labelled with fluorescent dye rhodamine-conjugated and functionalized with poly-amidoamine dendrimers to create a multi-functional scaffold for cell proliferation and transfection with

miRNAs.⁶⁸ These studies could pave a way of controlled delivery of genes in bone engineering.

12. Discussion

The human body is affected due to damaging of parts/organs. To fulfil this innovative requirement, the replacement of these parts/organs is essential to minimize the risk and to satisfy biomedical needs. Tissue engineering is used to regenerate and replace tissue/organs damaged by injury, disease or any other congenital anomalies. These damaged tissue/organs are repaired with scaffolds. AM is used to manufacture scaffold having biocompatible properties as tissue they regenerate. It allows manufacturing of precise and cheap 3D scaffolds in lesser time without any requirement of tools and fixture. 3D printing creates surgical guides which can provide improved efficiency than that of the existing one. Medical model printed by this technology is useful for training, surgery planning, and diagnosis. It is a suitable technique used in many industries to save cost and production time. In orthopaedics, bone tissue engineering is a developing field used to combine tissue engineering, material science and regenerative medicine to repair bone. This technique is used in clinical practice to restore significant bone defects. This bone defect regeneration is a significant challenge in the orthopaedics, which is now being taken up through AM technologies.

13. Limitations and future scope

There are various issues in 3D printing technologies such as material changing option, the strength of the part, post-processing cost, skilled human resources and layer thickness changing option. It is only beneficial for the manufacturing of a customised product. So, only limited products are being manufactured by this technology. Post-processing is also an essential requirement, which involves extra cost.

In future, 3D printing will become a versatile tool for bone tissue engineering for the customised patient match. It provides an opportunity to print with varied materials which are still a big challenge. By changing input material, a qualitative change in the scaffold can be evaluated for different cells/tissues. In upcoming years, its demand will increase to make custom medical devices to accomplish the goal. This technology will create technological change by printing multiple objects with multiple materials, including smart medical implants. This technology will come up with some significant innovations in future, the printing & testing of mixed and new materials.

14. Conclusion

Bone tissue engineering is an innovative approach which is directly used to repair bone defects/tissue for transplantation. In this process, biomaterials play an important role to support regenerative cells and regeneration of tissue. Additive manufacturing is a disruptive innovation, which uses different types of scanning, printing and associated software support. It is used to manufacture complex shapes customised scaffolds from images in lesser time and cost. In bone tissue engineering, it is successfully used to repair and regeneration of bone. It provides essential knowledge of bone biology and its development. In the medical field, this technology is introduced to manufacture customised patient-specific parts. In orthopaedics, it is developed in many ways for the manufacturing of customised human hard tissue in a cost-effective manner. It is also suitable for the manufacturing of living tissue, stem cells, hearing aids, craniofacial disfigurement and dental delivery devices. 3D printing is concerned with the

Table 2
3D Printing applications in bone tissue engineering.

S No	Applications	Description	References
1	Repair of bone defects	<ul style="list-style-type: none"> • 3D printing is applicable to repair the bone defects with the formation of new tissues • This technology is used to fulfil different problems faced during complex surgery by creating an accurate replica of the bone • Used for the manufacturing of patient-specific implants, prosthetics, cutting guides, fixation tools and devices 	Bendtsen et al., 2017 ⁴⁰ ; Qasim et al., 2019 ⁴¹
2	Cell planting	<ul style="list-style-type: none"> • For clinical applications, cell plantation becomes an essential part of creating new bone • Provides good biodegradability, biocompatibility and requisite mechanical strength • AM helps to fulfil the requirement of the bone scaffold by printing them from CT imaging data • Seems to have application in stem cell research, cancer cell biology and other analysis based on the cell planting 	Kim et al., 2013 ⁴² ; Xu et al., 2015 ⁴³
3	Biological functions	<ul style="list-style-type: none"> • Tissue engineering accepts the design determinants of the biological structures for making proper biological functions • Bone scaffolds are made as per bionic performance design and bionic structure design • 3D printed customised bone increase the comfort, efficiency and reliability of the patient outcome after performing the precise surgery 	Park et al., 2012 ⁴⁴ ; Demirtaş et al., 2017 ⁴⁵
4	Restoration of the functions of tissue	<ul style="list-style-type: none"> • This technology is used to repair the bone defect and restoring the functions of tissue • It is suitable to fulfil the need for tissue engineering with accuracy and speed • Meets the mechanical properties which successfully perform intended function • Increase properties after implanted in the patient body 	Wang et al., 2016 ⁴⁶ ; Yu et al., 2016 ⁴⁷ ; Byambaa, 2017 ⁴⁸
5	Functioning scaffolds	<ul style="list-style-type: none"> • Tissue regeneration is an interdisciplinary approach towards creating functioning scaffolds which support cellular activity • A most critical challenge is the development of bone tissue with required growth and surface properties • 3D printing provides a fundamental change in bone tissue engineering by employing layer by layer technique which overcomes the problems of the traditional method for the making of scaffolds 	Do et al., 2015 ⁴⁹ ; Ma et al., 2019 ⁵⁰
6	A functional gradient of cells	<ul style="list-style-type: none"> • This technology achieves fully functioning 3D printed tissue with a functional gradient of cells • This technological advancement allows creating robust designing of scaffolds with new materials • Applicable in medical research such as 3D liver models, vascularised constructs, cardiac tissue and other clinical applications 	Xia et al., 2013 ⁵¹ ; Dong et al., 2017 ⁵² ; Tao et al., 2019 ⁵³
7	Bone graft	<ul style="list-style-type: none"> • For bone repair and generation, a bone graft is utilized as a better approach, using data captured with the help of CT and MRI scan • Bone graft processes become convenient if AM technologies are used accurately 	Kim et al., 2013 ⁴² ; Nandi et al., 2018 ⁵⁴
8	Scaffold with desired mechanical properties	<ul style="list-style-type: none"> • Mechanical strength is a significant challenge in scaffolds which is fulfilled by 3D printing with new innovative materials • By using post-processing approaches in 3D printed scaffold, mechanical strength can further be improved as per the requirements • By decreasing layer thickness, the strength of the part gets increased 	Duan et al., 2010 ⁵⁵ ; Barak et al., 2018 ⁵⁶
9	Biocompatible material printing	<ul style="list-style-type: none"> • 3D printing is a promising technology in which a wide variety of bimetallic materials, biopolymers, bioceramics and biocomposites are used for bone tissue engineering • These materials are suitable for cell adhesion, growth and are helpful in the regeneration of bone tissues • The biodegradable polymer composite scaffolds can satisfy the requirements of bone tissue engineering • Polymer like PLA is used as a biocompatible material which is used as copolymers with other polymers like poly-ε-caprolactone and polyglycolic acid for bone regeneration 	Luo et al., 2013 ⁵⁷ ; Haleem and Javaid, 2019 ⁵⁸ ; Turnbull et al., 2017 ⁵⁹
10	Treatment	<ul style="list-style-type: none"> • Scaffold materials and autologous cells are used in bone tissue engineering for better treatment of bone defects • Successfully implantation in the patient body after loading of bone cells improves treatment • 3D printing efficiently undertakes the plantation of living cells and tissues 	Venkatesan et al., 2015 ⁶⁰ ; Javaid and Haleem, 2019 ⁶¹ ; Mastrogiacomo et al., 2019 ⁶²

creation of substitute implantable bone for critical application. During orthopaedics and craniofacial surgery, these are the most common defects as bone loss due to infection, trauma and tumour resection. In future, 3D printing will create disruptive innovation for the bone tissue engineering in which cells and bioactive molecules will be directly seeded into 3D biomaterials scaffolds.

Declaration of competing interest

None.

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