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REVIEW ARTICLE

Natural biopolymer for 3D printing

Shona Sara Shaji†, Kaladhar Kamalasanan*,†, Sapa Harika, Ankit Tiwari, Bhavana Raj

Department of Pharmaceutics, Amrita School of Pharmacy, Amrita Institute of Medical Sciences and Research Centre, AIMS Health Sciences Campus, Amrita Vishwa Vidyapeetham, Kochi 682041, Kerala, India

*** Corresponding author:** Kaladhar K, kaladhark@pharmacy.aims.amrita.edu

† Equal contributor

ABSTRACT

Three-dimensional (3D) bioprinting is a promising technological approach for various applications in the biomedical field. Natural polymers, which comprise the majority of 3D printable "bioinks", have played a crucial role in various 3D bioprinting technologies during the layered 3D manufacturing processes in the last decade. However, the polymers must be customized for printing and effector function needs in cancer, dental care, oral medicine and biosensors, cardiovascular disease, and muscle restoration. This review provides an overview of 3D bio-printed natural polymers—commonly employed in various medical fields—and their recent development.

Keywords: 3D bioprinting; natural polymers; bio-ink; tissue engineering; regenerative medicine

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1. Introduction

Natural biopolymers have many applications in various biomedical fields, but complex fabrication techniques need to be adopted to create complex systems like biological systems. So, one of the latest advances in technology, 3D bioprinting, is widely utilized in tissue engineering, regenerative medicine, cancer, and dental care to create scaffolds, drug delivery systems[1], and complex tissue structures that closely resemble natural organs and tissues[2]. This technology uses various biomaterials, including metals, ceramics, polymers, and live cells[3], to create complicated designs with superior mechanical properties that are impossible using traditional production methods[4–6]. Proteins, polysaccharides, and aliphatic polyesters are bio-based polymeric materials created through microbial, plant, or animal synthesis[7]. These biopolymers can be either biodegradable (like starch) or non-biodegradable (like polyethylene), which makes them distinct from other biopolymers[8]. In particular, to create biodegradable and biocompatible scaffolds for the biomedical industry, Additive Manufacturing (AM) has extensively utilized polycaprolactone (PCL) and polylactide (PLA) polymers[9]. Compared to conventional materials, the applications of these sustainable biomaterials based on biopolymers have risen considerably in the previous ten years[10]. Upon the implantation of biodegradable materials, the degradation can be carefully controlled to generate harmless components; the functional characteristics of tissues or organs created by additive manufacturing have been influenced by the biocompatibility of bioactive materials to

retain adhesion, strength, and interaction; and native cells must adhere to the biomaterials for implantation.

To produce planned 3D geometries with the appropriate bio-ink qualities, each bioprinting technology has its ideal requirements, including the required resolution and a high ratio embedded system of viable cells[11] (**Table 1**). An ideal bioink should have the desired physicochemical properties, such as proper mechanical, rheological, chemical, and biological characteristics[12]. These characteristics should result in (i) the creation of tissue constructs having sufficient mechanical strength and robustness while maintaining the mechanics that match the tissue, ideally in a tuneable way; (ii) resembling the natural microenvironment of the tissues through biocompatibility and, if necessary, biodegradability: (iii) ability to be chemically altered to fulfill tissue-specific requirements; and (iv) high shape fidelity structures can be printed with tunable gelation and stabilization[13]. Additionally, biomaterials require adherence of the native cells to maintain adhesion, viability[14], and interaction[15]. Some of these polymers can be chemically altered to increase their biocompatibility, biodegradability, and non-toxicity[16]. Natural biopolymers commonly used are chitosan[17], alginate, collagen, silk fibroin, and hyaluronic acid[18].

3D printing offers complex and accurate printing possibilities to develop complex systems like natural systems such as skin, dental constructs, the heart, lungs, and liver (**Figure 1**). As well as to engineer the whole or part of the organ that the reconstructive or restorative function can create. However, the polymers must be tailored for printing[19] and effector function requirements. This review generally analyses six important polymers concerning bioprinting requirements for tissue reconstruction, such as skin, cornea, heart patches, cancer, and dental care.

Herein, we highlight some natural polymers and their recent advancement in 3D printing (**Table 1, Figure 1**).

Table 1. Natural polymers with current applications in 3D printing.

Sl. no	Type of natural polymer	Current application	3D printing technique	Advantages	Reference
1.	Collagen	Cosmetic, medical cosmetology, photographic, biomedical, food, leather	Extrusion, inkjet printing	Excellent biocompatibility, controllable printability, and cell loading properly	[20]
2.	Chitosan	Bone regeneration, cartilage regeneration	Extrusion, stereolithography	Biodegradability, biocompatibility, low cost, and non-immunogenicity	[21]
3.	Alginate	Wound healing, tissue engineering	Extrusion	Excellent printability and biocompatibility, relatively low cost, low toxicity, as well as rapid gelation	[22]
4.	Gelatin	Scaffolds, biomedical, cosmetic	Extrusion, stereolithography	Biocompatible and easily available	[22]
5.	Hyaluronic acid	Biomedicine, tissue regeneration, cosmetics, nutricosmetic	Extrusion	High degree compatibility, good water absorption, easy to attain any shape and size	[23]
6.	Cellulose	Construction, pulp and paper making, textile	DIW, inkjet printing, FDM	Cost-effectiveness and desirability	[24]

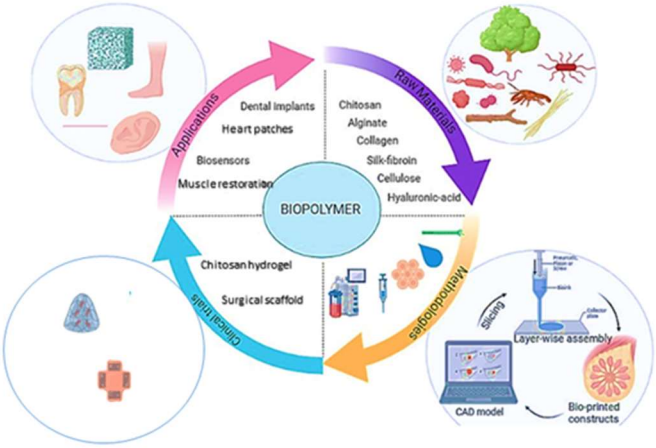

Figure 1. Natural biopolymer with recent applications in various fields like cancer, tissue engineering, oral medicine, biosensors, cardiovascular disease, and dental implants using 3D printing methodologies with potential for clinical translation.

2. Classification

Chitosan: The polysaccharide substance chitosan is produced when chitin is deacetylated, found in the skeleton of crustaceans, is widely used in biomedical applications[25,26]. Bergonzi et al. demonstrated 3D-printed chitosan/alginate hydrogels for the controlled release[27] of silver sulfadiazine in wound healing applications[28], both these polymers are FDA-approved for wound healing applications. In recent years, chitosan hydrogels have become a potential biomaterial for drug delivery applications and are under clinical trials[29]. For instance, to enhance cellular penetration, Ji et al. demonstrated the fabrication of porous chitosan scaffolds for soft tissue engineering using dense gas CO_2 for skin and cartilage regeneration, where the hydrogels created enabled cellular penetration and proliferation within the 3D system, indicating that these are promising materials for tissue engineering applications[30]. However, it is limited due to mechanical durability, swelling strength, gelation kinetics, contamination, sterility, and biodegradation control, which restrict them from being used in applications for drug delivery[31–35].

Alginate: Alginate, a heteropolysaccharide that is abundantly found in the cell walls of brown seaweed and the capsules of the bacterium *Pseudomonas* sp. and *Azotobacter* sp., can create a gel when divalent cations are added[36,37]. Additionally, various cross-linking techniques have been employed to prepare hydrogels for various uses in the biomedical field[38,39]. Recently, Madadian et al. demonstrated 3D printable albumin alginate foam for wound dressing application[40]. The Alginate (Alg)/Tr-calcium silicate (C3S) bone scaffolds were produced using an extrusion-based 3D printing technique. To enhance their physical and biological qualities, the scaffolds were covered in gelatin methacryloyl (GelMA), which is the superior bioink with biocompatible viscose hydrogel[41].

Collagen: A natural protein known as collagen is present throughout the bodies of all animals in the extracellular matrix (ECM) , particularly humans—the fibrous composition with dimeric peptides in primary receptors (integrins), which is present in collagen scaffolds[42]. For instance, Heo et al. observed that adding mesenchymal stem cells (MSCs) and umbilical vein endothelial cells (UVECs) to collagen hydrogels significantly increased cell survival, osteogenic differentiation, and vascular ingrowth[43]. Additionally, the combination of collagen with other organic biopolymers aids in forming fibrous polymeric scaffolds, which have outstanding strength and durability thanks to their cross-linked structure[44,45]. Moreover, due to their porosity, design, and surface characteristics, collagen sponges are also employed as a material for wound dressing.

Silk fibroin (SF): Silk fibroin is a naturally occurring proteinic polymer with a high strength-to-density ratio derived from silkworms, spiders, and cocoons of Bombyx mori[46]. Due to its excellent biocompatibility, mechanical solid performance, ease of processing, and adequate supply from the established sericulture industry, silkworm has been widely used. It comprises two major proteins—silk fibroin and sericin, which have been discussed extensively in various fields, recently[47]. For tissue engineering, hydrogels made of regenerated silk fibroin (RSF) are the best materials because they can be manufactured in the water phase, have low immunogenicity, and have excellent mechanical properties. For example, Gong et al. developed a two-step method for preparing a 3D printable and robust RSF hydrogel. They found that the weak, chemically cross-linked hydrogel's shear-thinning characteristics make it appropriate for 3D printing[48]. Tumour resection and local radiotherapy are part of standard early-stage breast cancer treatment to achieve long-term remission[49]. Hydrogels made of silk are now used to release doxorubicin and other possible anticancer medications[50]. Clinical trials for silk fibroin have been reported using innovative biomedical devices like SilkVoice®, Derma Silk, EPIFIBROIN, and SERI® Surgical Scaffold[51].

Cellulose: A polysaccharide that is renewable and biodegradable is widely present in natural biological sources, including plants (such as bamboo, wood, bast, and cotton) and microorganisms (such as algae, bacteria, and fungi)[52]. The primary types of cellulose utilized in tissue engineering scaffolds nowadays are nitrocellulose and cellulose derivatives[53], which are also frequently employed as conditioning agents for other natural polymer inks like alginate[54], [Alg], and gelatin[55]. Nevertheless, cellulose is challenging to treat because of its low solubility in organic solvents and melting difficulties due to strong hydrogen bonds[56].

Hyaluronic acid: Hyaluronic acid is a newly discovered and adaptable linear polysaccharide made up of non-sulfated glycosaminoglycan linkages that occur naturally in the body[57]. Recently, a study showed that when hyaluronic acid was added to alginate for islet transplantation, it improved the encapsulated cells' survival rate and minimized the body's immunological inflammatory reaction following the transplant[58]. However, pure hyaluronic acid is not printable and cannot be used in bio-inks for 3D printing[59].

Biopolymers have various applications using 3D printing technology, specifically in cancer, dental care, muscle restoration, cardiovascular diseases, and biosensors **Figure 1**.

Given below are various natural polymers with printing parameters, including printer type, scaffold dimension, time for printing, and temperature (**Table 2**). This is mainly being utilised for reconstructive purposes.

Table 2. Various natural polymers with printing parameters, including printer type, scaffold dimension, time for printing, and temperature.

Natural polymer	Printer type	Scaffold dimension	Time for printing	Temperature for printing	Reference
Collagen	Direct inkjet writing printer	5 × 5 × 1 mm		37 °C	[60]
Chitosan	3D bioprinter (Youni Technology Co., Ltd., organization 2500 X,Shenzhen,China	13 × 6 × 4 mm	4 mm/min	4 °C	[61]
Gelatin	BioBots 3D printer (Allevi Philadelphia, PA USA)	22.20 × 11.20 × 0.80 mm	4 mm/s	4 °C	[62]
Alginate	Extrusion-based 3D printing system	83 ± 14 mm	4 mm/min	25–30 °C	[63,64]
Cellulose	Modified fused deposition modeling (FDM) 3D printer	100–1000 nm	4 mm/min	55–75 °C	[65]
Silk fibroin					

Given the 3D printed materials for reconstructive purposes, the futuristic restorative and regenerative systems are analyzed to concerning specific applications.

3. Applications

Some of the applications that are in the process of clinical translation are reviewed further.

3.1. Cancer

Breast cancer is the most often diagnosed and the primary cause of cancer death in women, accounting for 19.3 million new cases of the disease each year[66,67]. Biopolymers have been utilized to model cancer tissues, notably breast cancer tissues, because of their excellent biocompatibility, bioactivity, and capacity to create hydrogels[68]. Two significant classes being used are; alginate, chitosan, cellulose, and hyaluronic acid, which are examples of polysaccharides, and gelatin, collagen, and silk fibroin are examples of proteins. They are shown in **Figure 2**. Collagen is partially hydrolysed to produce gelatin. Chen et al.[69] reported that compared to 2D cell cultures, the development of MCF-7 cells in a 3D cross-linked collagen structure led to a prolonged cell proliferation time and an upregulation of pro-angiogenic growth factors. Chemical transformations happen in prolonged culture, for example, amide groups are degraded into carboxyl groups, which might produce varying densities of carboxyl groups[70]. Gelatin-based scaffolds have been used to grow breast cancer cells. According to some reports, adding more gelatin causes the cells to proliferate more, resulting in larger spheroids[71]. Spiders and silkworms produce a protein-based fibre known as silk fibroin[72]. The comparatively high strength (0.1 to 1 MPa) and excellent cell adhesion property of silk fibroin make it a promising material to use as a cancer cell tissue model[73]. Additionally, silk fibroin can be employed to enhance and fine-tune the mechanical characteristics of 3D scaffolds. The addition of silk fibroin to a chitosan scaffold enabled the increase of chitosan's compressive modulus to 0.6 MPa and the reduction of chitosan's rate because of the chemical cross-linking between the amino groups in chitosan and the carboxyl groups in silk fibroin[74]. In another work, Liu et al.[75] demonstrated that the alginate in alginate-collagen hydrogels improved the network's porosity, allowing spheroids to migrate into the scaffold and grow there[76]. The second-most prevalent biopolymer on earth is chitin. It is primarily present in the fungi and yeast cell walls and the exoskeletons of insects and crustaceans. Taira et al.[77] recently demonstrated that using electrodeposition-based printing, a chitosan (DA 0.2)/gelatin hydrogel was produced with successful encapsulation and proliferation of human breast cancer cells (MCF-7) inside the hydrogel. The ECM (extracellular matrix) surrounding contains large amounts of hyaluronic acid, promoting tumour growth[78]. The interactions between cells and hyaluronic acid that encourage cell proliferation, differentiation, and migration can be mimicked by the presence of HA in cancer tissue models. This natural polymer and 3D printing is a research tool for faster drug discovery and the development of medical devices.

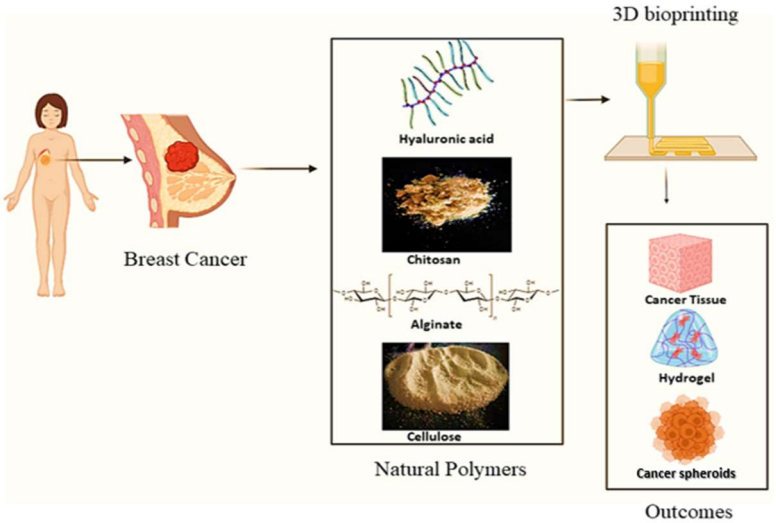

Figure 2. Schematic representation of commonly used natural polymers for breast cancer by 3D bioprinting produces outcomes like cancer tissue model, hydrogel, and cancer spheroids.

3.2. Heart patches

Globally, cardiovascular diseases are the leading cause of death. Myocardial infarction (MI) causes blood flow to become obstructed, which results in oxygen deprivation in the heart muscles and, ultimately, cell death[79,80]. Hydrogels (those made from natural polymers) are a dependable tool for regenerative medicine and have emerged as a promising alternative for heart tissue regeneration due to their hydrophilic nature and structural resemblance to the extracellular matrix[81]. Loureiro et al.[82] demonstrated a three-dimensionally Printed Hydrogel Cardiac Patch for infarct regeneration based on natural polysaccharides where gellan gum and konjac glucomannan acted as functional ink, representing that mechanical, physicochemical, and biological aspects of 3D printed objects are suitable for heart tissue regeneration, as illustrated in **Figure 3**. Cardiomyocytes produced from human pluripotent stem cells (hPSC-CMs) are an essential tool for in vitro modelling of the cardiac microenvironment and have significant potential for tissue engineering applications[83].

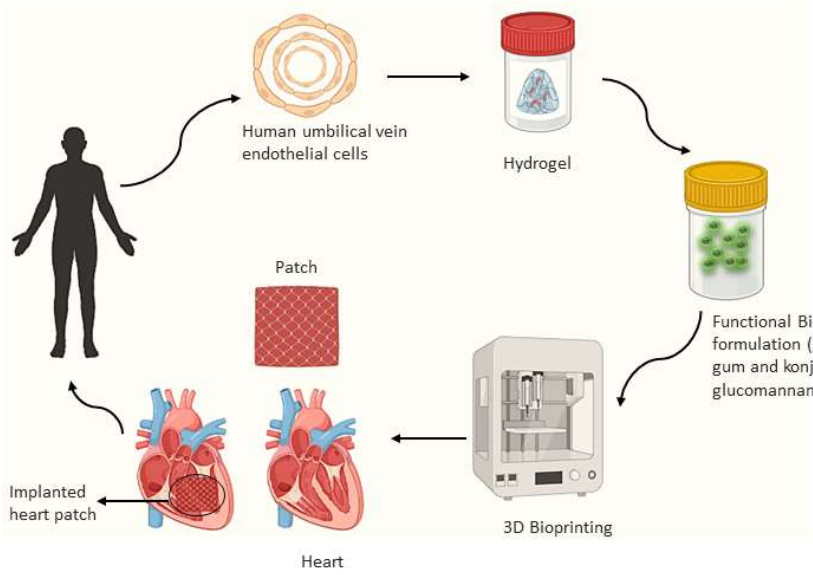

Figure 3. 3D printed hydrogel cardiac patch using the extrusion printing process formulated by functional bioink based on natural polysaccharides gellan gum (GG) and konjac glucomannan (KGM) and human umbilical vein endothelial cells to integrate with cardiac patch.

To increase tissues' mechanical properties, a new method with biocompatible reinforcement of engineered heart tissue is made from differentiated induced pluripotent stem cells (iPSCs) with hydrogel based on the extracellular matrix (ECM)[84]. Hydrogel is crucial for developing self-organized vascular networks, as evidenced by the ability of EVCs to construct 3D networks in artificial matrices. Except for those produced from the hematological system, all normal tissue-derived cells are anchorage-dependent and require surface/cell culture support for optimal growth[85]. To create EVC spheroid-filled cardiac patches, the authors used a 3D bioprinting technique that relies on anchorage-dependent heart tissue[86].

Here, choosing the composition of the bio-inks is a crucial stage in the printing process. As a result, the chosen biomaterials must be printable, have high structural integrity and reproducibility, and simultaneously, resemble the extracellular matrix of the human heart tissue to encourage cell growth and differentiation[87].

3.3. Dental care

Seaweeds are macroalgae that can be classified as being red (Rhodophyta), brown (Phaeophyceae), or green (Chlorophyta) in color based on appearance and can be explored as natural biomaterials over synthetics concerning biodegradability and reusability[88]. The use of natural biomaterials derived from seaweeds in healthcare is growing, particularly for dental applications[89]. Carrageenan and alginates, two biopolymers derived from seaweed, are frequently employed in drug delivery gels, as shown in **Figure 4**[90]. Alginate, a

natural polysaccharide, is commonly used in dentistry to create impressions. The alginate biopolymers may adsorb other molecules and water, which diffuse outward, making them appropriate for creating bio-inks[91]. The development of bioink using natural biopolymers offers promising benefits in this area. Dentin-derived bio-ink research for 3D printing in dentistry has shown encouraging results. It is also helpful for applications involving craniofacial tissue engineering[92].

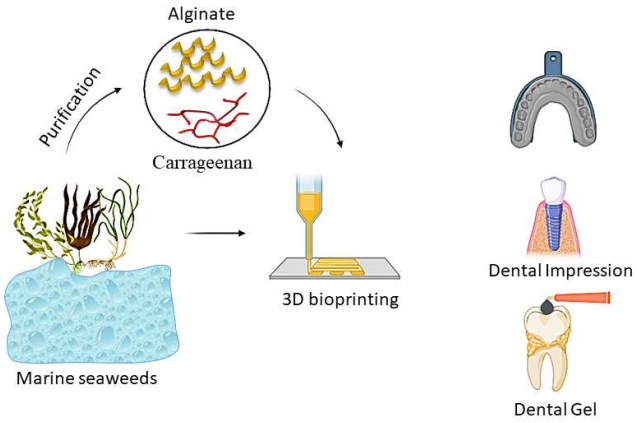

Figure 4. Applications of marine seaweeds after converting into alginate and carrageenan for 3D printing of dental impressions and drug delivery gel.

Some frequent applications in the dentistry industry include the creation of scaffolds and the making of impressions. Also, these polymers are used in food and medicine delivery applications in addition to dentistry uses[93–96]. Regarding root canal treatments, the body's clotting and pulp-evoked bleeding influence how the tooth remodels following the procedure; however, fabrication-inspired techniques, including novel bio-inks, have shown encouraging results. For instance, Athirasala et al.[97] described the creation of a unique bioink called Alg-Dent by combining printable alginate hydrogels with various dentin matrix compositions. Other applications like dental implants and removable dentures require particular care to avoid such microbial colonization due to their susceptibility to bacterial and fungal illness; the combination of the oligomer and triclosan showed promising anti-microbial efficacy against Porphyromonas gingivalis and Streptococcus mutants. This demonstrates that alginate oligomers can be utilized for dental care and, when paired with triclosan, could lower the amount of triclosan in oral care products[98]. However, seaweed-derived biopolymers are extensively used in dental care. The ability to produce nanofibers, notably from seaweed biopolymers, has scope for improvement.

3.4. Oral medicine and Biosensor

3D printing technology can be used to customize oral drugs and sensors[99]. Shi et al.[100] developed 5-fluorouracil oral tablets produced by a drop-on-powder 3D printing process for cancer treatment. Damiati et al.[101] developed a biosensor to detect CD133, a tumor marker in liver cancer cells. In another work, Tripathy et al.[102] demonstrated a biosensor that uses electrochemistry to identify single-point DNA mutations, which can lead to several hereditary diseases, including cancer. The benefits of 3D printing include altering dosage form and shape, which can be used to change release characteristics. For example, Windolf et al.[103] examined how the surface area to volume ratio (SA/V) affects the release profiles of 3D printed dosage forms, made predictions about them, and found that not all drug releases from oral dosage forms need to be investigated in drug release experiments, and their release profiles can be fine-tuned. These natural polymers need to be identified with the application of various parameters for the printing process (**Table 2**).

3.5. Muscle restoration

The 30%–40% of a person's total body mass of skeletal muscle is responsible for stabilizing and mobility of the skeleton, guarding the entrances and exits to the respiratory, digestive, and urinary systems, producing heat, and defending interior organs[104–106]. Natural polymers come from living things like plants, animals, bacteria, or biological systems[107]. The commonly used natural polymers for muscle restoration are cellulose, keratin, alginate, silk fibroin, starch, and chitosan, as shown in **Figure 5**[108] The fundamental building blocks of these polymers include proteins, polypeptides, and polysaccharides, which, when designed as scaffolds, can imitate their diverse activities in the native ECM (extracellular matrix).

Adult skeletal muscle stem cells, termed satellite cells, multiply, develop, and fuse at the impairment site after it happens to fill in and repair the gap that the lesion has left behind[109,110]. However, volumetric muscle loss (VML) shown in **Figure 5**, which is more severe and the body cannot restore, is brought on by acute traumas, congenital anomalies, tumor ablation, and denervation. This results in muscle weakening[111,112]. Tissue engineering (TE), an alternative therapy approach, employs cells seeded onto biomaterials to produce a viable, functioning substitute tissue[113]. Soft biomaterials called hydrogels have a high water content, are biodegradable, biocompatible, and can release medications[114–116]. Natural hydrogels are preferred because they cause a minimal inflammatory response and are frequently parts of the extracellular matrix (ECM), which has been demonstrated to promote skeletal muscle regeneration[117].

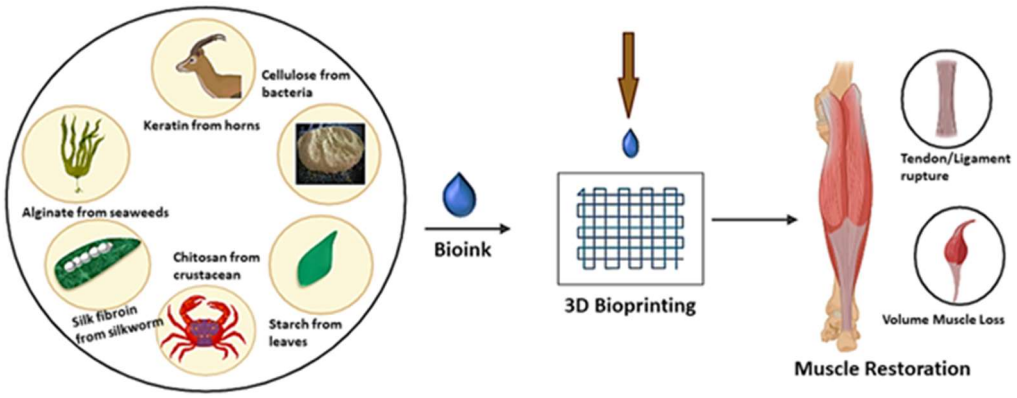

Figure 5. Natural polymers like keratin from horns, alginate from seaweeds, silk fibroin, chitosan from crustaceans, starch from leaves, and cellulose from bacteria 3D printed for muscle restoration applications like tendon/ligament rupture and volume muscle loss.

3.6. Bone

An interdisciplinary method called "bone tissue engineering" uses biomaterials in combination with cells to help tissues regain their necessary activities[118]. Commonly used materials for bone tissue engineering are ceramics, polymers, resins, silica, metal, polyether ether ketone (PEEK), graphene, and other materials. Bone tissue engineering can benefit from disruptive innovation brought about by inkjet 3D printing technology. It makes the necessary cell distribution within the scaffold easier. For effective regeneration and portion repair, it also makes use of bioink[119,120]. This bioink comes in viscous fluid and hydrogel forms. 3D constructions with a lot of cells are made with bioink[121]. Three hydrogels that are frequently used in applications involving bone tissue are collagen, gelatin, and alginate[122]. Because of its inherent biocompatibility and biomimetic qualities, alginate is a widely utilised polymer in bioprinting[123]. It has been demonstrated that alginate properly encapsulates cells and creates a safe environment, which increases the vitality of the cells after printing. Alginate is less frequently used in bone bioprinting, nevertheless, due to its comparatively poor mechanical qualities[121]. Gelatin methacrylol, or GelMA, is another typical example. It resembles the extracellular matrix environment and can cross-link when exposed to UV light[124,125]. However, GelMA is not used because it is difficult to print[122]. According to a recent study by Sawyer et al., human mesenchymal

stromal cells (hMSCs) encapsulated in a mixture of collagen, gelatin, and alginate could be optimized for bioprinting[126]. A bioink needs to be highly printable, but it also needs to keep its cells viable both during and after printing[127]. Similar to native bone tissue, a wide variety of cell types are presented for a bioprinted construct. Osteoinduction, which needs mesenchymal stem cells (MSCs) to develop into osteoblasts, usually with the aid of bone morphogenetic proteins (BMPs), is a crucial part of a successful bone graft. Certain materials, such as calcium phosphate ceramics, have the potential to naturally produce osteogenic differentiation[128]. Some of the commonly used bioinks for bone tissue engineering are given below in **Table 3**.

Table 3. Several hydrogels like alginate, gelatin, chitosan, collagen, and hyaluronic acid in bone tissue engineering with advantages and limitations.

Sl. no	Hydrogel	Advantage	Limitation	Reference
1.	Alginate	• Low cost • Simple to assemble 3D structures • optimal biocompatibility • simple gelling • cross linking, this material is appropriate for 3D printing.	• Bioinnate • Short-term, restricted stability • Fast mechanical property loss as a result of in vitro culturing • Restricted capacity for 3D shape	[129–135]
2.	Gelatin	• Quicken the gelling process • Biodegradable • With the ability to reversibly gel thermally	• Inadequate mechanical qualities • High rate of deterioration	[136,137]
3.	Chitosan	• Constituent is similar to native tissue's extracellular matrix • Harmless byproducts • Stimulates the growth of cell adhesion	• Delayed gelation rate • Inadequate mechanical quality • Potentially interfere with the printing of Ph-sensitive molecules and cells	[138–140]
4.	Collagen	• Less immunogenicity • Good biocompatibility • Regulate cell adhesion • Differentiation	• Poor mechanical properties • Loss of viscosity and slow gelation	[141–143]
5.	Hyaluronic acid	• Highly hydrophilic • Anti-microbial properties • Visco-elastic properties	• Poor mechanical strength	[144,145]

3.7. Skin

Millions of people have skin wounds that are non-healing, necessitating more expensive medical care. The use of 3D-printed skin offers a fantastic way to save lives[146]. These technologies extrude the hydrogels and cells that make up the materials for skin printing[147]. It builds the skin structure layer by layer, which can assist in providing burn patients with skin transplants made from their cells[148]. With this method, burn and accident victims can receive skin grafts[149] for making skin structure bioink is the initial stage in the skin 3D bioprinting process[150]. Even though tissue engineering has advanced significantly over the years, only a few bioinks possess tissue-matching properties and the capacity to encourage tissue growth[151]. Acellular dermal matrix (ADM), agarose, alginate, chitosan (CS), and silk fibroin are examples of further natural biomaterials as shown in **Figure 6**. Natural biomaterials include components found in extracellular matrix (ECM), such as collagen, gelatin, fibrin, and hyaluronic acid (HA). While natural materials are biocompatible, their extended gelation times and poor mechanical qualities are drawbacks. Shi et al.[152] reported a novel bioink that uses extrusion bioprinting to 3D bioprint biological skin tissues. This bioink is constructed of gelatin methacrylate (GelMA) and collagen that has been doped with tyrosinase. Their findings showed that bioink can use 3D bioprinting to create stable, living objects[152]. In another work, Ullah et al.[153] demonstrated hydrogel-based bio-ink for skin tissue engineering with high crosslinking and thermal stability. However, for applications

where they come into direct contact with cells, fibrin hydrogels don't have structural stability, and their high viscosity makes them challenging for printing[154].

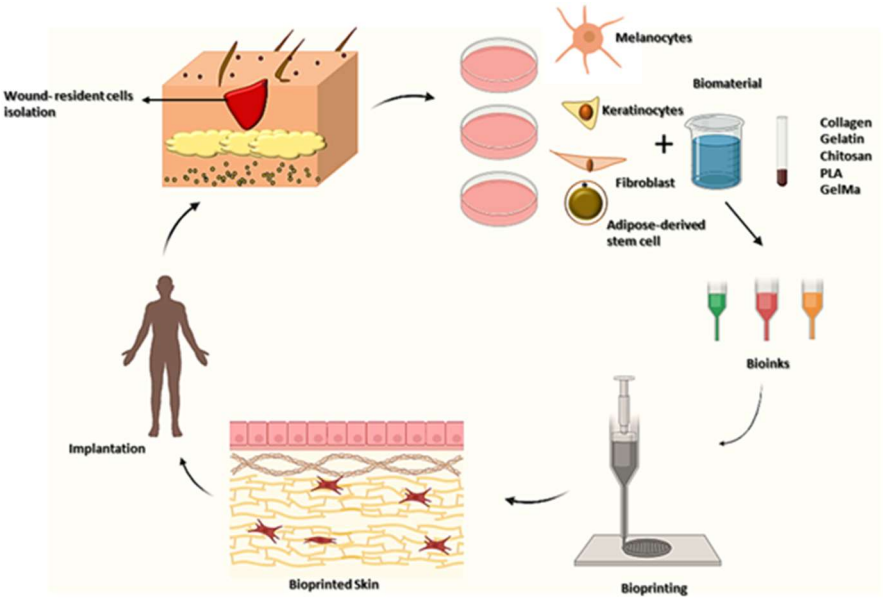

Figure 6. Schematic representation of bioprinted skin for wound healing application using wound cell isolation incorporated with biomaterial and converting to bio-ink for implantation.

3.8. Neural

Neural tissue, comprising the peripheral and central nervous systems, is essential for controlling bodily actions and physiological processes in humans[155]. Neural tissue engineering scaffolds can be created utilizing methods like phase separation and electrospinning, and they are now mostly used for PNS repair[156,157]. By layering living cells and nanomaterials, bioprinting can produce 3D neural tissue constructions for transplantation[158]. Liu et al.[159] created neural tissue constructs using hydrogels loaded with neuron stem cells using micro-extrusion bioprinting to treat spinal cord injuries. Additionally, by layer-by-layer depositing living cells, biomolecules, and biomaterials, 3D printing has progressed to the point that it can now be used to manufacture complex tissues or organs (such as skin, bone, and cartilage). This has given rise to a novel fabrication platform known as 3D bioprinting[160]. For instance, Gao et al.[161] demonstrated a work where, cell-filled spinal scaffolds were 3D bioprinted with a bioink made of hydrogels, poly(3,4-ethylenedioxythiophene): sulfonated lignin (PEDOT: LS), and neural stem cells. This bio-ink may encourage the stem cells in vitro neuronal differentiation and the regeneration of the spinal cord in vivo[161].

In addition to the naturally occurring hydrogels that are commonly employed, neural tissue engineering also makes use of printable natural materials like cellulose[162]. For instance, Kuzmenko et al.[163] used carbon nanotubes and cellulose nanofibril hydrogels to accomplish 3D printing. Compared to other tissues that have been extensively researched, such as skin, bones, heart tissue, and cartilaginous structures, very few studies have concentrated on the application of 3D bioprinting in the synthesis of neural tissue[164]. Fantini et al. developed a novel bioink that aids in the maturation of induced pluripotent stem cells (iPSCs) into neural stem cells (NSCs) and maintains their proliferation[165]. Thus, this constructs the generation of a more complex and realistic neural tissue 3D model for the study of neurodegenerative diseases.

4. Conclusion

The research and development of natural polymers for various applications using 3D printing technology, specifically in cancer, dental care, muscle restoration, cardiovascular diseases, biosensors, bone, neural, and

skin, are rapidly growing. However, the properties of natural biopolymers need to be fine-tuned by suitable structural modifications concerning this application to make it available for 3D printing. For that, in this review, the general properties of polymers for this application are being analyzed. In addition, these polymers need to be converted into bio-inks and fine-tuned for different printing applications. The existing information about developing 3D constructs and exploring bioink has reached clinical translation. Two major classes of biopolymers that can be used in cancer models are polysaccharides (alginate, chitosan, cellulose, and hyaluronic acid) and proteins (gelatin, collagen, and silk fibroin), which are later translated for 3D construct models like cancer tissues, hydrogels, and cancer cells. Meanwhile, cardiac patches represent a promising alternative for heart tissue regeneration, considering their hydrophilic nature and structural similarity to the extracellular matrix. Some frequent applications in the dentistry industry include the creation of scaffolds, making impressions, and drug delivery gels, where naturally derived marine seaweeds are widely used. 3D printing technology can also be used to customize oral medicines and sensors, but specific printing parameters are needed to customize them. Muscle restoration is another application where soft biomaterials like hydrogels are preferred. Nevertheless, future studies still require direction.

Author contributions

Conceptualization, KK; methodology KK and SSS; writing—KK, SSS and XX; writing—review and editing, SSS, SH and AT; visualization, SSS; supervision, KK; All authors have read and agreed to the published version of the manuscript.

Conflict of interest

The authors declare no conflict of interest.

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