Nanosilicate-Polysaccharide Composite Hydrogels for Bone Scaffolds

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The revolutionary technology of 3D printing has emerged, allowing us to create realistic models of bones known as anthropomorphic phantoms. These phantoms mimic the spatial, physical, and biological characteristics of bone tissue, enabling us to study and develop innovative techniques in various scientific disciplines. Nanosilicate-polysaccharide composite hydrogels are a well-studied class of materials in regenerative medicine that combine good 3D printability, staining, and biological properties, making them an excellent candidate material for complex bone scaffolds.

3D printing scaffolds phantoms

1. Introduction

Bones are fascinating structures in the human body, providing support as well as structure, mobility, and protection. Understanding the intricate properties of bone tissue is crucial for advancements in fields such as pathology, histology, and radiology. In recent years, the revolutionary technology of 3D printing has emerged, allowing us to create realistic models of bones known as anthropomorphic phantoms. These phantoms mimic the spatial, physical, and biological characteristics of bone tissue, enabling us to study and develop innovative techniques in various scientific disciplines.

- Each type of phantom requires specific materials that closely simulate the desired properties of bone tissue. Spatial and biomechanical properties are represented by high-fidelity anatomical models [1] or benchmark devices for biomechanical testing [2][3] that require materials with minimal thermal deformation (thermo- or photopolymers), and can be 3D printed with FDM (fused deposition modeling), SLS (selective laser sintering), or SLA (stereolithography) at optimal spatial resolutions.

- Biological and optical properties are represented by bioscaffolds [4] or organ-on-chip devices [5] made of biomaterials such as hydrogel biopolymers with optimal cell culturing characteristics and staining properties that do not impede histological examination and analysis; these can be 3D printed with extrusion-based 3D printing.

- Radiological properties are represented by imaging phantoms [6], which require materials with an atomic mass and X-ray attenuation coefficient similar to that of natural bone (for instance, thermopolymers or polymer-inorganic clay composites); these can be 3D printed with FDM.

2. Nanosilicate-Polysaccharide Composite Hydrogels for Bone Scaffolds

2.1. Composite Polysaccharide–Nanosilicate Hydrogels

Hydrogels are a class of hydrophilic materials composed of polymers and water [7]. These polymers can form insoluble fiber networks mimicking the structure of the extracellular matrix of certain human tissues [8]. Polymerization of monomeric units is achieved by means of a crosslinking mechanism based on thermal, light, or ionic interactions [9]. Organic and inorganic additives (such as metallic or clay nanoparticles [10][11], carbon nanomaterials [12], growth factors [13], pharmacological substances [14], etc.) can substantially modify the initial properties of the pure material [15]. Certain combinations of polymers and additives are especially promising for the cultivation of specific mesenchymal tissues, including bone [16], cartilage [17], ligaments [18], muscles [19], blood vessels [20], and more. Laponite is a nanosilicate clay composed of 10 nm crystals with a discoid shape [21]. When dissolved in water, the two sides of the discoids acquire opposite polarities and spontaneously assemble into a "house of cards" configuration [22]. When added to a polysaccharide hydrogel, Laponite significantly improves both its 3D printability and its osteogenic properties [23]. These properties make nanosilicate composite hydrogels a promising tissue-equivalent material for the generation of complex three-dimensional structures with high spatial, biological, and radiological fidelity.

2.2. 3D Printing of Bioscaffolds

Composite hydrogels can be 3D printed into complex and highly porous three-dimensional bioscaffolds [4]. They are currently being developed from highly biocompatible polymers such as alginate [24], cellulose [25], silk, chitosan [26], hyaluronic acid [27], collagen [28], fibrin [29], etc., and modified with additives [12] to further improve their biological properties. The bioscaffold can be inoculated with cells and cultivated in a bioreactor with cell culture medium to establish a 3D cell culture with specific spatial characteristics [30][31]. The materials of the scaffold, the cell culturing media, the conditions in the bioreactor, and the cell type all determine the fate of the cells, and can be used to simulate a broad spectrum of physiological or pathological conditions in a biological phantom. Bioscaffolds can be used to generate new implantable synthetic tissues [32] and simulate rare pathological conditions [33], as well as in oncological diseases [34] and drug testing [35].

The main goal of bioengineering is the creation of synthetic tissue and organ transplants that can replace damaged organs and tissues [36]. This achievement will eliminate the shortage of organs for transplantation, which is the main issue in regenerative medicine. There are several factors that determine the ideal implantable bioscaffold [37]:

- Biocompatibility: the scaffold must provide the necessary base for adequate cellular adhesion, proliferation, and differentiation [38]. If the scaffold is implantable, it should not cause any inflammatory or immune reaction, which disrupts tissue regeneration and may cause rejection by the recipient.

- Bioresorption: the materials of the scaffold must be bioresorbable and eventually replaced by a newly generated extracellular matrix [39]. The byproducts of biodegradation should be nontoxic and easy to eliminate from the organism without interference with other organs and systems.

- Mechanical properties: the scaffold should possess mechanical properties corresponding to those of the tissue in which it will be implanted [40] and must preserve its integrity from the moment of implantation to the completion of the remodeling process. This condition is especially important for bone and cartilage engineering.

- Scaffold architecture: the scaffold should possess a porous structure specific to the engineered tissue, with interconnected spaces occupying a sufficient part of the total volume [41]. High porosity ensures adequate cell migration, diffusion of nutrients, and elimination of waste products. Adequate vascularization of the scaffold prevents necrosis, inflammation, and rejection of the implant [42]. Another key concern is cell adhesion, as cells bind to chemical groups (ligands) that are naturally present only in extracellular fibrillar glycoproteins. In non-natural materials, active adhesion sites can be engineered by adding binding sequences (such as Arg-Gly-Asp, RGD) or by other means to facilitate cell adhesion [43].

- Radiological properties: as an implantable structure, the bioscaffold should be controlled using imaging methods. This requires tissue-equivalent radiological properties that ensure proper control over scaffold implantation [44].

- Histological properties: staining qualities must ensure that the engineered matrix does not interfere with histological and cytological analysis during scaffold development and testing [45].

- Manufacturing technology: bioscaffold production with 3D printing or other spatially controlled technology requires high reproducibility as well as proper quality control and certification [46].

Composite nanosilicate hydrogels possess properties that fit well with the listed paradigm and that can be manufactured into reproducible complex porous bone-mimicking bioscaffolds through extrusion-based 3D printing. This makes them a promising material for bone tissue engineering.

2.3. 3D Printing of Imaging Phantoms

Medical imaging using CT, MRI, or ultrasound plays a vital role in diagnostics and research. To ensure accurate and high-quality images while maintaining patient safety, medical imaging phantoms have traditionally been employed. Thanks to their known material composition and simple geometries, such phantoms have become essential for quality assurance and standardization. However, traditional phantoms have limitations such as restricted material usage and limited geometric complexity. While more sophisticated alternatives exist, their high cost makes them less accessible.

The rapid advancement of 3D printing technology in recent years has led to a significant breakthrough in the domain of medical imaging phantoms [47][48][49]. The 3D printing of sophisticated geometries using diverse materials can facilitate the development of affordable anthropomorphic phantoms. These phantoms are crafted using tissue-equivalent materials to realistically and accurately depict organs specific to each imaging modality employed. Simple shapes are no longer a limitation, as intricate anatomical structures can be printed with diverse materials.

Anthropomorphic phantoms can be used for standard quality assurance procedures as well as for protocol optimization tasks, image reconstruction algorithm optimization, and testing of new emerging techniques.

One of the most accessible and popular 3D printing technologies is fused filament fabrication (FFF), more commonly known by its trademark name of fused deposition modeling (FDM). The specifics of the process mean that FFF printers can easily be modified and augmented for printing with various materials. Such is the case with bioprinting, in which the typical extrusion system is replaced by a motor-driven syringe [50]. These improvements have led to the next big challenge in modern biophysics and radiology, namely, the development of universal and multimodal anthropomorphic phantoms.

2.4. Development of Complex Multipurpose Biological–Radiological Phantoms

The different phantoms represent only specific properties of the target tissue: either biological, simulated by bioscaffolds and 3D cell cultures, or radiological, simulated by imaging phantoms. Certain advanced phantoms can represent several submodalities of the main property; for example, there are complex X-ray/CT/CBCT/Angio-CT phantoms. Recently, the possibility of a new kind of phantom has emerged: a complex cell-laden bioscaffold with a porous structure that represents the morphological, physiological, histological, and radiological properties of bone tissue. This type of device can simulate a wide range of physiological and pathological conditions, including osseous callus formation, bone remodeling, osteoporosis, osteosclerosis, bone cysts, osteodegenerative conditions, primary or metastatic bone tumors, etc. Their high radiological fidelity could make possible the development of specific imaging algorithms for the detection and differential radiological diagnosis of these conditions.

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