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Review

Shape memory polymers in osteochondral tissue engineering

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Abstract

Bone and cartilage injuries are significantly increasing with population aging. Tissue engineering is considered an alternative and promising approach for alleviating osteochondral tissue injuries along with available therapeutic modalities. 3D- and 4D-printing fabrication protocols have been used to facilitate the production of bone/cartilage scaffolds that are similar to bone and cartilage microenvironments. In this regard, advanced biomaterials, including smart polymers and stimuli-responsive polymers are the first essential elements for improved bone/cartilage regeneration. Shape-memory polymers, are stimuli-responsive materials and are available in permanent and temporary structures. The application of shape-memory scaffolds can lead to providing in vivo-like conditions and improve cell bioactivity and phenotype acquisition. In this review article, we tried to highlight stimuli-responsive polymers and their application in osteochondral tissue engineering.

Introduction

According to data, the number of patients with different osteochondral pathologies is increasing because of population aging and the current treatments are not satisfactorily sufficient to restore tissue function. Tissue engineering is a promising modality for alleviating bone and cartilage injuries and defects. Progressive methods related to biomaterials synthesis and the application of cutting-edge fabrication technologies have led to an improved engineered scaffold synthesis.[1] Shape memory materials (SMMs) refer to substrates that can recover their original shape from a remarkable (sometimes maybe plastic) deformation following a specific stimulation. They can alter between a permanent and a temporary form during applying and removing external stimuli indefinitely.[2] SMMs are classified into three groups: shape memory alloys (SMAs), shape memory polymers (SMPs), and shape-memory hybrids (SMHs).[3,4] Among these, SMPs are used widely due to their low cost and great deformation capacity.[5,6] Several external stimulations such as stress, light, heating (above the glass transition or melting temperature), contact with certain chemicals, and electric or magnetic field can be used to dictate permanent shape in SMPs instead of temporary and dormant shapes (Figure 1).[7-9] Several classes of SMPs are commonly available based on different stimulation to which they respond.[2,3] In thermo-responsive SMPs, temperature changes are critical factors that correlate with a glass transition temperature (Tg).[3,10] Studies have indicated that the immersion of solvent-sensitive SMPs in an appropriate solvent such as water can decrease the T_g values and acts as a plasticizer on the polymer chains.[11] Along with these, light is another stimulating object for SMPs that can easily control spatial and precise applications.[12,13] In general, UV light (using photo-cross-linking/isomerization effect) or near-infrared (NIR) light are used to generate photo-responsive SMPs because of the photothermal effect.[14] Noteworthy, only NIR light is appropriate for *in vivo* uses because of its high tissue infiltration capacity.[15] The basis of this technology is based on dropping several photothermal nanofillers such as carbon nanotubes, gold nanorods, and graphene into thermo-responsive

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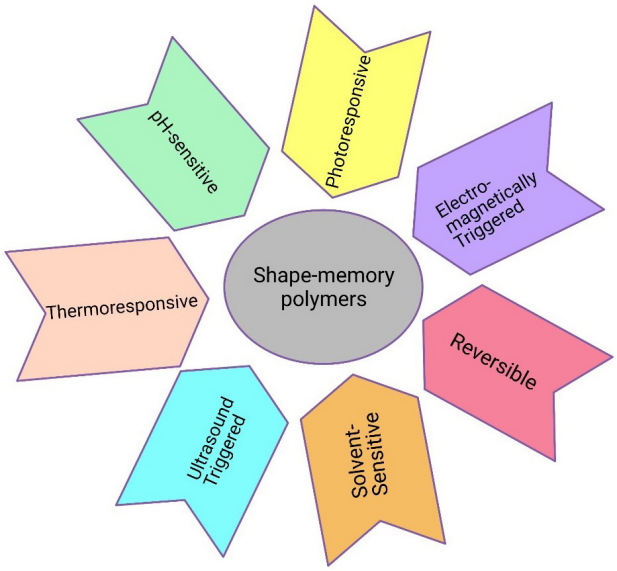

Figure 1. Various types of shape-memory polymers regarding several stimulus methods

SMP substrates. This method results in the production of NIR-photo-responsive SMP composites which absorb NIR light and transform the optical energy into heat.[16-18] Nevertheless, the challenging issue is the nanofillers poor biodegradability rate.[19,20] An electromagnetic field is another stimulating factor for SMPs, which is based on the induction of magnetic nanoparticles loaded in thermo-responsive shape-memory polymer composite matrixes.[15,21,22] Along with these modalities, ultrasound-responsive SMPs have also been investigated to be applied as indirect thermos-induction. For this purpose, ultrasound is used to transfer heat and elicit the shape-memory effects.[23,24] This technique enables us to remotely control the geometric features of shape-memory, which makes this method attractive for biomedical applications. The modulation of pH is another approach for the regulation of shape-memory in SMPs.[25] More recently, researchers have focused on the application of reversible shape-memory effects in different materials. Such an effect may be relevant to the switch of two morphologies without using other modalities.[26,27] Direct evidence has shown that shape-memory behavior is closely based on entropic elasticity.[2] The most entropically stable state of a formless polymer chain can lead to the coiled configuration, so the release of the force will result in returning to a more entropically favorable, coiled form (original form). The polymer has a "memory" of its original shape, and slippage of the polymer chains ("forgetting" the original state) occurs during the long-time implementation of the force (hours instead of seconds).[28,29] Physical or chemical cross-linking of the SMPs prevents the polymer slippage and fixes the chains in a permanent state, which is thermodynamically stable.[26,28] The application of scaffolds with shape-memory features and simultaneous 3 and 4D printing approaches has assisted in sophisticated bone grafting. Here, we highlighted the multiple shape-memory modalities available for engineered cartilage and bone tissues. The aim of this study was to investigate the advantages and disadvantages of using SMPs in cartilage and bone restoration according to the previous *in vitro* and *in vivo* studies. Also, 4D printing, as a novel technology attributed to the 3D printing of SMPs, and its applications in cartilage/bone regeneration is introduced.

SMPs in cartilage tissue engineering

Traumatic and destructive injuries to cartilage may lead to permanent disability. Osteoarthritis, cancers, excessive workload, repetitive stress, and inflammatory responses can lead to osteochondral tissue defects. The time-consuming healing procedure of cartilage is related to its dense avascular and aneural structure without a lymphatic system. Considering the avascular nature of cartilage, the distribution of fibrinogen, growth factors, and different cell types are limited to the defect site.[30,31] Regarding the limited self-regenerative capacity of articular cartilage, appropriate replacement systems are required to support normal tissue function physically, mechanically, histologically, and biologically. The cartilage's unique ability to create a load-bearing surface, almost friction-free and painless, depends on the complex interaction between cells and the extra-cellular matrix (ECM).[32] Adults' cartilage tissue is composed of relatively low chondrocyte numbers within the dense ECM. The chondrocytes are encapsulated in a dense matrix and cannot migrate easily to the repair area, so the matrix prevents the migrating cells from entering the synovial fluid.[33] Limitations of existing clinical treatments for osteochondral tissue defects have improved the association of regenerative medicine with novel engineering methods to restore the damaged tissue. Up to now, an extensive range of polymers have been designed in line with cartilage regeneration, but they are not completely efficient. SMPs provide a conducive and smart microstructure for chondrocyte adhesion, proliferation, and ECM simulation to completely and effectively fill the large cartilage defects. Several studies have recently been performed to this end. In an experiment conducted by Zhang et al, they developed pH-driven shape-memory nanocomposite hydrogel films with graphene oxide and chitosan. They induced self-assembly using the water evaporation technique and promoted cross-linking in an alkaline solution. Ultrastructural analysis revealed the existence of layered brick-and-mortar microstructure with significant mechanical features, making them appropriate scaffolds for artificial cartilage regeneration.[34] Combination of the smartness of SMPs with the biocompatibility of biological agents also has been proposed to increase the healing properties of the material. In a non-invasive approach, He et al developed a shape-memory hyaluronic acid-based cryogel grafted with adhesion peptides

(arginylglycylaspartic acid, RGD). They seeded the gels with chondrocytes and injected them to fill the cartilage defect. The gels proved to maintain shape-memory as they contracted and then returned to their original shape following injection.[35] Fifteen-day incubation of cells on these scaffolds exhibited enhanced cell proliferation and production of cartilage ECM glycosaminoglycans compared to hyaluronic acid-based hydrogels. Besides, immunohistochemical staining exhibited the generation of collagen type II and glycosaminoglycans in chondrocytes seeded within the cryogels. It seems that injectable shape-memory cryogels composed of hyaluronic acid possess high-pore interconnectivity, which provides appropriate conditions for dynamic growths of chondrocytes and ECM production. Structural, physio-chemical, and degradation properties of the SMPs can be adjusted and improved efficiency. As another example, Almeida et al designed a porous, shape-memory scaffold consisting of covalently cross-linked alginate with carbodiimide chemistry for cartilage regeneration. While the structure of the construct was adjusted using a directional freezing procedure.[36] Polylactic acid (PLA) can be applied in SMP scaffold synthesis because of certain biodegradability and biocompatibility.[37-39] These kinds of scaffolds also exhibited favorable *in vivo* performance for cartilage regeneration. Uto et al provided a 3D tubular PLA scaffold filled with cartilaginous particles originating from the human-induced pluripotent stem cells of auricular frames composed of a helix-antihelix prepared for auricle regeneration. Thermo-responsive shape-memory PLA mesh scaffolds filled with auricular chondrocyte pellets subcutaneously were transplanted in nude rats. Data showed that regenerative cartilage was observed in all samples eight weeks after transplantation, which maintained their cartilage features. According to the in vivo results, the regenerated cartilage maintained the shape and features of cartilage for one year.[40] Xuan et al designed bio-functionalized thermo-responsive shape-memory scaffolds with suitable elasticity, shape recovery, and permanent state generated by covalent network using polyglycerol sebacate (PGS). Besides, the existence of crystallized poly(1,3-propylene sebacate) (PPS) provides a reciprocal switch between permanent and temporary states. Also, the release of kartogenin (KGN) is proportional to the degradation rate, which can give us information about the chondrogenic capacity over a prolonged period. The results showed that the application of scaffolds composed of PPS-PGS-KGN could lead to non-invasive surgical procedures and improve cartilage defects *in vivo*. Also, the presence of KGN encouraged bone marrow mesenchymal stem cells' orientation toward chondrocytes while inhibiting the osteogenic capacity in a dose-dependent manner.[41] Previously, Jiang et al assessed the chondrogenic effects of 3D collagen and denatured collagen scaffolds with shape-memory properties on New Zealand rabbits' cartilage defects. Based on data, collagen scaffolds transplanted into the defect areas promoted cartilage synthesis and subchondral bone compared to the denatured collagen scaffolds.[42] These data showed that maintaining the triple-helical structure of collagen is an important index for the chondrogenic capacity of the scaffold. Integration of synthetic and natural polymers and also producing physiologically adapted shape-memory behavior are the other advantages that can be provided by SMPs. Innovatively, Chen et al fabricated a scaffold composed of dispersed gelatin/ poly(lactic-co-glycolic acid) (PLGA) fibers, hyaluronic acid, and polyethylene oxide solution to transform gelatin/PLGA electrospun fibers into 3D printing inks. They declared that the synthesized scaffold possessed appropriate elasticity with water-induced shape-memory properties. Besides, the combination of 3D printing ink with chondrocytes showed suitable cartilage regeneration *in vivo*.[43] A summary of studies based on shape-memory constructs for cartilage regeneration is collected in Table 1.

SMPs can be provided with several cartilage ECM mimicking structures, including collagen, alginate, hyaluronic acid, and gelatin, as well as different synthetic polymers. Shape-memory scaffolds are easily loaded with primary chondrocytes and mesenchymal cells, and stay viable while recovering their final permanent form after injection in the defect site. SMPs support cell attachment, proliferation, and differentiation offering a more stimulating and flexible microenvironment for cell adhesion, proliferation, and matrix biosynthesis. The shape-memory characterization of these scaffolds makes them suitable for being injected into the joint space, resulting in a non-invasive restoration of articular cartilage.

SMP in bone tissue engineering

Trauma, congenital defects, or tumor excision cause sized defects in bones, which will not be cured without intervention.[44] To enhance or hasten bone regeneration, grafting is a key clinical technique, but auto-grafts are accompanied by some pitfalls and downsides. For instance, damages and persistent pains could nerve result from autografts, while limited osseointegration, adverse immune response, disease transmission, and risk of infection can occur in allografts.[45,46] As a consequence, it is not completely applicable to use auto-grafts and allografts for bone defects.[47] Tissue engineering has been fascinated by noteworthy attention as an alternative approach to restore the function and structure of injured bone tissue.[48] Some vital features of each scaffold should be considered for efficient transplantation and outcome. To this end, transplant scaffolds should closely fit bone defects and be osteoconductive so that it permits osseointegration, hence allowing bone reconstitution either on the surface or inside the structures (Figure 2). Importantly, the transplant scaffold should provide physical forces and

Table 1. Summary of the selected studies using shape-memory constructs for cartilage regeneration

Chemical composition	Trigger	Fabrication method	Form	Notable characteristics	Ref
Cryogel consisting of hyaluronic acid	-	Cryogel technique	3D products	• Providing an Injectable scaffold for cartilage regeneration • Providing transport for nutrients, byproducts, and growth factors • Providing appropriate substrate for chondrocytes attachment • Offering proper substrate with RGD conjugates for chondrocytes growth	35
Anisotropic Alginate/ Collagen I or II Scaffolds	Mechanically compressed	Freezing technique	Cylindrical hydrogels	• Arranged pores • Increased production of sGAG and collagen	36
PLA mesh scaffolds + auricular chondrocytes	Thermo-responsive	Woven threads	3D auricular structures	• Simulation of the auricular structure	40
PPS/PGS/KGN cell-free scaffolds	Body temperature-responsive	The modified salt fusion process	Ternary scaffold	• Excellent bioactivity resulting in cell-free applications • The existence of widespread free hydroxyl groups leads to easily functionalization	41
Native collagen and denatured collagen scaffolds	Hydration	Casting process	3D scaffold	• Collagen scaffolds provide shape-memory properties • Chondrocyte-seeded scaffolds are more cartilaginous than cell-free scaffolds	42
Gel/PLGA and hyaluronic acid (HA)/ polyethylene oxide (PEO)	Water-inductive shape memory	3D printing and freeze-drying	Rectangular-shaped scaffold	• 3D printing of electrospun fibers with precise outlines and great pores which are elastic in the wet media • 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC)/ N-hydroxysuccinimide (NHS) crosslinking method increased the steadiness of the scaffolds	43

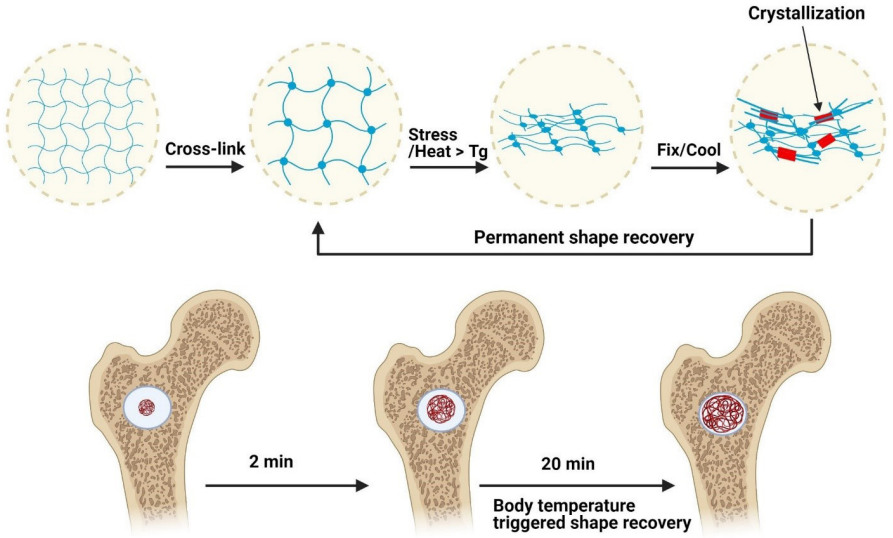

Figure 2. The schemes of covalently cross-linked SMP application in bone tissue engineering and fitting the irregular defects after 2 minutes and 20 minutes. The *in vivo* test can be performed in the water bath at 37ºC. The programming process classically involves polymer deformation above the respective transition temperature, in presence of stimulating forces, to the desired temporary shape

suitable mechanical stimulation to the juxtaposed bone tissue. According to clinical data, mechano-transduction is a critical factor in bone regeneration and can regulate bone osteogenesis and resorption.[49-51] To specifically shape and form a transplant scaffold for filling the bone cavity and appropriately connecting with the surrounding tissue, new intelligent methods are extremely demanded. Traditional synthetic scaffolds have limitations due to their post-fabrication shaping, mold shape, and complexity of computer-aided solid freeform fabrication methods.[48,52] In line with the comments, it seems that moving toward injectable composites such as hydrogels, ceramics, and glass-ceramics would be helpful to shape *in situ* solid scaffolds within the bone cavities and to reduce the invasiveness of the surgical procedure.[53,54] Unfortunately, the molecular identity of these materials can lead to the lack of suitable physicochemical properties, mechanical strength in which some of these materials are brittle and possess inadequate porosity and interconnectivity, reducing osteoblasts migration, bone ingrowth, and angiogenesis.[53,55,56] As a novel approach, SMPs are proposed as a new brand of stimuli-responsive materials that are at the center of attention for clinical applications.[57-59] In load-bearing tissues like hip or joints, the highest strength and minimal wearing of biomaterial are essential requirements for the scaffold design.[60] Therefore, the

physicochemical features of the transplant scaffold and juxtaposed tissues should match.[61,62] As a consequence, for bone implants, Young's modulus values of around 1 and 7 GPa are normally required.[62,63] Both natural and synthetic polymers can be applied for the fabrication of strong SMP scaffolds in bone regeneration. Hu et al assessed a combination of PLA-TMC (trimethylene) and natural polymer chitosan (CS). 3D PLA-TMC and PLA-TMC/CS scaffolds were developed using the solvent/non-solvent method. Results showed that PLA-TMC/CS scaffold exhibited great biocompatibility, appropriate mechanical properties, rapid shaping, and low biodegradability with excellent shape-memory effect. Experiments have shown that PLA-TMC/CS scaffolds can enhance the proliferation of murine osteoblastic lineage MC3T3-E1 cells and alkaline phosphatase (ALP) activity.[64] Other substrates like porous bio-polymers and foam materials have also been used as shape-memory scaffolds for bone defects. In an experiment conducted by Xie et al, a polyurethane (PU)/hydroxyapatite-based SMP foam was produced via the gas-foaming method. Using thermal stimulation, these polymers can be expanded to fill the bone defects. Results indicated PU-hydroxyapatite SMP has excellent biocompatibility and appropriate bone ingrowth. Micro-CT imaging and histological examination revealed rapid bone reconstitution and angiogenesis 12 weeks after surgery.[65] Previously, a thermo-responsive SMP scaffold was fabricated and coated with polydopamine by a revised solvent-casting particulate leaching technique that included photo-crosslinking of polycaprolactone (PCL) diacrylate accompanied by a NaCl salt template.[52] This scaffold exhibited suitable interconnectivity and an appropriate modulus of 4.4 MPa and can properly fill irregular bone defects. Of note, the coating of SMP with polydopamine does not affect the porosity, shape-memory properties, and physicochemical capacity. Besides, surface hydrophilicity was considerably augmented because of the presence of amine and hydroxyl groups. The coating of SMP with polydopamine supports hydroxyapatite mineralization, osteoblast adhesion, dynamics growth, and expression of certain genes associated with ECM synthesis and osteogenesis. In another work conducted by Zhao et al, it was shown that PCL/SMP foam could be considered the self-fitting substrate to heal craniomaxillofacial bone defects with an excellent shaped scaffolding and SMP composite [SMPC (PCL and Fe_2O_3)]. Their functions have been recently investigated using both in vivo and in vitro experiments. Results indicated enhanced cell attachment and proliferation rate.[66] Liu et al studied cross-linked PCL nano-hydroxyapatite for osteogenesis. To have scaffolds with highly interconnected pores, they used the sugar leaching technique. To enrich the scaffold with distinct factors such as bone morphogenetic protein-2 (BMP-2), both BMP-2 and calcium alginate was located on the pore wall surface. Data showed that these scaffolds had notable shape-memory recovery from the compact status. *In vitro* and *in vivo* analyses indicated appropriate cytocompatibility, porosity, and enhanced bone regeneration after grafting into the mandibular bone defect in the rabbit.[67]

Fibrous materials are the other types of SMP that are fabricated via the electrospinning approach. Bao et al studied electrospun scaffolds consisting of poly(D,L-lactide-co-trimethylene carbonate) (PLMC) with notable shape-memory features. The results showed that fiber diameter, SMP mechanical features, and the glass transition temperature are associated with ratios of D, L-lactic acid (DLLA) and trimethylene carbonate TMC in the PLMC structure. Shape recovery ratios of $R_r > 94\%$ and shape fixity ratios of $R_f > 98\%$ exhibited suitable shape-memory features either in 2D or 3D nanofibrous PMLC scaffolds with the potential to increase ALP activity, apatite deposition, and osteoblast attachment.[68]

Complex bone defects are also challenging in the regeneration of bone tissue. As an example, the occurrence of various fractures in a single bone, fragmentation, segmental defects, and open fractures coinciding with soft tissue injury can complicate the repair process.[69] Synthetic polymers have been examined for their potential to face these challenges.[70] Baker et al investigated *in vivo* application of SMPs composed of tertbutyl acrylate and butyl acrylate in a segmental bone defect in a mouse model . According to the data, SMP implants demonstrate promise strength to fill complex bone defects as synthetic load-bearing bone scaffolds (Table 2).[71]

Together, the application of SMPs can increase and orchestrate differentiation signaling pathways related to osteogenic differentiation. SMPs can be used as injectable, adjustable, and self-fitting implants for repairing bone defects. Furthermore, incorporating bioactive materials to the structure accompanies by the shape-memory feature of the scaffold, and improves and hastens osteointegration.

Fabrication of SMP using 4D printing

4D printing, a rapidly developing new capacity of research, is an inclusive system utilizing both smart materials and a 3D printing process.[2,75] The advantage of the 4D printing system correlates with time-dependent control of printed materials' shape.[76,77] For the first time, 4D printing technology was introduced by Tibbits in February 2013.[78,79] Like 3D printing, 4D technology is dependent on novel and smart biomaterials which alter in response to external stimuli.[1] After the synthesis procedure, product configuration can be changed after exposure to stimulators. SMPs, SMAs, shape-memory hydrogels, and liquid crystal elastomers are smart materials that can be modulated using 4D printing.[80-83] Some studies have used 4D technology to synthesize porous bone scaffolds.

Table 2. Summary of selected studies using shape-memory constructs for bone regeneration

Chemical composition	Trigger	Fabrication method	Form	Notable characteristics	Ref
PLA/PCL-based poly(urethane) + iron oxide NP + PEG/gelatin	Thermo/ moisture-responsive	Microextrusion-based low-temperature fuse deposition manufacturing (LFDM) platform	Scaffold	• Osteogenesis • biodegradability • 3D production	72
Poly(ε-caprolactone) acrylate + Hydroxyapatite (HA) nanoparticles	Thermo-responsive	Thiolene click reaction of thiol-modified HA particles with functional acrylate-terminated PCL	2D films	• PCL-HA structures exhibited excellent shape memory properties, shape fixing, and recovery ratio	73
Poly(*N*-acryloyl glycinamide) + Nanoclay	Thermo-responsive	Hydrogen bonding monomer (*N*-acryloyl glycinamide) (NAGA) was mixed with nanoclay (Laponite XLG and UV light irradiation was used to polymerize NAGA-Clay pre-gel which resulted in the fabrication of stiff PNAGA-Clay composite hydrogels	Scaffold	• 3D production • Improved osteogenesis • In vivo investigation: rat	74
Poly (lactic acid-co-trimethylene carbonate) and natural polymer chitosan	Thermo-responsive	Solvent/nonsolvent method	3D scaffolds	• Improved stability • Great shape memory properties • Increased ALP activity • Increased MC3T3-E1 cells attachment and growth	64
Polyurethane/hydroxyapatite	Thermo-responsive	Gas foaming technique	Foam	• Excellent *in-vivo* stability • Defect filling ability • Great bioactivity and osteogenesis • Fast neovascularization and bone regeneration at 12 weeks after surgery • Improved osteoconductivity	65
Poly(e-caprolactone) (PCL) diacrylate + polydopamine coating	Thermo-responsive	Photocrosslinking of PCL diacrylate 33 using a SCPL method + fused salt template	Dense scaffold	• Ability to fill the irregular bone defect • Pores with high interconnectivity • Increased mineralization • Improved bioactivity and osteoconductivity • Enhancing osteoblast attachment and proliferation • Osteogenic gene expression and ECM deposition	52
Polylactic acid/Fe_3O_4	Magnetic field-responsive	-	3D scaffold	• Mechanical stability • Exceeding improvement in cell attachment and growth	66
c-PCL and hydroxyapatite nanoparticles + coating calcium alginate layer and BMP-2 on the surface of the pore wall	Body temperature-responsive	Sugar leaching technique:chemical cross-linking	Dense scaffold	• Used both in -vitro and in-vivo • Improved shape memory properties • Excessive cytocompatibility • Improved osteogenesis in the rabbit mandibular bone defect	67
Poly(D,L-lactide-co-trimethylene carbonate)	Thermo-responsive	Electrospinning method	2-D and 3-D forms of nanofibrous scaffolds	• Composition of the construction blocks indicate fiber quality, glass transition temperature, and SMP mechanical characterization • Outstanding shape memory properties • Superb osteoblast attachment, proliferation and osteogenesis	68

Furthermore, shape-memory scaffolds are controllable from outside of the body after implanting into the human body. In a study, Zhang et al developed the PLA/Fe_3O_4 composite filaments, and shape-memory potential can be controlled by remote stimulation. They also prepared a 4D-printed composite containing 15% Fe_3O_4 in the shape of spinal bones. According to their findings, shape-memory features can be induced using a magnetic field at 27.5 kHz. The synthesized products can acquire primary structures for a few seconds. Temperatures on the surface of 4D-printed structures reached physiological ranges during the shape recovery procedure, which makes it suitable for biomedical applications.[75] This system has great potential in medical treatment using an external magnetic field to control implanted scaffold in the body. Senatov et al printed a porous PLA/HA (20:3 w/w) scaffold with shape-memory capacity for bone regeneration by fused deposition modeling. The use of hydroxyapatite particles in the structure increased the polymer's Tg from 53°C to 57.1°C coincided with the increased recovery stress of the stent.[84] These kinds of printable SMP can be used as self-fitting substrates for the regeneration of bone defects. *In vitro* investigations showed quick attachment of mesenchymal stem cells on the surface of the scaffold, and a regular cell network was detectable using histological staining. In immunofluorescence staining, $CD105^+$cells displayed excellent viability, proliferation, and suitable interaction of these cells with the material surface triggered

the formation of vascular units after implantation into the target sites. In cartilage and bone tissue engineering, mechanical loading is an important factor as these tissues' physical properties vary dynamically by changing the imposed forces.[85] This characteristic can be an essential issue in designing 4D SMP scaffolds and have a promising potential of being replaced by expensive and complicated bioreactors. Hendrickson et al. designed a shape-memory PU SMP indicated with two approaches to induce pore network arrangements (0/90° and 0/45°) (Figure 3).[86] *In vitro* tests in temporary shape after recovery of the permanent shape of the scaffold showed that plated cells were considerably more extended. Thus, shape recovery can generate mechanical induction, which can influence the morphology of cells and nuclei.[86] Miao et al. prepared epoxidized acrylate resin using UV light polymerization of soybean oil for SLA. They pointed out that the shape-memory effect is associated with the temperature affecting the cross-linking rate. Higher temperatures can help the scaffolds to retain their primary shapes, while low temperatures can result in a temporary shape of the scaffold because of cross-linker freezing.[87] In contrast to polyethylene glycol diacrylate-based scaffolds, improved mesenchymal stem cell attachment and proliferation were detected in epoxidized acrylate scaffolds. Also, no substantial difference was detected between the effects of PCL and PLA. These scaffolds have the potential to be used as advanced 4D printable biomaterials in bone tissue engineering.[88] Most of the available tissue engineering methods for bone and cartilage regeneration do not consider osteochondral joint forces in the physiological state.[89] Therefore, it is necessary to focus on the micro and nanostructure of scaffolds using computational analysis to increase the clinical dependability of scaffolds and evaluate the load-bearing potential of scaffolds under real physio-mechanical forces.[90] Also, these investigations can help predict the mechanism of injury and interactions between the defect and the designed biomaterials and *in vivo* cyclic forces. Consequently, using the 4D bioprinting technology lights the way for defect repair in bone/cartilage tissue engineering.[72-74,91]

Conclusion

SMP has become a dynamic field of research with a lot of application potential in regenerative medicine. Recently, developments in the areas of biomaterials and methodologies have brought significant achievements for cartilage and bone regeneration. However, major challenges still exist in the generation of stable and appropriate functional bone and cartilage implants to substitute for large bone/cartilage defects. These challenges are related to technical approaches, clinical assessments, vascularization, mechanical stability, and defect fitting. On the other hand, in bone/cartilage tissue, cyclic mechanical loading and the simulation of the real physical condition is the most important object. Bioreactors are used to replicate biophysical conditions, which are expensive and usually are not user-friendly. Novel technical procedures like 3D and 4D bioprinting and using computational modeling pave the way towards precise personalized tissue engineering of bone/cartilage. Smart biomaterials which are responsive to stimuli would

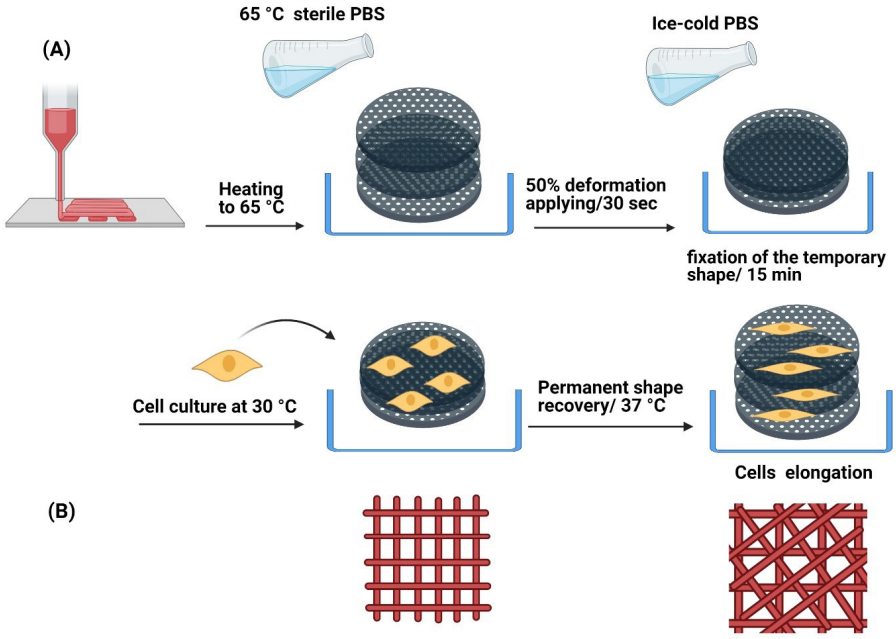

Figure 3. Schematic of a 4D scaffold fabrication. A shape-memory PU SMP indicated two approaches to induce pore network arrangements (0/90° and 0/45°). Seeded cells are considerably more extended in temporary shape after recovery of the permanent shape of the scaffold. Thus, shape recovery can generate mechanical induction which can influence the morphology of cells and nuclei[86]

be potential requirements of these novel methodologies and can help to produce a new generation of porous and mechanically stable biomaterials.

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Author Contributions
SS, RR, SFK, SN, ABK collected data and prepared the draft. HTN supervised the study.

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Ethical Approval
Not applicable.

Conflict of Interest
All authors declare that they have no competing interests.

References

1. Qasim M, Chae DS, Lee NY. Advancements and frontiers in nano-based 3D and 4D scaffolds for bone and cartilage tissue engineering. Int J Nanomedicine. 2019;14:4333-51. doi: 10.2147/ijn.s209431.
2. Delaey J, Dubruel P, Van Vlierberghe S. Shape-memory polymers for biomedical applications. Adv Funct Mater. 2020;30(44):1909047. doi: 10.1002/adfm.201909047.
3. Montoya C, Du Y, Gianforcaro AL, Orrego S, Yang M, Lelkes PI. On the road to smart biomaterials for bone research: definitions, concepts, advances, and outlook. Bone Res. 2021;9(1):12. doi: 10.1038/s41413-020-00131-z.
4. Huang WM, Ding Z, Wang CC, Wei J, Zhao Y, Purnawali H. Shape memory materials. Mater Today. 2010;13(7-8):54-61. doi: 10.1016/s1369-7021(10)70128-0.
5. Hu J, Zhu Y, Huang H, Lu J. Recent advances in shape-memory polymers: structure, mechanism, functionality, modeling and applications. Prog Polym Sci. 2012;37(12):1720-63. doi: 10.1016/j.progpolymsci.2012.06.001.
6. Hardy JG, Palma M, Wind SJ, Biggs MJ. Responsive biomaterials: advances in materials based on shape-memory polymers. Adv Mater. 2016;28(27):5717-24. doi: 10.1002/adma.201505417.
7. Xie H, Shao J, Ma Y, Wang J, Huang H, Yang N, et al. Biodegradable near-infrared-photoresponsive shape memory implants based on black phosphorus nanofillers. Biomaterials. 2018;164:11-21. doi: 10.1016/j.biomaterials.2018.02.040.
8. Koerner H, Price G, Pearce NA, Alexander M, Vaia RA. Remotely actuated polymer nanocomposites--stress-recovery of carbon-nanotube-filled thermoplastic elastomers. Nat Mater. 2004;3(2):115-20. doi: 10.1038/nmat1059.
9. Liu C, Qin H, Mather PT. Review of progress in shape-memory polymers. J Mater Chem. 2007;17(16):1543-58. doi: 10.1039/b615954k.
10. Migneco F, Huang YC, Birla RK, Hollister SJ. Poly(glycerol-dodecanoate), a biodegradable polyester for medical devices and tissue engineering scaffolds. Biomaterials. 2009;30(33):6479-84. doi: 10.1016/j.biomaterials.2009.08.021.
11. Xiao R, Huang WM. Heating/solvent responsive shape-memory polymers for implant biomedical devices in minimally invasive surgery: current status and challenge. Macromol Biosci. 2020;20(8):e2000108. doi: 10.1002/mabi.202000108.
12. Hribar KC, Metter RB, Ifkovits JL, Troxler T, Burdick JA. Light-induced temperature transitions in biodegradable polymer and nanorod composites. Small. 2009;5(16):1830-4. doi: 10.1002/smll.200900395.
13. Lendlein A, Jiang H, Jünger O, Langer R. Light-induced shape-memory polymers. Nature. 2005;434(7035):879-82. doi: 10.1038/nature03496.
14. Habault D, Zhang H, Zhao Y. Light-triggered self-healing and shape-memory polymers. Chem Soc Rev. 2013;42(17):7244-56. doi: 10.1039/c3cs35489j.
15. Weissleder R. A clearer vision for in vivo imaging. Nat Biotechnol. 2001;19(4):316-7. doi: 10.1038/86684.
16. Yi DH, Yoo HJ, Mahapatra SS, Kim YA, Cho JW. The synergistic effect of the combined thin multi-walled carbon nanotubes and reduced graphene oxides on photothermally actuated shape memory polyurethane composites. J Colloid Interface Sci. 2014;432:128-34. doi: 10.1016/j.jcis.2014.06.060.
17. Cheng Z, Wang T, Li X, Zhang Y, Yu H. NIR-Vis-UV light-responsive actuator films of polymer-dispersed liquid crystal/graphene oxide nanocomposites. ACS Appl Mater Interfaces. 2015;7(49):27494-501. doi: 10.1021/acsami.5b09676.
18. Zhang H, Xia H, Zhao Y. Optically triggered and spatially controllable shape-memory polymer–gold nanoparticle composite materials. J Mater Chem. 2012;22(3):845-9. doi: 10.1039/c1jm14615g.
19. Wang Z, Liu S, Ma J, Qu G, Wang X, Yu S, et al. Silver nanoparticles induced RNA polymerase-silver binding and RNA transcription inhibition in erythroid progenitor cells. ACS Nano. 2013;7(5):4171-86. doi: 10.1021/nn400594s.
20. Qu C, Wang L, He J, Tan J, Liu W, Zhang S, et al. Carbon nanotubes provoke inflammation by inducing the pro-inflammatory genes IL-1β and IL-6. Gene. 2012;493(1):9-12. doi: 10.1016/j.gene.2011.11.046.
21. Mohr R, Kratz K, Weigel T, Lucka-Gabor M, Moneke M, Lendlein A. Initiation of shape-memory effect by inductive heating of magnetic nanoparticles in thermoplastic polymers. Proc Natl Acad Sci U S A. 2006;103(10):3540-5. doi: 10.1073/pnas.0600079103.
22. Buckley PR, McKinley GH, Wilson TS, Small W, Benett WJ, Bearinger JP, et al. Inductively heated shape memory polymer for the magnetic actuation of medical devices. IEEE Trans Biomed Eng. 2006;53(10):2075-83. doi: 10.1109/tbme.2006.877113.
23. Li G, Yan Q, Xia H, Zhao Y. Therapeutic-ultrasound-triggered shape memory of a melamine-enhanced poly(vinyl alcohol) physical hydrogel. ACS Appl Mater Interfaces. 2015;7(22):12067-73. doi: 10.1021/acsami.5b02234.
24. Bhargava A, Peng K, Stieg J, Mirzaeifar R, Shahab S. Focused ultrasound actuation of shape memory polymers; acoustic-thermoelastic modeling and testing. RSC Adv. 2017;7(72):45452-69. doi: 10.1039/c7ra07396h.
25. Persi E, Duran-Frigola M, Damaghi M, Roush WR, Aloy P, Cleveland JL, et al. Systems analysis of intracellular pH vulnerabilities for cancer therapy. Nat Commun. 2018;9(1):2997. doi: 10.1038/s41467-018-05261-x.
26. Behl M, Zotzmann J, Lendlein A. Shape-memory polymers and shape-changing polymers. In: Lendlein A, ed. Shape-Memory Polymers. Berlin, Heidelberg: Springer; 2010. p. 1-40. doi: 10.1007/12_2009_26.
27. Wang K, Jia YG, Zhu XX. Two-way reversible shape memory polymers made of cross-linked cocrystallizable random copolymers with tunable actuation temperatures. Macromolecules. 2017;50(21):8570-9. doi: 10.1021/acs.macromol.7b01815.
28. Leng J, Du S. Shape-Memory Polymers and Multifunctional

Composites. CRC Press; 2010.
29. Bastiaansen CWM, Meyer HEH, Lemstra PJ. Memory effects in polyethylenes: influence of processing and crystallization history. Polymer. 1990;31(8):1435-40. doi: 10.1016/0032-3861(90)90147-q.
30. Wang M, Yuan Z, Ma N, Hao C, Guo W, Zou G, et al. Advances and prospects in stem cells for cartilage regeneration. Stem Cells Int. 2017;2017:4130607. doi: 10.1155/2017/4130607.
31. Filardo G, Perdisa F, Roffi A, Marcacci M, Kon E. Stem cells in articular cartilage regeneration. J Orthop Surg Res. 2016;11(1):42. doi: 10.1186/s13018-016-0378-x.
32. Jeznach O, Kołbuk D, Sajkiewicz P. Injectable hydrogels and nanocomposite hydrogels for cartilage regeneration. J Biomed Mater Res A. 2018;106(10):2762-76. doi: 10.1002/jbm.a.36449.
33. Huang YZ, Xie HQ, Silini A, Parolini O, Zhang Y, Deng L, et al. Mesenchymal stem/progenitor cells derived from articular cartilage, synovial membrane and synovial fluid for cartilage regeneration: current status and future perspectives. Stem Cell Rev Rep. 2017;13(5):575-86. doi: 10.1007/s12015-017-9753-1.
34. Zhang Y, Zhang M, Jiang H, Shi J, Li F, Xia Y, et al. Bio-inspired layered chitosan/graphene oxide nanocomposite hydrogels with high strength and pH-driven shape memory effect. Carbohydr Polym. 2017;177:116-25. doi: 10.1016/j.carbpol.2017.08.106.
35. He T, Li B, Colombani T, Joshi-Navare K, Mehta S, Kisiday J, et al. Hyaluronic acid-based shape-memory cryogel scaffolds for focal cartilage defect repair. Tissue Eng Part A. 2021;27(11-12):748-60. doi: 10.1089/ten.TEA.2020.0264.
36. Almeida HV, Sathy BN, Dudurych I, Buckley CT, O'Brien FJ, Kelly DJ. Anisotropic shape-memory alginate scaffolds functionalized with either type I or type II collagen for cartilage tissue engineering. Tissue Eng Part A. 2017;23(1-2):55-68. doi: 10.1089/ten.TEA.2016.0055.
37. Barmouz M, Hossein Behravesh A. Shape memory behaviors in cylindrical shell PLA/TPU-cellulose nanofiber bio-nanocomposites: analytical and experimental assessment. Compos Part A Appl Sci Manuf. 2017;101:160-72. doi: 10.1016/j.compositesa.2017.06.014.
38. Song JJ, Srivastava I, Kowalski J, Naguib HE. Fabrication and characterization of a foamed polylactic acid (PLA)/thermoplastic polyurethane (TPU) shape memory polymer (SMP) blend for biomedical and clinical applications. In: Behavior and Mechanics of Multifunctional Materials and Composites 2014. Vol 9058. San Diego: SPIE; 2014. p. 86-98. doi: 10.1117/12.2046494.
39. Zhang W, Zhang F, Lan X, Leng J, Wu AS, Bryson TM, et al. Shape memory behavior and recovery force of 4D printed textile functional composites. Compos Sci Technol. 2018;160:224-30. doi: 10.1016/j.compscitech.2018.03.037.
40. Uto S, Hikita A, Sakamoto T, Mori D, Yano F, Ohba S, et al. Ear cartilage reconstruction combining induced pluripotent stem cell-derived cartilage and three-dimensional shape-memory scaffold. Tissue Eng Part A. 2021;27(9-10):604-17. doi: 10.1089/ten.TEA.2020.0106.
41. Xuan H, Hu H, Geng C, Song J, Shen Y, Lei D, et al. Biofunctionalized chondrogenic shape-memory ternary scaffolds for efficient cell-free cartilage regeneration. Acta Biomater. 2020;105:97-110. doi: 10.1016/j.actbio.2020.01.015.
42. Jiang LB, Su DH, Liu P, Ma YQ, Shao ZZ, Dong J. Shape-memory collagen scaffold for enhanced cartilage regeneration: native collagen versus denatured collagen. Osteoarthritis Cartilage. 2018;26(10):1389-99. doi: 10.1016/j.joca.2018.06.004.
43. Chen W, Xu Y, Liu Y, Wang Z, Li Y, Jiang G, et al. Three-dimensional printed electrospun fiber-based scaffold for cartilage regeneration. Mater Des. 2019;179:107886. doi: 10.1016/j.matdes.2019.107886.
44. Spicer PP, Kretlow JD, Young S, Jansen JA, Kasper FK, Mikos AG. Evaluation of bone regeneration using the rat critical size calvarial defect. Nat Protoc. 2012;7(10):1918-29. doi: 10.1038/nprot.2012.113.
45. Giannoudis PV, Dinopoulos H, Tsiridis E. Bone substitutes: an update. Injury. 2005;36 Suppl 3:S20-7. doi: 10.1016/j.injury.2005.07.029.
46. Xie C, Reynolds D, Awad H, Rubery PT, Pelled G, Gazit D, et al. Structural bone allograft combined with genetically engineered mesenchymal stem cells as a novel platform for bone tissue engineering. Tissue Eng. 2007;13(3):435-45. doi: 10.1089/ten.2006.0182.
47. Neovius E, Engstrand T. Craniofacial reconstruction with bone and biomaterials: review over the last 11 years. J Plast Reconstr Aesthet Surg. 2010;63(10):1615-23. doi: 10.1016/j.bjps.2009.06.003.
48. Hollister SJ. Porous scaffold design for tissue engineering. Nat Mater. 2005;4(7):518-24. doi: 10.1038/nmat1421.
49. Albrektsson T, Johansson C. Osteoinduction, osteoconduction and osseointegration. Eur Spine J. 2001;10(Suppl 2):S96-101. doi: 10.1007/s005860100282.
50. Senturk B, Cubuk MO, Ozmen MC, Aydin B, Guler MO, Tekinay AB. Inhibition of VEGF mediated corneal neovascularization by anti-angiogenic peptide nanofibers. Biomaterials. 2016;107:124-32. doi: 10.1016/j.biomaterials.2016.08.045.
51. Sikavitsas VI, Temenoff JS, Mikos AG. Biomaterials and bone mechanotransduction. Biomaterials. 2001;22(19):2581-93. doi: 10.1016/s0142-9612(01)00002-3.
52. Zhang D, George OJ, Petersen KM, Jimenez-Vergara AC, Hahn MS, Grunlan MA. A bioactive "self-fitting" shape memory polymer scaffold with potential to treat cranio-maxillo facial bone defects. Acta Biomater. 2014;10(11):4597-605. doi: 10.1016/j.actbio.2014.07.020.
53. Adhikari R, Gunatillake PA, Griffiths I, Tatai L, Wickramaratna M, Houshyar S, et al. Biodegradable injectable polyurethanes: synthesis and evaluation for orthopaedic applications. Biomaterials. 2008;29(28):3762-70. doi: 10.1016/j.biomaterials.2008.06.021.
54. Hayashi C, Kinoshita A, Oda S, Mizutani K, Shirakata Y, Ishikawa I. Injectable calcium phosphate bone cement provides favorable space and a scaffold for periodontal regeneration in dogs. J Periodontol. 2006;77(6):940-6. doi: 10.1902/jop.2006.050283.
55. Stancu IC, Lungu A, Iovu H. Hydrogels for bone regeneration. In: Dubruel P, Van Vlierberghe S, eds. Biomaterials for Bone Regeneration. Woodhead Publishing; 2014. p. 62-86. doi: 10.1533/9780857098104.1.62.
56. Liu J, Mao K, Liu Z, Wang X, Cui F, Guo W, et al. Injectable biocomposites for bone healing in rabbit femoral condyle defects. PLoS One. 2013;8(10):e75668. doi: 10.1371/journal.pone.0075668.
57. Lendlein A, Langer R. Biodegradable, elastic shape-memory polymers for potential biomedical applications. Science. 2002;296(5573):1673-6. doi: 10.1126/science.1066102.
58. Yakacki CM, Gall K. Shape-memory polymers for biomedical applications. In: Lendlein A, ed. Shape-Memory Polymers. Berlin, Heidelberg: Springer; 2010. p. 147-75. doi: 10.1007/12_2009_23.
59. Langer R, Tirrell DA. Designing materials for biology and medicine. Nature. 2004;428(6982):487-92. doi: 10.1038/nature02388.
60. Hastings GW. Structural considerations and new polymers for biomedical applications. Polymer. 1985;26(9):1331-5. doi:

10.1016/0032-3861(85)90308-8.
61. Thomas S, Grohens Y, Ninan N. Nanotechnology Applications for Tissue Engineering. William Andrew; 2015.
62. Safranski DL, Smith KE, Gall K. Mechanical requirements of shape-memory polymers in biomedical devices. Polymer Reviews. 2013;53(1):76-91. doi: 10.1080/15583724.2012.752385.
63. Prasadh S, Wong RCW. Unraveling the mechanical strength of biomaterials used as a bone scaffold in oral and maxillofacial defects. Oral Sci Int. 2018;15(2):48-55. doi: 10.1016/S1348-8643(18)30005-3.
64. Hu X, He J, Yong X, Lu J, Xiao J, Liao Y, et al. Biodegradable poly (lactic acid-co-trimethylene carbonate)/chitosan microsphere scaffold with shape-memory effect for bone tissue engineering. Colloids Surf B Biointerfaces. 2020;195:111218. doi: 10.1016/j.colsurfb.2020.111218.
65. Xie R, Hu J, Hoffmann O, Zhang Y, Ng F, Qin T, et al. Self-fitting shape memory polymer foam inducing bone regeneration: a rabbit femoral defect study. Biochim Biophys Acta Gen Subj. 2018;1862(4):936-45. doi: 10.1016/j.bbagen.2018.01.013.
66. Zhao W, Huang Z, Liu L, Wang W, Leng J, Liu Y. Porous bone tissue scaffold concept based on shape memory PLA/Fe3O4. Compos Sci Technol. 2021;203:108563. doi: 10.1016/j.compscitech.2020.108563.
67. Liu X, Zhao K, Gong T, Song J, Bao C, Luo E, et al. Delivery of growth factors using a smart porous nanocomposite scaffold to repair a mandibular bone defect. Biomacromolecules. 2014;15(3):1019-30. doi: 10.1021/bm401911p.
68. Bao M, Lou X, Zhou Q, Dong W, Yuan H, Zhang Y. Electrospun biomimetic fibrous scaffold from shape memory polymer of PDLLA-co-TMC for bone tissue engineering. ACS Appl Mater Interfaces. 2014;6(4):2611-21. doi: 10.1021/am405101k.
69. Reichert JC, Saifzadeh S, Wullschleger ME, Epari DR, Schütz MA, Duda GN, et al. The challenge of establishing preclinical models for segmental bone defect research. Biomaterials. 2009;30(12):2149-63. doi: 10.1016/j.biomaterials.2008.12.050.
70. Felfel RM, Ahmed I, Parsons AJ, Haque P, Walker GS, Rudd CD. Investigation of crystallinity, molecular weight change, and mechanical properties of PLA/PBG bioresorbable composites as bone fracture fixation plates. J Biomater Appl. 2012;26(7):765-89. doi: 10.1177/0885328210384532.
71. Baker RM, Tseng LF, Iannolo MT, Oest ME, Henderson JH. Self-deploying shape memory polymer scaffolds for grafting and stabilizing complex bone defects: a mouse femoral segmental defect study. Biomaterials. 2016;76:388-98. doi: 10.1016/j.biomaterials.2015.10.064.
72. Wang YJ, Jeng US, Hsu SH. Biodegradable water-based polyurethane shape memory elastomers for bone tissue engineering. ACS Biomater Sci Eng. 2018;4(4):1397-406. doi: 10.1021/acsbiomaterials.8b00091.
73. Tian G, Zhu G, Xu S, Ren T. A novel shape memory poly(ε-caprolactone)/hydroxyapatite nanoparticle networks for potential biomedical applications. J Solid State Chem. 2019;272:78-86. doi: 10.1016/j.jssc.2019.01.029.
74. Zhai X, Ma Y, Hou C, Gao F, Zhang Y, Ruan C, et al. 3D-printed high strength bioactive supramolecular polymer/clay nanocomposite hydrogel scaffold for bone regeneration. ACS Biomater Sci Eng. 2017;3(6):1109-18. doi: 10.1021/acsbiomaterials.7b00224.
75. Zhang F, Wang L, Zheng Z, Liu Y, Leng J. Magnetic programming of 4D printed shape memory composite structures. Compos Part A Appl Sci Manuf. 2019;125:105571. doi: 10.1016/j.compositesa.2019.105571.
76. Gladman AS, Matsumoto EA, Nuzzo RG, Mahadevan L, Lewis JA. Biomimetic 4D printing. Nat Mater. 2016;15(4):413-8. doi: 10.1038/nmat4544.
77. González-Henríquez CM, Sarabia-Vallejos MA, Rodriguez-Hernandez J. Polymers for additive manufacturing and 4D-printing: materials, methodologies, and biomedical applications. Prog Polym Sci. 2019;94:57-116. doi: 10.1016/j.progpolymsci.2019.03.001.
78. Tibbits S. 4D printing: multi-material shape change. Archit Des. 2014;84(1):116-21. doi: 10.1002/ad.1710.
79. Tibbits S. Printing products. Rochelle, NY: Mary Ann Liebert, Inc; 2016.
80. Kirillova A, Maxson R, Stoychev G, Gomillion CT, Ionov L. 4D biofabrication using shape-morphing hydrogels. Adv Mater. 2017;29(46):1703443. doi: 10.1002/adma.201703443.
81. Ambulo CP, Burroughs JJ, Boothby JM, Kim H, Shankar MR, Ware TH. Four-dimensional printing of liquid crystal elastomers. ACS Appl Mater Interfaces. 2017;9(42):37332-9. doi: 10.1021/acsami.7b11851.
82. Caputo MP, Berkowitz AE, Armstrong A, Müllner P, Solomon CV. 4D printing of net shape parts made from Ni-Mn-Ga magnetic shape-memory alloys. Addit Manuf. 2018;21:579-88. doi: 10.1016/j.addma.2018.03.028.
83. Ge Q, Sakhaei AH, Lee H, Dunn CK, Fang NX, Dunn ML. Multimaterial 4D printing with tailorable shape memory polymers. Sci Rep. 2016;6:31110. doi: 10.1038/srep31110.
84. Senatov FS, Niaza KV, Zadorozhnyy MY, Maksimkin AV, Kaloshkin SD, Estrin YZ. Mechanical properties and shape memory effect of 3D-printed PLA-based porous scaffolds. J Mech Behav Biomed Mater. 2016;57:139-48. doi: 10.1016/j.jmbbm.2015.11.036.
85. Saghati S, Nasrabadi HT, Khoshfetrat AB, Moharamzadeh K, Hassani A, Mohammadi SM, et al. Tissue engineering strategies to increase osteochondral regeneration of stem cells; a close look at different modalities. Stem Cell Rev Rep. 2021;17(4):1294-311. doi: 10.1007/s12015-021-10130-0.
86. Hendrikson WJ, Rouwkema J, Clementi F, van Blitterswijk CA, Farè S, Moroni L. Towards 4D printed scaffolds for tissue engineering: exploiting 3D shape memory polymers to deliver time-controlled stimulus on cultured cells. Biofabrication. 2017;9(3):031001. doi: 10.1088/1758-5090/aa8114.
87. Miao S, Cui H, Nowicki M, Lee SJ, Almeida J, Zhou X, et al. Photolithographic-stereolithographic-tandem fabrication of 4D smart scaffolds for improved stem cell cardiomyogenic differentiation. Biofabrication. 2018;10(3):035007. doi: 10.1088/1758-5090/aabe0b.
88. Miao S, Zhu W, Castro NJ, Nowicki M, Zhou X, Cui H, et al. 4D printing smart biomedical scaffolds with novel soybean oil epoxidized acrylate. Sci Rep. 2016;6:27226. doi: 10.1038/srep27226.
89. Wang MO, Vorwald CE, Dreher ML, Mott EJ, Cheng MH, Cinar A, et al. Evaluating 3D-printed biomaterials as scaffolds for vascularized bone tissue engineering. Adv Mater. 2015;27(1):138-44. doi: 10.1002/adma.201403943.
90. Geris L. In vivo, in vitro, in silico: computational tools for product and process design in tissue engineering. In: Geris L, ed. Computational Modeling in Tissue Engineering. Berlin, Heidelberg: Springe; 2013. p. 1-15. doi: 10.1007/8415_2012_144.
91. Rychter P, Pamula E, Orchel A, Posadowska U, Krok-Borkowicz M, Kaps A, et al. Scaffolds with shape memory behavior for the treatment of large bone defects. J Biomed Mater Res A. 2015;103(11):3503-15. doi: 10.1002/jbm.a.35500.