SYSTEMATIC REVIEW

Plant-based scaffolds and osteogenesis: A systematic review

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Abstract

Objective: To enhance the understanding and usefulness of decellularised plant tissues as scaffolds for bone tissue engineering, and to review the recent advances in plant-based scaffolds for bone regeneration.

Method: The systematic review was conducted from February to June 2022, and comprised literature search on PubMed and Science Direct databases for articles published in the English language between 2013 and June, 2022. The search was conducted using key words 'plant-based scaffolds and osteogenesis' and 'decellularised plant and scaffolds and bone tissue engineering' and 'plant root-based scaffold and bone formation'. Full-text articles and short communication covering both in vitro and in vivo studies focussing on plant-based scaffolds for osteogenesis were included. Additional references that satisfied the inclusion criteria were found on Google Scholar and included in the review. Quality evaluation of the studies included was done independently by two researchers, and the risk of bias was calculated and categorised as low, medium and high risk.

Results: Of the 564 articles, 10(1.77 %) were included; 6(60%) purely in vitro studies, and 4(40%) studies having both in vitro and in vivo components. Decellularized plant tissue alone was used as three-dimensional scaffold materials in 5(50%) studies, but in 4(40%) studies, plant-based composite scaffolds were used. The risk of bias was medium in 7(70%) studies.

Conclusions: Plant-based scaffolds promote osteogenesis, but more in vivo research on plant-based studies is needed to label it as suitable scaffolds for bone tissue engineering.

Keywords: Plant-based scaffolds, Osteogenesis, Decellularised Plant, bone tissue engineering, Plant root-based scaffold, Bone formation. (JPMA 74: 1990; 2024) **DOI: https://doi.org/10.47391/JPMA.10485**

Introduction

Other than the debates related to civilisation, culture and age, bone lesions are widespread in different communities, and their frequency increases with age. Furthermore, bone disorders and injuries from accidents and traumas are widespread in most countries, and fractured bones do not heal on their own in majority of cases.[1] Trauma, bone resection because of amputation surgery, and repair of congenital deformities are all common bone illnesses and defects in clinical practice.[2] Bone tissue transplantation, including allograft and autograft transplantation, is the ongoing treatment strategy.[1] Despite the fact that autograft grafts are the gold standard treatment, this approach has significant unresolved issues that are dependent on the method's nature.[1] Transmission of diseases and graft rejection are also important issues with allografts. In order to develop new therapies, researchers are looking into tissue engineering and cell therapy as alternatives.[1] Scaffolds with an important role in bone tissue engineering should ideally be characterised by high porosity microstructure with pore size typically ranging 100-200µm, which facilitates cell adhesion and migration while maintaining the desired mechanical performance under controlled degradation and biocompatibility promoting cell growth and differentiation.[3] Bone tissue engineering has earned recognition as a comprehensive method for modifying biomaterials to improve therapeutic effectiveness.[4] Due to the high occurrence of major segmental bone defects arising from trauma, tumours resections, or infection/inflammation, the demand for new-generation scaffolds for bone tissue engineering is currently enormous.[4]

Synthetic polymer-based scaffolds are reproducible, have a known chemical composition, and may be tailored to meet the needs of the application. Despite their limited reproducibility, naturally-derived polymer scaffolds have a better biological response, strong biocompatibility, and environmental safety when compared to synthetic scaffolds.[5] Tissue decellularisation is a suitable alternative to materials processing because the biomimetic native structure can be kept while cellular components are removed to produce three-dimensional (3D) acellular scaffolds for tissue engineering applications.[6] For cell adhesion, proliferation and differentiation, decellularised matrix provides an appropriate morphological and biochemical microenvironment.[7] Plant-derived tissues have recently attracted a lot of attention as a reasonable alternative to animal sources for obtaining decellularised scaffolds for tissue engineering applications.[8] Plant tissues,

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Submission complete: 05-10-2023 **Review began:** 11-12-2023

Acceptance: 31-08-2024 **Review end:** 27-07-2024

in reality, have good biocompatibility and cyto-compatibility, in addition to their easy availability, low cost, ease of application, and lack of ethical concerns.[9] So, does the use of plant-based materials as scaffolds lead to better healing in patients with bone defects via new bone formation compared to the rest? The current systematic review was planned to find an answer to the question by reviewing both in vitro and in vivo research to see the potential usage of plant-based materials as scaffolds in bone tissue engineering.

Materials and Methods

The systematic review was conducted from February to June 2022, and comprised literature search on PubMed and Science Direct databases for articles published in the English language between 2013 and June, 2022. The search was conducted using key words 'plant-based scaffolds and osteogenesis' and 'decellularised plant and scaffolds and bone tissue engineering' and 'plant root-based scaffold and bone formation'. The studies identified were imported into endnote X7 software to remove duplicates.

Full-text articles and short communication covering both in vitro and in vivo studies focussing on plant-based scaffolds for osteogenesis were included. Narrative reviews, grey literature, books, publications in languages other than English, articles whose full texts were unavailable, abstracts presented at conferences, studies older than 2013, and

Table-1: Summary of the selected in vitro studies regarding effectiveness of plant-based scaffolds for osteogenesis.

Reference	Material/ of Scaffold/ Type of Study	Specifications of scaffold	Tests performed/ Methodology	Conclusion/ Remarks
Salehi, 2020[1]	Decellularized Spinach leaf scaffold	**Atomic force microscopy (AFM)**: Ra of scaffold = 11.50nm **Scanning electron microscopy (SEM):** Pore size: 50µm, Pore shape: polygon, irregularities. Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR): Bands in range of protein bands ➠ probably related to the type of residual plant proteins **Brunauer-Emmett-Teller (BET):** Specific surface area of scaffold= 28.540m/g Pore volume to weight ratio: High (0.06255cm/g). Mechanical properti	Cell culturing, 4′,6-diamidino-2-phenylindole (DAPI) staining, Alzarine Red S (ARS) staining, Alkaline phosphatase (ALP) & calcium content assays. Contact angle assay, Reverse transcription polymerase chain reaction (RT-PCR)	Cells expansion without aggregation, cell growth increased; Exhibited stem cell proliferation & mineralization.
Salehi, 2021[2]	Decellularized Cabbage 3-D cellulose scaffold. In Vitro study	**SEM:** Regular geometric grids & some pores. **AFM:** Close peaks & surface roughness. **ATR-FTIR & BET assay:** Scaffold's specific surface area = 6.789m/g, Total Pore volume = 0.1766cm/G, Mean pore diameter = 10nm, Contact angle = 70.88° (Hydrophilic).	Cell culturing, (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) (MTT) assay, Alizarin red-S (ARS) staining, ALP activity, Calcium content, Contact angle, Cell proliferation assay, RT-PCR.	Biocompatible & non- toxic, Bone Marrow Mesenchymal Stem Cells (BM-MSCs) showed proliferation & mineralization (high expression of Runt-related transcription factor 2 (Runx2) & Collagen-1 (Col-I)
Selvakum, 2016[4]	Electro-spun Aloe vera (Al) wrapped mesoporous hydroxyapatite (mHA)	Average pore size of nanocrystal ⟶ ~ 2.2nm for (mHA) & ~ 6.2nm for (Al-mHA). SPU/Al-mHA scaffold: Contact angle 86°	MTT assay, Antibacterial activity test.	Improved biocompatibility, antimicrobial activity & biodegradability; Rapid & strong mineralization & high osteoconductive
Contessi Negrini, 2020[12]	Decellularized apple, carrot & celery scaffolds	**SEM:** 3D highly porous structure. Carrot derived scaffolds have a heterogenous structure with Average pore size ➠ 70±12µm in central region & 130±26µm in peripheral region. Carrot derived scaffold showed Compression modulus=43.4±5.22kPa & Cell viability= 85.0±7.2%	Cell culturing, Direct contact test, Alamar blue assay, cell viability assay, ALP assay	Carrots supported MC3T3-E1 pre-osteoblast adhesion, proliferation and osteogenic differentiation. Viable cells adhered to decellularized carrot tissue and showed Osteogenic differentiation (increased ALP activity)
Ji, 2014[13]	Electro-spun Nano-scaffolds Naringin enriched Polycaprolactone (PCL) & Poly Ethylene Glycol (PEG)-b-PCL.	**FTIR spectra:** PCL/Naringin: Fiber diameter: 367 ± 38nm. Contact angle: 137 ±2.1◦ PCL/PEG-b-PCL/Naringin: Fiber diame	Drug release & Degradation assay, Cell culturing, Colorimetric MTS assay, ARS staining.	PCL/PEG-b-PCL/naringin showed calcium presence, faster degradation (losing 55% weigh), Controlled naringin release & enhanced functions of MC3T3-E1 osteoblasts & suppressed formation of osteoclasts. Pure PCL showed <10% weight loss during 60 days
Kang, 2018[14]	Mesoporous magnesium silicate/polycaprolactone/wheat protein (mMS/PCL/WP) composite scaffold by Sol – gel technique.	SEM: Interconnected rectangular 450µm wide pores & regular strands, increased surface roughness.	Mesoporous magnesium silicate/polycaprolactone/wheat protein (mMS/PCL/WP) composite scaffold by Sol – gel technique.	SEM: Interconnected rectangular 450µm wide pores & regular strands, increased surface roughness.

Atomic force microscopy (AFM); Scanning electron microscopy (SEM); Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR); Brunauer-Emmett-Teller (BET); 4′,6-diamidino-2-phenylindole (DAPI) staining, Alzarine Red S (ARS) staining, Alkaline phosphatase (ALP; Reverse transcription polymerase chain reaction (RT-PCR); (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) (MTT); Alizarin red-S (ARS); Aloe vera (Al); mesoporous hydroxyapatite (mHA); , Bone Marrow Mesenchymal Stem Cells (BM-MSCs); Runt-related transcription factor 2 (Runx2); Collagen-1 (Col-I); Polycaprolactone (PCL); Poly Ethylene Glycol (PEG); Mesoporous magnesium silicate/polycaprolactone/wheat protein (mMS/PCL/WP).

duplicate studies were excluded. Studies focussing on any other aspect of plant-based scaffolds were also excluded.

Preferred Reporting Items for Systematic reviews (PRISMA) guidelines were adhered to.[10] Following the guidelines of Cochrane statistical collaboration review[11] quality evaluation of the studies was done independently by two researchers and the risk of bias outcomes were calculated and described as having low, medium, and high risk. The risk was identified on the basis of plain plant-based scaffold, calculated scaffold's parameters, fibre diameter, pore size, contact angle, mechanical properties, anti-bacterial potential, biocompatibility and osteogenic potential of the tested plant-based scaffolds.

All the references in the selected publications were checked and compared to the ones found during the original search. Additional references that satisfied the inclusion criteria were searched on Google Scholar and subsequently included in the review.

Results

Of the 564 articles screened, 10(1.77%) were included (Figure). There was 3(30%) studies from the year 2021, followed by 2(20%) from 2018, and 1(10%) each from 2014, 2016, 2019, 2020 and 2022.

Among the selected studies, 6(60%) were purely in vitro studies (Table 1),[1,2,4,5,12,13] while 4(40%) studies had both in vitro and in vivo components (Table 2).[3,14-16] Decellularised plant tissue alone was used as 3D scaffold materials in 5(50%) studies in which the sources of these scaffolds included cabbage, bamboo stem, spinach leaf, broccoli, sweet pepper, carrot, persimmon, jujube, and celery, while

Table-2: Summary of the selected mixed studies regarding effectiveness of plant-based scaffolds for osteogenesis.

Reference	Material/ of Scaffold/ Type of Study	Specifications of scaffold	Tests performed/ Methodology	Remarks
Huang, 2022[3]	Lotus root scaffolds (LRS) & mineralized LRS. (implantation of scaffolds in 5mm bone defects in Sprague–Dawley (SD) Rat model for 1-2 months)	**Scanning electron microscope (SEM):** Pores size ~100μm & ellipsoid starch grains (about 30μm × 60μm) with parallel striped pattern on LRS hole wall. Mineralized LRS: Irregular crystals on hole wall	In-vitro: Live/dead cells staining assay; (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) (MTT) assay; 2,2-Diphenyl-1-picrylhydrazyl (DPPH) assay, & Reverse transcription polymerase chain reaction (RT-PCR); In-vivo: Reactive oxygen species (ROS) test, Scratch test assay; enzyme-linked immunosorbent assay (ELISAs), Bone defect micro-CT images, Quantitative analysis, histological & immunohistochemical examinations.	Mineralized LRS showed > biocompatibility, angiogenesis after 24h, early & > osteogenic differentiation in vitro & > bone mineral density, bone volume & trabecular thickness, earlier & > bone formation after 1-month implantation in vivo.
Sasayama, 2018[15]	Epigallocatechin gallate – modified gelatin sponge scaffold (vhEGCG-GS) by Aqueous synthesis technique. (Rat congenital cleft jaw model)	**SEM:** Pores size vhEGCG-GS:10μm, vhGS: 100μm. **Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR) spectroscopy:** Existence of EGCG identified. **Contact angle:** vhGS: 110.4° (Hydrophobic), vhEGCG-GS: 3.8°(Hydrophilic) **Zeta potential:** vhGS: +0.24mV vhEGCG-GS: -0.54mV	In vitro cell attachment assay, cell culturing, centrifugation, ceiling culture technique.	vhEGCG-GS: Higher cell attachment; augmented cell adhesion; Precipitation of Calcium phosphate enabled rADSC & rDFAT cells to induce ossification in rat congenital cleft jaw model.
Mohan, 2021[16]	Decellularized Bamboo stem (Subcutaneous implantation in rat model)	**SEM:** Pore size = 20-150 μm **Contact angle** reduced from 93.41 to 22.48 ° (hydrophilic). **Compressive strength:** 1.52 MPa Oxidized bamboo scaffold treated with 0.1 & 0.5% sodium periodate showed: Enhanced hydrophilicity & protein adsorption but lower mechanical strength	Deoxyribonucleic acid (DNA) assay, Alkaline phosphatase (ALP) activity, Mechanical testing, Degradation study, Protein adsorption study, cell adhesion, cell viability assay & osteogenic differentiation.	DNA<50ng indicates completion of decellularization. Oxidized bamboo scaffold treated with 0.1 & 0.5% sodium periodate showed: Stimulated adhesion, viability & osteogenic differentiation of mesenchymal stem cells (MSCs). Ozone depletion potential (ODP) 0.5% scaffold induced angiogenesis, good biocompatibility & biodegradation in rat model.
Lee, 2019[17]	Decellularized apple, broccoli, sweet pepper, carrot,persimmon & jujube. (Rat calvarial deficiency model	SEM: Apple scaffold pores in diameter = 200μm Decellularized scaffolds have pores of various shapes & sizes. Carrot & persimmon both are similar to apple in pore shape & size.	Cell culturing, ARS, Von kossa staining, RT-PCR, cell viability assay & cellular immunostaining	Once seeded with Human-induced pluripotent stem cells (hiPSCs), only cells cultivated in apple scaffolds survived & proliferated well. Bone implants promoted healing of bone defects & have capacity to provide grafted bone with needed bone supply.

Lotus root scaffolds (LRS); Sprague–Dawley (SD); Scanning electron microscope (SEM): (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) (MTT); 2,2-Diphenyl-1-picrylhydrazyl (DPPH); : Reactive oxygen species (ROS); enzyme-linked immunosorbent assay (ELISAs); Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR); Epigallocatechin gallate – modified gelatin sponge scaffold (vhEGCG-GS); Deoxyribonucleic acid (DNA; Alkaline phosphatase (ALP); Mesenchymal stem cells (MSCs); Ozone depletion potential (ODP); Human-induced pluripotent stem cells (hiPSCs).

Table-3: Quality assessment of the studies included in the systematic review for the risk of bias outcomes.

Reference	Plant-based Scaffold	Calculated Parameters of Scaffolds							Score
		Fiber diameter	Pore Size	Contact angle	Mechanical Properties	Anti-bacterial Potential	Biocompatibility	Osteogenic Potential	
Salehi A, 2020[1]	yes	-	yes	yes	-	-	yes	yes	5/8
Salehi A, 2021[2]	yes	yes	-	yes	-	-	yes	yes	5/8
Huang, 2022[3]	yes	-	yes	-	-	-	yes	yes	4/8
Selvakumar, 2016[4]	yes	-	yes	yes	-	yes	yes	yes	6/8
Contesssi Negrini, 2020[5]	yes	-	yes	-	yes	-	yes	yes	5/8
Ji, 2014[13]	yes	yes	-	yes	-	-	-	yes	4/8
Kang, 2018[14]	yes	-	yes	yes	yes	-	yes	yes	6/8
Sasayama, 2018[15]	yes	-	yes	yes	-	-	-	yes	4/8
Mohan, 2021[16]	yes	-	yes	yes	yes	-	yes	yes	6/8
Lee, 2019[17]	yes	-	yes	-	-	-	yes	yes	4/8

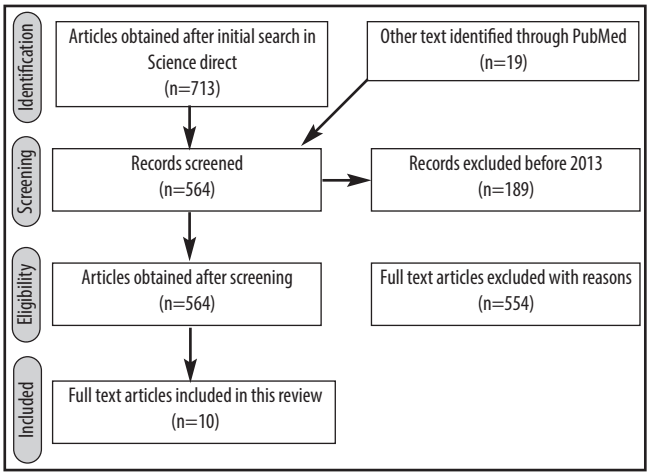

Figure: Preferred Reporting Items for Systematic reviews (PRISMA) flow chart.

lotus root was also used as a scaffold in 1(10%) study.[1-3,5,15,16] In the rest of the studies, plant-based composite scaffolds included mesoporous magnesium silicate (mMS)/polycaprolactone (PCL)/wheat protein (WP), epigallocatechin gallate-modified gelatin sponge scaffold (vhEGCG-GS), aloe vera-wrapped mesoporous hydroxyapatite, naringin-incorporated PCL, polyethyleneglycol-block-poly-caprolactone (PEG-b-PCL) and mineralised lotus root scaffold.[3]

Among all the studies 3 30%) showed high risk of bias[4,13,15] and 7(70%) showed medium bias(Table 3).[1-3,5,12,14,16]

Discussion

The majority of the studies included in this systematic review were conducted in vitro and involved plant-based scaffolds. Pure plant-based scaffolds prepared by decellularisation technique were employed in 5 studies[1,2,5,15,16] whereas composites of plant and other materials were being used as scaffolds in 5 studies.[3,4,12-14] Among these, mMS/PCL/WP composite scaffold was manufactured by a rapid prototyping technique[14] vhEGCG-GS was prepared by aqueous synthesis technique[14] aloe vera-wrapped mesoporous hydroxyapatite scaffolods, naringin incorporated PCL and PEG-b-PCL nano-scaffolds were synthesised by electrospinning[4,12] and modified lotus root scaffolds were prepared by simple mineralisation.[3] All the studies included in this systematic review showed osteogenic potential.

The scaffolds mentioned in the studies were characterised using scanning electron microscopy (SEM)[1-3,5,13-16] and atomic force microscopy (AFM)(1, 2) to visualise their pore size and arrangement, Brunauer-Emmett-Teller (BET) analysis[1,2] to know the specific interactive surface area of scaffolds, attenuated total reflectance- Fourier transform infrared spectroscopy (ATR-FTIR)[1,2,12,14] to assess changes in chemical structure upon scaffold modification, contact angle to measure scaffold's wettability[1,2,4,12,14,15] and zeta potential to assess surface interactions.[14]

In vitro testing worth mentioning in relation to the mentioned bone forming scaffolds in this systematic review included live/dead cells staining assay which assesses osteoblast viability[3] alamar blue assay[5] 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) assay[2-4] to assess cytocompatibility, 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) assay for evaluation of anti-oxidant in plant extract[3] reverse transcription polymerase chain reaction (RT-PCR) to analyse bone-related gene expression[1-3,16] and alkaline phosphatase[18] activity assay[17] to assess osteoblastic activity.[1,2,5,15] In vivo testing included in relation to the mentioned scaffolds were reactive oxygen species (ROS) test to detect the presence of ROS, which plays a significance role in cellular vitality[3] cell differentiation, inflammation and cell signalling, enzyme-linked immunosorbent assay (ELISA) to detect harmful products in body[3] bone defect micro-computed tomography (CT) images for bone density analysis,[3] quantitative analysis and histological examination to see new bone formation.[3]

Salehi et al. in 2021 tested decellularised cabbage leaves as potential scaffolds for bone regeneration. Such scaffolds

exhibited surface roughness, porosity and hydrophilicity. It proved to be a biocompatible and non-toxic substrate for bone morphogenic mesenchymal stem cells (BM-MSCs) and resulted in osteogenic differentiation, BM-MSCs proliferation and mineralisation. BM-MSCs grown on the decellularised cabbage scaffold also expressed high runt-related transcription factor 2 (Runx2) and Collagen type I (Col-I).[2] In another study, decellularised spinach leaves were employed as a scaffold, and cells grew on the surface of the scaffold and did not aggregate, resulting in greater cell proliferation and mineralisation. Thus, the decellularised spinach leaf scaffold provided a suitable substrate for BM-MSCs during cellular attachment, growth and proliferation as well as improved osteogenic differentiation.[1]

In another study, a decellularisation of bamboo stem was done to yield a plant-based scaffold which was then implanted subcutaneously in a rat model. Oxidised bamboo scaffold treated with 0.1% and 0.5% sodium periodate showed enhanced hydrophilicity and protein adsorption, but lower mechanical strength. It stimulated adhesion, viability and osteogenic differentiation of MSCs. It also exhibited good biocompatibility and biodegradation.[15]

In two in vitro studies, different fruits and vegetables, including apple, broccoli, sweet pepper, carrot, persimmon, jujube and celery, were decellularised and their potential to be used as scaffolds for bone tissue engineering was assessed.[5,16] These decellularised scaffolds exhibited pores of various shapes and sizes. The apple scaffold had pores of about 200µm in diameter. Carrot and persimmon both were similar to apple in pore shape and size.[16] Carrot-derived scaffolds had a heterogenous structure with average pore size of 70±12µm in the central region and 130±26µm in the peripheral region.[5] Decellularised carrot scaffold supported osteoblastic cell line MC3T3-E1 pre-osteoblast adhesion (viable cells=85.0±7.2%), proliferation and osteogenic differentiation.[5]

A 2021 study attempted to create bone-like tissue by seeding pluripotent stem cells (hiPSCs) that had been stimulated to differentiate as osteoblasts in culture onto the decellularised scaffolds of apple, broccoli, sweet pepper, carrot, persimmon and jujube.[16] The apple scaffolding with regular pores appeared to provide the greatest construct. The resulting bone-like tissue was implanted in a rat calvarial deficiency model, where it assisted in the formation of calcified tissue.[16] This method easily facilitates the creation of mineralised bone, depending on the regularity and sizing of scaffold pores. The bone implants promote healing of bone defects in vivo and have the capacity to provide grafted bone with needed bone supply.

The ability of decellularised apple, carrot and celery-derived tissues to act as scaffolds for the regeneration of adipose tissue, bone tissue and tendons was demonstrated in a 2020 study. Only carrots supported MC3T3-E1 pre-osteoblast adhesion, proliferation and osteogenic differentiation, whereas apple and celery supported adipose and tendon regeneration.[5] Thus, a possible application of decellularised carrot scaffolds could be bone-fillers in non-load-bearing sites.

A 2018 study proposed new micro/macro porous composite scaffolds made from mMS, PCL, and WP using a fast-prototyping technique.[13] Increased surface roughness, hydrophilicity and biodegradability were seen with the addition of WP. The mMS/PCL/WP composite scaffold was non-toxic, promoted MSC proliferation and osteogenic differentiation aided by decreased mechanical strength[13] thus, appearing to be a potential candidate for cell-based bone regeneration.

In a 2014 study, naringin-incorporated electro-spun nanoscaffold containing PCL and PEG-b-PCL were prepared. These nanoscaffolds showed controlled release of naringin, improved the functions of MC3T3-E1 osteoblasts, inhibited the production of osteoclasts, faster degradation losing 55% weight when compared to pure PCL which showed <10% weight-loss during 60 days. This indicated that naringin incorporated electro-spun nanoscaffold containing PEG-b-PCL might be employed as a guided bone regeneration (GBR) scaffold for the repair of osteoporotic bone defects.[12]

In a 2016 study, anti-infective scaffold was fabricated by electro-spinning and ornamented by segmented polyurethane (SPU) with two-dimensional (2D) aloe vera-wrapped mesoporous hydroxyapatite (Al-mHA) nanorods.[4] This Al-mHA composite scaffold showed improved biocompatibility, antibacterial activity, enhanced biodegradability, rapid mineralisation and high osteoconductive capacity appropriate for GBR.[4]

The vhEGCG-GS was prepared by aqueous synthesis technique in a 2018 study, which, when employed as a scaffold in rat congenital cleft jaw model, allowed rat adipose-derived stem cells (rADSCs) and rat dedifferentiated fat (rDFAT) cells to induce ossification,[14] thus showing its potential as effective scaffold material for repair of bone defects via bone tissue engineering.

In a 2022 study, the osteogenic properties of lotus root scaffolds and mineralised lotus root scaffolds were examined both in vitro and in vivo for bone tissue engineering. The lotus root scaffolds exhibited good natural microstructure, but mineralised lotus root scaffolds

showed superior biocompatibility, angiogenesis after 24h, early and greater osteogenic differentiation in vitro, and earlier, greater and superior bone formation after 1-month implantation in vivo. Thus, lotus root-based scaffolds have the ability to regenerate bone because they have an appropriate natural architecture, excellent biocompatibility, particular bioactivities and minimal manufacturing costs.[3]

Despite the fact that more research is needed to confirm the feasibility of in vivo implant of decellularised plants and their integration with the host organism. decellularised plant tissues can sustain cell adhesion, proliferation and functionality, which is critical for the regeneration of selected functional human tissues, implying that different plant tissues could be used for versatile tissue engineering applications. In addition to this, various composite plant-based scaffolds can be prepared with improved mechanical, rheological and bone regeneration potentials via promoting cellular attachment, proliferation, differentiation and enhanced mineralisation.

In the studies analysed, quality assessment was done by measuring various parameters, including plant-based scaffold, fibre diameter, pore size, contact angle, mechanical properties, anti-bacterial potential, biocompatibility and osteogenic potential. Three studies covered most of the parameters.[4] The studies having a bias risk of 4/8 cover the least parameters.[16] All of the studies included in this review had osteogenic potential.

Due to limited availability of tissue donors and other issues with tissue transplantation, such as the risk of virus infection, it has been attempted to construct tissues using a variety of natural and synthetic scaffolds.[18,19] Plants can be used as an alternative to animal and human-harvested decellularised tissues because of their low cost, abundant and accessible availability, and lack of ethical concerns. Plant-based scaffolds include cellulose, which is resistant to degradation by mammalian cells and so has a high durability.[20] They offer superior cellular attachment and proliferation, good water retention and transport, a large surface area, interconnected porosity and pre-existing vascular networks, and thy are not expensive.[20]

The majority of the studies included in this review demonstrated that plant-based scaffolds may be used to promote osteogenesis. However, more work has to be done in vivo before clinical trials on patients may be conducted to employ these plant-based scaffolds for osteogenesis in individuals with various bone deformities, bone resorption, bone malignancies, and patients with periodontitis.

The current systematic review has limitations as it relied on the published studies available on Science Direct and PubMed databases only, and ignored the studies published before 2013 as well as those that did not meet the inclusion criteria. The systematic review also did not meet the international prospective register of systematic reviews (PROSPERO) inclusion criteria and was not eligible for registration with PROSPERO. Majority of the studies included were purely in vitro, and only a few had both in vitro and in vivo components. Finally, it lacked the inclusion of any clinical trials.

Conclusions

The majority of the studies in the current systematic review were conducted in vitro and demonstrated that plant-based scaffolds could be employed to promote osteogenesis. To determine the efficacy of these scaffolds and their involvement in osteogenesis, more in vivo research must be conducted because there has not been much done in this area so far. Future research should investigate additional plant-based materials and their various characteristics in order to synthesise materials more effectively.

Disclaimer: None.

Conflict of Interest: None.

Source of Funding: None.

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