



iJRASET

International Journal For Research in
Applied Science and Engineering Technology



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 8 Issue: XI Month of publication: November 2020

DOI: <https://doi.org/10.22214/ijraset.2020.32351>

www.ijraset.com

Call:  08813907089

E-mail ID: ijraset@gmail.com

Prospects of Polycaprolactone-Based Nanocomposite Porous Scaffolds for Sustained Release of Flavonoids in Bone Tissue Engineering

R. Ranjith¹, M. C. John Milton², B. Jothi Kumar³, D. Elamparithi⁴, V. Moorthy⁵, J. Ganesh⁶, S. Balraj⁷

^{1, 2, 6, 7} P.G. and Research Department of Advanced Zoology and Biotechnology, Loyola College, Chennai, Tamil Nadu, India

³Tamilannai Siddha Maruthuvamanai, Madurai, Tamil Nadu, India

^{4, 5}Department of Biotechnology, Annai College of Arts and Science, Kumbakonam, Tamil Nadu, India

Abstract: Current therapeutic limitations of bone graft substitutes drive the development of multifunctional grafts where the bioactive flavonoids with osteogenic potential can be incorporated into three-dimensional porous bone regeneration scaffolds. The aim of this review is to explore the possibility of fabricating nanocomposite scaffolds incorporated with widely available and non-toxic flavonoid formulation for repair and regeneration of bone tissue. This review summarizes different bone regeneration strategies based on porous scaffolds and provides an overview of the most widely preferred methods for obtaining porous polycaprolactone scaffolds. Further, we intend to outline flavonoids and their classification, and have listed a few of them used for in vitro bone tissue engineering applications. Finally, we aim to show the prospects of flavonoids to be incorporated into nanocomposite porous scaffolds based on polycaprolactone for sustained release of flavonoids to enhance bone regeneration.

Keywords: Nanocomposite scaffolds, Polycaprolactone, Flavonoids, Bone tissue engineering, Drug delivery, Porous scaffolds

I. INTRODUCTION

Bone is a mineralized connective tissue that performs numerous important functions in the body including hematopoiesis, vital organs protection, mineral homeostasis and musculoskeletal mechanical support [1], [2]. The bone's self-renewal capability allows spontaneous scar-free healing [3], [4]. For patients with critical size bone defects, intrinsic bone self-healing ability is limited and invasive surgical procedures are needed to restore the structural integrity of the damaged bone [4]–[6]. Autografts (from the person's own body) and allografts (from human cadavers or living donors) are widely used to treat large bone defects, but they show the following drawbacks, such as donor-site morbidity and limited availability, and the risk of disease transmission [7]–[10]. Metallic implants (joint prostheses, plates and screws) commonly used to provide structural and mechanical assistance for joint arthroplasties and long bone and spine fractures, but these are limited due to non-degradability, poor host tissue integration, high rigidity, extrusion and infection [11]. In order to overcome conventional implant limitations, bone tissue engineering (BTE) has emerged as a promising strategy for bone reconstitution [12], [13]. BTE strategies include either the use of scaffold alone, or a combination of scaffold and bioactive molecules, or a combination of scaffold and harvested cells (stem cells are harvested from various types of tissues), or a combination of scaffold, harvested cells and bioactive molecules, or a combination of scaffold and differentiated cells (differentiated osteoblasts from harvested cells), or a combination of scaffold, differentiated cells and bioactive molecules (Fig. 1) [2], [6], [14]–[18].

II. POLYCAPROLACTONE (PCL): A BIOMATERIAL FOR BTE APPLICATIONS

PCL is synthetic biodegradable aliphatic polyester approved by the Food and Drug Administration, used as scaffolds or as bone tissue supports in medical applications [19]–[21].

III. SYNTHESIS OF PCL

PCL can be synthesized either by polycondensation of 6-hydroxyhexanoic acid (forms low molecular weight PCL) or by radical ring-opening polymerization of 2-methylene-1,3-dioxepane (forms amorphous PCL) or by ring-opening polymerization (ROP) of ϵ -caprolactone (forms high molecular weight PCL with low polydispersity) (Fig. 2) [22]–[24]. High molecular weight PCL is needed for scaffolds and implants to maintain the material's stability for months or years; hence, ROP is the preferred method of synthesizing PCL with high molecular weight and low polydispersity [25], [26]. The ROP process includes four general mechanisms (anionic, cationic, monomer-activated and coordination-insertion), each affecting the resulting molecular weight and its distribution, the end group composition and the copolymer structure [26]. Different catalytic systems, such as metal, organic and enzymatic, support the mechanisms involved in ROP, where stannous (II) 2-ethylhexanoate is the most frequently used catalyst with high efficacy and low toxicity in ROP [20], [23].

IV. PHYSICOCHEMICAL CHARACTERISTICS AND BIODEGRADATION OF PCL

PCL, a biodegradable and biocompatible polymer with a glass transition temperature of around -60°C and a melting point of 59°C – 64°C [27], [28]. At ambient temperature, PCL is soluble in dichloromethane, chloroform, benzene, carbon tetrachloride, toluene, 2-nitropropane and cyclohexanone; slightly soluble in ethyl acetate, acetone, 2-butanone, acetonitrile and dimethylformamide; and insoluble in water, alcohols, diethyl ether and petroleum ether [23]. Depending on molecular weight, degree of crystallinity and conditions of degradation, PCL degrades within several months to several years [20], [29]. From the degradation studies reported in the literature it can be inferred that PCL undergoes a two-stage degradation process, (i) non-enzymatic hydrolytic breakdown of ester bonds in the amorphous region results in mass loss and subsequent increase in crystallinity; (ii) intracellular degradation occurs when the molecular weight of PCL is low (less than 3000) and highly crystalline [20], [30]. The biodegradation of polymer leads to fragmented materials by the action of living organisms until the complete decomposition to carbon dioxide and water [31].

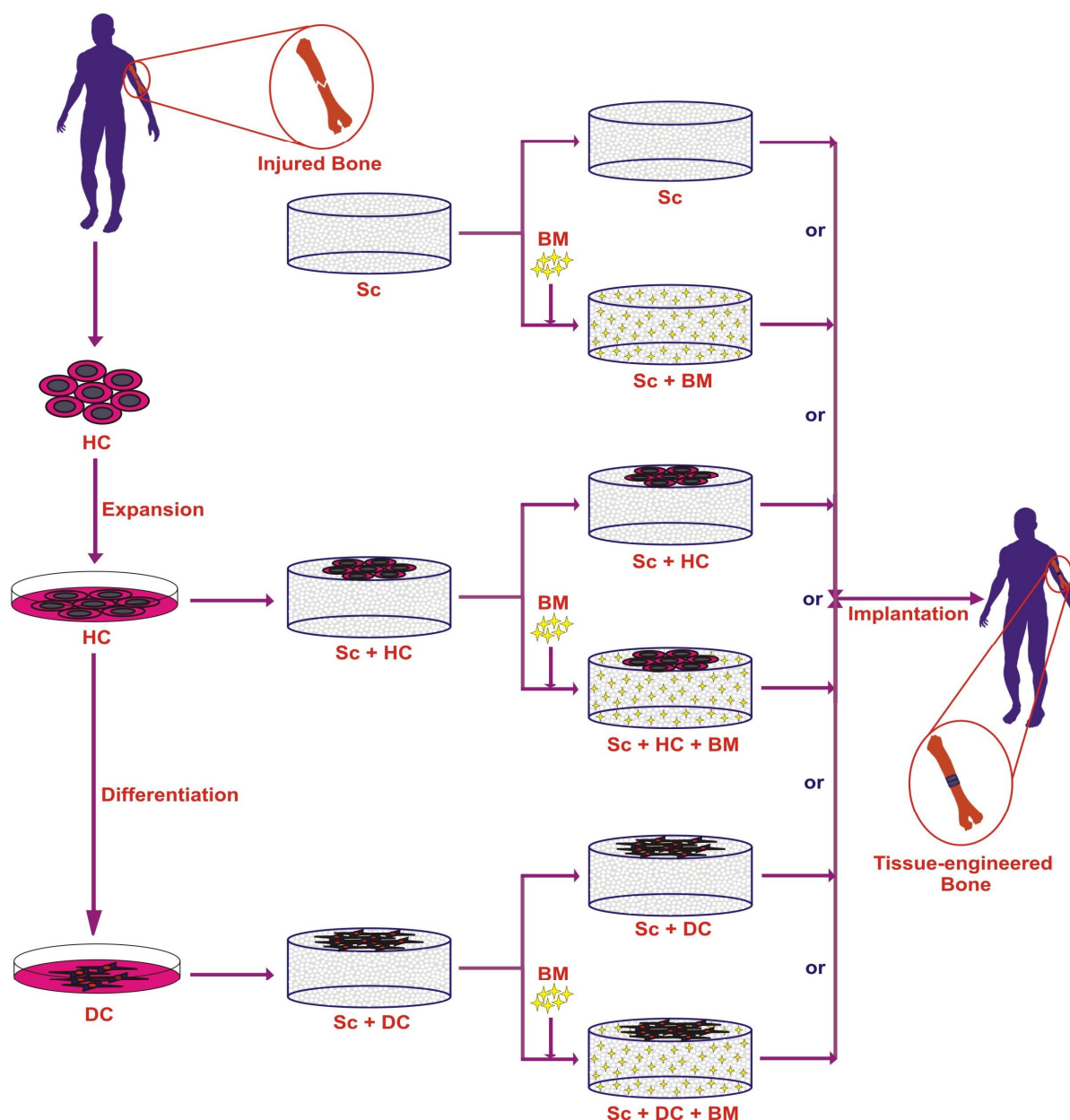


Fig. 1 Schematic illustration of porous scaffold-assisted different bone tissue engineering strategies. Where, Sc – Scaffold; HC – Harvested cells; DC – Differentiated cells; BM – Bioactive molecules.

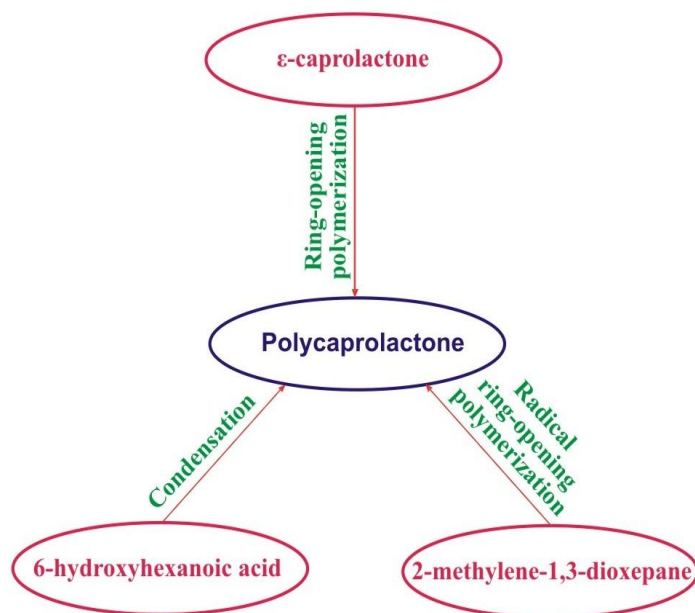


Fig. 2 Schematic showing methods of polycaprolactone synthesis.

V. POROUS PCL SCAFFOLD FABRICATION METHODS

Different methods have been reported for preparing porous PCL scaffolds [32], and Fig. 3 shows few commonly used fabrication methods.

A. Solvent Casting and Particulate Leaching

Solvent casting and particulate leaching is one of the most common techniques used for the preparation of porous PCL scaffolds [33]. This technique consists of four steps, (i) homogeneous dispersion of porogens in a polymer solution; (ii) pouring the mixture obtained into the mold; (iii) removal of the solvent from the solution by evaporation; and (iv) immersion of the polymer/porogen composite into water or a suitable solvent to leach porogens [34]. Scaffold pore size and porosity can be controlled by the size and amount of porogens used [35]–[37]. In addition to its simplicity, some challenges are associated with solvent casting and particulate leaching method, such as the solvent removal mechanism may lead to dimensional shrinkage or solvent trapping inside the samples [34].

B. Gas Foaming and Particulate Leaching

Gas foaming technique allows to fabricate solvent-free porous scaffolds, and pores are formed by gas expansion in a polymer [36], [38]. Carbon dioxide, a low-toxic and non-flammable gas used as a porogen gas [36]. In this method, there are three basic steps, (i) polymer plasticization because of the diffusion of carbon dioxide into the polymer matrix with increasing pressure; (ii) nucleation of gas bubbles due to depressurization and supersaturation; and (iii) nucleation growth because of the diffusion of gas from the surrounding polymer [39]. However, interconnectivity remains limited and is mostly combined with particulate leaching to increase porous interconnectivity [38].

C. Freeze Extraction

Freeze extraction, a simple and energy-efficient process does not require special equipment to prepare the scaffolds and allows modification of certain factors affecting the structure [40], [41]. This process involves three basic steps, (i) pour the polymer solution prepared into a mold and freeze; (ii) immerse the frozen polymer solution into a suitable pre-cooled extractive solvent (a polymer non-solvent) at a temperature equal to or less, to extract the solvent used to dissolve the polymer; (iii) after extraction, dry the scaffold obtained at room temperature to eliminate the extractive solvent [42]–[45]. By modifying the polymer concentration, solvent / non-solvent ratio, freezing temperature and time, and the cooling rate may lead to porous structures with distinctive morphologies adapted to the target application [43], [46], [47].

D. Extrusion-based Cryogenic 3D Printing

Extrusion-based cryogenic 3D printing is considered an advanced 3D printing process for fabricating BTE scaffolds with the capability to deliver biomolecules [48]. There are three basic steps in this method, (i) prepare the printing ink by dissolving the polymer in a suitable solvent and loading it into a syringe; (ii) fix the polymer solution loaded syringe with motor-driven piston in the extrusion chamber to adjust flow rate (BTE scaffolds can be printed and stabilized by adjusting the processing parameters (extrusion velocity, platform speed, cryogenic substrate temperature and nozzle diameter)); (iii) lyophilize the printed scaffold to eliminate the residual solvent and to obtain dried scaffolds [48], [49].

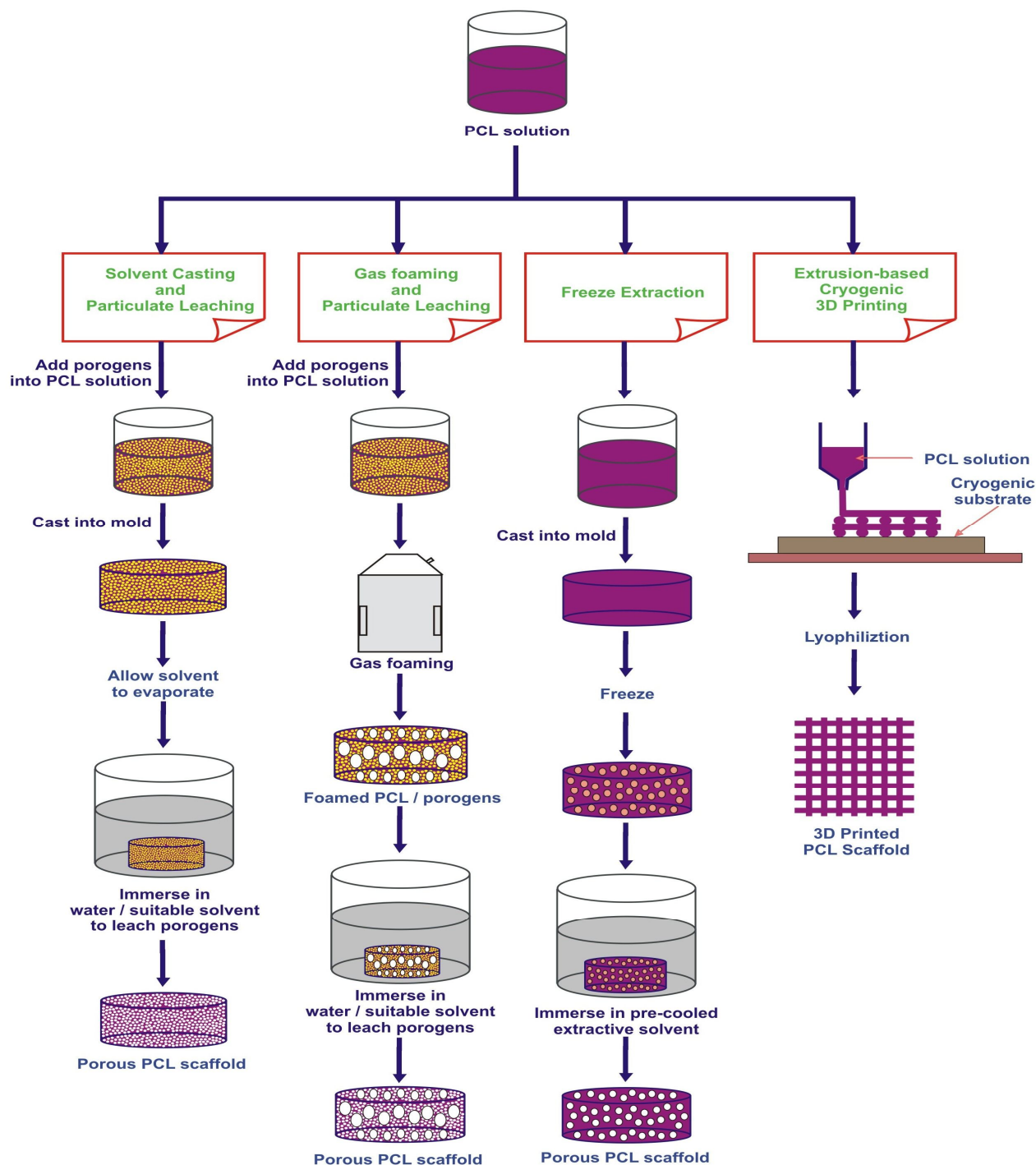


Fig. 3 Scheme of various methods to fabricate porous polycaprolactone scaffolds. Where, PCL – Polycaprolactone.

VI. FLAVONOIDS

Flavonoids are naturally available biologically active compounds that are an excellent source for drug discovery [50], [51]. The basic structure of flavonoids consists of two benzene rings (ring A and ring B), linked by three-carbon-ring (ring C) [52], [53]. Flavonoids are classified into different subclasses (Fig. 4), such as flavanones (naringin, hesperetin and naringenin), flavonols (quercetin, myricetin and kaempferol), flavones (apigenin, luteolin, chrysin and rutin), flavanols (catechin, epicatechin and epigallocatechin), isoflavones (genistein and daidzein) and anthocyanidins (cyanidin and delphinidin) [54]–[56]. Concerns regarding their wide-ranging bioactive benefits including anti-oxidant, anti-inflammatory, anti-viral/bacterial, anti-cancer, cardioprotective, anti-aging, anti-diabetic, have long been gained increasing attention and well-supported by several studies [52], [57], [58]. Table 1 lists a few flavonoids used for *in vitro* BTE applications.

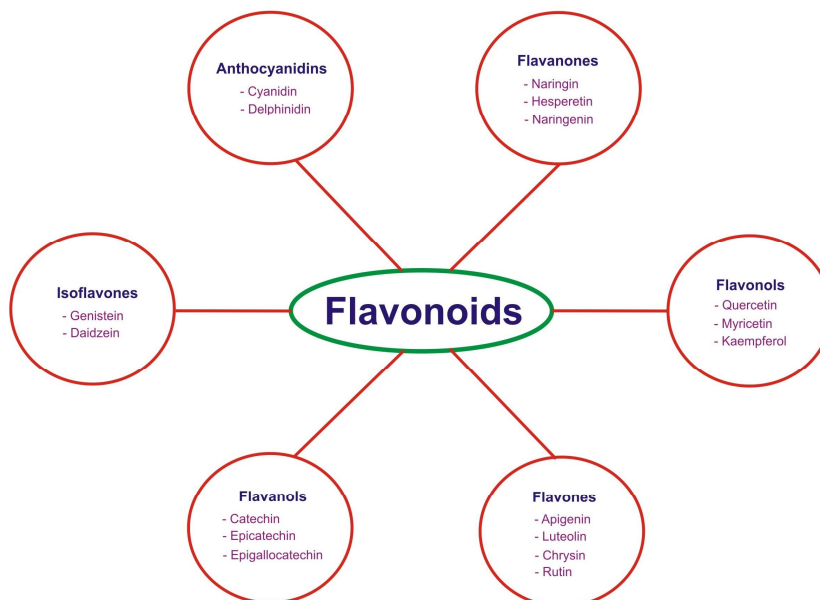


Fig. 4 Scheme showing classes and subclasses of flavonoids.

TABLE I

List Of A Few Flavonoids Used For *In Vitro* Bone Tissue Engineering Applications

Flavonoids	Cell line	References
Naringin	Human amniotic fluid-derived stem cells	[59]
	Human periodontal ligament stem cells	[60]
	Human bone marrow mesenchymal stem cells	[61], [62]
Hesperetin	Primary rat osteoblasts	[63]
	Human mesenchymal stem cells	[64]
Quercetin	Rat bone marrow-derived mesenchymal stem cells	[65]
	human adipose tissue-derived stromal cells	[66]
	Mouse adipose-derived stem cells	[67]
	Mouse bone marrow mesenchymal stem cells	[68]
	Mouse bone marrow derived mesenchymal stem cells	[69]
Myricetin	MG-63, hFOB	[70]
	Human bone marrow stromal cells	[71]
	Human periodontal ligament stem cells	[72]
Kaempferol	Human mesenchymal stromal cells	[73]
	Rat bone marrow stromal cells	[74]
	MC3T3-E1	[75]

Apigenin	Human fetal bone marrow-derived mesenchymal stem cells	[76]
Chrysin	MC3T3-E1	[77]
Rutin	Mouse bone marrow derived mesenchymal stem cells	[69]
	Periodontal ligament stem cells	[78]
Catechin	Human mesenchymal stem cells	[79]
Genistein	MG-63	[80]
	Human bone marrow cells	[81]
	Rat calvarial osteoblasts	[82]
Daidzein	Human mesenchymal stem cells	[83]
	MG-63	[84]

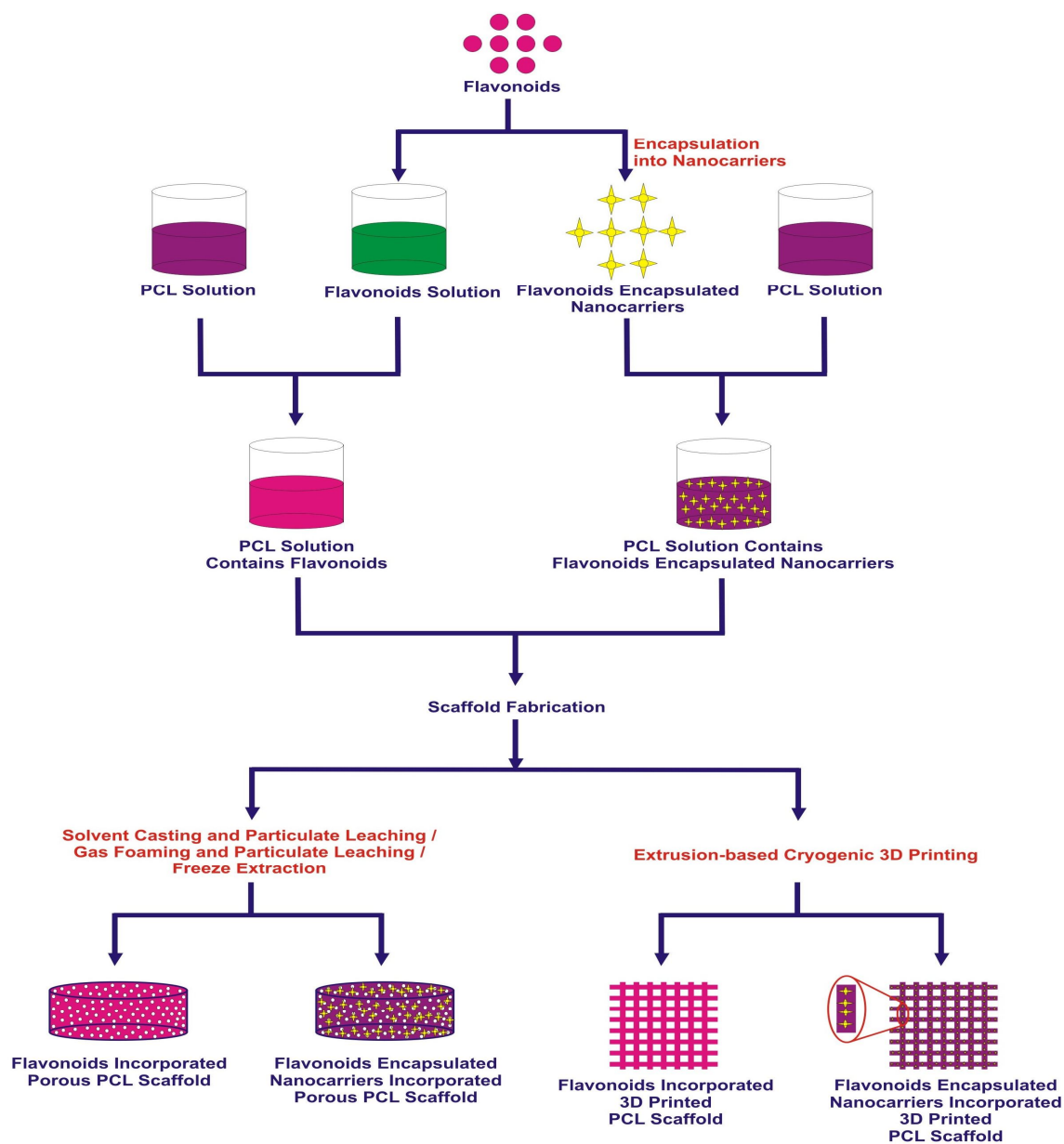


Fig. 5 Schematic showing the prospects of flavonoids to be incorporated into polycaprolactone porous scaffolds for sustained flavonoids release. Where, PCL – Polycaprolactone.

VII. PROSPECTS OF FLAVONOIDS RELEASE FROM POLYCAPROLACTONE-BASED NANOCOMPOSITE POROUS SCAFFOLD

The porous scaffolds fabricated using various methods could provide physical support for proliferation and differentiation of the cells, however, physical support alone is not adequate for ideal BTE applications [85]. A new generation of tissue engineering scaffolds strives to further enhance tissue regeneration by delivering bioactive molecules locally that are essential for natural bone formation [86]. The incorporation of bioactive flavonoids in porous scaffolds enables site-specific controlled drug delivery, which could further enhance bone repair or regeneration efficiency [85], [87]–[89].

Flavonoid delivery methods can rely on the incorporation of flavonoids within the scaffold or the use of nanoparticles as flavonoid reservoirs (Fig. 5) [86]. Drug delivery systems using various types of nanocarriers could improve the bioavailability, solubility, pharmacokinetics and site-specific controlled delivery of the encapsulated drugs [90]–[92]. Nanotechnology approaches in BTE help to overcome some of the limitations associated with inadequate mechanical strength of scaffolds [93]. The incorporation of nanoparticles into scaffolds has shown improvements in the mechanical strength of scaffolds [94], [95]. The encapsulation of flavonoids within nanoparticles, which then get delivered by scaffolds, would allow effective control of their release and achieve the sustained, long-term release profiles required for BTE applications [86], [96], [97].

VIII. CONCLUSION

Designing scaffolds that can mimic the extracellular matrix's three-dimensional structure and deliver bioactive molecules which favorably influence and regulate the cell response is of great importance in BTE applications [98], [99]. The use of porous scaffolds has become a more important aspect for treating bone defects [100]. Porous scaffold based on PCL have been extensively used for BTE applications due to its biodegradability, cost efficacy, tissue compatible and so on [101]. In this review, we highlighted different strategies based on porous scaffolds used for BTE. We also outlined flavonoids and its classification, and listed a few of them used in BTE applications. A growing interest has recently emerged in evaluating the potential of flavonoids to enhance bone repair or regeneration [82]. Further, we have attempted to show the possible ways for sustained flavonoids release from PCL-based nanocomposite porous scaffolds to enhance bone regeneration. The PCL porous scaffold with flavonoids encapsulated nanoparticles could be used in BTE applications as a promising bone grafting material.

REFERENCES

- [1] R. Florencio-Silva, G. R. da S. Sasso, E. Sasso-Cerri, M. J. Simões, and P. S. Cerri, "Biology of Bone Tissue: Structure, Function, and Factors That Influence Bone Cells," *Biomed Res. Int.*, vol. 2015, p. 17 pages, 2015.
- [2] Robert E. Guldberg and Angel O. Duty, "Design parameters for engineering bone regeneration," in *Functional tissue engineering*, Farshid Guilak, David L. Butler, Steven A. Goldstein, and David Mooney, Eds. Springer, 2003, pp. 146–161.
- [3] M. Matsumoto, C. Bigueti, A. Fonseca, and P. Saraiva, "Bone Tissue Healing Dynamics: From Damage to Reconstruction," *J. Mol. Signal. Updat.*, vol. 1, pp. 33–40, 2016.
- [4] R. Shi, Y. Huang, C. Ma, C. Wu, and W. Tian, "Current advances for bone regeneration based on tissue engineering strategies," *Front. Med.*, vol. 13, no. 2, pp. 160–188, 2019.
- [5] A. Roffi, G. S. Krishnakumar, N. Gostynska, E. Kon, C. Candrian, and G. Filardo, "The Role of Three-Dimensional Scaffolds in Treating Long Bone Defects: Evidence from Preclinical and Clinical Literature—A Systematic Review," *Biomed Res. Int.*, vol. 2017, p. 8074178, 2017.
- [6] X. Bai, M. Gao, S. Syed, J. Zhuang, X. Xu, and X.-Q. Zhang, "Bioactive hydrogels for bone regeneration," *Bioact. Mater.*, vol. 3, no. 4, pp. 401–417, 2018.
- [7] A. P. Moreno Madrid, S. M. Vrech, M. A. Sanchez, and A. P. Rodriguez, "Advances in additive manufacturing for bone tissue engineering scaffolds," *Mater. Sci. Eng. C*, vol. 100, pp. 631–644, 2019.
- [8] A. Mansour, M. A. Mezour, Z. Badran, and F. Tamimi, "Extracellular Matrices for Bone Regeneration: A Literature Review," *Tissue Eng. Part A*, 2017, doi: 10.1089/ten.tea.2017.0026.
- [9] H. Xu et al., "Icariin loaded-hollow bioglass/chitosan therapeutic scaffolds promote osteogenic differentiation and bone regeneration," *Chem. Eng. J.*, vol. 354, pp. 285–294, 2018.
- [10] E. J. Sheehy, D. J. Kelly, and F. J. O'Brien, "Biomaterial-based endochondral bone regeneration: a shift from traditional tissue engineering paradigms to developmentally inspired strategies," *Mater. Today Bio*, vol. 3, p. 100009, 2019.
- [11] L. Roseti et al., "Scaffolds for Bone Tissue Engineering: State of the art and new perspectives," *Mater. Sci. Eng. C*, vol. 78, no. Supplement C, pp. 1246–1262, 2017.
- [12] A. De Mori, M. Peña Fernández, G. Blunn, G. Tozzi, and M. Roldo, "3D Printing and Electrospinning of Composite Hydrogels for Cartilage and Bone Tissue Engineering," *Polymers (Basel)*, vol. 10, no. 3, p. 285, 2018.
- [13] J. E. Phillips and A. J. García, "Retroviral-Mediated Gene Therapy for the Differentiation of Primary Cells into a Mineralizing Osteoblastic Phenotype," pp. 333–354, 2008.
- [14] G. Vozzi, C. Corallo, and C. Daraio, "Pressure-activated microsyringe composite scaffold of poly(L-lactic acid) and carbon nanotubes for bone tissue engineering," *J. Appl. Polym. Sci.*, vol. 129, no. 2, pp. 528–536, 2013.
- [15] K. S. Masters and W. L. Murphy, *Tissue Engineering*, Second Edi. John Wiley & Sons, Inc., 2006.

- [16] M. Rodríguez-Vázquez, B. Vega-Ruiz, R. Ramos-Zúñiga, D. A. Saldaña-Koppel, and L. F. Quiñones-Olvera, "Chitosan and Its Potential Use as a Scaffold for Tissue Engineering in Regenerative Medicine," *Biomed Res. Int.*, vol. 2015, p. 15 pages, 2015.
- [17] D. Marolt, M. Knezevic, and G. Vunjak-Novakovic, "Bone tissue engineering with human stem cells," *Stem Cell Res. Ther.*, vol. 1, no. 2, p. 10, 2010.
- [18] J. Ge, M. Zhai, Y. Zhang, J. Bian, and J. Wu, "Biocompatible Fe₃O₄/chitosan scaffolds with high magnetism," *Int. J. Biol. Macromol.*, vol. 128, pp. 406–413, 2019.
- [19] U. Aydemir Sezer, D. Arslantunali, E. A. Aksoy, V. Hasirci, and N. Hasirci, "Poly(ϵ -caprolactone) composite scaffolds loaded with gentamicin-containing β -tricalcium phosphate/gelatin microspheres for bone tissue engineering applications," *J. Appl. Polym. Sci.*, vol. 131, no. 8, p. 40110, 2014.
- [20] D. Mondal, M. Griffith, and S. S. Venkatraman, "Polycaprolactone-based biomaterials for tissue engineering and drug delivery: Current scenario and challenges," *Int. J. Polym. Mater. Polym. Biomater.*, vol. 65, no. 5, pp. 255–265, 2016.
- [21] F. Mohanty and S. K. Swain, "Chapter 18 - Bionanocomposites for Food Packaging Applications," A. E. Oprea and A. M. B. T.-N. A. in F. Grumezescu, Eds. Academic Press, 2017, pp. 363–379.
- [22] S. Mallakpour and N. Nouruzi, "Polycaprolactone/metal oxide nanocomposites: an overview of recent progress and applications," in *Biodegradable and Biocompatible Polymer Composites: Processing, Properties and Applications*, N. G. Shimpi, Ed. Woodhead Publishing, 2017, pp. 223–255.
- [23] M. Labet and W. Thielemans, "Synthesis of polycaprolactone: a review," *Chem. Soc. Rev.*, vol. 38, no. 12, pp. 3484–3504, 2009.
- [24] V. Guarino, G. Gentile, L. Sorrentino, and L. Ambrosio, "Polycaprolactone: Synthesis, Properties, and Applications," *Encyclopedia of Polymer Science and Technology*, pp. 1–36, 2017.
- [25] M. Contardi et al., "Low molecular weight ϵ -caprolactone-p-coumaric acid copolymers as potential biomaterials for skin regeneration applications," *PLoS One*, vol. 14, no. 4, p. e0214956, 2019.
- [26] N. Siddiqui, S. Asawa, B. Birru, R. Baadhe, and S. Rao, "PCL-Based Composite Scaffold Matrices for Tissue Engineering Applications," *Mol. Biotechnol.*, vol. 60, no. 7, pp. 506–532, 2018.
- [27] P. A. Gunatillake and R. Adhikari, "Biodegradable synthetic polymers for tissue engineering," *Eur. Cells Mater.*, vol. 5, pp. 1–16, 2003.
- [28] B. Azimi, P. Nourpanah, M. Rabiee, and S. Arbab, "Poly (ϵ -caprolactone) Fiber: An Overview," *J. Eng. Fiber. Fabr.*, vol. 9, no. 3, pp. 74–90, 2014.
- [29] R. A. Gross and B. Kalra, "Biodegradable Polymers for the Environment," *Science (80-.)*, vol. 297, no. 5582, pp. 803–807, 2002.
- [30] M. A. Woodruff and D. W. Hutmacher, "The return of a forgotten polymer—Polycaprolactone in the 21st century," *Prog. Polym. Sci.*, vol. 35, no. 10, pp. 1217–1256, 2010.
- [31] A. Heimowska, M. Morawska, and A. Bocho-Janiszewska, "Biodegradation of poly(ϵ -caprolactone) in natural water environments," *Polish J. Chem. Technol.*, vol. 19, no. 1, pp. 120–126, 2017.
- [32] N. Thadavirul, P. Pavasant, and P. Supaphol, "Development of polycaprolactone porous scaffolds by combining solvent casting, particulate leaching, and polymer leaching techniques for bone tissue engineering," *J. Biomed. Mater. Res. Part A*, vol. 102, no. 10, pp. 3379–3392, 2014.
- [33] Q. Yang, L. Chen, X. Shen, and Z. Tan, "Preparation of Polycaprolactone Tissue Engineering Scaffolds by Improved Solvent Casting/Particulate Leaching Method," *J. Macromol. Sci. Part B*, vol. 45, no. 6, pp. 1171–1181, 2006.
- [34] S. Taherkhani and F. Moztaazadeh, "Fabrication of a poly(ϵ -caprolactone)/starch nanocomposite scaffold with a solvent-casting/salt-leaching technique for bone tissue engineering applications," *J. Appl. Polym. Sci.*, vol. 133, no. 23, p. 43523, 2016.
- [35] J. F. Mano et al., "Natural origin biodegradable systems in tissue engineering and regenerative medicine: present status and some moving trends," *J. R. Soc. Interface*, vol. 4, no. 17, pp. 999–1030, 2007.
- [36] P. Chocholata, V. Kulda, and V. Babuska, "Fabrication of Scaffolds for Bone-Tissue Regeneration," *Materials (Basel)*, vol. 12, p. 568, 2019.
- [37] Q. L. Loh and C. Choong, "Three-Dimensional Scaffolds for Tissue Engineering Applications: Role of Porosity and Pore Size," *Tissue Eng. Part B. Rev.*, vol. 19, no. 6, pp. 485–502, 2013.
- [38] D. W. Hutmacher, T. B. F. Woodfield, and P. D. Dalton, "Chapter 10 - Scaffold Design and Fabrication," in *Tissue Engineering*, Second ed., C. A. Van Blitterswijk and J. B. T. De Boer, Eds. Oxford: Academic Press, 2014, pp. 311–346.
- [39] N. Annabi, A. Fathi, S. M. Mithieux, A. S. Weiss, and F. Dehghani, "Fabrication of porous PCL/elastin composite scaffolds for tissue engineering applications," *J. Supercrit. Fluids*, vol. 59, pp. 157–167, 2011.
- [40] S. I. A. Razak et al., "A Conductive polylactic acid/polyaniline porous scaffold via freeze extraction for potential biomedical applications," *Soft Mater.*, vol. 14, no. 2, pp. 78–86, 2016.
- [41] M. Budnicka, D. Kołbuk, P. Ruśkowski, and A. Gadomska-Gajadur, "Poly-L-lactide scaffolds with super pores obtained by freeze-extraction method," *J. Biomed. Mater. Res. Part B Appl. Biomater.*, 2020, doi: 10.1002/jbm.b.34642.
- [42] A. R. Sarasam, A. I. Samli, L. Hess, M. A. Ihnat, and S. V. Madihally, "Blending Chitosan with Polycaprolactone: Porous Scaffolds and Toxicity," *Macromol. Biosci.*, vol. 7, no. 9-10, pp. 1160–1167, 2007.
- [43] R. Akbarzadeh and A.-M. Yousefi, "Effects of Processing Parameters in Thermally Induced Phase Separation Technique on Porous Architecture of Scaffolds for Bone Tissue Engineering," *J. Biomed. Mater. Res. Part B Appl. Biomater.*, vol. 102, no. 6, pp. 1304–1315, 2014.
- [44] M.-H. Ho et al., "Preparation of porous scaffolds by using freeze-extraction and freeze-gelation methods," *Biomaterials*, vol. 25, no. 1, pp. 129–138, 2004.
- [45] Ming-Hua Ho, Da-Ming Wang, Hsyue-Jen Hsieh, and Juin-Yih Lai, "Preparation of Polylactide Scaffolds," in *Macroporous Polymers: Production Properties and Biotechnological/Biomedical Applications*, Bo Mattiasson, Ashok Kumar, and Igor Yu. Galaev, Eds. 2009, pp. 117–127.
- [46] B. Thavorniyutikarn, N. Chantarapanich, K. Sithiseripratip, G. A. Thouas, and Q. Chen, "Bone tissue engineering scaffolding: computer-aided scaffolding techniques," *Prog. Biomater.*, vol. 3, no. 2, pp. 61–102, 2014.
- [47] N. Sultana and T. H. Khan, "Polycaprolactone Scaffolds and Hydroxyapatite/Polycaprolactone Composite Scaffolds for Bone Tissue Engineering," *J. Bionanosci.*, vol. 7, pp. 169–173, 2013.
- [48] C. Wang et al., "3D printing of bone tissue engineering scaffolds," *Bioact. Mater.*, vol. 5, no. 1, pp. 82–91, 2020.
- [49] W. Zhang et al., "Fabrication and characterization of porous polycaprolactone scaffold via extrusion-based cryogenic 3D printing for tissue engineering," *Mater. Des.*, vol. 180, p. 107946, 2019.
- [50] A. Tawani and A. Kumar, "Structural Insight into the interaction of Flavonoids with Human Telomeric Sequence," *Sci. Rep.*, vol. 5, p. 17574, 2015, <http://dx.doi.org/10.1038/srep17574>.

- [51] G. Hussain et al., "Role of Plant-Derived Flavonoids and Their Mechanism in Attenuation of Alzheimer's and Parkinson's Diseases: An Update of Recent Data," *Molecules*, vol. 23, no. 4, 2018, doi: 10.3390/molecules23040814.
- [52] T. Wang, Q. Li, and K. Bi, "Bioactive flavonoids in medicinal plants: Structure, activity and biological fate," *Asian J. Pharm. Sci.*, vol. 13, no. 1, pp. 12–23, 2018.
- [53] H. Khan et al., "Evidence and prospective of plant derived flavonoids as antiplatelet agents: Strong candidates to be drugs of future," *Food Chem. Toxicol.*, 2018, doi: 10.1016/j.fct.2018.02.014.
- [54] S. Kumar and A. K. Pandey, "Chemistry and biological activities of flavonoids: an overview.," *Sci. World J.*, vol. 2013, p. 16 pages, 2013.
- [55] A. N. Panche, A. D. Diwan, and S. R. Chandra, "Flavonoids: an overview," *J. Nutr. Sci.*, vol. 5, p. e47, 2016.
- [56] J. Čvorović, L. Zibera, S. Fornasaro, F. Tramer, and S. Passamonti, "Chapter 22 - Bioavailability of Flavonoids: The Role of Cell Membrane Transporters," in *Polyphenols: Mechanisms of Action in Human Health and Disease (Second Edition)*, R. R. Watson, V. R. Preedy, and S. B. T.-P. M. of A. in H. H. and D. (Second E. Zibadi, Eds. Academic Press, 2018, pp. 295–320.
- [57] H. Isoda, H. Motojima, S. Onaga, I. Samet, M. O. Villareal, and J. Han, "Analysis of the erythroid differentiation effect of flavonoid apigenin on K562 human chronic leukemia cells," *Chem. Biol. Interact.*, vol. 220, pp. 269–277, 2014.
- [58] R. Chen, Q.-L. Qi, M.-T. Wang, and Q.-Y. Li, "Therapeutic potential of naringin: an overview," *Pharm. Biol.*, vol. 54, no. 12, pp. 3203–3210, 2016.
- [59] M. Liu, Y. Li, and S.-T. Yang, "Effects of naringin on the proliferation and osteogenic differentiation of human amniotic fluid-derived stem cells," *J. Tissue Eng. Regen. Med.*, vol. 11, no. 1, pp. 276–284, 2017.
- [60] L. Yin et al., "Effects of Naringin on Proliferation and Osteogenic Differentiation of Human Periodontal Ligament Stem Cells In Vitro and In Vivo.," *Stem Cells Int.*, vol. 2015, p. 9 pages, 2015.
- [61] H. Wang et al., "Naringin enhances osteogenic differentiation through the activation of ERK signaling in human bone marrow mesenchymal stem cells," *Iran. J. Basic Med. Sci.*, vol. 20, no. 4, pp. 408–414, 2017.
- [62] Peng-Zhang et al., "Effects of naringin on the proliferation and osteogenic differentiation of human bone mesenchymal stem cell," *Eur. J. Pharmacol.*, vol. 607, no. 1–3, pp. 1–5, 2009.
- [63] A. Trzeciakiewicz et al., "Hesperetin stimulates differentiation of primary rat osteoblasts involving the BMP signalling pathway," *J. Nutr. Biochem.*, vol. 21, no. 5, pp. 424–431, 2010.
- [64] D. Xue et al., "The role of hesperetin on osteogenesis of human mesenchymal stem cells and its function in bone regeneration," *Oncotarget*, vol. 8, no. 13, pp. 21031–21043, 2017.
- [65] Y. Zhou et al., "The Effect of Quercetin on the Osteogenesis Differentiation and Angiogenic Factor Expression of Bone Marrow-Derived Mesenchymal Stem Cells," *PLoS One*, vol. 10, no. 6, p. e0129605, 2015.
- [66] Y. J. Kim, Y. C. Bae, K. T. Suh, and J. S. Jung, "Quercetin, a flavonoid, inhibits proliferation and increases osteogenic differentiation in human adipose stromal cells," *Biochem. Pharmacol.*, vol. 72, no. 10, pp. 1268–1278, 2006.
- [67] C. Zhou and Y. Lin, "Osteogenic differentiation of adipose-derived stem cells promoted by quercetin," *Cell Prolif.*, vol. 47, no. 2, pp. 124–132, 2014.
- [68] X.-G. Pang, Y. Cong, N.-R. Bao, Y.-G. Li, and J.-N. Zhao, "Quercetin Stimulates Bone Marrow Mesenchymal Stem Cell Differentiation through an Estrogen Receptor-Mediated Pathway," *Biomed Res. Int.*, vol. 2018, p. 4178021, 2018.
- [69] S. Srivastava, R. Bankar, and P. Roy, "Assessment of the role of flavonoids for inducing osteoblast differentiation in isolated mouse bone marrow derived mesenchymal stem cells," *Phytomedicine*, vol. 20, no. 8, pp. 683–690, 2013.
- [70] Y.-L. Hsu, J.-K. Chang, C.-H. Tsai, T.-T. C. Chien, and P.-L. Kuo, "Myricetin induces human osteoblast differentiation through bone morphogenetic protein-2/p38 mitogen-activated protein kinase pathway," *Biochem. Pharmacol.*, vol. 73, no. 4, pp. 504–514, 2007.
- [71] X. Ying et al., "Myricetin enhances osteogenic differentiation through the activation of canonical Wnt/ β -catenin signaling in human bone marrow stromal cells," *Eur. J. Pharmacol.*, vol. 738, pp. 22–30, 2014.
- [72] H.-Y. Kim, S.-Y. Park, and S.-Y. Choung, "Enhancing effects of myricetin on the osteogenic differentiation of human periodontal ligament stem cells via BMP-2/Smad and ERK/JNK/p38 mitogen-activated protein kinase signaling pathway," *Eur. J. Pharmacol.*, vol. 834, pp. 84–91, 2018.
- [73] T. Mazaki et al., "In vitro and in vivo enhanced osteogenesis by kaempferol found by a high-throughput assay using human mesenchymal stromal cells," *J. Funct. Foods*, vol. 6, pp. 241–247, 2014.
- [74] S. Tsuchiya et al., "Kaempferol-immobilized titanium dioxide promotes formation of new bone: effects of loading methods on bone marrow stromal cell differentiation in vivo and in vitro," *Int. J. Nanomedicine*, vol. 13, pp. 1665–1676, 2018.
- [75] Y. Wang, H. Chen, and H. Zhang, "Kaempferol promotes proliferation, migration and differentiation of MC3T3-E1 cells via up-regulation of microRNA-101," *Artif. Cells, Nanomedicine, Biotechnol.*, vol. 47, no. 1, pp. 1050–1056, 2019.
- [76] X. Zhang et al., "Apigenin promotes osteogenic differentiation of human mesenchymal stem cells through JNK and p38 MAPK pathways," *Mol. Cell. Biochem.*, vol. 407, no. 1, pp. 41–50, 2015.
- [77] W. Zeng, Y. Yan, F. Zhang, C. Zhang, and W. Liang, "Chrysin promotes osteogenic differentiation via ERK/MAPK activation," *Protein Cell*, vol. 4, no. 7, pp. 539–547, 2013.
- [78] B. Zhao, Y. Xiong, Y. Zhang, L. Jia, W. Zhang, and X. Xu, "Rutin promotes osteogenic differentiation of periodontal ligament stem cells through the GPR30-mediated PI3K/AKT/mTOR signaling pathway," *Exp. Biol. Med.*, vol. 245, no. 6, pp. 552–561, 2020.
- [79] Y. J. Wei et al., "Catechin stimulates osteogenesis by enhancing PP2A activity in human mesenchymal stem cells," *Osteoporos. Int.*, vol. 22, no. 5, pp. 1469–1479, 2011.
- [80] C. Morris, J. Thorpe, L. Ambrosio, and M. Santin, "The Soybean Isoflavone Genistein Induces Differentiation of MG63 Human Osteosarcoma Osteoblasts," *J. Nutr.*, vol. 136, no. 5, pp. 1166–1170, 2006.
- [81] N. Okumura, T. Yoshikawa, J. Iida, A. Nonomura, and Y. Takakura, "Osteogenic Effect of Genistein on In Vitro Bone Formation by Human Bone Marrow Cell Culture - For Development of Advanced Bio-Artificial Bone," *Key Eng. Mater.*, vol. 284–286, pp. 667–670, 2005.
- [82] H.-P. Ma et al., "Icariin is more potent than genistein in promoting osteoblast differentiation and mineralization in vitro," *J. Cell. Biochem.*, vol. 112, no. 3, pp. 916–923, 2011.

- [83] A. L. Strong et al., "Design, Synthesis, and Osteogenic Activity of Daidzein Analogs on Human Mesenchymal Stem Cells," ACS Med. Chem. Lett., vol. 5, no. 2, pp. 143–148, 2014.
- [84] X. Jin et al., "Daidzein stimulates osteogenesis facilitating proliferation, differentiation, and antiapoptosis in human osteoblast-like MG-63 cells via estrogen receptor-dependent MEK/ERK and PI3K/Akt activation," Nutr. Res., vol. 42, pp. 20–30, 2017.
- [85] B. Guo, B. Lei, P. Li, and P. X. Ma, "Functionalized scaffolds to enhance tissue regeneration," Regen. Biomater., vol. 2, no. 1, pp. 47–57, 2015.
- [86] T.-M. De Witte, L. E. Fratila-Apachitei, A. A. Zadpoor, and N. A. Peppas, "Bone tissue engineering via growth factor delivery: from scaffolds to complex matrices," Regen. Biomater., pp. 197–211, 2018.
- [87] B. Sundaram and M. C. John Milton, "Porous Polycaprolactone Scaffold Engineered with Naringin Loaded Bovine Serum Albumin Nanoparticles for Bone Tissue Engineering," Biosci. Biotechnol. Res. Asia, vol. 14, no. 4, pp. 1355–1362, 2017.
- [88] K. S. Washington and C. A. Bashur, "Delivery of Antioxidant and Anti-inflammatory Agents for Tissue Engineered Vascular Grafts," Frontiers in Pharmacology, vol. 8, p. 659, 2017.
- [89] A. I. Yaremenko et al., "Prospectives for using artificial scaffolds in oral and craniofacial surgery: literature review," Cell. Ther. Transplant., vol. 7, no. 1, pp. 21–27, 2018.
- [90] A. Z. Wilczewska, K. Niemirowicz, K. H. Markiewicz, and H. Car, "Nanoparticles as drug delivery systems," Pharmacol. Reports, vol. 64, no. 5, pp. 1020–1037, 2012.
- [91] F. ud Din et al., "Effective use of nanocarriers as drug delivery systems for the treatment of selected tumors," Int. J. Nanomedicine, vol. 12, pp. 7291–7309, 2017.
- [92] S. Vieira, S. Vial, R. L. Reis, and J. M. Oliveira, "Nanoparticles for bone tissue engineering," Biotechnol. Prog., vol. 33, no. 3, pp. 590–611, 2017.
- [93] P. Jayaraman, C. Gandhimathi, J. R. Venugopal, D. L. Becker, S. Ramakrishna, and D. K. Srinivasan, "Controlled release of drugs in electrosprayed nanoparticles for bone tissue engineering," Adv. Drug Deliv. Rev., vol. 94, pp. 77–95, 2015.
- [94] J. Corona-Gomez, X. Chen, and Q. Yang, "Effect of Nanoparticle Incorporation and Surface Coating on Mechanical Properties of Bone Scaffolds: A Brief Review," Journal of Functional Biomaterials, vol. 7, no. 3, p. 18, 2016.
- [95] S. Ullah, I. Zainol, and R. H. Idrus, "Incorporation of zinc oxide nanoparticles into chitosan-collagen 3D porous scaffolds: Effect on morphology, mechanical properties and cytocompatibility of 3D porous scaffolds," Int. J. Biol. Macromol., vol. 104, no. Part A, pp. 1020–1029, 2017.
- [96] Z. Wang et al., "BMP-2 encapsulated polysaccharide nanoparticle modified biphasic calcium phosphate scaffolds for bone tissue regeneration," J. Biomed. Mater. Res. A, vol. 103, no. 4, pp. 1520–1532, 2015.
- [97] S. Zhang et al., "Polyethylenimine-coated albumin nanoparticles for BMP-2 delivery," Biotechnol. Prog., vol. 24, no. 4, pp. 945–956, 2008.
- [98] S. Azizian, A. Hadjizadeh, and H. Niknejad, "Chitosan-gelatin porous scaffold incorporated with Chitosan nanoparticles for growth factor delivery in tissue engineering," Carbohydr. Polym., vol. 202, pp. 315–322, 2018.
- [99] M. Ansari, "Bone tissue regeneration: biology, strategies and interface studies," Prog. Biomater., vol. 8, no. 4, pp. 223–237, 2019.
- [100] Y. Zhang et al., "Polymer Fiber Scaffolds for Bone and Cartilage Tissue Engineering," Adv. Funct. Mater., vol. 29, no. 36, p. 1903279, 2019.
- [101] R. Dwivedi et al., "Polycaprolactone as biomaterial for bone scaffolds: Review of literature," J. Oral Biol. Craniofacial Res., vol. 10, no. 1, pp. 381–388, 2020.



10.22214/IJRASET



45.98



IMPACT FACTOR:
7.129



IMPACT FACTOR:
7.429



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Call : 08813907089  (24*7 Support on Whatsapp)