Review
ISSN 2413-0516

Regenerative Cardiology: Emerging Hopes Towards Restoring the Heart

Fatemeh Ghanbari[1,2], Nasim Naderi[3], Shahram Rabbani[2], Ali Vasheghani Farahani[4], Hossein Ghanbari[2,5*]

[1]School of Medicine, Tehran University of Medical Sciences, Tehran, Iran.
[2]Research Center for Advanced Technologies in Cardiovascular Medicine, Cardiovascular Diseases Research Institute, Tehran University of Medical Sciences, Tehran, Iran.
[3]Department of Heart Failure and Transplantation, Rajaei Cardiovascular Medical and Research Center, Iran University of Medical Sciences, Tehran, Iran.
[4]Cardiac Primary Prevention Research Center, Cardiovascular Diseases Research Institute, Tehran University of Medical Sciences, Tehran, Iran.
[5]Regenerative Nanomedicine Research Group, Department of Medical Nanotechnology, School of Advanced Medical Technologies in Medicine, Tehran University of Medical Sciences, Tehran, Iran.
*Correspondence to: Hossein Ghanbari (E-mail: hghanbari@tums.ac.ir)
(Submitted: 20 March 2025 – Revised version received: 02 April 2025 – Accepted: 30 April 2025 – Published online: 26 June 2025)

Abstract
Cardiovascular diseases, particularly myocardial infarction and heart failure, continue to affect the lives of millions and pose significant global health challenges. Despite all efforts, a definitive cure to prevent the progression of heart disease has remained elusive, and all current therapies fail to address the root issue: the negligible regenerative capacity of the adult human heart. For over twenty years, regenerative cardiology has sought to overcome this deficit by taking cues from naturally regenerating species such as zebrafish, and by integrating diverse therapeutic modalities—including cell-based therapies, cell-free and direct reprogramming approaches, biomaterial-assisted tissue engineering, and others—to restore myocardial structure and function following injury. This review aims to provide an overview of the latest progress in the field with particular attention to the potential advantages and the main challenges and limitations of the emerging regenerative strategies. While significant challenges remain, continued interdisciplinary research holds great promise for translating regenerative approaches into available clinical practice for ischemic heart disease and heart failure.
Keywords: Regenerative cardiology, cell therapy, cell-free therapies, cardiac patch, myocardial infarction

Introduction

Affecting approximately 1 in 13 individuals and claiming 20.5 million lives in 2021 alone, cardiovascular diseases (CVD) remain the leading cause of mortality worldwide.[1,2] This trend is projected to continue over the coming decades, driven by an aging global population and persistent prevalence of risk factors.[3,4] Among the various forms of CVD, ischemic heart disease (IHD) and myocardial infarction (MI), in particular, are the most lethal and, when considering the costs of healthcare services, medications, and lost productivity due to death, impose a significant economic burden on healthcare systems worldwide.[5–7]

Myocardial infarction, by definition, results from an acute imbalance in oxygen supply, most commonly due to atherosclerotic blockage of the coronary arteries, which leads to widespread myocardial damage and cardiomyocyte (CM) necrosis. The extent of this damage determines the severity of MI. It may cause sudden reductions in cardiac output and life-threatening arrhythmias that result in sudden cardiac death, and nonlethal cases can lead to chronically impaired cardiac function and eventual heart failure.[8–10] This deterioration is mainly due to the heart's limited regenerative capacity and the irreversibility of CM death, prompting the infarcted heart to undergo complex structural and histological changes through two main processes: (a) the formation of fibrotic scar tissue in both the damaged and surrounding healthy myocardium (cardiac fibrosis), and (b) hypertrophic and dilatory changes in heart morphology (cardiac remodelling). While these pathophysiological processes are initially compensatory, they ultimately establish a vicious cycle of mechanical stress and neurohormonal activation, resulting in progressive cardiac dysfunction.[11–14]

Initial management of MI aims to preserve the at-risk myocardium by focusing on rapid diagnosis and timely restoration of blood flow via prompt reperfusion with strategies such as percutaneous coronary intervention (PCI) alongside pharmacotherapy.[15] These interventions restore perfusion, limit infarct size, and successfully alleviate acute symptoms; however, they fail to regenerate lost myocardium. As a result, despite a reduction in short-term mortality, the long-term incidence of heart failure is, in turn, elevated.[16,17]

Chronic management of heart failure has also successfully improved patient symptoms and reduced hospital admissions, yet current medical therapies fail to address the underlying cause of myocardial dysfunction. With a five-year survival rate of just 50%, the long-term prognosis for heart failure remains poor.[18–20] Left ventricular assist devices (LVADs) can postpone the need for heart transplantation, but serious risks such as infection, bleeding, arrhythmia, and thrombosis complicate their use.[21] Ultimately, heart transplantation is the gold standard and only definitive treatment for end-stage heart failure; however, limitations such as donor shortages, transplant rejection, and the adverse effects of long-term immunosuppression have made this option inaccessible for many patients.[19,20]

With these shortcomings, developing practical therapeutic strategies that address the root cause of cardiac dysfunction—namely, the loss of viable myocardium—is becoming more and more necessary. Regenerative cardiology began over the last 20 years on the backbone of landmark findings such as the discovery of the natural regenerative capacity of zebrafish hearts and the adult mammalian heart's limited regenerative capacity, the development of stem cell-derived CMs, and the identification of molecular pathways that regulate CM proliferation. This review aims to present a comprehensive overview of the current landscape of regenerative cardiology and its main field of research, alongside its limitations and future potential for clinical applications.

Endogenous Regeneration: Potential for New Therapies

Natural Regenerative Capacity of the Human Heart

More than two decades ago, the discovery of the zebrafish heart's regenerative ability ushered in a wave of cardiac regeneration research. Since then, numerous species have been evaluated for their heart's regenerative potential.[22] Zebrafish can fully regenerate their heart without scar formation.[23] Similarly, certain amphibians, such as salamanders and newts, boast a lifelong ability to regenerate damaged myocardium and can survive the removal of up to one-quarter of a cardiac ventricle.[24] Alongside these findings, two other landmark discoveries in the early 2010s in mammalian cardiac regeneration, explained in the following, laid a theoretical foundation for further exploring the regenerative capacity of the adult human heart.[25]

In 2011, it was shown that immature mammalian hearts, similar to zebrafish, possess a transient regenerative ability via cardiomyocyte proliferation. Neonatal mice can fully regenerate up to 15% of the left ventricle following resection, with minimal hypertrophy or fibrosis, hallmarks of the impaired reparative response observed in the adult heart. However, this regenerative ability is rapidly lost within the first week of life, coinciding with cardiomyocyte cell cycle arrest. From this point onward, heart growth occurs predominantly through cardiomyocyte hypertrophy rather than hyperplasia.[26]

Around the same time, researchers discovered that the adult mammalian heart is not strictly a post-mitotic organ, as was previously believed; instead, it retains a small but measurable capacity for cardiomyocyte renewal.[27] Quantifying the cardiomyocyte turnover rate in humans has been challenging due to the lack of direct *in vivo* methods to measure cell division.[28] Nonetheless, using retrospective radiocarbon birth dating, it has been estimated that roughly 1% of cardiomyocytes are renewed annually at age 25, with the rate declining to around 0.3% by age 75.[29] The origin of these new cardiomyocytes remains debated, with proposed possibilities including the division of pre-existing CMs, differentiation of resident cardiac stem cells, or the migration and differentiation of bone marrow-derived stem cells.[30] Understanding the mechanisms underlying effective regeneration is essential for developing a clinically successful strategy for cardiac repair.

Hallmarks of Effective Cardiac Regeneration

A cornerstone of cardiac regeneration is the restoration of lost cardiomyocytes. In regenerative models like zebrafish and neonatal mice, this is achieved predominantly through the dedifferentiation and proliferation of pre-existing cardiomyocytes. Notably, cardiac stem cells do not appear to contribute directly to this process. However, cardiomyocyte replacement alone is insufficient for complete functional recovery. Effective regeneration requires the coordinated participation of various cell populations—including endothelial cells, epicardial-derived cells, and immune cells—which collectively regulate neovascularisation, extracellular matrix remodelling, and inflammation resolution and provide a supportive environment for CM proliferation.[31,32]

While these mechanisms are consistently observed in regenerative species, they are largely altered or absent in the adult mammalian heart.[32] As such, they provide not only therapeutic targets but also a valuable framework for evaluating the success of regenerative interventions. To address the lack of standard criteria in assessing these therapies, Bertero and Murry (2018) proposed five "hallmarks of cardiac regeneration": remuscularisation, electromechanical stability, angiogenesis and arteriogenesis, resolution of fibrosis, and immunological balance.[33] These hallmarks offer a conceptual and functional blueprint for both designing and assessing regenerative strategies. Table 1 provides an overview of their definitions and key clinical considerations.

Novel Therapeutic Approaches to Cardiac Regeneration

With the five hallmarks of effective regeneration in mind, several therapeutic strategies have emerged that aim to restore cardiac function in various ways. These approaches can be broadly categorised into these major areas:

1. Cell-based therapies: These methods aim to induce regeneration via transplantation of an exogenous supply of cells that are either pluripotent or multipotent stem cells, cardiac progenitor cells, or cardiomyocytes.[34]
2. Cell-free therapies and *in situ* regeneration: Intending to regenerate new cardiomyocytes directly within the infarcted myocardium while avoiding exogenous cell delivery, methods harnessing paracrine signalling by administering bioactive molecules have gained traction.[35] Another cell-free approach is direct cardiac reprogramming or *in situ* regeneration, which involves converting fibroblasts into functional cardiomyocyte-like cells by introducing transcription factors or other reprogramming agents.
3. Tissue engineering and biomaterial-based therapies: This category aims to overcome the limits of previous strategies by employing natural or synthetic biomaterials like hydrogels and other engineered drug delivery systems to enhance the integration, stability, and efficacy of therapeutic agents at the target site.[36]
4. Other Miscellaneous Therapies: Macrophage modification and lymphangiogenesis are newly emerging alternative strategies that potentially enhance cardiac regeneration (Figure 1).

Cell-Based Therapies: Exogenous Replacement of the Lost Myocardium

Since the discovery of stem cells and their capacity to differentiate into multiple cell types in the late 20th century, researchers have investigated their regenerative potential for various diseases, leading to the emergence of cell-based therapies as one of the earliest and most extensively studied approaches in cardiac regeneration.[37] The primary goal of cardiac cell therapy is to replace the population of lost cardiomyocytes with exogenously delivered cells capable of integrating with and differentiating into myocardial tissue.[38] To achieve this, various types of stem cells—the most common being mesenchymal stem cells (MSCs), embryonic stem cells (ESCs), and induced pluripotent stem cells (iPSCs)—have been explored with varying results.[39]

Table 1. **Hallmarks of cardiac regeneration (adapted from Bertero & Murry, 2018)**

Hallmark	Definition / Description	Key Strategies / Considerations
Remuscularisation	Restoration of cardiac muscle by generating new cardiomyocytes and preventing widespread cardiomyocyte death	– Stimulate endogenous cardiomyocyte proliferation – Reprogram resident stromal cells – Supplement with exogenous stem or progenitor cells or human pluripotent stem cells (hPSCs)-derived cardiomyocytes
Electromechanical stability	Integration of new cells into native electrical and mechanical systems to ensure proper cardiac function and contractility	– Induce rapid maturation of cellular excitation-contraction machinery – Suppress the automaticity of new cardiomyocytes – Ensure precise and efficient delivery of cells, molecules, and gene vectors
Angiogenesis and arteriogenesis	Formation of new blood vessels and arteries to support regenerating tissue	– Paracrine factor therapy (e.g., growth factors, exosomes) – Delivery of vasculogenic cells – Bioengineering of vascular networks for structured perfusion
Resolution of fibrosis	Regulation of extracellular matrix production to prevent excessive scarring that inhibits regeneration	– Modulate fibroblast activity – Use antifibrotic factors – Avoid excessive scarring to maintain electromechanical coupling and vascularisation, especially in cardiac patches.
Immunological balance	Regulation of immune responses to support regeneration while preventing adverse reactions such as immune rejection	– Clear necrotic cells and promote healing – Avoid chronic inflammation – Induce immune tolerance or use autologous/engineered cells to minimise the risk of rejection

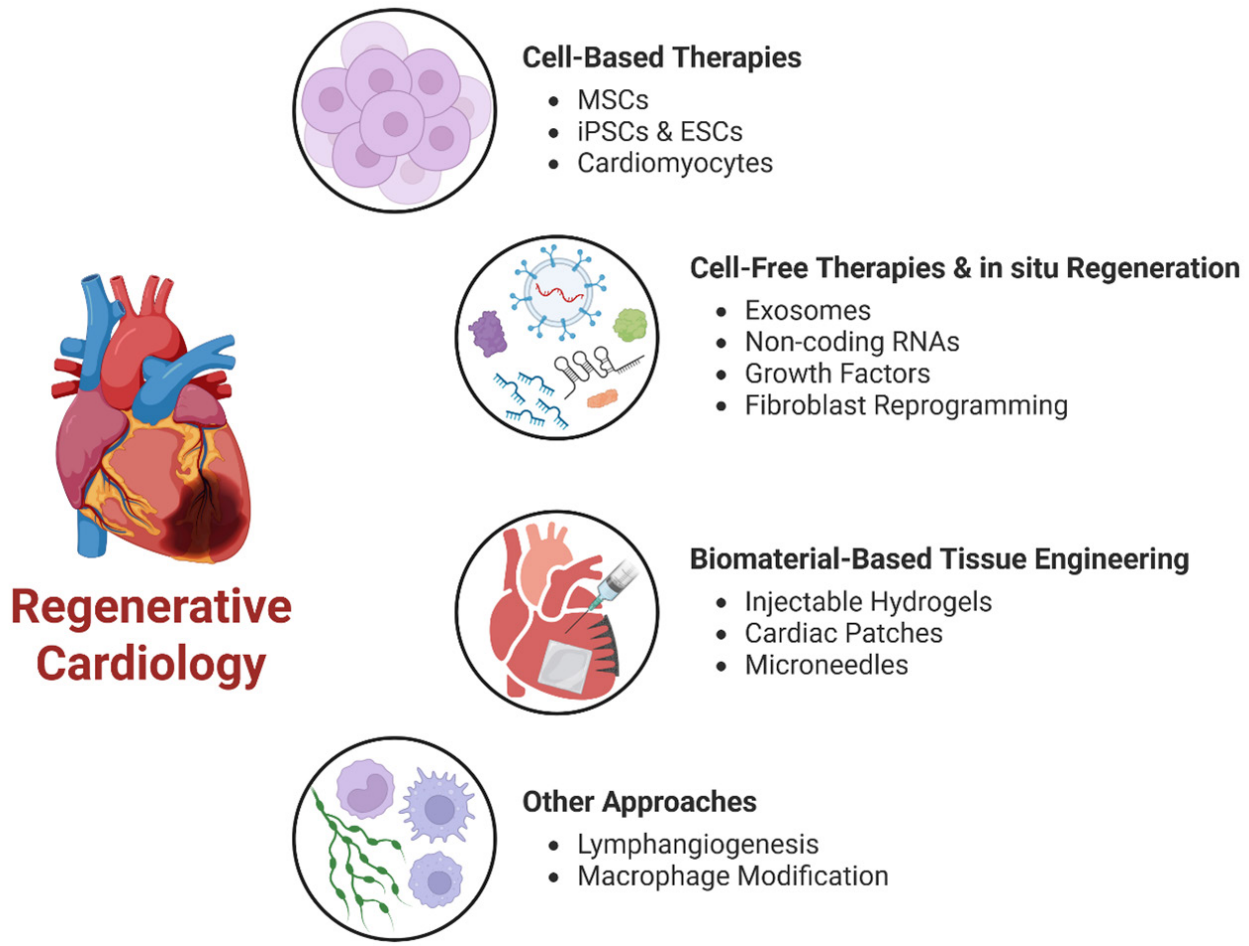

Fig. 1 **General classification of novel therapeutic strategies in regenerative cardiology.**

Cell Types and Mechanisms

Early investigations in stem cell therapy used adult sources of stem cells—skeletal myoblasts, bone marrow-derived mononuclear cells (BM-MNCs), and cardiosphere-derived cells (CDCs)—with initial preclinical trials showing modest improvements in left ventricular function. However, these findings were not reproducible, and subsequent studies yielded inconsistent results.[40,41] Early human clinical trials on skeletal myoblasts in patients with ischaemic cardiomyopathy, like the MAGIC trial, showed a trend of increased arrhythmias in cell-treated groups with no improvement in left ventricular ejection fraction (LVEF).[42] Other clinical trials assessing the effect of autologous BM-MNC transplantation in patients with ischaemic or non-ischaemic cardiomyopathy, like the 2012 FOCUS-CCTRN trial[43] and the 2021 MiHeart trials,[44] did not produce arrhythmias but also showed no improvement in left ventricular ejection fraction (LVEF) or LV systolic and diastolic volumes in treatment groups. These adverse results, combined with preclinical data indicating that essential components of bone marrow cells involved in cardiac repair are MSCs, redirected attention from whole bone marrow transplantation methods to using more refined stem cell options, like MSCs, for cardiac cell therapy.[45,46]

MSCs are multipotent, fibroblast-like cells that can be readily isolated from sources like bone marrow, adipose tissue, and umbilical cord blood. They are characterised by their ability to differentiate into adipocytes, chondrocytes, and osteoblasts.[47] Despite successful differentiation into cardiomyocyte-like cells *in vitro*, MSCs do not produce functionally beating cells, and *in vivo* applications have shown no effective cardiomyogenesis.[48] This has led researchers to conclude that their benefits likely stem from paracrine signalling pathways that induce regeneration via pro-angiogenic, anti-fibrotic, anti-apoptotic, and anti-inflammatory effects, rather than direct differentiation into cardiomyocytes.[49,50] Clinical trials using bone marrow-derived MSCs (BM-MSCs) in patients with ischemic cardiomyopathy and heart failure have produced mixed results. The MSC-HF trial, for example, found notable improvements in left ventricular end-systolic volume, LVEF, and stroke volume 12 months after autologous BM-MSCs were injected transendocardially into the heart. At the 4-year follow-up, patients in the treatment group reported fewer hospitalisations.[51] On the other hand, trials like TAC-HFT,[52] ixCELL-DCM[53] and CCTRN CONCERT-HF,[54] did not find any significant change in LVEF, left ventricular volumes, or scar size, though patients did report improved quality of life and lower hospitalisation rates. Clinical trials assessing the efficacy of umbilical cord-derived MSCs (UC-MSCs) in HF, such as the RIMECARD study,[55] have shown encouraging results, including improvements in ejection fraction at 3, 6, and 12 months, with no serious side effects reported.[45,46] Overall, MSC therapy is generally safe in humans; however, the heterogeneity of MSC sources, injection methods, sample sizes, cell dosage, and the timing of intervention and follow-up has limited the effective comparison of findings. Further investigation with larger trials is warranted for more conclusive results.[56]

Given the limited cardiomyogenic potential of adult stem cells and the inconsistent outcomes in clinical studies, regenerative cardiology has increasingly turned its attention to pluripotent stem cells (PSCs). PSCs, which include ESCs and iPSCs, can self-renew indefinitely and differentiate into most cell types, including atrial, nodal, and ventricular cardiomyocytes. ESCs, derived from developing blastocysts, are highly effective at transforming into specific cells, but their use is restricted by ethical concerns. iPSCs, which are genetically reprogrammed adult somatic cells with similar potency, bypass these ethical issues and offer the option of autologous transplants, reducing immune rejection risks.[40,57] Direct injection of undifferentiated ESCs into healthy or infarcted hearts in mice has only resulted in immune rejection and teratoma formation without evidence of cardiomyogenic differentiation.[58] Therefore, attention has shifted to producing PSC-derived cardiac progenitors (PSC-CPCs) or cardiomyocytes (PSC-CMs) *in vitro* before *in vivo* application. While PSC-CPCs and PSC-CMs in comparison have shown similar improvement in LVEF in a small animal study, PSC-CPCs did not result in effective engraftment and, like MSCs, their benefits likely resulted from paracrine signalling.[39,42] In contrast, PSC-CMs can remuscularise the heart with functional, force-generating cardiomyocytes, reduce scar formation, and improve cardiac function in small and large animal MI models. Even so, these cells exhibit phenotypic markers and electrophysiological properties of immature, embryonic cardiomyocytes, increasing the risk of arrhythmias as observed in most studies involving large animals like primates and pigs.[40,42,59] The ESCORT trial, the first in-human clinical trial to investigate the safety and feasibility of ESC-CPCs in severe LV dysfunction, reported improvements in systolic motion of the treated segments in the ventricular wall, with no evidence of tumour formation, arrhythmias, or symptomatic alloimmunisation in patients.[60] However, the small sample size (n = 6) necessitates the design of larger trials to validate findings.[38] Several other ongoing clinical trials on the efficacy of ESC-CMs and iPSC-CMs treatment are showing promising results in the early stages.[46] Nevertheless, the clinical application of stem cells remains constrained by several technical and safety challenges, including uncertain long-term outcomes, ethical issues associated with ESCs, risks of tumorigenicity, alongside variability in delivery techniques, dosing, timing, and individual patient responses.[35,38,39]

Delivery Strategies and Integration Challenges

Cardiac cell therapy is administered through three main routes of delivery: intramyocardial, intracoronary, or intravenous (systemic) injection. Intramyocardial administration is further divided into transendocardial or endovascular delivery and transepicardial injection.[61,62] Among these routes, transepicardial injection is the most direct yet invasive approach, allowing delivery of stem cells straight to the infarcted region during cardiac surgery. On the other hand, intravenous infusion is the safest route of cell delivery, but since cells are dispersed throughout the body, they rely on physiological homing signals secreted only acutely after MI to migrate to the injured myocardium.[62] The intracoronary route is also less invasive than transepicardial injection, yet it can potentially induce further ischemia, and cell delivery to target regions is limited by poor perfusion.[57]

The biggest obstacle to the successful application of cell therapy is the poor engraftment and low retention rate of transplanted cells.[61,63] For instance, in preclinical animal studies, only 11% of MSCs remain in the myocardium 90 minutes

after intramyocardial injection, dropping to just 0.6% after 24 hours. Most cells are rapidly washed away to non-target organs such as the lungs and liver.[34,50] Intracoronary or intravenous methods have even lower retention rates, and studies have shown that long-term retention is near zero regardless of the delivery method.[62]

To increase delivery efficiency and cell longevity, stem cells can be modified or labelled for increased infarct-targeting and binding capacity, or tissue-engineered into constructs like biomaterial-based scaffolds, such as cardiac patches and scaffold-free constructs like cell sheets.[34,64] These constructs are implanted onto the epicardial surface as epicardial patches or directly within the myocardium as interposition grafts. The epicardial layer may inhibit direct electrical coupling between the patch and host myocardium, hindering full integration of cells and limiting their action to paracrine signalling. In contrast, interposition grafts interface directly with the host myocardium without barriers, which facilitates remuscularisation but can result in arrhythmogenic complications.[62,65]

Limitations and Future Directions

Cell therapy shows promise for repairing heart damage in ischemic heart disease by encouraging tissue regeneration and boosting heart function, but several challenges still stand in the way of making it a reliable clinical option. Many clinical trials have drawn criticism for poor design or insufficient sample sizes, and the moderate improvements seen in early preclinical studies don't always hold up in larger clinical trials.[46] Outcomes vary significantly depending on patient differences, the types of stem cells used, and how they're delivered. Age, comorbidities, and the extent of heart damage all hinder standardisation. Ethical and safety concerns also limit the use of ESCs, and risks like tumour formation, immune rejection, and differing regulations across countries make them difficult to use in practice. iPSCs offer a less controversial alternative with lower risk of rejection, but they come with their own set of problems, including genetic instability and unintended side effects from the reprogramming process.[38] Other barriers to widespread clinical adoption of cell therapy include the complexities of growing, storing, and transporting viable stem cells. Maintaining cell viability and function during this process presents a significant obstacle for scaling up treatments. These issues have contributed to a growing interest in cell-free alternatives as a more practical and safer path to heart repair.[34] While cell therapy is still central to cardiac regeneration research, its future success likely depends on improving cell retention, standardising protocols, and combining therapies—like pairing cell delivery with engineered tissue scaffolds or using cell-free techniques to enhance results (Figure 2).

Cell-Free Therapies and in Situ Regeneration

Given the accumulating evidence favouring paracrine signalling as the primary benefactor of cell-based therapy, other therapeutic methods emerged intending to leverage these paracrine effects to foster the activation of endogenous repair pathways. These cell-free therapies lead to improved function through the stimulation of angiogenesis, regulation of fibrosis, mitigation of inflammation and apoptosis, and/or recruitment of tissue resident stem and/or progenitor cells.[66] Paracrine signalling molecules, such as growth factors, noncoding RNAs, and extracellular vesicles, offer significant advantages to cell therapy: they are easier to standardise and mass produce, carry lower immunogenic and tumorigenic risks, and are more compatible with drug delivery systems.[36] Alternatively, direct cardiac programming is another cell-free approach to remuscularising the heart that has also emerged, which induces *in situ* regeneration via delivery of transforming factors to purposefully reprogram non-CM cell populations, like fibroblasts, into cardiomyocytes (Figure 3).[67]

Growth Factors

Growth factors (GFs) are signalling proteins that regulate essential processes such as cell growth, adhesion, proliferation, migration, and others.[68] For example, neuregulin 1 (NRG1) and its receptors play critical roles in heart development and repair. In adult mouse models, activating this pathway promotes cardiomyocyte proliferation and improves cardiac function. A clinical trial with NRG1 showed benefits in heart failure patients, but some in vivo studies questioned its effect on cardiomyocyte proliferation, calling attention to the need for further validation.[69] Angiogenic growth factors like vascular endothelial growth factor (VEGF) and fibroblast growth factor (FGF) can improve cardiac function by stimulating the formation of new blood vessels when administered after MI,[70] which has been corroborated in several animal model studies and human clinical trials.[71–73] However, results in clinical trials have been inconsistent[74,75] and adverse effects such as nitric oxide-mediated hypotension and formation of aberrant and leaky vessels have limited clinical translation to patients.[70] Granulocyte-colony stimulating factor (G-CSF) and hepatocyte growth factor (HGF) promote the recruitment of circulating progenitor cells to the heart and help prevent cardiomyocyte apoptosis. Insulin-like growth factor 1 (IGF-1) enhances the proliferation and survival of cardiac stem and progenitor cells, supporting functional recovery.[70] The limited success of growth factor therapy in clinical trials is likely due to their rapid clearance, low stability, and non-specific distribution in the target tissue.[36] To address these challenges, biomaterial scaffolds and gene-based delivery methods, like synthetic modified RNA, have been developed to sustain growth factor release and enhance therapeutic outcomes.[69]

Noncoding RNAs

Noncoding RNAs (ncRNAs) regulate gene expression and include microRNAs (miRNAs) and long noncoding RNAs (lncRNAs). These molecules are key regulators of cardiac repair after MI and influence processes like cardiomyocyte proliferation, apoptosis, inflammation, and angiogenesis.[40] miRNAs such as miR-19a/19b boost cardiomyocyte proliferation and inhibit apoptosis, while other miRNAs modulate inflammation and facilitate angiogenesis.[76,77] lncRNAs often act as competing endogenous RNAs (ceRNAs) by sequestering miRNAs, thereby modulating their activity. They also influence autophagy, support cell proliferation through pathways like PI3K/Akt, and participate in epigenetic regulation by modifying histones to suppress unwanted gene expression. lncRNAs are considered promising therapeutic targets for enhancing cardiac regeneration and post-MI recovery.[78,79] Noncoding RNAs are degraded by RNases in extracellular fluids, therefore like GFs, efficient and targeted delivery is their main challenge for clinical application.[36] Similar to GFs, strategies like hydrogel scaffolds or exosome-mediated

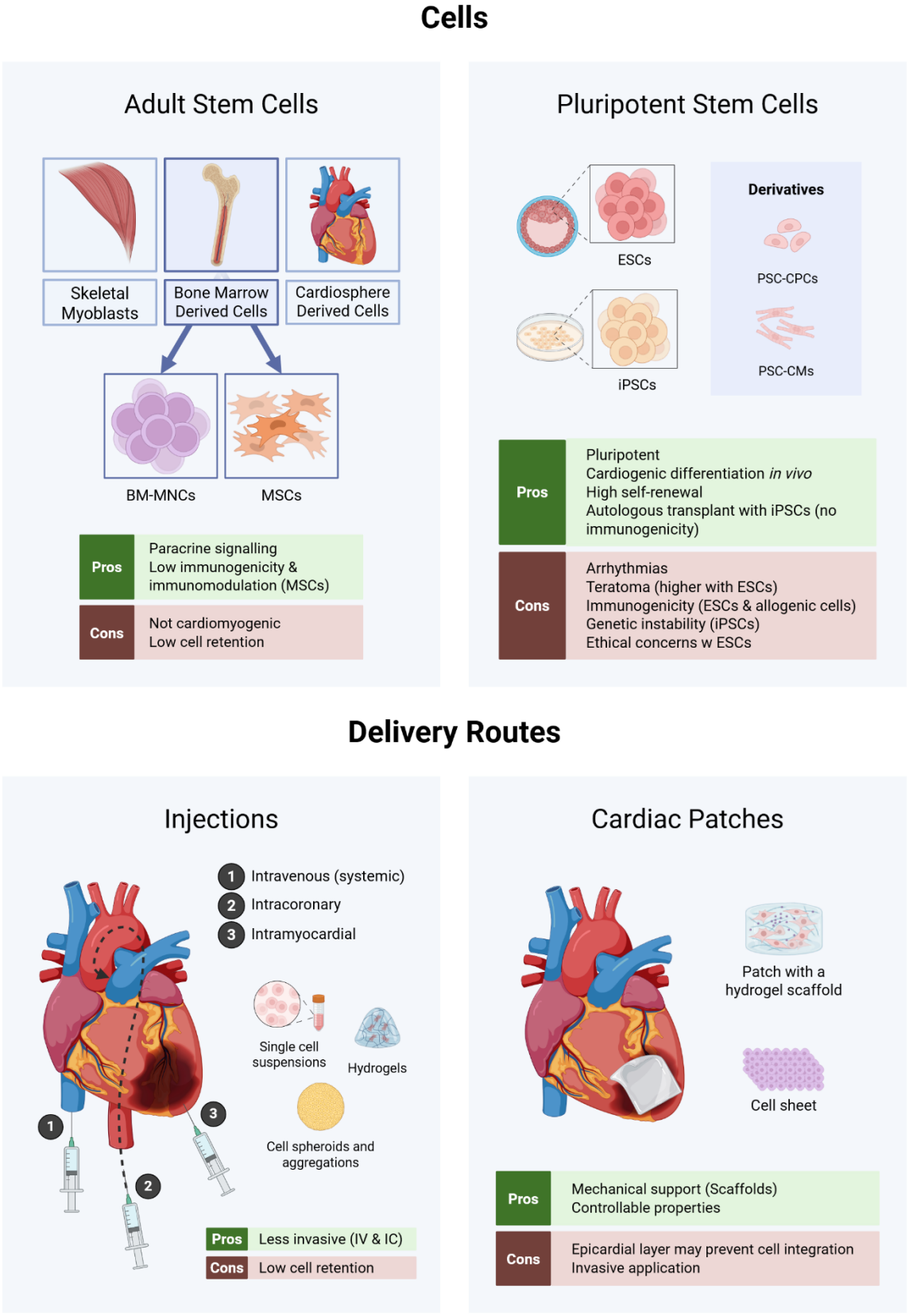

Fig. 2 **The cells and delivery methods used in cell therapy, and the pros and cons of each technique. BM-MNCs, bone marrow-derived mononuclear cells; MSCs, mesenchymal stem cells; ESCs, embryonic stem cells; iPSCs, induced pluripotent stem cells; PSC-CPCs, pluripotent stem cell-derived cardiac progenitors; PSC-CMs, pluripotent stem cell-derived cardiomyocytes; IV, intravenous; IC, intracoronary.**

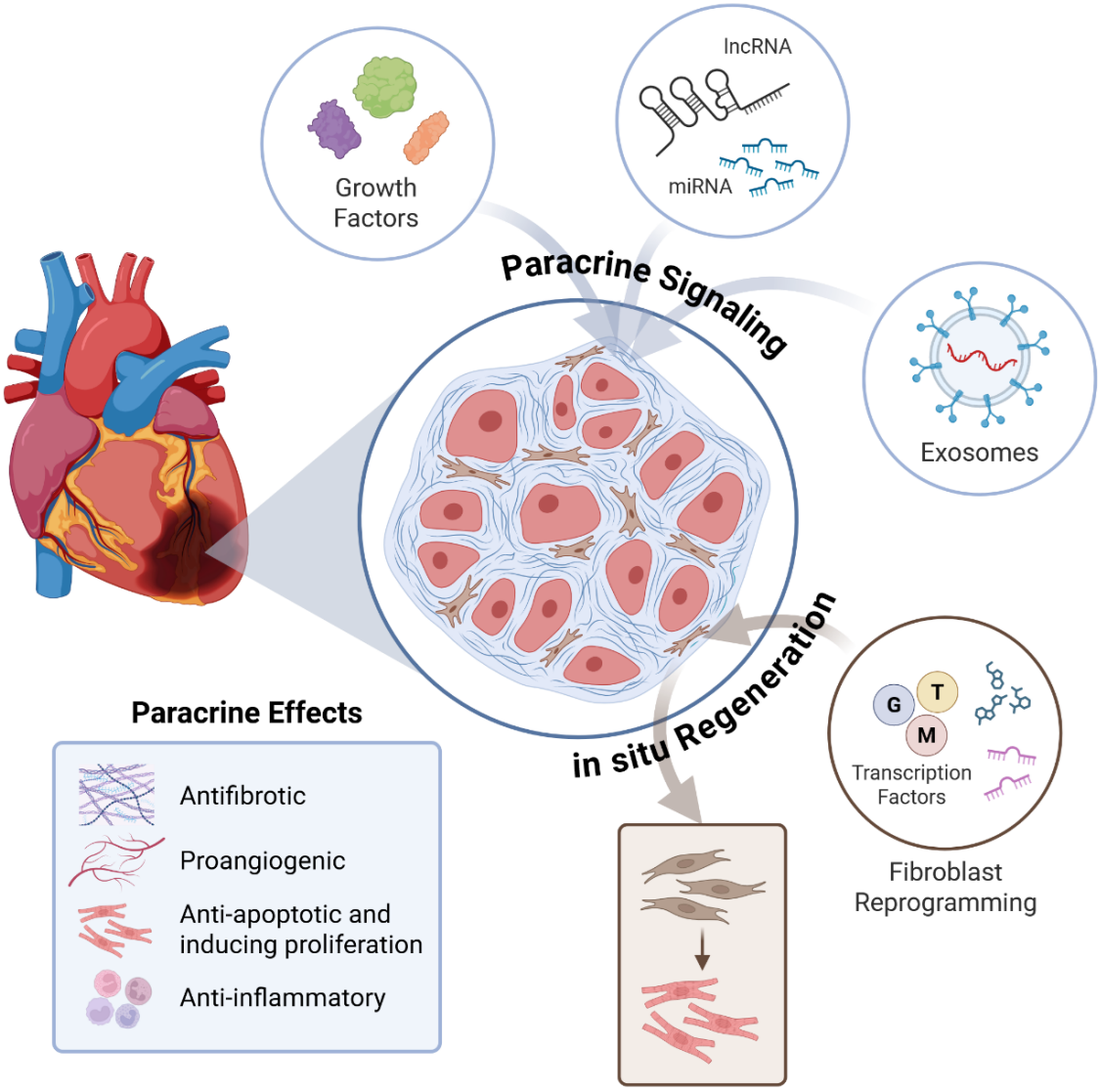

Fig. 3 **Cell-free therapeutic approaches for cardiac regeneration.**

transport have shown promise in sustained local delivery, enhancing their regenerative effect.[69] In previous works we studied the efficacy of exosome-mediated miR-126 and miR-146a delivery on cardiac tissue regeneration. Results showed a synergistic effect on cell migration, angiogenesis, and cardiac function.[80]

Extracellular Vesicles and Exosomes

Exosomes are a type of extracellular vesicle (EV), typically ranging from 40 to 160 nanometres in size, and are secreted by many cell types, including endothelial and stem cells. They serve as intercellular messengers carrying a variety of substances like proteins, lipids, metabolites, DNA, and noncoding RNAs. Compared to synthetic delivery systems, exosomes offer several advantages: they're less toxic, can pass easy through biological membranes, have low immunogenicity, and can deliver multiple therapeutic agents at once.[81,82] Studies on exosomes have had good results. For example, injecting exosomes derived from human endometrial MSCs, encapsulated in fibrin gel, significantly promoted angiogenesis in the myocardium and improved heart regeneration and function after MI.[83] In clinical trials, the ESCORT trial, which focused on ESC-CPCs highlighted the role of stem cell secretions on patient outcomes. This prompted a follow-up clinical trial on exosome delivery dubbed the SECRET-HF trial. Results showed that in patients with non-ischaemic dilated cardiomyopathy, the intravenous delivery of EVs derived from iPSC-CPCs—mainly exosomes—was safe, well tolerated, and improved symptoms with no adverse events after 6 months.[84] In a study in mice, intramyocardial injection of exosomes derived from iPSCs Cs 48 hours after reperfused MI led to better cardiac repair than iPSCs themselves in comparison. Treated mice showed improved heart function, more blood vessel growth, and reduced cell death and hypertrophy. Because they're cell-free, iPSC-derived exosomes offer a potentially safer option for treating ischemic heart damage.[85] The therapeutic benefits of exosomes can be improved by optimising their contents, targeting, and delivery, either through engineering the source cells or directly modifying the exosomes using chemical or physiological methods.[86] Despite this, limitations still remain

in exosome therapy. Like cytokines, exosomes are quickly taken up by cells, which shortens their half-life in the body and narrows their therapeutic window, therefore limiting any long-lasting benefit.[36,87]

Direct Cardiac Programming

During the process of fibrosis following myocardial infarction, dead cardiomyocytes are replaced by fibroblasts.[88] Direct cardiac programming first began as a concept in regenerative cardiology in 2010, when Ieda et al. reported that postnatal cardiac or dermal fibroblasts can be directly reprogrammed to differentiated cardiomyocyte-like cells using a specific combination of three transcription factors, Gata4, Mef2c, and Tbx5 (GMT).[89] This finding was replicated in other studies, including one in 2012, where *in vivo* delivery of GMT caused the development of induced cardiomyocytes (iCMs) in a murine model of MI.[90] However, only a small percentage (~0.01–0.1%) of iCMs transduced with GMT factors successfully developed organised sarcomere structures, and other studies revealed that reprogramming with GMT alone resulted in partial or inefficient cellular conversion, prompting ongoing efforts over the past decade to enhance reprogramming efficacy.[67] Further studies focused on increasing conversion efficiency by modification or addition of transcription factors to the original GMT combination, the most important of which demonstrated that GMT factors plus Hand2 (GHMT) resulted in more efficient reprogramming of cardiac fibroblasts into beating iCMs than the use of GMT alone.[91]

Pro-fibrotic signalling pathways also play a critical antagonistic role in the process of cardiac reprogramming. Zhao et al. demonstrated that inhibition of these networks using microRNAs and small-molecule inhibitors targeting TGF-β and Rho-associated kinase pathways enhanced the conversion rate of iCMs up to 60%. At the same time, their overactivation attenuates cardiac reprogramming.[92] However, Cardiac fibroblasts play a vital role in maintaining myocardial integrity and regulating cardiac remodelling following MI, and impairing pro-fibrotic pathways can worsen infarct size, ventricular dilation, and mechanical instability, thereby increasing the risk of adverse outcomes such as rupture. Consequently, anti-fibrotic therapies must be precisely timed, as early intervention may compromise structural support, while late treatment may be ineffective due to the maturation of fibrotic tissue. Further research is needed to understand better the effects of fibroblast inhibition during MI.[93]

Despite recent progress, cardiac reprogramming still faces significant challenges, particularly in achieving efficient reprogramming of human fibroblasts—arguably the most critical barrier in the field. Early studies revealed that factor combinations such as GMT and GHMT failed to activate cardiac gene expression in human cardiac or dermal fibroblasts, and even with the addition of supplementary factors, efficiency remains low and inconsistent across cell types and experimental conditions.[67] *In vivo* delivery of reprogramming factors also presents difficulties, often relying on invasive techniques with limited precision. While non-integrative systems like modified mRNAs offer a safer alternative, the long-term stability of reprogrammed phenotypes is uncertain, and viral vectors carry risks of mutagenesis and oncogenesis.[67,93] Furthermore, iCMs tend to exhibit immature and heterogeneous characteristics, raising concerns about arrhythmogenic potential and poor functional integration. The underlying molecular mechanisms, particularly at the epigenetic and signalling levels, remain poorly defined, complicating protocol optimisation and reproducibility.[93] These limitations highlight the need for improved factor combinations, deeper insights into molecular mechanisms, and the development of safer, more efficient delivery platforms before successful clinical implementation.[67]

Cardiac Tissue Engineering and Biomaterial-Based Therapies

In light of the major hurdles associated with both cell-based and acellular therapies, such as low cell retention, rapid degradation of therapeutic agents, and off-target effects,[94] cardiac tissue engineering has emerged as a multidisciplinary approach seeking to develop 3-dimensional (3D) cell culture systems to provide both an accurate model for *in vitro* study of heart function, disease mechanisms, and preclinical drug screening as well as a ready source of functional tissues for *in vivo* therapeutic transplantation.[95,96]

Biomaterials serve as a basis for tissue engineering approaches,[97] and using biomaterial-based scaffolds and delivery systems helps overcome the main clinical limitations of cell-based and cell-free therapies and enhances their efficacy. Biomaterials can enhance cell retention and promote electrical and mechanical coupling into the host myocardium, increase the half-life of bioactive molecules *in vivo* while allowing sustained release, in addition to improving compromised cardiac function when used in isolation.[96,98] Currently, the most commonly used biomaterials for cardiac applications include injectable hydrogels, cardiac patches, and, recently, microneedles (Figure 4).[98]

Injectable Hydrogels

Ideal biomaterials used in cardiac therapies should be biocompatible and biodegradable, and mimic the chemical, biological, and electrical properties of the native myocardium. Their mechanical properties must adequately provide mechanical support to damaged tissue while maintaining the heart's contractile function. Additionally, these biomaterials should provide an environment that increases the bioavailability and function of their therapeutic contents and facilitates their controlled release.[96,99] Hydrogels are three-dimensional cross-linked networks of hydrophilic polymer chains capable of absorbing, swelling, and retaining large volumes of water. Their soft, porous shape, high water content, and highly controllable mechanical features and microscale structure grant hydrogels a close resemblance to living tissue, making them some of the most ideal biomaterials for engineering scaffolds and drug delivery systems.[100,101]

Hydrogels are produced from natural polymers such as collagen, alginate, gelatin, chitosan, hyaluronic acid, and fibrin, or synthetic polymers like polycaprolactone and polyurethane.[99] They are used in cardiac applications primarily in the form of cardiac patches or injectable hydrogels; injectable hydrogels deliver therapeutics *in situ* to the injured myocardium via intramyocardial, intracoronary or intravenous routes, and hydrogel-based cardiac patches are *in vitro* fabricated scaffolds containing cells or bioactive molecules that are surgically attached to the epicardial surface of the heart.[102] Despite not being bioactive, hydrogels have been shown to improve heart function in animal studies, even in the absence of any other therapeutic agent (Table 2).[103]

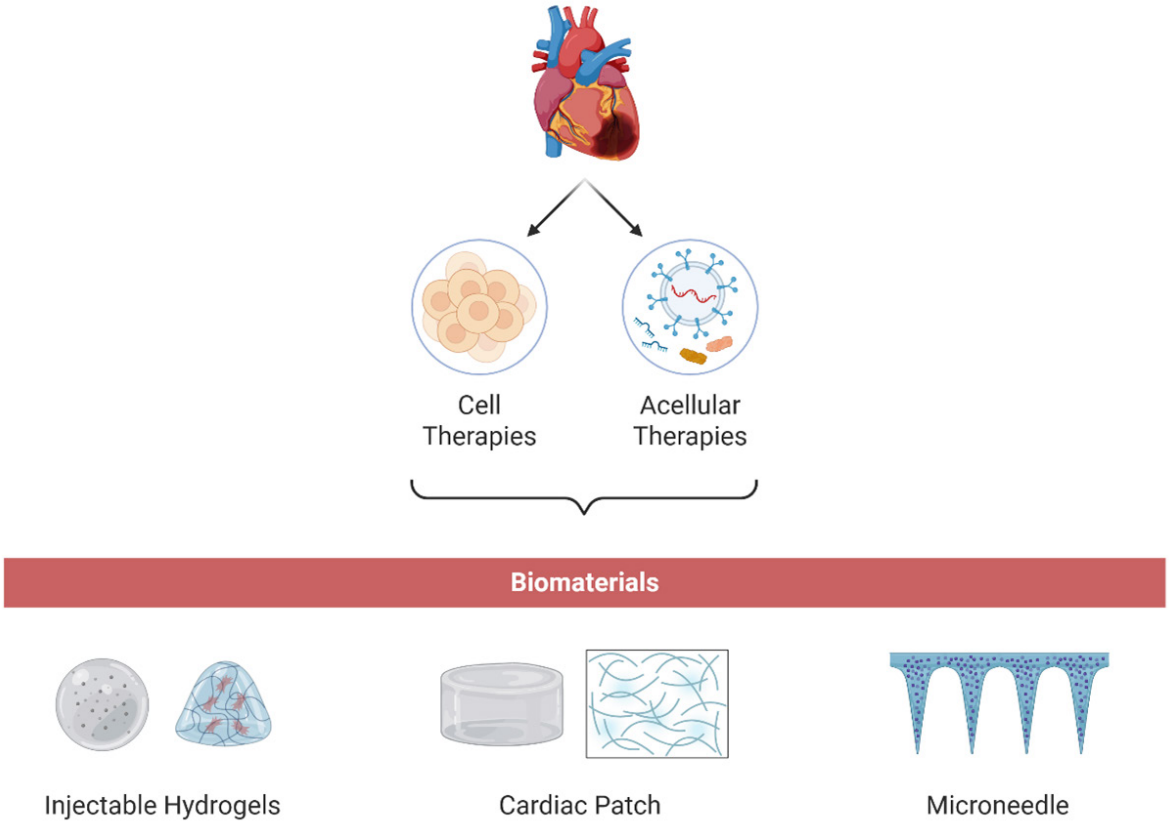

Fig. 4 **Biomaterials used in cardiac regeneration.**

Table 2. **Advantages and disadvantages of some of the most common types of hydrogels**

Type	Hydrogel	Advantages	Disadvantages	Gelation/cross-linking method	References
Natural Polymers	Collagen	Biocompatible Biodegradable Easy mixing easily with other types of hydrogels	Slow and inefficient cross-linking Low mechanical stability	Thermal	(99, 103, 104)
	Fibrin	Biocompatible Biodegradable Availability	Low mechanical stability Fast degradation *in vivo*	Peptide-self assembly	
	Hyaluronic acid	Biocompatible, biodegradable	Modifications needed for cross-link	Redox inhibitor	
	Alginate	Biocompatible Non-thrombogenic Rapid gelation, ECM-like structure, low cost	Low mechanical stability Poor long-term durability and uncontrollable degradation	Ionic	
Synthetic Polymers	Polyethylene glycol	Biocompatible Biodegradable Fast polymerisation times Good mechanical stability Allows photo cross-linking	Low cell adhesion Non-injectable Non-degradable Photo cross-linking process can affect cell viability	Peptide-self assembly	
	Polyvinyl alcohol	Biocompatible Easily modifiable	Gelling process potentially harmful for the patient	Chemical cross-linking	
Hybrid	Gelatin methacryloyl	Highly biocompatible Non-immunogenic	Requires UV light for polymerisation	Photo-polymerisation	

Cardiac Patches

Cardiac patches are disc-shaped scaffolds designed for the localised delivery of therapeutic agents. They are designed to attach to the epicardial surface of the infarcted myocardium as an alternative delivery approach to the potentially damaging method of intramyocardial injection. Cardiac patches enhance cell retention, allow controlled release of therapeutics, and create a suitable microenvironment for cell growth and proliferation.[99] Cardiac patches consist of two main components: therapeutic agents (like cells, bioactive molecules such as growth factors and mRNAs, drugs, or other engineered materials) which play a main role in inducing cardiac regeneration; and a structural scaffold, which provides mechanical and functional support and facilitates the interaction between therapeutic agents and the host tissue.[104,105]

The structural scaffold of cardiac patches is constructed using various synthetic or natural materials. The most common types of biomaterials used are decellularized ECM (dECM), collagen, alginate, and polyethylene glycol.[19] dECM is produced by removing cellular components from tissue using methods that preserve the composition and structure of ECM. These methods support new cell adhesion and growth, however, any residual cellular debris not fully cleared may trigger host immune responses.[106] Hydrogel patches have diverse properties depending on their materials and, due to their porous structure, can be specifically designed to release therapeutic agents in a controlled, sustained manner. They can be produced using simple and low-cost methods like freeze–thaw cycles and photo-polymerisation, or with advanced techniques such as 3D printing and electrospinning, which allow for precise architecture and complex structures. By incorporating materials like conductive nanostructures, hydrogels can be adapted for a wide range of cardiac therapeutic applications.[106,107]

Cardiac patches are typically implanted onto the epicardium using sutures or bio-adhesives and release their therapeutic agents from the surface of the heart, where these agents permeate through the epicardium to the myocardium. Regardless of differences in structure and implantation, cardiac patches successfully provide mechanical support and stimulate regeneration in animal MI models. They reduce the infarct size and reverse adverse ventricular remodelling while improving ejection fraction (EF) and other cardiac functions via anti-inflammatory, anti-apoptotic, pro-angiogenic, and anti-fibrotic processes.[106] However, the epicardial layer may act as a barrier, hindering effective diffusion and electrochemical interaction between the patch and the myocardium. Moreover, since the inner layers of the ventricular wall are more affected during MI, epicardial patches may not offer optimal therapeutic coverage.[19] One promising solution is the development of microneedle patches.

Microneedles

Microneedle patches represent a novel generation of engineered medical tools that, through the use of extremely fine needles ranging in length from 100 to 1000 micrometres, offer an innovative approach to therapeutic delivery. These microneedles, which garnered interest over two decades ago as a means of transdermal drug delivery, enhance delivery efficiency by penetrating through the tissue layer barriers in a minimally invasive manner. The success of this technology in improving transdermal drug delivery has expanded its application to other tissues and organs. In recent years, microneedles have also been explored for use in cardiac regenerative therapies.[108,109]

In cardiac applications, microneedles can create microscopic channels through the epicardium for precise delivery of agents directly into myocardial tissue, circumventing the limitations of cardiac patches. Also, compared to intramyocardial hydrogel injection, the smaller needle pores reduce the risk of cardiac wall damage and inflammatory responses.[19,110] Fabricated from natural or synthetic biomaterials like polyvinyl alcohol (PVA), hyaluronic acid, or gelatin methacryloyl using moulding or 3D printing, microneedles were first used for cardiac regeneration in 2018 in conjunction with cell therapy. Results confirmed their safety and enhanced therapeutic efficacy.[110] Since then, they have been evaluated for delivering cells, growth factors, miRNAs, nanoparticles, and other therapeutic agents for treating MI, with positive results.[111-113]

Despite these advances, cardiac microneedle technology remains in its early stages. In general, further research is required in all biomaterial-based therapeutics to standardise treatment protocols, ensure material safety, clinical feasibility, reproducibility of results, and scalability for mass production. Continued development of biomaterial constructs with improved or tuneable mechanical properties is much needed in cardiac tissue engineering and myocardial repair. In addition, finetuning their biomimetic properties, such as by enhancing electrical conductivity using electroconductive biomaterials,[114,115] can further improve performance.[107]

Other Emerging Strategies: Beyond Conventional Regeneration

Macrophage Polarisation and Immune Modulation

Over the past decade, macrophages have emerged as central regulators of cardiac regeneration, transitioning from their traditional role as debris-clearing phagocytes to active participants in myocardial repair. Advances in single-cell transcriptomics and genetic fate mapping have revealed the heterogeneity and functional plasticity of macrophage subpopulations, distinguishing between resident and monocyte-derived macrophages.[116] Cardiac-resident macrophages originate from yolk-sac progenitors and populate the heart prenatally, maintaining homeostasis via immunosurveillance and clearing dead cells and debris in the myocardium, and performing critical functions in healthy myocardium, such as facilitating electrical conduction by coupling with cardiomyocytes through gap junctions.[117]

Studies using regenerative models such as neonatal mice and zebrafish have revealed important distinctions in macrophage responses following cardiac injury when compared to non-regenerative adult mammalian models. In zebrafish, depletion of macrophages significantly impaired heart regeneration[118] and cardiac-resident macrophages dominated, tending to be less inflammatory, more angiogenic, and less fibrotic, whereas in adult mammals, monocyte-derived macrophages dominated and often adopt a pro-fibrotic phenotype.[116,119] While exhibiting reparative properties similar to neonatal macrophages, cardiac-resident macrophages in adult hearts are rapidly lost due to necrosis after MI. Monocyte-derived macrophages mediate post-MI inflammatory

responses by differentiating into two main phenotypes, M1 and M2, with the former acting pro-inflammatory and the latter anti-inflammatory.[120,121]

Strategies to bias macrophage polarisation toward regenerative phenotypes are under active investigation. Dental pulp MSC-derived extracellular vesicles (MSC-EVs) shift macrophages to an M2-like state, reducing infarct size in rat models.[122] Similarly, interleukin 4/13 administration post-MI improves ejection fraction by enhancing macrophage efferocytosis and angiogenesis.[123] Human iPSC-derived primitive macrophages, cultured in a medium enriched with pro-regenerative growth factors, has demonstrated efficacy in promoting adult cardiomyocyte proliferation and functional recovery in murine models. However, challenges remain in standardizing vesicle production and ensuring targeted delivery.[124] Preclinical studies suggest that expanding resident macrophage populations before injury could prime the heart for regeneration. In zebrafish, early clodronate-mediated depletion of resident macrophages leads to long-term regenerative deficits, highlighting their irreplaceable role.[118] In addition, pharmacological agents that enhance resident macrophage survival or function, such as CSF1R agonists, are also being explored.[116]

Even with these promising results, research in macrophage-mediated cardiac regeneration is still in its early stages. Cardiac macrophage plasticity and phenotypes and their specific function in regeneration have yet to be accurately established, and it is currently unknown whether macrophage polarisation can successfully induce efficient regeneration. The interspecies differences in macrophage function and heterogeneity of resident macrophages complicate targeting specific pro-regenerative subtypes. Taking this into account, research in this topic can help clarify the mechanisms of cardiac repair.[120,121]

Lymphangiogenesis

Another newly emerging hot spot in regenerative cardiology is lymphangiogenesis. The lymphatic vasculature of the heart has an essential role in heart physiology, regulating tissue fluid balance, lipid metabolism, transportation of extravasated proteins, and immune function.[125] Myocardial injury leads to dysfunction of these processes and, therefore, results in fluid imbalance and cardiac dysfunction. Thus, therapy promoting lymphangiogenesis may improve myocardial tissue repair post-MI.[125] The lymphatic system's role in cardiac regeneration has been studied in adult zebrafish models of cardiac injury in the past few years. Research has shown different healing responses according to the type of injury; with no signs of cardiac lymphangiogenesis following apical resection and, in contrast, enlargement and migration of lymphatic vessels into the wound site after cryoinjury.[126] Selective stimulation of cardiac lymphangiogenesis via VEGF-C limited remodelling of lymphatic precollectors post-MI and improved myocardial fluid balance and reduced inflammation, fibrosis, and dysfunction as a result.[127] These results are debatable, with one research group reporting that genetic blockade of lymphangiogenesis in mice with the ablation of VEGFR3 signalling does not impair LVEF and cardiac function two weeks following MI.[128] Another research group showed that while impairing the lymphatic network via deletion of the endothelial adhesion protein VE-cadherin does not cause cardiac dysfunction in mice, it causes increased cardiac infarct size and fibrosis after MI.[129] This suggests that dysfunction of the lymphatic vasculature may impact cardiac function post-MI and exacerbate heart failure.[130] Further studies are needed to better understand the molecular mechanisms of lymphatic transport and its role in heart disease, especially in mammalian hearts, and more effort should be focused on gaining insight into the potential role of lymphangiogenesis in cardiac regeneration.[131]

Future Directions

Despite the challenges in cardiac regeneration approaches, emerging novel strategies to overcome current obstacles have opened new horizons to the field. For instance, the development of nanotechnology-based delivery systems capable of a sustainable and controlled release of multiple bioactive agents may facilitate *in situ* regeneration strategies in the future. While our understanding of endogenous regeneration mechanisms of cardiac tissue enhances, endeavouring innovative and combined methods to trigger *in situ* regeneration of cardiac tissue is expected. Adopting cardiac regeneration mechanisms of other species into the human heart, direct reprogramming techniques based on *in situ* genetic and epigenetic manipulation of the cells, and advancement in targeted delivery of different bioactive agents to the site of interest are some areas of research that offer great potential in restoring the heart. However, the field is still in the early stage of development and requires more innovative multidisciplinary approaches.

Conclusion

Several cardiac regeneration strategies have been studied to restore damaged cardiac tissue during the last decades. Cell-based therapies continue to be a significant area of research in regenerative cardiology, dedicating more focus on developing targeted cell therapies with more cell retention potential and better cell survival at the injury site. Cell-free therapies and *in situ* regeneration strategies on the other hand, have gained more research attention in recent years, owing to their great therapeutic and regenerative potential and safer features. Cardiac tissue engineering and biomaterial-based approaches are also growing fields, providing new hope for researchers as well as patients suffering from cardiovascular diseases. Although there are several obstacles that should be overcome, the results of recent innovative approaches hold great promise in the field of regenerative cardiology.

Acknowledgment

The author would like to acknowledge the support provided by Tehran University of Medical Sciences for this work (grant number: 62977-264-2-1402). We would also like to acknowledge that AI was used to edit some parts of the manuscript.

Conflict of Interest

The authors declare no conflict of interest.

References

1. Lindstrom M, DeCleene N, Dorsey H, Fuster V, Johnson CO, LeGrand KE, et al. Global Burden of Cardiovascular Diseases and Risks Collaboration, 1990–2021. J Am Coll Cardiol. 2022;80(25):2372–425.
2. Roth GA, Mensah GA, Johnson CO, Addolorato G, Ammirati E, Baddour LM, et al. Global Burden of Cardiovascular Diseases and Risk Factors, 1990–2019: Update From the GBD 2019 Study. J Am Coll Cardiol. 2020;76(25):2982–3021.
3. Collaborators GBDF. Burden of disease scenarios for 204 countries and territories, 2022–2050: a forecasting analysis for the Global Burden of Disease Study 2021. Lancet. 2024;403(10440):2204–56.
4. Chong B, Jayabaskaran J, Jauhari SM, Chan SP, Goh R, Kueh MTW, et al. Global burden of cardiovascular diseases: projections from 2025 to 2050. Eur J Prev Cardiol. 2024.
5. The top 10 causes of death [Internet]. Geneva: World Health Organization; [updated 7 August 2024. Available from: https://www.who.int/news-room/fact-sheets/detail/the-top-10-causes-of-death.
6. Salari N, Morddarvanjoghi F, Abdolmaleki A, Rasoulpoor S, Khaleghi AA, Hezarkhani LA, et al. The global prevalence of myocardial infarction: a systematic review and meta-analysis. BMC Cardiovasc Disord. 2023;23(1):206.
7. Rittiphairoj T, Bulstra C, Ruampatana C, Stavridou M, Grewal S, Reddy CL, et al. The economic burden of ischaemic heart diseases on health systems: a systematic review. BMJ Glob Health. 2025;10(2).
8. Thygesen K, Alpert JS, Jaffe AS, Chaitman BR, Bax JJ, Morrow DA, et al. Fourth Universal Definition of Myocardial Infarction (2018). Circulation. 2018;138(20):e618–e51.
9. Anderson JL, Morrow DA. Acute Myocardial Infarction. N Engl J Med. 2017;376(21):2053–64.
10. Gabriel-Costa D. The pathophysiology of myocardial infarction-induced heart failure. Pathophysiology. 2018;25(4):277–84.
11. Frantz S, Hundertmark MJ, Schulz-Menger J, Bengel FM, Bauersachs J. Left ventricular remodelling post-myocardial infarction: pathophysiology, imaging, and novel therapies. Eur Heart J. 2022;43(27):2549–61.
12. Bostan MM, Statescu C, Anghel L, Serban IL, Cojocaru E, Sascau R. Post-Myocardial Infarction Ventricular Remodeling Biomarkers-The Key Link between Pathophysiology and Clinic. Biomolecules. 2020;10(11).
13. Frey N, Katus HA, Olson EN, Hill JA. Hypertrophy of the heart: a new therapeutic target? Circulation. 2004;109(13):1580–9.
14. Heusch G, Libby P, Gersh B, Yellon D, Bohm M, Lopaschuk G, et al. Cardiovascular remodelling in coronary artery disease and heart failure. Lancet. 2014;383(9932):1933–43.
15. Zafari AM, Yang EH. Myocardial Infarction Treatment & Management: Approach Considerations, Prehospital Care and Initial Management, Emergency Department Care and In-Hospital Management: Medscape; [Available from: https://emedicine.medscape.com/article/155919-treatment.
16. McMurray JJ, Pfeffer MA. Heart failure. Lancet. 2005;365(9474):1877–89.
17. Frangogiannis NG. Pathophysiology of Myocardial Infarction. Compr Physiol. 2015;5(4):1841–75.
18. Dzau VJ, Antman EM, Black HR, Hayes DL, Manson JE, Plutzky J, et al. The cardiovascular disease continuum validated: clinical evidence of improved patient outcomes: part II: Clinical trial evidence (acute coronary syndromes through renal disease) and future directions. Circulation. 2006;114(25):2871–91.
19. Liu T, Hao Y, Zhang Z, Zhou H, Peng S, Zhang D, et al. Advanced Cardiac Patches for the Treatment of Myocardial Infarction. Circulation. 2024;149(25):2002–20.
20. Garbern JC, Lee RT. Heart regeneration: 20 years of progress and renewed optimism. Dev Cell. 2022;57(4):424–39.
21. Miller L, Birks E, Guglin M, Lamba H, Frazier OH. Use of Ventricular Assist Devices and Heart Transplantation for Advanced Heart Failure. Circ Res. 2019;124(11):1658–78.
22. Nakamura M, Huang GN. Why some hearts heal and others don't: The phylogenetic landscape of cardiac regenerative capacity. Semin Cell Dev Biol. 2025;170:103609.
23. Poss KD, Wilson LG, Keating MT. Heart regeneration in zebrafish. Science. 2002;298(5601):2188–90.
24. Kikuchi K, Poss KD. Cardiac regenerative capacity and mechanisms. Annu Rev Cell Dev Biol. 2012;28:719–41.
25. Ma S, Yan J, Chen L, Zhu Y, Chen K, Zheng C, et al. A Bibliometric and Visualized Analysis of Cardiac Regeneration Over a 20-Year Period. Front Cardiovasc Med. 2021;8:789503.
26. Porrello ER, Mahmoud AI, Simpson E, Hill JA, Richardson JA, Olson EN, et al. Transient regenerative potential of the neonatal mouse heart. Science. 2011;331(6020):1078–80.
27. Senyo SE, Steinhauser ML, Pizzimenti CL, Yang VK, Cai L, Wang M, et al. Mammalian heart renewal by pre-existing cardiomyocytes. Nature. 2013;493(7432):433–6.
28. Auchampach J, Han L, Huang GN, Kuhn B, Lough JW, O'Meara CC, et al. Measuring cardiomyocyte cell-cycle activity and proliferation in the age of heart regeneration. Am J Physiol Heart Circ Physiol. 2022;322(4):H579–H96.
29. Bergmann O, Bhardwaj RD, Bernard S, Zdunek S, Barnabe-Heider F, Walsh S, et al. Evidence for cardiomyocyte renewal in humans. Science. 2009;324(5923):98–102.
30. Riching AS, Song K. Cardiac Regeneration: New Insights Into the Frontier of Ischemic Heart Failure Therapy. Front Bioeng Biotechnol. 2020;8:637538.
31. Brezitski KD, Goff AW, DeBenedittis P, Karra R. A Roadmap to Heart Regeneration Through Conserved Mechanisms in Zebrafish and Mammals. Curr Cardiol Rep. 2021;23(4):29.
32. Rolland L, Jopling C. The multifaceted nature of endogenous cardiac regeneration. Front Cardiovasc Med. 2023;10:1138485.
33. Bertero A, Murry CE. Hallmarks of cardiac regeneration. Nat Rev Cardiol. 2018;15(10):579–80.
34. Li Z, Hu S, Cheng K. Chemical Engineering of Cell Therapy for Heart Diseases. Acc Chem Res. 2019;52(6):1687–96.
35. Csobonyeiova M, Beerova N, Klein M, Debreova-Cehakova M, Danisovic L. Cell-Based and Selected Cell-Free Therapies for Myocardial Infarction: How Do They Compare to the Current Treatment Options? Int J Mol Sci. 2022;23(18).
36. Bar A, Cohen S. Inducing Endogenous Cardiac Regeneration: Can Biomaterials Connect the Dots? Front Bioeng Biotechnol. 2020;8:126.
37. Carvalho AB, de Carvalho AC. Heart regeneration: Past, present and future. World J Cardiol. 2010;2(5):107–11.
38. Ali SA, Mahmood Z, Mubarak Z, Asad M, Sarfraz Chaudhri MT, Bilal L, et al. Assessing the Potential Benefits of Stem Cell Therapy in Cardiac Regeneration for Patients With Ischemic Heart Disease. Cureus. 2025;17(1):e76770.
39. Muller P, Lemcke H, David R. Stem Cell Therapy in Heart Diseases - Cell Types, Mechanisms and Improvement Strategies. Cell Physiol Biochem. 2018;48(6):2607–55.
40. Pezhouman A, Nguyen NB, Kay M, Kanjilal B, Noshadi I, Ardehali R. Cardiac regeneration - Past advancements, current challenges, and future directions. J Mol Cell Cardiol. 2023;182:75–85.
41. Braunwald E. Cell-Based Therapy in Cardiac Regeneration: An Overview. Circ Res. 2018;123(2):132–7.
42. Selvakumar D, Clayton ZE, Chong JJH. Robust Cardiac Regeneration: Fulfilling the Promise of Cardiac Cell Therapy. Clin Ther. 2020;42(10):1857–79.
43. Perin EC, Willerson JT, Pepine CJ, Henry TD, Ellis SG, Zhao DX, et al. Effect of transendocardial delivery of autologous bone marrow mononuclear cells on functional capacity, left ventricular function, and perfusion in chronic heart failure: the FOCUS-CCTRN trial. JAMA. 2012;307(16):1717–26.
44. Martino H, Brofman P, Greco O, Bueno R, Bodanese L, Clausell N, et al. Multicentre, randomized, double-blind trial of intracoronary autologous mononuclear bone marrow cell injection in non-ischaemic dilated cardiomyopathy (the dilated cardiomyopathy arm of the MiHeart study). Eur Heart J. 2015;36(42):2898–904.
45. Banerjee MN, Bolli R, Hare JM. Clinical Studies of Cell Therapy in Cardiovascular Medicine: Recent Developments and Future Directions. Circ Res. 2018;123(2):266–87.
46. Zhang JJ, Pogwizd SM, Fukuda K, Zimmermann WH, Fan C, Hare JM, et al. Trials and tribulations of cell therapy for heart failure: an update on ongoing trials. Nat Rev Cardiol. 2025;22(5):372–85.
47. Margiana R, Markov A, Zekiy AO, Hamza MU, Al-Dabbagh KA, Al-Zubaidi SH, et al. Clinical application of mesenchymal stem cell in regenerative medicine: a narrative review. Stem Cell Res Ther. 2022;13(1):366.
48. Gupta S, Sharma A, S A, Verma RS. Mesenchymal Stem Cells for Cardiac Regeneration: from Differentiation to Cell Delivery. Stem Cell Rev Rep. 2021;17(5):1666–94.
49. Zhu D, Cheng K. Cardiac Cell Therapy for Heart Repair: Should the Cells Be Left Out? Cells. 2021;10(3).
50. Huang K, Hu S, Cheng K. A New Era of Cardiac Cell Therapy: Opportunities and Challenges. Adv Healthc Mater. 2019;8(2):e1801011.
51. Mathiasen AB, Qayyum AA, Jorgensen E, Helqvist S, Kofoed KF, Haack-Sorensen M, et al. Bone marrow-derived mesenchymal stromal cell treatment in patients with ischaemic heart failure: final 4-year follow-up of the MSC-HF trial. Eur J Heart Fail. 2020;22(5):884–92.
52. Heldman AW, DiFede DL, Fishman JE, Zambrano JP, Trachtenberg BH, Karantalis V, et al. Transendocardial mesenchymal stem cells and

mononuclear bone marrow cells for ischemic cardiomyopathy: the TAC-HFT randomized trial. JAMA. 2014;311(1):62–73.
53. Patel AN, Henry TD, Quyyumi AA, Schaer GL, Anderson RD, Toma C, et al. Ixmyelocel-T for patients with ischaemic heart failure: a prospective randomised double-blind trial. Lancet. 2016;387(10036):2412–21.
54. Bolli R, Mitrani RD, Hare JM, Pepine CJ, Perin EC, Willerson JT, et al. A Phase II study of autologous mesenchymal stromal cells and c-kit positive cardiac cells, alone or in combination, in patients with ischaemic heart failure: the CCTRN CONCERT-HF trial. Eur J Heart Fail. 2021;23(4):661–74.
55. Bartolucci J, Verdugo FJ, Gonzalez PL, Larrea RE, Abarzua E, Goset C, et al. Safety and Efficacy of the Intravenous Infusion of Umbilical Cord Mesenchymal Stem Cells in Patients With Heart Failure: A Phase 1/2 Randomized Controlled Trial (RIMECARD Trial [Randomized Clinical Trial of Intravenous Infusion Umbilical Cord Mesenchymal Stem Cells on Cardiopathy]). Circ Res. 2017;121(10):1192–204.
56. Seyihoglu B, Orhan I, Okudur N, Aygun HK, Bhupal M, Yavuz Y, et al. 20 years of treating ischemic cardiomyopathy with mesenchymal stromal cells: a meta-analysis and systematic review. Cytotherapy. 2024;26(12):1443–57.
57. Abouzid MR, Umer AM, Jha SK, Akbar UA, Khraisat O, Saleh A, et al. Stem Cell Therapy for Myocardial Infarction and Heart Failure: A Comprehensive Systematic Review and Critical Analysis. Cureus. 2024;16(5):e59474.
58. Nussbaum J, Minami E, Laflamme MA, Virag JA, Ware CB, Masino A, et al. Transplantation of undifferentiated murine embryonic stem cells in the heart: teratoma formation and immune response. FASEB J. 2007;21(7):1345–57.
59. Wang J, An M, Haubner BJ, Penninger JM. Cardiac regeneration: Options for repairing the injured heart. Front Cardiovasc Med. 2022;9:981982.
60. Menasche P, Vanneaux V, Hagege A, Bel A, Cholley B, Parouchev A, et al. Transplantation of Human Embryonic Stem Cell-Derived Cardiovascular Progenitors for Severe Ischemic Left Ventricular Dysfunction. J Am Coll Cardiol. 2018;71(4):429–38.
61. Wulfse M, Vervoorn MT, Amelink J, Ballan EM, de Jager SCA, Sluijter JPG, et al. Past Trends and Future Directions of Cardiac Regenerative Medicine - A Systematic Analysis of Clinical Trial Registries. J Cardiovasc Transl Res. 2025;18(1):209–20.
62. Watanabe T, Hatayama N, Guo M, Yuhara S, Shinoka T. Bridging the Gap: Advances and Challenges in Heart Regeneration from In Vitro to In Vivo Applications. Bioengineering (Basel). 2024;11(10).
63. Pagano F, Picchio V, Chimenti I, Sordano A, De Falco E, Peruzzi M, et al. On the Road to Regeneration: "Tools" and "Routes" Towards Efficient Cardiac Cell Therapy for Ischemic Cardiomyopathy. Curr Cardiol Rep. 2019;21(11):133.
64. Cambria E, Pasqualini FS, Wolint P, Gunter J, Steiger J, Bopp A, et al. Translational cardiac stem cell therapy: advancing from first-generation to next-generation cell types. NPJ Regen Med. 2017;2:17.
65. Gibbs CE, Marchiano S, Zhang K, Yang X, Murry CE, Boyle PM. Graft-host coupling changes can lead to engraftment arrhythmia: a computational study. J Physiol. 2023;601(13):2733–49.
66. Menasche P. Cell therapy trials for heart regeneration - lessons learned and future directions. Nat Rev Cardiol. 2018;15(11):659–71.
67. Garry GA, Bassel-Duby R, Olson EN. Direct reprogramming as a route to cardiac repair. Semin Cell Dev Biol. 2022;122:3–13.
68. Arunadevi N, Jone Kirubavathy S. Three-dimensional approaches based on nanotechnology towards wound management. In: Solanki PR, Kumar A, Pratap Singh R, Singh J, Rb Singh K, editors. Nanotechnological Aspects for Next-Generation Wound Management: Academic Press; 2024. p. 245–80.
69. Hashimoto H, Olson EN, Bassel-Duby R. Therapeutic approaches for cardiac regeneration and repair. Nat Rev Cardiol. 2018;15(10):585–600.
70. Segers VF, Lee RT. Protein therapeutics for cardiac regeneration after myocardial infarction. J Cardiovasc Transl Res. 2010;3(5):469–77.
71. Schumacher B, Stegmann T, Pecher P. The stimulation of neoangiogenesis in the ischemic human heart by the growth factor FGF: first clinical results. J Cardiovasc Surg (Torino). 1998;39(6):783–9.
72. Yanagisawa-Miwa A, Uchida Y, Nakamura F, Tomaru T, Kido H, Kamijo T, et al. Salvage of infarcted myocardium by angiogenic action of basic fibroblast growth factor. Science. 1992;257(5075):1401–3.
73. Rasanen M, Sultan I, Paech J, Hemanthakumar KA, Yu W, He L, et al. VEGF-B Promotes Endocardium-Derived Coronary Vessel Development and Cardiac Regeneration. Circulation. 2021;143(1):65–77.
74. Stewart DJ, Kutryk MJ, Fitchett D, Freeman M, Camack N, Su Y, et al. VEGF gene therapy fails to improve perfusion of ischemic myocardium in patients with advanced coronary disease: results of the NORTHERN trial. Mol Ther. 2009;17(6):1109–15.
75. Henry TD, Annex BH, McKendall GR, Azrin MA, Lopez JJ, Giordano FJ, et al. The VIVA trial: Vascular endothelial growth factor in Ischemia for Vascular Angiogenesis. Circulation. 2003;107(10):1359–65.
76. Gao F, Kataoka M, Liu N, Liang T, Huang ZP, Gu F, et al. Therapeutic role of miR-19a/19b in cardiac regeneration and protection from myocardial infarction. Nat Commun. 2019;10(1):1802.
77. Zhu K, Liu D, Lai H, Li J, Wang C. Developing miRNA therapeutics for cardiac repair in ischemic heart disease. J Thorac Dis. 2016;8(9):E918-E27.
78. Fang Y, Xu Y, Wang R, Hu L, Guo D, Xue F, et al. Recent advances on the roles of LncRNAs in cardiovascular disease. J Cell Mol Med. 2020;24(21):12246–57.
79. Dalil D, Bahanesteh A, Rezaei M, Afzali A, Vanaki A, Varamini A. The Role of Long Noncoding RNAs in Cardiomyocyte Proliferation and Heart Regeneration After Myocardial Infarction: A Systematic Review. Cell Transplant. 2024;33:9636897241266725.
80. Shafei S, Khanmohammadi M, Ghanbari H, Nooshabadi VT, Tafti SHA, Rabbani S, et al. Effectiveness of exosome mediated miR-126 and miR-146a delivery on cardiac tissue regeneration. Cell Tissue Res. 2022;390(1):71–92.
81. Wei Q, Xiao Y, Du L, Li Y. Advances in Nanoparticles in the Prevention and Treatment of Myocardial Infarction. Molecules. 2024;29(11).
82. Reiss AB, Ahmed S, Johnson M, Saeedullah U, De Leon J. Exosomes in Cardiovascular Disease: From Mechanism to Therapeutic Target. Metabolites. 2023;13(4).
83. Sepehri M, Rabbani S, Ai J, Bahrami N, Ghanbari H, Namini MS, et al. Therapeutic potential of exosomes derived from human endometrial mesenchymal stem cells for heart tissue regeneration after myocardial infarction. Regen Ther. 2025;28:451–61.
84. Menasche P, Renault NK, Hagege A, Puscas T, Bellamy V, Humbert C, et al. First-in-man use of a cardiovascular cell-derived secretome in heart failure. Case report. EBioMedicine. 2024;103:105145.
85. Adamiak M, Cheng G, Bobis-Wozowicz S, Zhao L, Kedracka-Krok S, Samanta A, et al. Induced Pluripotent Stem Cell (iPSC)-Derived Extracellular Vesicles Are Safer and More Effective for Cardiac Repair Than iPSCs. Circ Res. 2018;122(2):296–309.
86. Han C, Yang J, Sun J, Qin G. Extracellular vesicles in cardiovascular disease: Biological functions and therapeutic implications. Pharmacol Ther. 2022;233:108025.
87. Das D, Jothimani G, Banerjee A, Dey A, Duttaroy AK, Pathak S. A brief review on recent advances in diagnostic and therapeutic applications of extracellular vesicles in cardiovascular disease. Int J Biochem Cell Biol. 2024;173:106616.
88. Talman V, Ruskoaho H. Cardiac fibrosis in myocardial infarction-from repair and remodeling to regeneration. Cell Tissue Res. 2016;365(3):563–81.
89. Ieda M, Fu JD, Delgado-Olguin P, Vedantham V, Hayashi Y, Bruneau BG, et al. Direct reprogramming of fibroblasts into functional cardiomyocytes by defined factors. Cell. 2010;142(3):375–86.
90. Qian L, Huang Y, Spencer CI, Foley A, Vedantham V, Liu L, et al. In vivo reprogramming of murine cardiac fibroblasts into induced cardiomyocytes. Nature. 2012;485(7400):593–8.
91. Sadahiro T, Ieda M. Direct Cardiac Reprogramming for Cardiovascular Regeneration and Differentiation. Keio J Med. 2020;69(3):49–58.
92. Zhao Y, Londono P, Cao Y, Sharpe EJ, Proenza C, O'Rourke R, et al. High-efficiency reprogramming of fibroblasts into cardiomyocytes requires suppression of pro-fibrotic signalling. Nat Commun. 2015;6:8243.
93. Andrianto A, Mulia EPB, Luke K. Direct cardiac reprogramming: basics and future challenges. Mol Biol Rep. 2023;50(1):865–71.
94. Portillo Esquivel LE, Zhang B. Application of Cell, Tissue, and Biomaterial Delivery in Cardiac Regenerative Therapy. ACS Biomater Sci Eng. 2021;7(3):1000–21.
95. Sharma V, Dash SK, Govarthanan K, Gahtori R, Negi N, Barani M, et al. Recent Advances in Cardiac Tissue Engineering for the Management of Myocardium Infarction. Cells. 2021;10(10).
96. Pomeroy JE, Helfer A, Bursac N. Biomaterializing the promise of cardiac tissue engineering. Biotechnol Adv. 2020;42:107353.
97. Huyer LD, Montgomery M, Zhao Y, Xiao Y, Conant G, Korolj A, et al. Biomaterial based cardiac tissue engineering and its applications. Biomed Mater. 2015;10(3):034004.
98. Gil-Cabrerizo P, Scacchetti I, Garbayo E, Blanco-Prieto MJ. Cardiac tissue engineering for myocardial infarction treatment. Eur J Pharm Sci. 2023;185:106439.
99. Tariq U, Gupta M, Pathak S, Patil R, Dohare A, Misra SK. Role of Biomaterials in Cardiac Repair and Regeneration: Therapeutic Intervention for Myocardial Infarction. ACS Biomater Sci Eng. 2022;8(8):3271–98.
100. Xu Y, Yu Y, Guo Z. Hydrogels in cardiac tissue engineering: application and challenges. Mol Cell Biochem. 2025;480(4):2201–22.
101. Saludas L, Pascual-Gil S, Prosper F, Garbayo E, Blanco-Prieto M. Hydrogel based approaches for cardiac tissue engineering. Int J Pharm. 2017;523(2):454–75.

102. Rakshit P, Giri TK, Mukherjee K. Progresses and perspectives on natural polysaccharide based hydrogels for repair of infarcted myocardium. Int J Biol Macromol. 2024;269(Pt 2):132213.
103. Sun X, Nunes SS. Overview of hydrogel-based strategies for application in cardiac tissue regeneration. Biomed Mater. 2015;10(3):034005.
104. Zhang X, Zhao G, Ma T, Simmons CA, Santerre JP. A critical review on advances and challenges of bioprinted cardiac patches. Acta Biomater. 2024;189:1–24.
105. Mei X, Cheng K. Recent Development in Therapeutic Cardiac Patches. Front Cardiovasc Med. 2020;7:610364.
106. Li M, Wu H, Yuan Y, Hu B, Gu N. Recent fabrications and applications of cardiac patch in myocardial infarction treatment. View. 2021;3(2):20200153.
107. Liu Z, Zheng Z, Xie J, Wei H, Yu CY. Hydrogel-based cardiac patches for myocardial infarction therapy: Recent advances and challenges. Mater Today Bio. 2024;29:101331.
108. Panda A, Matadh VA, Suresh S, Shivakumar HN, Murthy SN. Non-dermal applications of microneedle drug delivery systems. Drug Deliv Transl Res. 2022;12(1):67–78.
109. Lee K, Goudie MJ, Tebon P, Sun W, Luo Z, Lee J, et al. Non-transdermal microneedles for advanced drug delivery. Adv Drug Deliv Rev. 2020; 165–166:41–59.
110. Tang J, Wang J, Huang K, Ye Y, Su T, Qiao L, et al. Cardiac cell-integrated microneedle patch for treating myocardial infarction. Sci Adv. 2018;4(11):eaat9365.
111. Yuan J, Yang H, Liu C, Shao L, Zhang H, Lu K, et al. Microneedle Patch Loaded with Exosomes Containing MicroRNA-29b Prevents Cardiac Fibrosis after Myocardial Infarction. Adv Healthc Mater. 2023;12(13):e2202959.
112. Lim S, Park TY, Jeon EY, Joo KI, Cha HJ. Double-layered adhesive microneedle bandage based on biofunctionalized mussel protein for cardiac tissue regeneration. Biomaterials. 2021;278:121171.
113. Wang Q, Cao S, Zhang T, Lv F, Zhai M, Bai D, et al. Reactive oxide species and ultrasound dual-responsive bilayer microneedle array for in-situ sequential therapy of acute myocardial infarction. Biomater Adv. 2024;162:213917.
114. Shokraei N, Asadpour S, Shokraei S, Nasrollahzadeh Sabet M, Faridi-Majidi R, Ghanbari H. Development of electrically conductive hybrid nanofibers based on CNT-polyurethane nanocomposite for cardiac tissue engineering. Microsc Res Tech. 2019;82(8):1316–25.
115. Ahmadi P, Nazeri N, Derakhshan MA, Ghanbari H. Preparation and characterization of polyurethane/chitosan/CNT nanofibrous scaffold for cardiac tissue engineering. Int J Biol Macromol. 2021;180:590–8.
116. Alvarez-Argote S, O'Meara CC. The Evolving Roles of Cardiac Macrophages in Homeostasis, Regeneration, and Repair. Int J Mol Sci. 2021;22(15).
117. Suku M, Forrester L, Biggs M, Monaghan MG. Resident Macrophages and Their Potential in Cardiac Tissue Engineering. Tissue Eng Part B Rev. 2022;28(3):579–91.
118. Wei KH, Lin IT, Chowdhury K, Lim KL, Liu KT, Ko TM, et al. Comparative single-cell profiling reveals distinct cardiac resident macrophages essential for zebrafish heart regeneration. Elife. 2023;12.
119. Constanty F, Wu B, Wei KH, Lin IT, Dallmann J, Guenther S, et al. Border-zone cardiomyocytes and macrophages regulate extracellular matrix remodeling to promote cardiomyocyte protrusion during cardiac regeneration. Nat Commun. 2025;16(1):3823.
120. Lafuse WP, Wozniak DJ, Rajaram MVS. Role of Cardiac Macrophages on Cardiac Inflammation, Fibrosis and Tissue Repair. Cells. 2020;10(1).
121. Molinaro C, Scalise M, Leo I, Salerno L, Sabatino J, Salerno N, et al. Polarizing Macrophage Functional Phenotype to Foster Cardiac Regeneration. Int J Mol Sci. 2023;24(13).
122. Amaro-Prellezo E, Gomez-Ferrer M, Hakobyan L, Ontoria-Oviedo I, Peiro-Molina E, Tarazona S, et al. Extracellular vesicles from dental pulp mesenchymal stem cells modulate macrophage phenotype during acute and chronic cardiac inflammation in athymic nude rats with myocardial infarction. Inflamm Regen. 2024;44(1):25.
123. Bakhshian Nik A, Alvarez-Argote S, O'Meara CC. Interleukin 4/13 signaling in cardiac regeneration and repair. Am J Physiol Heart Circ Physiol. 2022;323(5):H833–H44.
124. Xiao Y, Zhang H, Liu X, Xu P, Du H, Wang J, et al. Medium from human iPSC-derived primitive macrophages promotes adult cardiomyocyte proliferation and cardiac regeneration. Nat Commun. 2025;16(1):3012.
125. Bai L, Wang Y, Du S, Si Y, Chen L, Li L, et al. Lymphangiogenesis: A new strategy for heart disease treatment (Review). Int J Mol Med. 2024;53(4).
126. Klaourakis K, Vieira JM, Riley PR. The evolving cardiac lymphatic vasculature in development, repair and regeneration. Nat Rev Cardiol. 2021;18(5):368–79.
127. Henri O, Pouehe C, Houssari M, Galas L, Nicol L, Edwards-Levy F, et al. Selective Stimulation of Cardiac Lymphangiogenesis Reduces Myocardial Edema and Fibrosis Leading to Improved Cardiac Function Following Myocardial Infarction. Circulation. 2016;133(15):1484–97; discussion 97.
128. Keller TCSt, Lim L, Shewale SV, McDaid K, Marti-Pamies I, Tang AT, et al. Genetic blockade of lymphangiogenesis does not impair cardiac function after myocardial infarction. J Clin Invest. 2021;131(20).
129. Harris NR, Nielsen NR, Pawlak JB, Aghajanian A, Rangarajan K, Serafin DS, et al. VE-Cadherin Is Required for Cardiac Lymphatic Maintenance and Signaling. Circ Res. 2022;130(1):5–23.
130. Liu X, Cui K, Wu H, Li KS, Peng Q, Wang D, et al. Promoting Lymphangiogenesis and Lymphatic Growth and Remodeling to Treat Cardiovascular and Metabolic Diseases. Arterioscler Thromb Vasc Biol. 2023;43(1):e1–e10.
131. Wang YC, Meng WT, Zhang HF, Zhu J, Wang QL, Mou FF, et al. Lymphangiogenesis, a potential treatment target for myocardial injury. Microvasc Res. 2023;145:104442.