

# Nanomedicine for Diagnosis and Treatment of Cardiac Fibrosis

Yanay Montano-Peguerio<sup>1</sup>, Hugo E Verdejo<sup>1,2</sup>, Jaime A Riquelme<sup>1</sup>, Marcelo J Kogan<sup>1,3</sup>, Sergio Lavandero<sup>1,4</sup>

<sup>1</sup>Advanced Center for Chronic Diseases (ACCDiS), Facultad de Ciencias Químicas y Farmacéuticas and Facultad Medicina, Universidad de Chile, Santiago, Chile; <sup>2</sup>División de Enfermedades Cardiovasculares, Facultad de Medicina, Pontificia Universidad Católica de Chile, Santiago, Chile;

<sup>3</sup>Departamento de Química Farmacológica y Toxicológica, Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile, Santiago, Chile;

<sup>4</sup>Cardiology Division, University of Texas Southwestern Medical Center, Dallas, TX, USA

Correspondence: Marcelo J Kogan; Sergio Lavandero, Advanced Center for Chronic Diseases (ACCDiS), Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile, Santiago, 8380494, Chile, Email mkogan@ciq.uchile.cl; slavander@uchile.cl

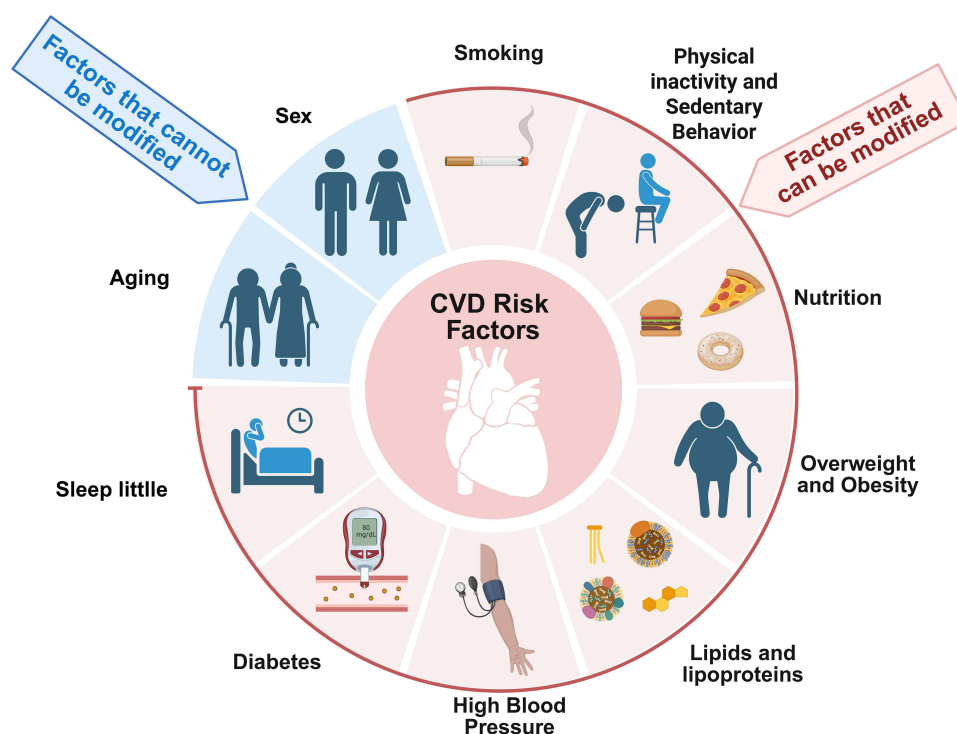
**Abstract:** Cardiovascular diseases (CVDs) are the leading cause of death worldwide. Although different risk factors condition the evolution and outcome of CVDs, a limited number of common pathological pathways converge in adverse cardiac remodeling. Among these, cardiac fibrosis has been postulated not only as a mechanism for the onset and development of CVDs but also as a predictor of disease severity. Tools to diagnose subclinical cardiac fibrosis and therapies targeting cardiac fibrosis constitute an essential line of research in CVDs. To date, magnetic resonance imaging (MRI) is a sensitive tool for diagnosing cardiac fibrosis, but sampling errors can occur. Moreover, its availability is limited by the high cost of the equipment. For this reason, nanotechnology offers a promising approach for developing nanosystems directed at molecular targets of cardiac damage, to provide both affordable and readily available diagnostics and the possibility of targeted therapy. This review will showcase major research advances on the role of nanoparticles in cardiac fibrosis. It will discuss the advantages and limitations of nanoparticles, and the standards for advancing them to the clinical phase of treatment and diagnosis of cardiac fibrosis.

**Keywords:** cardiac fibrosis, diagnostic imaging, therapy, nanoparticles, gold colloid

## Introduction

Cardiovascular diseases (CVDs) is the leading cause of death worldwide. According to the GBD (*Global Burden of Disease*) report, the prevalence of CVDs worldwide increased from 271 million cases in 1990 to 523 million in 2019. Mortality has also increased, from 12 million to 18 million cases. Most CVD deaths occur in low- and middle-income countries, accounting for 80% of all cases. The incidence is higher in males than in females.<sup>1</sup> The costs related to these diseases exceed 250 billion dollars per year. There are risk factors that influence the onset and progression of CVDs.<sup>2</sup> Age and sex are risk factors that have been described as unmodifiable and that increase the risk of CVDs. However, other factors can be modified, such as smoking, physical activity, sedentary lifestyle, nutrition, sleep, overweight and obesity, hypercholesterolemia, diabetes, and hypertension, as shown in Figure 1.<sup>3,4</sup> In the United States (US), clinical risk factors are projected to increase from 2020 to 2050: hypertension (51.2% to 61.0%), diabetes (16.3% to 26.8%), and obesity (43.1% to 60.6%). Additionally, unhealthy lifestyle habits, including bad sleep, sedentary lifestyle, smoking, and inadequate diet, increase. This estimate correlated with an increase in the prevalence of CVDs by the year 2050.<sup>2</sup> The data on incidence, mortality, economic costs, and mental and physical disability that compromise the social performance of patients demonstrate the complexity of CVDs. This background makes CVDs a subject of constant research to solve this global health problem.

Cardiac fibrosis has been identified as a predictor of the severity of CVDs, such as heart failure and ischemic heart disease. Therefore, it is postulated as a risk factor that should be diagnosed and treated to act on the progression of CVDs. Currently, there is no approved treatment aimed explicitly at cardiac fibrosis. It is therefore approached from



**Figure 1** Primary risk factors for developing cardiovascular disease. Created in BioRender. Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.

a pharmacological approach (treating the symptoms involved) and an approach based on risk factor intervention (modifying unhealthy lifestyles). One of the current limitations of therapy is that it is not directed at targets identified in the pathophysiological mechanisms of the disease. In addition, there are no robust, sensitive, and accessible diagnostic methods to follow the evolution of the disease, nor to evaluate the efficacy of antifibrotic therapy.<sup>5,6</sup> Therefore, this article discusses the usefulness of nanoparticles (NPs) for the design of treatments and diagnostic tools for cardiac fibrosis, based on molecular targets involved in the CVDs.

## The Role of Cardiac Fibrosis in Cardiovascular Disease

Cardiac fibrosis is a significant pathological condition that arises from various cardiovascular risk factors. The convergence of these risk factors leads to a cascade of molecular and cellular events that culminate in the activation of cardiac fibroblasts, which, due to stimuli of a biochemical or mechanical nature, migrate to the damaged area of the heart and differentiate into extracellular matrix (ECM)-secreting cardiac myofibroblasts. Initially, this response can be reparative. However, if sustained over time, the excessive deposition of ECM components, primarily collagen types I and III, results in impaired cardiac function and increased morbidity and mortality associated with heart failure.<sup>7–9</sup> Transforming growth factor beta (TGF- $\beta$ ) activates profibrotic signaling pathways. It promotes the differentiation of fibroblasts into myofibroblasts, and they produce ECM excessively.<sup>10–12</sup> Activation of TGF- $\beta$  signaling is often exacerbated by hyperglycemia and oxidative stress, leading to increased fibrosis and subsequent cardiac dysfunction.<sup>13,14</sup>

Cardiac fibrosis can present itself in diverse ways, depending on the stimulus that triggers it. In response to an acute injury that causes a significant death of cardiomyocytes, such as the one observed in myocardial infarction or certain forms of myocarditis, an inflammatory response is triggered, which signals fibroblasts to activate, migrate, and proliferate in the affected area. In the presence of inflammatory cytokines, cardiac fibroblasts differentiate into myofibroblasts, a phenotype that secretes collagen as a component of the ECM. This replacement fibrosis, in which dead cells in the wound area are replaced or substituted by collagen, is typically reparative and aims to maintain the structural integrity of the heart. However, if the replacement is too extensive, it may lead to loss of contractile force and systolic dysfunction.

Chronic injuries that extend over time, such as sustained pressure overload, chronic inflammation, and aging, promote diffuse-type fibrosis. Prolonged activation of these fibrogenic stimuli leads to deposition of ECM component proteins interstitially, perivascularly, or both, without significant cardiomyocyte death.<sup>9,15,16</sup> In this form of fibrosis, the imbalance between increased production of ECM proteins and decreased ECM turnover causes stiffening of the ventricles, reducing heart distensibility and impairing diastolic function.<sup>15,17,18</sup>

## The Role of the Cardiac Fibroblast in Cardiac Fibrosis

Cardiac fibroblasts (CFs) are key components of the heart's connective tissue. These cells lack a basement membrane and can produce substantial ECM proteins.<sup>8,19,20</sup> In infarcted and stressed hearts, cardiac fibroblasts differentiate into myofibroblasts, acquiring a highly active phenotype characterized by a prominent endoplasmic reticulum and the expression of contractile proteins such as  $\alpha$ -smooth muscle actin ( $\alpha$ -SMA). This contractile protein is absent in fibroblasts of healthy hearts, making  $\alpha$ -SMA a specific marker of myofibroblasts in hypertrophic and fibrotic myocardium.<sup>18,21</sup> Cardiac myofibroblasts have a higher proliferative rate, migration capacity, and contractile capacity, and can secrete higher amounts of ECM proteins than CFs.<sup>22,23</sup>

The regulation of myocardial collagen turnover is primarily mediated by resident CFs and their differentiation into myofibroblasts. This process is directly stimulated by mechanical stretching and the influence of autocrine and paracrine factors, including aldosterone, angiotensin II (Ang-II), TGF- $\beta$ , platelet-derived growth factor (PDGF), and endothelin-1 (ET-1).<sup>24</sup> These factors bind to their respective cell surface receptors, activating intracellular signaling pathways that drive fibrogenesis. The fibrotic response within the myocardium varies depending on the nature of the injury and the involvement of distinct cell types. Immune cells such as macrophages, lymphocytes, mast cells, and eosinophils play a pivotal role in activating CFs through the secretion of cytokines, growth factors, and ECM proteins. Similarly, vascular endothelial cells and pericytes activate CFs by releasing mediators that stimulate fibrotic processes. Additionally, stressed or injured cardiomyocytes release fibrogenic mediators and damage-associated molecular patterns (DAMPs), which further drive inflammation and CFs activation, perpetuating the fibrotic response.<sup>8,25,26</sup> Pressure overload induces an inflammatory reaction in the myocardium. There are increased levels of inflammatory cytokines (IL-1, IL-6, IL-13, and IL-21), which also influence the activation and function of CFs.<sup>27,28</sup> Inflammatory response increased vascular permeability and remodeling of the ECM. The environment of damage and remodeling in the heart favors the effect of increased permeability and retention (EPR).<sup>29</sup>

## Diagnostic and Treatment Strategies for Cardiac Fibrosis

Research to develop tools for diagnosing and treating cardiac fibrosis has focused on identifying molecular targets. In addition, they agree on the need to know and characterize the fibrotic environment in detail to obtain better results in therapies. In the healthy heart, there is a homeostasis between the production of ECM components and their degradation. When the heart is subjected to sustained stress, the balance is broken, and the initial reparative response becomes pathological.

## Imaging Tools for the Diagnosis of Cardiac Fibrosis

A significant limitation in diagnosing cardiac fibrosis is the lack of a readily available and sensitive method to detect it in its early stages. While myocardial biopsy remains the gold standard for definitively diagnosing cardiac fibrosis by quantifying collagen content in the myocardial tissue, it is an invasive procedure with inherent drawbacks. Its sensitivity is limited by the focal and heterogeneous nature of the fibrotic response, making it less effective in identifying diffuse or early-stage fibrosis.<sup>23,30,31</sup> Echocardiography is also performed clinically, but this measurement provides results of heart functionality. However, it has a low sensitivity and specificity for diagnosing cardiac fibrosis.<sup>30</sup>

Magnetic resonance imaging (MRI) has emerged as an essential tool for the early diagnosis of cardiac fibrosis, providing critical insights into myocardial structure and function. The ability of MRI to detect myocardial fibrosis, mainly through techniques such as late gadolinium enhancement (LGE), allows for the identification of fibrotic changes before significant functional impairment occurs. One of the most important advantages of cardiac MRI is its non-invasive nature, which facilitates the assessment of myocardial fibrosis without the need for invasive procedures. Studies have

shown that LGE-MRI can effectively visualize areas of fibrosis, correlating with clinical outcomes and disease severity, particularly in diabetic patients.<sup>32</sup> Furthermore, the role of MRI extends to monitoring conditions such as hypertrophic cardiomyopathy, where fibrosis is linked to poorer prognoses.<sup>33</sup> Advanced MRI techniques, such as T1 mapping, further enhance the capability to quantify myocardial fibrosis. This method allows for a more nuanced understanding of the fibrotic process, including the differentiation between focal and diffuse fibrosis.<sup>34</sup>

One significant limitation of MRI in cardiac fibrosis is its spatial resolution.<sup>7</sup> While late gadolinium enhancement (LGE) MRI effectively visualizes myocardial scars, it struggles with the resolution needed to assess smaller structures, particularly in the left atrium.<sup>35</sup> This limitation can lead to underdiagnosis or misinterpretation of fibrotic changes in the atrial myocardium, which is critical given the role of atrial fibrosis in arrhythmogenesis. Another limitation is the reliance on contrast agents, which can pose risks for specific patient populations. The use of gadolinium-based contrast agents is standard in LGE MRI; however, these agents can cause nephrogenic systemic fibrosis in patients with severe renal impairment. The interpretation of MRI findings can also be complicated by diffuse myocardial fibrosis, which may not be as readily detectable as focal fibrosis. While T1 mapping techniques have been developed to quantify diffuse fibrosis, the accuracy of these measurements can be influenced by various factors, including edema or inflammation, which can alter T1 values. Moreover, MRI is not universally accessible and may be limited by factors such as cost, availability of equipment, and the need for specialized personnel to perform and interpret the studies. In many clinical settings, particularly in low-resource environments, these barriers can impede the widespread adoption of MRI for cardiac fibrosis assessment. Integrating MRI with other imaging modalities, such as positron emission tomography (PET) and echocardiography, can comprehensively evaluate cardiac health, particularly in complex cases involving multiple comorbidities.<sup>36</sup>

Similarly, PET offers exceptional detection sensitivity; however, its reliance on radioactive tracers presents notable limitations. PET employs ionizing radiation, with positron emissions producing two gamma rays at 511 keV, essential for imaging and diagnosis. Although the radiopharmaceutical doses are typically low, repeated exposure over time poses potential risks, making it less ideal for long-term patient monitoring. Consequently, PET is not well-suited for the routine follow-up of patients with cardiac fibrosis, particularly when considering the cumulative effects of radiation exposure.<sup>37</sup>

Computed tomography (CT) is a much more accessible technology for most healthcare systems. It offers high resolutions but does not differentiate healthy tissue from fibrotic tissue.<sup>38</sup> Due to their unique optical and radiographic properties, gold nanoparticles (AuNPs) have emerged as a promising solution to overcome this disadvantage. Gold efficiently scatters visible light and exhibits a high X-ray attenuation coefficient at clinically relevant energy levels, generating high-contrast X-ray images that are particularly useful for disease diagnosis. X-ray images use high-energy electromagnetic radiation to create images of internal structures interacting with matter at clinically relevant energy levels. The detection of contrast agents by measuring the characteristic X-ray attenuation profiles has been used to detect AuNPs in vivo.<sup>39</sup> CT allows 3D reconstruction of X-ray images by rotating the detector and the X-ray source around the imaged body, providing detailed anatomical insights. The X-ray images are considered safe and cost-effective when radiation doses are appropriately controlled for patient safety.<sup>38,39</sup>

These advances highlight the transformative potential of integrating nanotechnology and imaging to diagnose and treat cardiac fibrosis. The advantages and limitations of all these methods for imaging cardiac fibrosis are shown in Table 1. This is an advantage of AuNPs over other types of NPs; it provides high contrast for soft tissues and chemical stability. Because soft tissues have a low ability to stop X-rays, AuNPs target soft tissue and increase the visualization of the structure. This way, damage to cardiac structures, eg, cardiac fibrosis, can be detected. NPs increase the spatial resolution of the images obtained, improve diagnostic specificity and accuracy and enhances the use of noninvasive techniques to diagnose cardiac fibrosis.<sup>30,40–44</sup>

## Molecular Targets for the Study of New Treatments for Cardiac Fibrosis

Several drugs currently used for treating CVDs, such as angiotensin I-converting enzyme inhibitors (ACEI), AT1 receptor antagonists, and  $\beta$ -blockers, exert an antifibrotic effect.<sup>45</sup> The antifibrotic effect has not been well demonstrated. However, ongoing research is shifting toward more specific therapeutic targets. These molecular targets are shown in Table 2 and discussed here.

**Table 1** Summary of Advantages and Limitations of Imaging Technologies for Cardiac Fibrosis Diagnosis

| Detection Methods                     | Advantages                                                                                                                                                      | Limitations                                                                                                                          |
|---------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------|
| Myocardial biopsy                     | <ul style="list-style-type: none"> <li>• High sensitivity</li> <li>• Certain diagnosis</li> <li>• High specificity</li> </ul>                                   | <ul style="list-style-type: none"> <li>• Invasive sampling</li> <li>• Sampling error</li> </ul>                                      |
| Echocardiography (ECHO)               | <ul style="list-style-type: none"> <li>• Non-invasive</li> <li>• Highly available</li> <li>• Low cost</li> </ul>                                                | <ul style="list-style-type: none"> <li>• Low sensitivity</li> <li>• Low specificity</li> </ul>                                       |
| Magnetic resonance imaging (MRI)      | <ul style="list-style-type: none"> <li>• High resolution</li> <li>• High sensitivity</li> </ul>                                                                 | <ul style="list-style-type: none"> <li>• High cost</li> <li>• Sampling error</li> </ul>                                              |
| Positron emission tomography (PET)    | <ul style="list-style-type: none"> <li>• Non-invasive</li> <li>• High detection sensitivity</li> <li>• Certain diagnosis</li> <li>• High Specificity</li> </ul> | <ul style="list-style-type: none"> <li>• High cost</li> <li>• Uses ionizing radiation</li> <li>• Uses radioactive tracers</li> </ul> |
| Computed tomography/ X-Ray (CT/X-Ray) | <ul style="list-style-type: none"> <li>• Non-invasive</li> <li>• Accessible cost</li> <li>• High resolutions</li> </ul>                                         | <ul style="list-style-type: none"> <li>• A contrast agent is required to detect fibrosis.</li> <li>• Ionizing radiation</li> </ul>   |

**Table 2** Molecular Targets for the Study of New Diagnosis Tools and Treatments of Cardiac Fibrosis

| Target                                            | In vivo Model of Cardiac Injury Induction                                           | Species                                                          | Aim of the Study                                                                                                     | Reference |
|---------------------------------------------------|-------------------------------------------------------------------------------------|------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------|-----------|
| <b>TGF-<math>\beta</math></b>                     | Hereditary progressive cardiac conduction disease.                                  | Scn5a <sup>+/-</sup> mice                                        | To identify the molecular pathway involved in developing fibrosis and prevent its activation.                        | [46]      |
| <b>Smad7</b>                                      | Transverse aortic constriction.                                                     | MFS7KO mice                                                      | To evaluate whether Smad7 induction in the heart restricts TGF- $\beta$ signaling, thereby regulating fibrosis.      | [47]      |
| <b>IL-11</b>                                      | Genetic deletion of the IL-11 receptor gene Il11ra1.                                | C57BL/6 Il11ra1 <sup>-/-</sup> , KO, Il11ra1 <sup>+/-</sup> mice | To evaluate whether IL-11 is a possible therapeutic strategy to treat fibrotic diseases.                             | [48,49]   |
| <b><math>\beta</math>2 - adrenergic receptors</b> | Stimulation with ISO s.c 5 mg/kg/7 days.                                            | BALB/c mice                                                      | To investigate the role of -AR-mediated cardiac inflammation and fibrosis in increasing vascular permeability.       | [50]      |
| <b>ROS</b>                                        | Administration of low doses of ISO (20 mg/kg/day, s.c.) for 10 days.                | Mice                                                             | To evaluate the possible protective effect of nanoceria (NC) in cardiac lesioning.                                   | [51]      |
| <b>Osteopontin</b>                                | Myocardial infarction was induced by left anterior descending artery ligation.      | C57BL/6 mice                                                     | Design a nanosystem for early detection and assessment of myocardial fibrosis.                                       | [52]      |
| <b>Type I Collagen</b>                            | Myocardial infarction was induced by left anterior descending artery ligation.      | C57BL/6 mice                                                     | To evaluate the effect of the nanosystem targeting type I collagen on inhibiting fibrotic scarring.                  | [53]      |
| <b>FAP</b>                                        | Cardiac injury and fibrosis induced by administered AngII/PE with osmotic minipump. | Mice                                                             | To demonstrate the efficacy of redirected T-cell immunotherapy to specifically target pathological cardiac fibrosis. | [54–56]   |

**Abbreviations:** TGF- $\beta$ , transforming growth factor-beta; Smad7, Mothers Against Decapentaplegic homolog 7; IL-11, interleukin 11; ROS, Reactive oxygen species; FAP, fibroblast activation protein; KO, Knockout.

Among these, TGF- $\beta$  is a potent pro-fibrotic factor, making it a key focus of fibrosis-related investigations. In mammals, the three isoforms of TGF- $\beta$  (TGF- $\beta$ 1, TGF- $\beta$ 2, and TGF- $\beta$ 3) compete for the same receptors, leading to potentially antagonistic effects. Notably, TGF- $\beta$ 1 has been implicated in promoting pathological fibrosis, whereas TGF- $\beta$ 3 exhibits antifibrotic effects by reducing collagen production and preventing scar formation.<sup>57</sup> Inhibitors targeting TGF- $\beta$ 1 signaling have been evaluated as potential treatments for cardiac fibrosis. Examples include GW788388, which explicitly inhibits ALK5 and T $\beta$ R1, and pirfenidone and tranilast, which target TGF- $\beta$ 1.<sup>58</sup> However, caution is warranted, as TGF- $\beta$  signaling plays a critical role in the biological processes of repair and homeostasis. Inhibiting this pathway may result in serious adverse effects, including increased mortality, as the reparative response of the heart is also hampered.<sup>59</sup>

Collagen deposition in the ECM is a hallmark of cardiac fibrosis, making collagen an ideal target for nanosystems development. NPs have been studying using collagen as a target.<sup>60</sup> Interleukin-11 (IL-11) and its receptor (IL11RA) are upregulated in fibroblasts following stimulation with TGF- $\beta$ 1. This transcriptional activation is essential for the fibrotic processes to manifest. Inhibition of IL-11 signaling effectively prevents these effects, making it a promising therapeutic target for treating cardiac fibrosis.<sup>48,49</sup> Similarly, the antifibrotic potential of Smad7 – an intracellular inhibitor of TGF- $\beta$  signaling – has been explored in the context of cardiac fibrosis. Murine models of pressure overload have demonstrated that Smad7 attenuates TGF-driven fibrotic pathways, offering another potential strategy for regulating cardiac fibrosis.<sup>47</sup>

Recent studies have identified fibroblast activation protein (FAP) as a reliable marker for cardiac myofibroblasts, as it is uniquely expressed in this cell population.<sup>22,26</sup> Building on its potential, FAP has been successfully utilized in the genetically a vaccine targeting FAP and engineered chimeric antigen receptor (CAR) T cells. Anti-FAP CAR T-cell nanosystems have been developed to recognize and eliminate FAP-expressing cells. This immunotherapy has already gained US Food and Drug Administration (FDA) approval for treating certain types of cancer.<sup>22,54,59,61</sup> A therapeutic vaccine against cardiac fibrosis was also developed. A FAP peptide vaccine was administered to C57BL/6J mice. This vaccine decreased cardiac fibrosis induced by Ang-II treatment. Moreover, it showed no adverse effects in the mice treated.<sup>22</sup> However, applying this technology to CVDs requires extensive evaluation, mainly due to the potential for triggering severe immune responses, such as cytokine release syndrome, which could lead to systemic complications.<sup>55,62,63</sup>

Nanotechnology has become valuable in exploring innovative strategies to prevent and treat cardiac fibrosis. Nanosystems offer significant potential in addressing fibrotic mechanisms by enabling targeted delivery and enhancing therapeutic agents' stability. One promising approach involves using PLGA-b-polyethylene glycol NPs loaded with TGF- $\beta$ 3. This nanosystem protects TGF- $\beta$ 3 from enzymatic degradation and rapid elimination, overcoming the inherent instability of this antifibrotic cytokine. In vitro studies using a human cardiac fibroblast cell line demonstrate the potential effect of TGF- $\beta$ 3 in mitigating fibrosis progression and even preventing its appearance.<sup>46</sup>

It is relevant to analyze the molecular mechanism of NPs targeting cardiac fibrosis, eg, lipid NPs targeting IL-11. The IL11RA1 siRNA-loaded NPs target cells in the heart, interact with the cell membrane, are endocytosed, and release their contents into the cell cytoplasm. Inhibition of IL-11 signaling with anti-IL11 or anti-IL11RA antibodies reduced cardiac fibrosis and cardiac dysfunction induced by pressure overload in a murine model. The non-canonical ERK-mediated GSK3 $\alpha$  pathway, whose activation is IL-11-dependent, was targeted. This is a cardiac fibrosis-inducing pathway independent of canonical TGF- $\beta$ -SMAD2/3 signaling. This strategy of inhibiting IL-11 or its receptor is highly relevant, as IL-11 is up-regulated in tissues from old mice and patient samples. So, without inhibiting the canonical TGF- $\beta$ -SMAD2/3 pathway, cardiac fibrosis can be treated using lipid NPs-IL11RA1 siRNA. Therefore, the advantages of using nanoparticles to develop diagnostic tools and treatments for cardiac fibrosis will be analyzed.<sup>64–66</sup>

Treatments for CVDs have been formulated with large molecules. A new field of research is aimed at designing treatments with disease-regulating RNAs. Non-coding RNAs (ncRNAs) do not code for proteins, but have functions, eg, gene regulation, interacting with mRNAs, regulation of cellular processes, and diseases. The ncRNAs can be microRNA (miRNAs) and long ncRNAs (lncRNAs).<sup>67–70</sup> They can be down- or up-regulated in pathophysiological processes, such as cardiac fibrosis. Researchers have identified miRNAs and lncRNAs that can promote or inhibit cardiac fibrosis (Table 3) in response to myocardial infarction, Ang-II-induced pressure overload, and TGF- $\beta$ .<sup>67,71–74</sup> Thus, miRNAs and lncRNAs are also emerging targets for treatments and biomarkers of cardiac fibrosis. Treatments with ncRNAs have some limitations during their development, such as bioavailability and instability. In addition, some lncRNAs do not

**Table 3** Summary of miRNA and lncRNA That May Inhibit or Enhance Cardiac Fibrosis

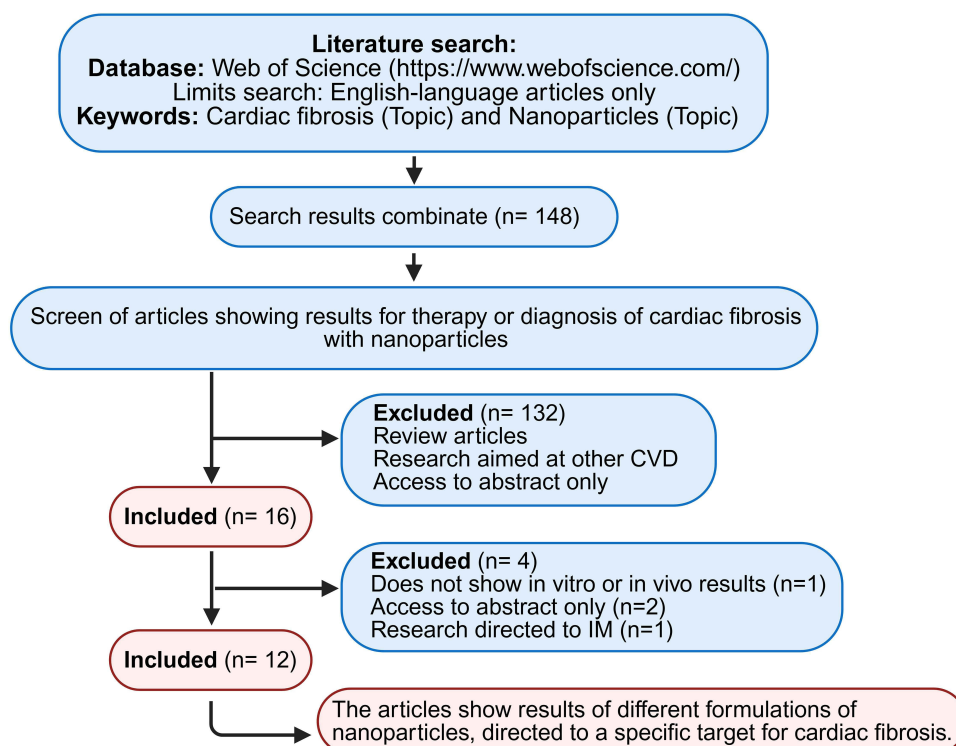
| miRNAs                                                                                   |                                                                  | lncRNAs                                                                                   |                            |
|------------------------------------------------------------------------------------------|------------------------------------------------------------------|-------------------------------------------------------------------------------------------|----------------------------|
| Promote Fibrosis                                                                         | Inhibit Fibrosis                                                 | Promote Fibrosis                                                                          | Inhibit Fibrosis           |
| miR-21<br><b>miR34a</b><br>miR-99b-3p<br>miR-125b<br>miR-130a<br>miR-199a<br>miR-212/132 | miR-26a<br>miR-29<br>miR-30c<br>miR-133<br>miR-Let7i<br>miR-1954 | lncRNA MALAT1<br>lncRNA MEG3<br>lncRNA H19<br>lncRNA WISPER<br>lncRNA PFL<br>lncRNA CHAST | lncRNA SAIL<br>lncRNA MHRT |

**Abbreviations:** miR, microRNA; lncRNAs, long non-coding RNAs.

conserve their sequence between species, which makes their preclinical evaluation difficult. In addition, it is necessary to evaluate their genotoxicity and target them to a specific site for their anti-fibrotic action. LNPs, PNP, and extracellular vesicles are technologies that can provide solutions for using miRNAs and lncRNAs for new and innovative anti-fibrotic treatments. It is essential to determine the therapeutic dose, since inadequate dosage, overexpression, or inhibition of miRNA and lncRNA can lead to adverse effects and undesired reactions resulting from the treatment. We must take advantage of scientific advances while ensuring patient safety.

## Nanoparticles for the Diagnosis and Treatment of Cardiac Fibrosis

We conducted a systematic review to assess the current research on NPs specifically targeting. The flow diagram shown in Figure 2 outlines the pre-established eligibility criteria for the review. Our analysis focused on answering a targeted research topic: “the development and application of NPs for the treatment or diagnosis of cardiac fibrosis”. Of the articles retrieved from literature databases, only 12 met the inclusion criteria defined in our study. Table 4 provides a summary of



**Figure 2** Example of different types of nanoparticle materials. Nanoparticle (NP). Created in BioRender. Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.

**Table 4** Nanoparticles Target Cardiac Fibrosis

| Nanoparticle Type (NP)                                                      | Target                          | Animal Model                                             | Species                                     | NP Dose and Route of Administration                                                    | Biodistribution                                                                                           | Analysis Techniques                                    | Ref        |
|-----------------------------------------------------------------------------|---------------------------------|----------------------------------------------------------|---------------------------------------------|----------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------|--------------------------------------------------------|------------|
| AuNPs-PEG 13 nm                                                             | $\beta 2$ -adrenergic receptors | ISO s.c 5 mg/kg/7 days                                   | Male BALB/c mice 12-week-old                | AuNPs-PEG i.v 810 $\mu\text{g/kg/7d}$                                                  | Specific targeting of 13 nm NP-PEGs to fibrotic heart tissues.                                            | Histology, TEM, IHC, ICP-MS                            | [104]      |
| AuNPs-PEG 13, 30, 50 nm                                                     | $\beta 2$ -adrenergic receptors | ISO s.c 200 mg/kg/3 days                                 | Male FVB/N mice 10–12 weeks old             | AuNPs-PEG i.v 540 $\mu\text{g/kg/3 d}$                                                 | 13 nm AuNPs accumulate more in the heart than 30 and 50 nm AuNPs.                                         | Histology, TEM, IHC, ICP-MS, WB                        | [105]      |
| AuNPs 5, 40, 100 nm                                                         | $\beta 2$ -adrenergic receptors | ISO i.p 20 mg/kg/mL/ 7 days)                             | Male Sprague-Dawley (SD) rats 6–7 weeks old | Au-NPs i.v 5, 40, 100 nm/1d                                                            | 5 nm Au-NPs cardiac protective effect. Low accumulation in the heart.                                     | Histology, TEM, ICP-MS, TUNEL Staining                 | [106]      |
| Mesoporous Silica (FH-NMP-LiMSNs-miR)                                       | Tenascin-C (TN-C)               | I/R injury                                               | C57BL/6 N mice 8-week-old                   | FH-NMP-LiMSNs-miR i.v 200 $\mu\text{L}$                                                | Specific orientation in the heart. Found in the liver and spleen.                                         | Histology, IHC, IF                                     | [107]      |
| Lipid NP Lutein-loaded (LNP) fused to MM (MMLNPs)                           | AP-I/IL-11                      | Transverse aortic constriction                           | Male C57BL/6 N mice 8–10-week-old           | 5 mg/kg i.v (by lutein content)                                                        | Increase circulation time. Increases accumulation in the heart. Found in spleen, liver, lung, and kidney. | Histology, IHC, IF, WB                                 | [108]      |
| Lipid NPs functionalized with a fluorescent probe (OPN@PFP-DiR NPs)         | Osteopontin                     | MI by permanent left coronary artery ligation            | C57BL/6 N mice                              | OPN@PFP-DiR NPs i.v 100 $\mu\text{L}$ (concentration of 100 $\mu\text{g mL}^{-1}$ DiR) | Cardiac orientation. Greater accumulation in the heart and liver.                                         | Ultrasound, infrared, Histology                        | [52]       |
| Liposome (GSL-NPs) functionalized with sonda fluorescent DiR                | Type I collagen                 | MI by left anterior descending artery ligation           | Male C57BL/6 mice (18–22 g)                 | GSL-NPs i.v (1mg/kg)                                                                   | Cardiac orientation. Greater accumulation in the heart.                                                   | Histology, IHC, IF, WB                                 | [53]       |
| Liposome (CNA35-GNR/PFP@PFPNPs) functionalized with sonda fluorescent DiR   | Type I and III collagen         | MI by permanent left anterior descending artery ligation | Male Sprague-Dawley (SD) rats 10-week-old   | CNA35-GNR /PFP@PFPNPs i.v 30 min                                                       | Detected in the heart from 30 minutes to 4 hours post-administration.                                     | Ultrasound, photoacoustics, CT, Histology, IHC, IF, WB | [38]       |
| FAP-CAR mRNA-loaded lipid nanoparticles (LNP)                               | FAP                             | Ang-II through an implanted 28-day osmotic minipump      | C57BL/6                                     | CD5/LNP-FAPCAR i.v 10 $\mu\text{g}$                                                    | Specific orientation in the heart.                                                                        | Flow cytometry, IF                                     | [55,61,62] |
| Polymeric nanocapsules (Mel/CHP@AuNPs) labeled with a fluorescent probe Dil | CHP                             | Transverse aortic constriction.                          | Male Sprague-Dawley rats 6–8-week-old       | 1,5 mL Mel/ CHP@AuNPs-Dil i. v                                                         | Cardiac orientation. Greater accumulation in the heart.                                                   | ICP-MS, RT-PCR, Histology                              | [109]      |
| GNP-TA-Lis                                                                  | ACE                             | –                                                        | Mice                                        | GNP-TA-Lis 1 mg/mL                                                                     | High accumulation in the heart one hour after administration                                              | CT/X-ray                                               | [110]      |

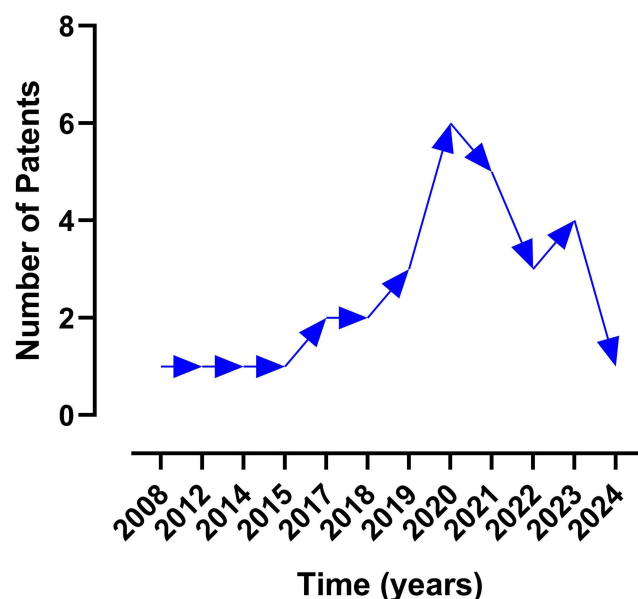
**Abbreviations:** AuNPs, Gold nanoparticles; TEM, Transmission electron microscopy; CT, Computerized tomography; ICP-MS, Inductively coupled plasma mass spectrometry; RT-PCR, reverse transcription-polymerase chain reaction; IHC, immunohistochemistry; IF, immunofluorescence; WB, Western blot; CHP, Cardiac homing peptide; ACE, angiotensin-converting enzyme; Ang-II, angiotensin II; i.v, intravenous via; i.p, intraperitoneal via; ISO, Isoproterenol; FAP, fibroblast activation protein; AP-I, activator protein I; IL-11, interleukin 11.

these investigations. Our findings indicate that while the field of nanotechnology research directed at cardiac fibrosis is active and growing, the number of studies remains limited. This is further evidenced by the relatively small number of patents related to cardiac fibrosis, as shown in Figure 3.<sup>75–103</sup> Among these, very few nanosystems have been explicitly designed for the therapy or diagnosis of cardiac fibrosis. Given the broad range of applications offered by nanotechnology, there is significant potential to expand its use in the biopharmaceutical domain, particularly for addressing this critical area of cardiovascular medicine.

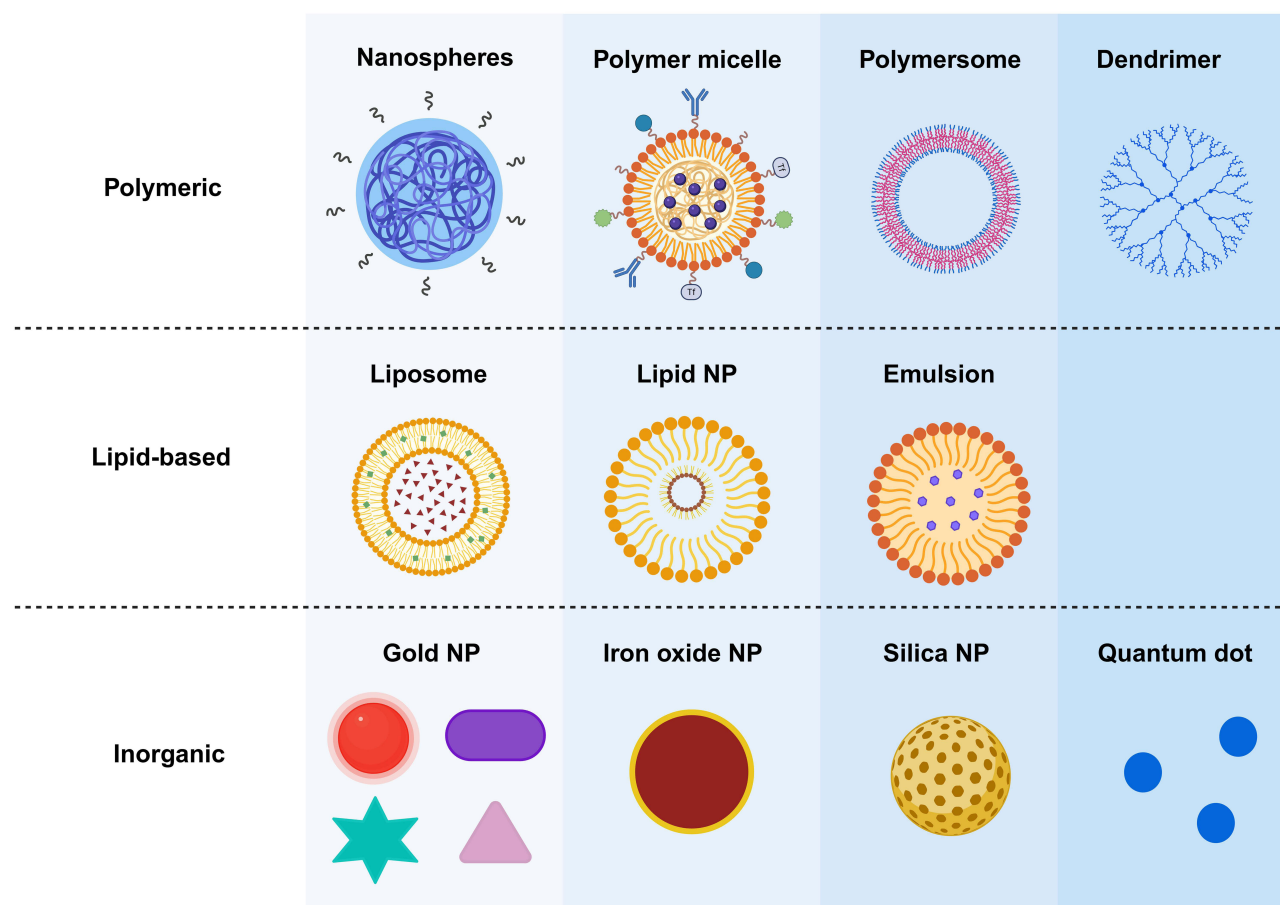
Nanotechnology is a multidisciplinary field that focuses on designing, synthesizing, and characterizing materials with precisely controlled sizes and shapes at the nanoscale ( $10^{-9}$  meters). At this scale, materials exhibit significant physical and chemical properties alterations, including electrical conductivity, mechanical strength, color, elasticity, and chemical reactivity. These unique properties allow for innovative applications, particularly in medicine.<sup>111–114</sup> Nanosystems offer several advantages, such as reducing toxicity, enhancing solubility, improving bioavailability, and increasing the concentration and distribution of therapeutic agents at the site of action. NPs are particularly intriguing due to their high surface-to-volume ratio and reduced size. This ratio amplifies their reactivity and functionality compared to larger particles of the same material. Moreover, NPs exhibit unique optical, magnetic, and electrical properties distinct from their bulk counterparts in solid-state chemistry (micrometer scale). These properties enable precise extracellular tracking and localization, making NPs valuable tools for diagnostics, drug delivery, targeted therapy, and controlled drug release.<sup>115–117</sup> There are diverse types of NPs as shown in Figure 4.<sup>112,117–119</sup> Research on NPs targeting cardiac fibrosis evaluates several types of NPs (Table 4), such as:

- Polymeric nanoparticles (PNPs)
- Lipidic nanoparticles (LNPs)
- Inorganic nanoparticles (AuNPs)

Platforms based on PNPs and LNPs show solid results that have allowed their application at the clinical level. Their scalability and stability have been demonstrated. Their efficacy in solving solubility and bioavailability problems of therapeutic candidates has been demonstrated. There is a considerable amount of research on AuNPs. However, their transfer to the clinic has not advanced as much as that of PNPs and LNPs. Therefore, it is desirable to outline the advantages of AuNPs to boost their clinical application, specifically in cardiac fibrosis. The benefits and limitations of



**Figure 3** Flow diagram for screening research articles aimed at the therapy or diagnosis of cardiac fibrosis with nanoparticles. Created in BioRender: Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.



**Figure 4** Patent applications containing nanoparticles in their composition and cardiac fibrosis.

these types of AuNPs will be described below. In addition, their application in nanomedicine directed to the heart, specifically to the therapy and diagnosis of cardiac fibrosis, will be discussed.

## Polymeric Nanoparticles

PNPs are colloidal particles widely applied in biomedicine. Because they can trap and deliver various molecules, for therapeutic and diagnostic purposes. PNPs are synthesized from the assembly of polymers that form micelles. The micelle structure can be an apolar/hydrophobic or reverse micelle with a polar/hydrophilic core. The polymers can be of natural origin (albumin, gelatin, alginate, chitosan) or synthetic (poly(d,l-lactic-co-glycolic acid) (PLGA), and they are biodegradable polymers. Polyacrylates, which are non-biodegradable polymers, are also used in a highly regulated manner and are at risk of bioaccumulation. PNPs can be synthesized by methods such as: dispersion of preformed polymers (solvent evaporation, nanoprecipitation, emulsification, salting, dialysis, supercritical fluid), polymerization of monomers (emulsion, interfacial polymerization), and coacervation of hydrophilic polymers.<sup>29,120,121</sup>

PNPs can preferentially accumulate in the region of damage by the EPR effect. It can be directed to a specific site and remain for a longer time, compared to non-EPR sites. This effect is also one of the limitations in the preclinical and clinical evolution of engineered nanosystems. The main challenges involve the treatment of reaching the specific target, being retained for the necessary time for the pharmacological effect, and being eliminated from the body. It is required to demonstrate the stability of PNPs over time, the loading capacity, and their sustained release, dependent on physiological conditions. The synthesis components of PNPs are biocompatible and of low toxicity. However, it is essential to consider the PNP administration route. Intramyocardial administration guarantees direct administration into the heart, but causes damage to the cardiac tissue, with the risk of arrhythmias.<sup>29,122</sup> Therefore, the intravenous route is considered less

invasive and safer. One of the most critical issues is to provide evidence that the formulated nanosystem is non-toxic, safe, and effective over time.

PNPs have been developed as innovative approaches to target cardiac fibrosis.<sup>122</sup> One example is a spermine-acetylated dextran (AcDXSp) nanosystem coated with tannic acid (TA) and  $\text{Fe}^{3+}$  ions. This system was designed to encapsulate drugs that stimulate cardiomyocyte proliferation in vitro and reduce the expression of pro-fibrotic genes, such as collagen-I and osteopontin. AcDXSp is used as an encapsulating agent, TA coating increases the retention of the nanocarrier, and  $\text{Fe}^{3+}$  has adhesive properties when coordinated with polyphenolic materials. It can also be applied to fibrotic pathway modulation and fibrosis detection with MRI.<sup>123</sup> Another example is PNPs from chitosan and sodium tripolyphosphate (TPP) to enhance the bioavailability of ginsenoside Rb3 (NpRb3). Ginsenoside Rb3 extracted from *Panax ginseng* has a triterpene structure. It affects the decrease of intracellular  $\text{Ca}^{2+}$  in ischemic-reperfusion-injured PC12 cells.<sup>124</sup> The efficacy of NpRb3 was tested in a rat model of cardiac fibrosis, where it effectively inhibited fibrosis progression. Mechanistic studies revealed that the antifibrotic effects of Rb3 were mediated through its activation of the peroxisome proliferator-activated receptor  $\alpha$  (PPAR $\alpha$ ).<sup>125</sup> It is necessary to demonstrate the safety and non-toxicity of NPPs. Because there are studies documenting that polymers such as chitosan and PLGA induce IL-1 $\beta$  and IL-18 secretion, and neutrophil degranulation, respectively. Furthermore, they tend to accumulate in regions of inflammatory damage, which is not necessarily the target for their therapeutic action.<sup>29</sup>

## Lipid Nanoparticles

LNPs are spherical vesicles widely used as drug vehicles. Their components include ionizable lipids, auxiliary lipids, cholesterol, and polyethylene glycol (PEG).<sup>126,127</sup> There is an excellent platform of LNPs, starting from: liposomes, liposomes encapsulating hydrophobic and hydrophilic drugs, antibody functionalized liposomes, sterically stabilized, and PEG functionalized liposomes. LNPs contain nucleic acids within lipid micelles and nucleic acids sandwiched between lipid bilayers. LNPs synthesis methods include: microemulsions, supercritical fluid, solvent evaporation, microfluidics, nanoprecipitation, high-pressure homogenizers, and ultrasonic homogenization.<sup>128,129</sup>

The FDA has approved several LNPs treatments. LNPs-based research has provided very comprehensive scientific support for clinical applications. Most of these are targeted cancer treatments such as Lipusu and CAELYX.<sup>130–132</sup> Notable applications of LNPs include the carriage of genetic materials such as plasmid DNA, mRNA, siRNA, and ncRNA. This potential has been shown in the treatment of heart failure and myocardial ischemia. What is the rationale for this use? Nucleic acid-based therapies act by expressing therapeutic proteins, editing genes, regulating, or silencing pathogenic genes.<sup>133</sup> However, they present challenges such as immunogenicity, difficulty crossing the cell membrane, scalability, stability, etc. Therefore, the use of LNPs protects mRNAs and ncRNAs from degradation. They facilitate their entry into the cell by endocytosis and cargo release into the cytoplasm in response to stimuli such as temperature, pH variation, magnetic field, and laser irradiation.<sup>129,134</sup>

Some of the applications of LNPs targeting CVDs include the formulation of vascular endothelial growth factor-A (VEGF-A) mRNA for the regenerative treatment of heart failure.<sup>135–137</sup> The formulation of LNP-mRNAs encoding Relaxin se (NCT05659264) to treat heart failure has also been taken to the clinical phase.<sup>133</sup> A thermosensitive nanosystem was synthesized, composed of liposomes loaded with angiotensin-(1-9) [Ang-(1-9)] and coated with AuNPs. With this nanosystem, it was possible to increase the half-life of the Ang-(1-9) peptide and its controlled and targeted release in the heart. Offering a technological solution to use Ang-(1-9) in treating hypertension and cardiac remodeling.<sup>138</sup> LNPs' vehiculization reduces cardiotoxicity, increases solubility, and circulation time. Allows scaling up formulations from laboratory to industrial scale. LNPs can be applied to the proteins identified in this review as targets for therapy and diagnosis of cardiac fibrosis. For example, LNPs loaded with siRNA against BRD4 were synthesized to inhibit CFs activity after cardiac injury. The LNPs were conjugated with anti-FAP antibodies to target activated CFs. This preclinical investigation demonstrated that the lipid nanosystem inhibited CFs activation, reduced fibrosis, and improved cardiac function after myocardial infarction. In addition, its safety and toxicity were evaluated in primates.<sup>139</sup>

## Gold Nanoparticles

AuNPs are among the most prominent inorganic NPs utilized in clinical applications due to their versatility and biocompatibility. They can be synthesized in various shapes, such as spheres, rods, stars, and cubes, with sizes ranging from 1 to 100 nm.

The size and shape of AuNPs significantly influence their optical properties, making them highly adaptable for diverse applications. These NPs can be coated with compounds such as polyethylene glycol, amino acids, proteins, and charged chains (positive, negative, neutral), and improve stability and function. Moreover, AuNPs can be functionalized with various biomolecules for targeting and therapeutic purposes, including antibodies, peptides, drugs, radioisotopes, genes, and carbohydrates, tailoring them for specific biomedical uses. Their exceptional biocompatibility and tunable surface properties have enabled their integration into cutting-edge medical technologies. AuNPs have been applied as optical biosensors, drug delivery platforms, components of laser-based therapies, and contrast agents for advanced imaging techniques.<sup>113,115,140,141</sup>

AuNPs exhibit an intrinsic property called surface plasmon resonance (SPR) or plasmon effect. This optical phenomenon exploits the interaction between an electromagnetic wave and conduction band electrons ( $e^-$ ) ("plasmons").<sup>115,142</sup> The collective oscillation of the  $e^-$  on the surface of the metal NPs, known as SPR, can be observed within the visible region of the spectrum. The electric field of the incident light on the AuNPs induces an electric dipole in the particle. The  $e^-$  moves through the electric field and returns to its basal state. In this process, energy is emitted, part of which is transformed into local heat, generating a photothermal effect. This thermal energy can be used for the selective destruction of tumor cells, disaggregation of toxic aggregates, recognition of biomolecules, and targeted and controlled drug delivery.<sup>114,115,143,144</sup>

## Potential Clinical Applications of AuNPs in Cardiac Fibrosis

These findings underscore the biocompatibility of AuNPs-PEGs under physiological and pathological conditions in the heart,<sup>50</sup> along with their potential cardioprotective effects,<sup>145</sup> raising considerable interest in applications in diagnosis, treatment, and theranostics of CVDs. Innovative examples include a gold nanosystem based on theranostic liposomes loaded with Ang-(1-9) and functionalized with the cardiac targeting peptide IMTP to promote site-directed delivery of this therapeutic agent to ischemic regions in a preclinical ex vivo model.<sup>138,145</sup> In addition, in clinical studies, AuNPs functionalized with specific antibodies have been successfully employed to quantify cardiac biomarkers such as myoglobin and cardiac troponin.<sup>146,147</sup>

One of the primary mechanisms AuNPs can exert their therapeutic effects is by modulating inflammatory responses associated with cardiac fibrosis. Research has shown that citrate-stabilized AuNPs can downregulate IL-1 $\beta$ -induced pro-inflammatory responses, pivotal in the pathogenesis of cardiac fibrosis.<sup>148</sup> By selectively targeting inflammatory pathways, AuNPs can potentially reduce the fibrotic response in cardiac tissue in response to chronic cardiac injuries. In addition to their anti-inflammatory effects, AuNPs can be engineered for targeted delivery of therapeutic agents. For example, biomimetic NPs composed of platelet and erythrocyte membranes have been developed to deliver JQ1. This small molecule inhibitor targets bromodomain and extra-terminal domain (BET) proteins specifically to cardiac fibroblasts in animal models of heart failure.<sup>149</sup> This targeted approach enhances the treatment's efficacy and minimizes systemic side effects.

Applying AuNPs in imaging techniques also holds promise for assessing subclinical cardiac fibrosis. Integrating AuNPs into imaging protocols can enhance the contrast and specificity of these techniques, allowing for better visualization of fibrotic tissue and early diagnosis and monitoring of disease progression in patients with heart failure. Moreover, by combining serum biomarkers of extracellular matrix turnover<sup>150</sup> with AuNP-based imaging techniques, clinicians may better understand a patient's fibrotic status and tailor treatments accordingly.

To address these challenges, alternative strategies have been developed to enhance the stability and biocompatibility of therefore. One use as a practical approach involves functionalizing AuNPs with PEG.<sup>151</sup> PEG provides excellent stability due to its steric effect in physiological environments and significantly reduces the toxicity associated with cetyltrimethylammonium bromide (CTAB<sup>+</sup>). It can also be used as a bridge to bind other molecules, peptides, antibodies, etc, and to direct the nanosystem to a specific target.<sup>113,152–154</sup> Key factors such as the shape, size, and surface charge of AuNPs significantly influence their arrival time, cellular uptake, and retention within different organs.<sup>111</sup> PEGylation also increases the circulation half-life of the NPs. When NPs reach the blood circulation, they can be recognized by red blood cells, white blood cells, platelets, plasma proteins, immunoglobulins, and complement proteins. In general, this interaction promotes the opsonization of NPs by the mononuclear phagocytic system and the secretion of proinflammatory cytokines. Therefore, the functionalization of NPs with PEG decreases immune recognition. Thus, PEG-NPs evade one of the major causes that prevent the delivery of NPs to their target site, so that they accumulate for the necessary time before being eliminated.<sup>29,155,156</sup>

Studying the biodistribution and toxicity of nanosystems under both physiological and pathological conditions is crucial to ensuring their safety and efficacy.<sup>105,157</sup> Preclinical toxicity studies, often conducted in mice and rats due to their high percentage of genetic homology with humans, provide valuable insights into the safety profile of AuNPs. These NPs have been administered via various routes, including intraperitoneal (i.p), intravenous (i.v), and intranasal methods. Results indicate that AuNPs do not cause acute damage to major organs such as the blood, liver, spleen, kidneys, testes, thymus, heart, lungs, or brain. Additionally, studies involving pregnant rats demonstrated that while AuNPs can cross the placental barrier, they do not cause harm to the offspring, further supporting their potential for safe biomedical applications.<sup>158,159</sup>

In murine studies, AuNPs size determines which organs are reached and in which organs they accumulate. Very small AuNPs (5 nm) have a low accumulation in the heart and are cleared from circulation *in vivo*.<sup>160</sup> Slightly larger AuNPs (10–15 nm) are detectable in the blood, liver, spleen, kidneys, testis, thymus, heart, lungs, and brain after i.v injection.<sup>161</sup> In contrast, larger AuNPs (100–250 nm) are primarily located in the blood, liver, and spleen.<sup>162</sup> After a low dose of lipopolysaccharide (LPS) and AuNPs 20, 100, and 500 nm were administered, it was demonstrated that systemic inflammation affects the biodistribution of AuNPs, and the distribution depended on the size of the AuNPs.<sup>163–165</sup> The accumulation of AuNPs-PEG (10, 30, and 50 nm) after repeated doses was studied in mouse hearts treated with isoproterenol (ISO), a  $\beta$ 2-adrenergic receptor agonist. ISO increases vascular permeability and produces inflammatory infiltration and fibrosis. Treatment with AuNPs in healthy mice did not induce inflammation, cardiac hypertrophy, or fibrosis and did not affect cardiac function. For example, 10 nm AuNPs-PEGs administered for two weeks induced reversible cardiac hypertrophy.<sup>105,166,167</sup> The internalization of AuNPs was studied in BALB/3T3 mouse fibroblast cell models treated with 5–15 nm AuNPs for 72 h. AuNPs were internalized by cell membrane invaginations without caveolin intervention and deposited in vesicles without entering other organelles. They reach the end of the endo/lysosomal pathway at 2 h. AuNPs of 5 nm ( $\geq 50 \mu\text{M}$ ) are cytotoxic to cardiac fibroblasts, but not those of 15 nm. This result reaffirms the importance of NPs size for cellular response and its biodistribution.<sup>106</sup>

## Pharmacokinetics, Biodistribution, and Toxicology of NPs

Several factors determine the pharmacokinetics and biodistribution of NPs, eg, route of administration, type of NPs, and physicochemical properties (shape, size, and surface chemistry). Shape and size determine the circulation time of NPs in the bloodstream and the elimination of half-life.<sup>106</sup> These factors determine their interaction with biological systems, cellular uptake, interaction with plasma proteins, and accumulation in tissues. NPs can cross cell membranes by active (endocytosis, phagocytosis) and passive transport mechanisms. NPs type, shape, size, charge, and surface chemistry are determining factors that favor an internalization mechanism.<sup>168</sup> Excretion of nanoparticles from the cellular interior is key to avoiding their accumulation and possible harmful effects on cells. However, rapid exocytosis may reduce their efficacy, while longer permanence may benefit drug delivery.<sup>169</sup> In the context of cardiac fibrosis, NPs can accumulate in the tissue by several mechanisms, for example, passive targeting, due to increased vascular permeability in the fibrosis zone of the heart, mediated by overstimulation of  $\beta$ -adrenoceptors.<sup>170</sup> As mentioned, NPs accumulate in fibrotic tissue due to the EPR effect, inflammation, and oxidative stress in cardiac fibrosis.<sup>50</sup> They can also accumulate by an active targeting mechanism, eg, NPs conjugated with a specific ligand, for recognition and interaction with overexpressed receptors in the fibrotic tissue, eg, AT1 receptor, FAP, collagens I/III, IL-11, etc.<sup>50,139,145–148</sup>

The metabolism of NPs involves their transformation and decomposition in the organism. Understanding the metabolic pathways of NPs is essential to predict their result in biological systems. Factors such as the surface chemistry of NPs can influence their metabolization and accumulation in tissues.<sup>149</sup> NPs can be sensitive to degradation by enzymes such as matrix metalloproteinases (MMP-2 and MMP-9). They can be retained in cardiac tissue for days before being eliminated. *In vitro* biotransformation studies in primary human fibroblasts have shown that AuNPs do not remain intact in the tissue. They can be degraded over time in lysosomes by the action of ROS (Reactive oxygen species). Moreover, this degradation was dependent on the size of the AuNP.<sup>150</sup> *In vitro*, treatment of hepatocytes with AuNPs results in a time- and dose-dependent increase in ROS production. ROS induces cell damage, affecting proteins, DNA, membranes, and organelles. It is necessary to study the potential toxicity of NPs, knowing that large-sized NPs accumulate in organs such as the liver, spleen, and kidneys, and that small-sized NPs can be taken up by cells.<sup>171</sup> These results reinforce the need to conduct long-term studies for AuNP treatments.

Finally, the clearance or process by which NPs are eliminated from the body is determined by the physicochemical properties of the NPs. Smaller NPs can be eliminated faster than larger ones. They can be filtered by the kidneys or absorbed by the reticuloendothelial system (RES).<sup>50</sup> NPs smaller than 6 nm can be eliminated through the urine, using the renal route, in a short time (hours). NPs larger than 6 nm, which are degradable, will be cleared by the hepatobiliary and renal routes. The clearance can occur from hours to weeks. On the other hand, very large NPs, which are not degradable, can be retained for a long time (months) by the mononuclear-phagocyte system.<sup>112,170</sup> It is known that NPs can be retained in cardiac tissue for days before they are eliminated. Therefore, it must be demonstrated that NP treatments are not toxic in the long term.

NPs have been the subject of intense research to evaluate their pharmacokinetic, biopharmacological, and toxicological profiles. To ensure their safety and efficacy in humans. AuNPs can be synthesized in different shapes and sizes, each with unique properties and applications. For example, spherical AuNPs are stable in a colloidal state, with sodium citrate commonly used as a stabilizing agent. In contrast, gold nanowires are stabilized in solution using CTAB<sup>+</sup>. However, these nanowires are unstable in culture media and when administered in vivo. Moreover, CTAB<sup>+</sup> is known to be toxic to human organisms, presenting a significant limitation for biomedical applications.<sup>172</sup> The toxicity of NPs depends mainly on their composition and size. The more biodegradable and biocompatible they are, the lower the risk of accumulation and long-term toxicity. It is also important to know and monitor the degradation of products resulting from the metabolism of NPs. It is necessary to have a better toxicokinetic profile of NPs to ensure the safe long-term use of NPs.<sup>173</sup>

## Limitations and Prospects for the Clinical Use of Nanoparticles for Cardiac Fibrosis

It is important to note that currently, 350 clinical studies are using NPs in various diseases.<sup>174</sup> Only three studies involve the word “fibrosis” but these trials are focused on liver fibrosis. Thus, we still have a long way to go before using NPs in the context of cardiac fibrosis can be a reality. Several factors influence the translation of nanodrugs to the clinic. Among them are high research costs, patent processes, and established regulations. Regulatory agencies such as the FDA, European Medicines Agency (EMA), and the Department of Biotechnology, Government of India (DBT), have established standards and guidelines for evaluating NPs in biomedicine.<sup>175</sup> To apply for registration with the regulatory agency, data must be provided on: Scientific rationale for nanodrug development, detailed description of all components, physicochemical characterization, titration methods, in vitro drug dissolution/release methods for quality testing, manufacturing process, and process control. In addition, stability studies, animal pharmacology data, animal toxicology data, and clinical trial data are required. The central objective is to ensure safety in the use of NPs. As described in this review, research exists for the therapy and diagnosis of cardiac fibrosis.

Large-scale production of NPs faces significant technical hurdles, eg, reproducibility of synthesis protocols, aggregation, contamination, and particle degradation. This interferes with throughput and production costs. The scale-up process aims to increase the size of production batches to support higher demands. Equipment scale-up mass and energy balance calculations are crucial in the scale-up of nanopharmaceutical production. However, there are no standardized optimization parameters to ensure uniformity of NPs. Titration methods for nanopharmaceuticals are not always established, and stability studies are limited.<sup>176,177</sup> These are obstacles to the acceptance of NPs in the pharmaceutical industry. Therefore, innovative strategies, such as green nanotechnology, are required to improve economic efficiency and reduce environmental impact.<sup>171,178,179</sup> These are challenges for nanomedicine to increase its use in the biopharmaceutical industry.

We might think that these data are sufficient, but they are still accumulating chemical, scale-up, pharmacological, and toxicokinetic data to be evaluated in clinical trials. Considering the decrease in size at the nm scale, systemic evaluation is necessary to demonstrate that it is not toxic for the organism. Although treatments using nanoparticles have already been approved, the regulations for their use are still being written. Additionally, pharmacovigilance and post-commercialization studies are pending tasks to demonstrate that the approved treatments are safe over time.<sup>180–183</sup>

It should be noted that there are clinical results of NPs for plasmonic photothermal therapy of atherosclerosis (NCT01270139). Long-term results demonstrate the safety of NANOM and the decrease in cardiovascular events.<sup>184,185</sup> Still, it is also an opportunity to develop this field further. Nevertheless, it is also essential to acknowledge the limitations that impede the translation from bench to bedside. Some chemicals used to synthesize AuNPs can harm the environment.<sup>186</sup> Critical knowledge gaps regarding AuNPs biodistribution, pharmacokinetics, and toxicological

profile remain.<sup>187</sup> Regarding this point, long-term studies using large animal models may provide valuable insights into these aspects. AuNPs offer a wide range of nanosystems that can be designed for specific targeting. Table 2 describes targets for the development of cardiac fibrosis. AuNPs targeting FAP, osteopontin, Collagen-I, and TGF- $\beta$  can be synthesized for diagnosis, therapy, or theranosis of cardiac fibrosis (Figure 5).

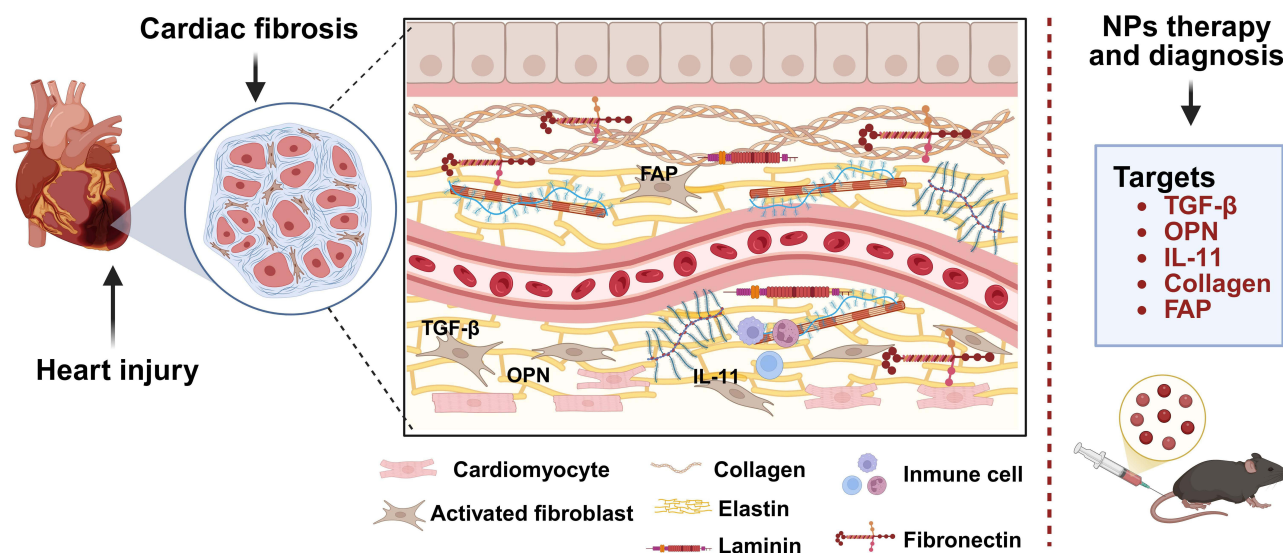
While it is crucial to overcome these challenges to harness the theranostic capacity of NPs, we have come a long way, with significant scientific advances in the last decades. In 1959, Richard Feynman gave a lecture called “There is Plenty of Room at the Bottom” that marked the origin of nanoscience.<sup>188</sup> To this day, nanomaterials have countless applications in our lives. The development and applications of nanotechnology have been so significant that it has become a tool for nanomedicine.

Nanotechnology has been applied in the rational design of drugs. Some milestones are targeted NPs with action in the heart, and their biodistribution and accumulation studies. NPs for cardiac reprogramming and regeneration, and the use of NPs-RNA to treat and detect CVDs, eg, cardiac infarction and cardiac fibrosis.<sup>38,104,105,107,111,160,189</sup> We can use it innovatively and consciously to diagnose and treat CVDs. The limits of the future development of nanomedicine will be our intellect. So, we can take advantage of the properties of nanomaterials and different types of NPs to provide innovative solutions to health problems. Figure 6 shows the evolution in the use of NPs targeting cardiac fibrosis. We must comply with ethical principles for the safe and effective use of nanosystems.

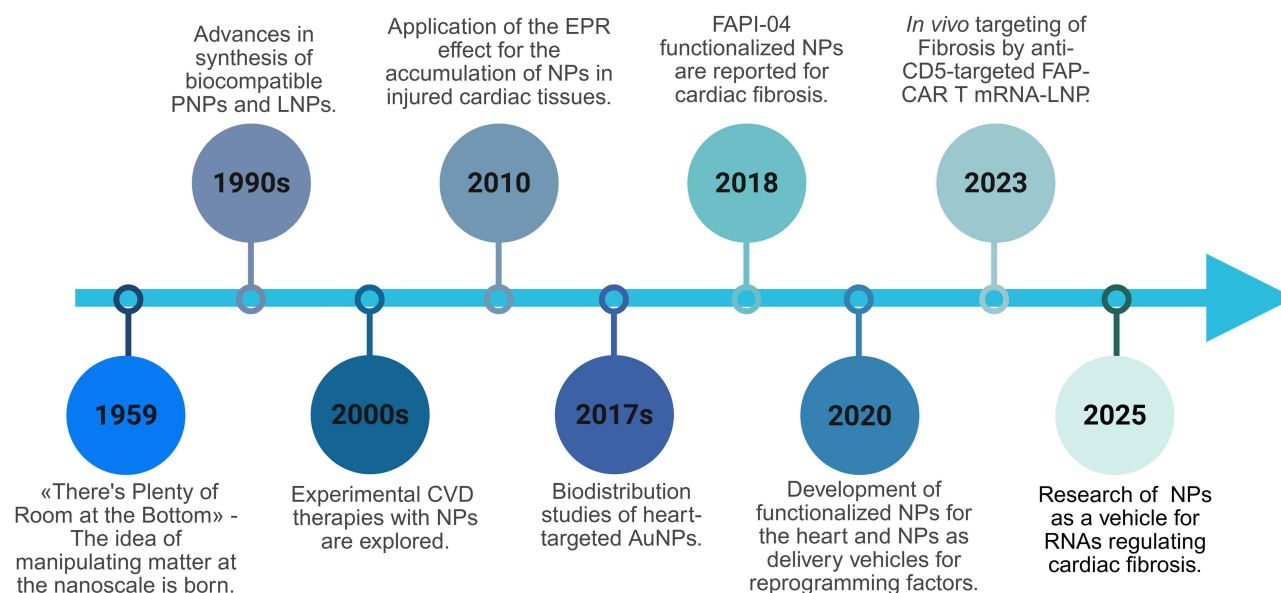
Overall, various challenges need to be addressed before nanotechnology can be used in the clinical arena. An important first step should be an increase in the quantity and quality of preclinical studies. These findings can be translated from bench to bedside. Small and large animal models of CVDs, such as hypertension or post-myocardial infarction, heart failure, may be used to assess the diagnostic accuracy of NPs. Showing more sensitivity than current imaging methods is essential to establish the potential superiority of NPs for early diagnosis of cardiac fibrosis. Moreover, the use of NPs for drug delivery to treat cardiac fibrosis must show increased efficacy and safety compared with the current anti-cardiac remodeling pharmacology. In other words, future preclinical studies must consider a comparison with currently available treatments and diagnostic tools.

## Future Directions and Emerging Trends in the Field of Nanotechnology for Cardiac Fibrosis

There are emerging technologies for the application of cardiac fibrosis. Fields such as transcriptomics and proteomics have identified transcripts and proteins that regulate the molecular mechanisms of cardiac fibrosis.<sup>23</sup> They have demonstrated



**Figure 5** Cardiac fibrosis can be detected and treated with nanoparticles directed to specific targets. Stress in the heart induces a fibrotic response. It involves the activation of Transforming growth factor-beta (TGF- $\beta$ ), osteopontin (OPN), fibroblast activation protein (FAP) immune cells, and collagen secretion. Nanosystems can be designed and synthesized for drug delivery to the pharmacological target. Created in BioRender. Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.



**Figure 6** Nanoscience and nanomaterials have evolved and are being investigated for application in CVD and cardiac fibrosis for therapy and diagnosis. Created in BioRender. Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.

convergence in the response of cardiac fibroblasts from different sources (adult primary CFs, fetal primary CFs, and induced pluripotent stem cell-derived CFs (hiPSC-CF), stimulated with TGF- $\beta$ 1. In all three cell types, the stimulus induced activation, differentiation, and migration of cardiac fibroblasts and remodeling of the ECM. This reaffirms the complexity in the signaling pathways of the fibrotic response, where TGF- $\beta$ 1, SMAD3, SMAD4, non-coding RNA activated by DNA damage (NORAD), insulin-like growth factor 1 (IGF1), extracellular signal-regulated kinase (ERK), and mitogen-activated protein kinase 1 (MAPK1) are upregulated. In addition, the role of protein-protein interactions in maintaining fibroblast activation is discussed. Targets such as ITGAV, CCN2, FAP, FN1, ITGB5, MMP2, SDC1, SERPINE1 and TNC are highlighted, which can modulate the phenotype of fibrosis, and create new treatments and means for its diagnosis.<sup>23,190–192</sup>

Complementary to these tools, other gene editing tools such as CRISPR/Cas have been used. CRISPR/Cas is based on the bacterial immune system. Researchers identified spaced DNA sequences from viruses, known as CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats). So, bacteria can defend themselves against viruses by this CRISPR copy of the virus, and with the action of the Cas cleavage protein. Based on this mechanism, the scientists decided to apply CRISPR/Cas to make gene editing in other cell types and apply it to disease therapies. The system contains the unique guide RNA (sgRNA), complementary to the DNA sequence to be recognized, and Cas9 (one of the most widely used Cas) makes two cuts in the DNA. The DNA double strand is broken, and the host cell binds and repairs the DNA.<sup>133</sup>

Thus, CRISPR/Cas9 has been used in genomic editing and epigenetic modifications. To remove, insert, or inhibit the expression of specific genes in the cardiovascular field.<sup>193,194</sup> Cellular and animal models have been developed, using genomic editing of cardiac cells, for example, pathogenic mutations in natural iPSCs and correction of mutations in iPSCs from diseased patients. Transgenic Myh6-Cas9 mouse models were administered with AAV-sgRNA-adenovirus (AAV), and CASA AV mice were administered with AAV-sgRNA-cTnT-Cre. Both approaches were effective in achieving cardiomyocyte-specific gene deletion. The CRISPR-Cas technique has been applied in the *in vivo* delivery of CRISPR-Cas components to *in vivo* therapy.<sup>195,196</sup> For the latter application, CRISPR-Cas delivery systems are needed. Thus, non-viral NPs (LNPs, PNPs, inorganic NPs) are one strategy to deliver DNA, mRNA, or protein from Cas9 and sgRNA into cells. LNPs are the most widely used due to their stability in plasma, low immunogenicity, more cost-effective production than AVV, and cellular internalization capacity. However, we must continue to address limitations such as limited biodistribution and high accumulation in the liver and spleen.<sup>196,197</sup>

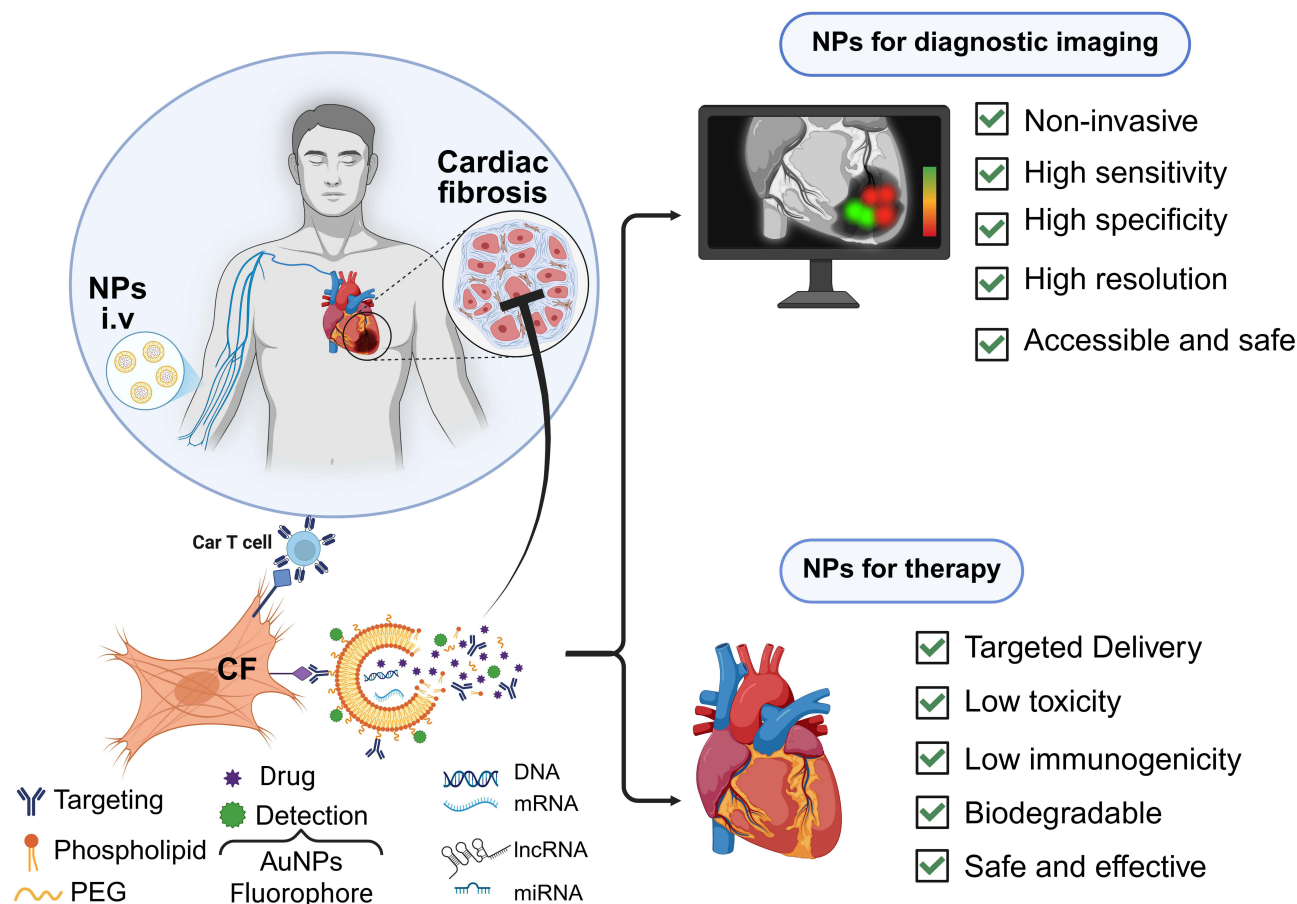
Although CRISPR-Cas technology is relatively low-cost, simple, and accurate, it must be considered that the specificity of the CRISPR-Cas chain is not total. Therefore, one of its limitations is precisely that it can induce cuts in other undesired sites. We must be rigorous in the studies and regulations that allow its safe and ethical use. To achieve greater efficacy in NPs-

CRISPR-Cas therapy, it is necessary to address specific targets on the cell surface of cardiomyocytes and cardiac fibroblasts.<sup>196</sup> The application of CRISPR-Cas for cardiac fibrosis has been limited so far. However, some research can be cited, eg, CRISPR-Cas9-mediated inactivation of the miR34a gene (decreased fibrosis, enhanced proliferation of cardiomyocytes, and improved heart function),<sup>198</sup> and CRISPR-Cas-mediated reprogramming of fibroblasts to cardiovascular progenitor cells. Differentiation of reprogrammed fibroblasts to endothelial cells, cardiomyocytes, and smooth muscle cells was achieved. In addition, scar size was reduced, and cardiac function was restored in a preclinical model of myocardial infarction.<sup>199</sup> We must apply the new tools described in research aimed at cardiac fibrosis and consider their advantages and limitations.

Due to the results obtained, it is important to mention the CAR T-cell engineered therapy again. FAP-CAR-T LNPs contain mRNA encoding a CAR and have a functionalized CD5 antibody to target the LNPs to T lymphocytes. Thus, the CAR binds to FAP which is expressed on activated cardiac fibroblasts. This nanosystem reduced fibrosis and restored cardiac function in preclinical assays.<sup>54,63,133,200,201</sup> Genetic engineering of T cells combined with nanotechnology has offered encouraging results targeting cardiac fibrosis.

## Conclusions

Due to their targeted drug delivery capabilities and potential applications in advanced imaging techniques, NPs represent a promising approach to address cardiac fibrosis from a diagnostic and theragnostic standpoint (Figure 7). However, its clinical application still faces significant challenges, such as the need for further evidence on its safety, toxicity, pharmacokinetics, and long-term efficacy. In addition, regulations are still under development, and studies in cardiac



**Figure 7** Nanoparticle platforms are a technology that can potentially target the diagnosis and therapy of cardiac fibrosis. Nanoparticles can be designed to recognize specific components of fibrotic tissue, allowing their selective accumulation in the damaged heart. Lipid and polymeric nanoparticles can carry therapeutic or imaging agents, facilitating targeted delivery strategies and monitoring techniques such as CT, X-ray, PET, or MRI, or multimodality imaging. Nanoparticle (NPs), intravenous via (i.v.), gold nanoparticle (AuNPs). Created in BioRender. Kogan, M. (2025) <https://BioRender.com/bu3pdqm>.

fibrosis are scarce compared to other pathologies. Technological advances, the design of selective and stable NPs in the body, and positive results in preclinical models reinforce the potential of NPs as key tools for effective and safe personalized medicine targeting cardiac fibrosis. Early detection of cardiac fibrosis due to risk factors such as age and hypertension can reduce CVD mortality, where fibrosis is a worse prognostic risk factor. The advantages of NPs can be applied to develop tools for early diagnosis and effective treatment of cardiac fibrosis.

## Acknowledgments

The authors thank ANID-FONDECYT grants number 1251140 and 1240443, Anillo ACT 210068, ANID-FONDAP grant number 15130011, FONDAP 1523A0008, and ANID National PhD number 21200473.

## Author Contributions

All authors contributed significantly to the conception, execution, data acquisition, analysis, and interpretation of the reported work. They have participated in the drafting and critical revision of the article. They have given their final approval for the version to be published. They have decided on the journal to which the article has been submitted. They agree to be responsible for all aspects of the work.

## Disclosure

The authors have declared that no competing interest exists.

## References

1. Roth GA, Mensah GA, Johnson CO, 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. doi:10.1016/j.jacc.2020.11.010
2. Joynt Maddox KE, Elkind MSV, Aparicio HJ, et al. Forecasting the burden of cardiovascular disease and stroke in the United States through 2050—prevalence of risk factors and disease: a presidential advisory from the American heart association. *Circulation*. 2024;150(4). doi:10.1161/CIR.0000000000001256.
3. Martin SS, Aday AW, Almarzooq ZI, et al. 2024 heart disease and stroke statistics: a report of US and global data from the American heart association. *Circulation*. 2024;149(8).
4. Martin SS, Aday AW, Allen NB, et al. 2025 heart disease and stroke statistics: a report of US and global data from the American heart association. *Circulation*. 2025;151(8). doi:10.1161/CIR.0000000000001303
5. Floris E, Cozzolino C, Marconi S, et al. A review of therapeutic strategies against cardiac fibrosis: from classical pharmacology to novel molecular, epigenetic, and biotechnological approaches. *Rev Cardiovasc Med*. 2023;24(8):226. doi:10.31083/j.rcm2408226/htm
6. Mamidi N, Franco De Silva F, Orash Mahmoudsalehi A. Advanced disease therapeutics using engineered living drug delivery systems. *Nanoscale*. 2025;17:7673–7696. doi:10.1039/D4NR05298F
7. Bertaud A, Joshkon A, Heim X, et al. Signaling pathways and potential therapeutic strategies in cardiac fibrosis. *Int J Mol Sci*. 2023;24:1756. doi:10.3390/ijms24021756
8. Travers JG, Kamal FA, Robbins J, Yutzey KE, Blaxall BC. Cardiac fibrosis: the fibroblast awakens. *Circ Res*. 2016;118:1021–1040. doi:10.1161/CIRCRESAHA.115.306565
9. Kurose H. Cardiac fibrosis and fibroblasts. *Cells*. 2021;10:1716. doi:10.3390/cells10071716
10. Chen Z, Zheng L, Chen G. 2-Arachidonoylglycerol attenuates myocardial fibrosis in diabetic mice via the TGF- $\beta$ 1/smad pathway. *Cardiovasc Drugs Ther*. 2023;37(4):647–654. doi:10.1007/s10557-021-07307-7
11. Jin L, Zhang J, Deng Z, et al. Mesenchymal stem cells ameliorate myocardial fibrosis in diabetic cardiomyopathy via the secretion of prostaglandin E2. *Stem Cell Res Ther*. 2020;11(1):122. doi:10.1186/s13287-020-01633-7
12. Li CJ, Lv L, Li H, Yu DM. Cardiac fibrosis and dysfunction in experimental diabetic cardiomyopathy are ameliorated by alpha-lipoic acid. *Cardiovasc Diabetol*. 2012;11(1):73. doi:10.1186/1475-2840-11-73
13. Zheng D, Dong S, Li T, et al. Exogenous hydrogen sulfide attenuates cardiac fibrosis through reactive oxygen species signal pathways in experimental diabetes mellitus models. *Cell Physiol Biochem*. 2015;36:917–929. doi:10.1159/000430266
14. Zheng X, Peng M, Li Y, et al. Cathelicidin-related antimicrobial peptide protects against cardiac fibrosis in diabetic mouse heart by regulating endothelial-mesenchymal transition. *Int J Biol Sci*. 2019;15:2393–2407. doi:10.7150/ijbs.35736
15. Passaro F, Tocchetti CG, Spinetti G, et al. Targeting fibrosis in the failing heart with nanoparticles. *Adv Drug Deliv Rev*. 2021;174:461–481.
16. Schimmel K, Ichimura K, Reddy S, Haddad F, Spiekerkoetter E. Cardiac fibrosis in the pressure overloaded left and right ventricle as a therapeutic target. *Front Cardiovascul Med*. 2022;9. doi:10.3389/fcvm.2022.886553
17. Sweeney M, Corden B, Cook SA. Targeting cardiac fibrosis in heart failure with preserved ejection fraction: mirage or miracle? *EMBO Mol Med*. 2020;12(10). doi:10.15252/emmm.201910865
18. Frangogiannis NG. Cardiac fibrosis. *Cardiovascul Res*. 2021;117:1450–1488. doi:10.1093/cvr/cvaa324
19. Vasquez C, Benamer N, Morley GE. The cardiac fibroblast: functional and electrophysiological considerations in healthy and diseased hearts. *J Cardiovasc Pharmacol*. 2011;57(4):380–388. doi:10.1097/FJC.0b013e31820cda19
20. Aguado-Alvaro LP, Garitano N, Pelacho B. Fibroblast diversity and epigenetic regulation in cardiac fibrosis. *Int J Mol Sci*. 2024;25:6004. doi:10.3390/ijms25116004

21. Talman V, Ruskoaho H. Cardiac fibrosis in myocardial infarction—from repair and remodeling to regeneration. *Cell Tissue Res.* **2016**;365:563–581. doi:10.1007/s00441-016-2431-9
22. Yoshida S, Hayashi H, Kawahara T, et al. A vaccine against fibroblast activation protein improves murine cardiac fibrosis by preventing the accumulation of myofibroblasts. *Circ Res.* **2025**;136:26–40. doi:10.1161/CIRCRESAHA.124.325017
23. Qi R, Lin E, Song J, Wang Y, Lin L. Proteomic insights into cardiac fibrosis: from pathophysiological mechanisms to therapeutic opportunities. *Molecules.* **2022**;27:8784. doi:10.3390/molecules27248784
24. Leask A. Getting to the heart of the matter. *Circ Res.* **2015**;116:1269–1276. doi:10.1161/CIRCRESAHA.116.305381
25. Fu X, Liu Q, Li C, Li Y, Wang L. Cardiac fibrosis and cardiac fibroblast lineage-tracing: recent advances. *Front Physiol.* **2020**;11. doi:10.3389/fphys.2020.00416
26. Liu M, López de Juan Abad B, Cheng K. Cardiac fibrosis: myofibroblast-mediated pathological regulation and drug delivery strategies. *Adv Drug Deliv Rev.* **2021**;173:504–519. doi:10.1016/j.addr.2021.03.021
27. Prabhu SD, Frangogiannis NG. The biological basis for cardiac repair after myocardial infarction. *Circ Res.* **2016**;119:91–112. doi:10.1161/CIRCRESAHA.116.303577
28. Lafuse WP, Wozniak DJ, Rajaram MVS. Role of cardiac macrophages on cardiac inflammation, fibrosis and tissue repair. *Cells.* **2021**;10:1–27.
29. Karam M, Fahs D, Maatouk B, Safi B, Jaffa AA, Mhanna R. Polymeric nanoparticles in the diagnosis and treatment of myocardial infarction: challenges and future prospects. *Mater Today Bio.* **2022**;14:100249. doi:10.1016/j.mtbio.2022.100249
30. Mandoli GE, D'ascenzi F, Vinco G, et al. Novel approaches in cardiac imaging for non-invasive assessment of left heart myocardial fibrosis. *Front Cardiovasc Med.* **2021**;8. doi:10.3389/fcvm.2021.614235
31. Hahn VS, Yanek LR, Vaishnav J, et al. Endomyocardial biopsy characterization of heart failure with preserved ejection fraction and prevalence of cardiac amyloidosis. *JACC Heart Fail.* **2020**;8:712–724. doi:10.1016/j.jchf.2020.04.007
32. Yang Z, Xu R, Wang J, et al. Association of myocardial fibrosis detected by late gadolinium-enhanced MRI with clinical outcomes in patients with diabetes: a systematic review and meta-analysis. *BMJ Open.* **2022**;12:e055374.
33. Ozawa K, Funabashi N, Takaoka H, et al. Characteristic myocardial strain identified in hypertrophic cardiomyopathy subjects with preserved left ventricular ejection fraction using a novel multi-layer transthoracic echocardiography technique. *Int J Cardiol.* **2015**;184:237–243. doi:10.1016/j.ijcard.2015.01.070
34. Arava D, Masarwy M, Khawaled S, Freiman M. Deep-learning based motion correction for myocardial T1 mapping. In: *2021 IEEE International Conference on Microwaves, Antennas, Communications and Electronic Systems (COMCAS).* **2021**.
35. McGann C, Akoum N, Patel A, et al. Atrial fibrillation ablation outcome is predicted by left atrial remodeling on MRI. *Circ Arrhythm Electrophysiol.* **2014**;7:23–30. doi:10.1161/CIRCEP.113.000689
36. Broncano J, Bhalla S, Gutierrez FR, et al. Cardiac MRI in pulmonary hypertension: from magnet to bedside. *RadioGraphics.* **2020**;40:982–1002. doi:10.1148/rq.2020190179
37. Vaquero JJ, Kinahan P. Positron emission tomography: current challenges and opportunities for technological advances in clinical and preclinical imaging systems. *Annu Rev Biomed Eng.* **2015**;17:385–414. doi:10.1146/annurev-bioeng-071114-040723
38. Li F, Chen L, Zhong S, et al. Collagen-targeting self-assembled nanoprobes for multimodal molecular imaging and quantification of myocardial fibrosis in a rat model of myocardial infarction. *ACS Nano.* **2024**;18:4886–4902. doi:10.1021/acsnano.3c09801
39. Mahan MM, Doiron AL. Gold nanoparticles as X-ray, CT, and multimodal imaging contrast agents: formulation, targeting, and methodology. *J Nanomater.* **2018**;2018:1–15. doi:10.1155/2018/5837276
40. Sollini M, Kirienko M, Gelardi F, Fiz F, Gozzi N, Chiti A. State-of-the-art of FAPI-PET imaging: a systematic review and meta-analysis. *Eur J Nucl Med Mol Imaging.* **2021**;48:4396–4414. doi:10.1007/s00259-021-05475-0
41. Varma M, Xuan HV, Fort E. Gold nanoparticles in cardiovascular imaging. *WIREs Nanomed Nanobiotechnol.* **2018**;10(1). doi:10.1002/wnan.1470
42. Mahan MM, Doiron AL. Gold nanoparticles as X-ray, CT, and multimodal imaging contrast agents: formulation, targeting, and methodology. *J Nanomater.* **2018**;2018:1–15.
43. Mayola MF, Thackeray JT. The potential of fibroblast activation protein-targeted imaging as a biomarker of cardiac remodeling and injury. *Curr Cardiol Rep.* **2023**;25:515–523. doi:10.1007/s11886-023-01869-8
44. Sun F, Wang C, Feng H, et al. Visualization of activated fibroblasts in heart failure with preserved ejection fraction with [18F]AIF-NOTA-FAPI-04 PET/CT Imaging. *Mol Pharm.* **2023**;20:2634–2641. doi:10.1021/acs.molpharmaceut.3c00075
45. Ren Z, Zhang Z, Ling L, Liu X, Wang X. Drugs for treating myocardial fibrosis. *Front Pharmacol.* **2023**;14. doi:10.3389/fphar.2023.1221881
46. Yabanoglu-Ciftci S, Baysal I, Eriksi A, Arica B, Ucar G. Transforming growth factor- $\beta$ 3 (TGF- $\beta$ 3) loaded PLGA-b-PEG nanoparticles: efficacy in preventing cardiac fibrosis induced by TGF- $\beta$ 1. *J Drug Deliv Sci Technol.* **2018**;48:223–234. doi:10.1016/j.jddst.2018.09.021
47. Humeres C, Shinde AV, Tuleta I, et al. Fibroblast Smad7 induction protects the remodeling pressure-overloaded heart. *Circ Res.* **2024**;135:453–469. doi:10.1161/CIRCRESAHA.123.323360
48. Schafer S, Viswanathan S, Widjaja AA, et al. IL-11 is a crucial determinant of cardiovascular fibrosis. *Nature.* **2017**;552:110–115. doi:10.1038/nature24676
49. Widjaja AA, Lim WW, Viswanathan S, et al. Inhibition of IL-11 signalling extends mammalian healthspan and lifespan. *Nature.* **2024**;632:157–165. doi:10.1038/s41586-024-07701-9
50. Yang C, Tian A, Wu J, et al. Gold nanoparticles for targeting the fibrotic heart: a probe indicating vascular permeability. *J Nanosci Nanotechnol.* **2019**;19:7546–7550. doi:10.1166/jnn.2019.16774
51. Kumari P, Saifi MA, Khurana A, Godugu C. Cardioprotective effects of nanoceria in a murine model of cardiac remodeling. *J Trace Elements Med Biol.* **2018**;50:198–208. doi:10.1016/j.jtemb.2018.07.011
52. Zhao X, Qin Y, Wang B, et al. A non-invasive osteopontin-targeted phase changeable fluorescent nanoprobe for molecular imaging of myocardial fibrosis. *Nanoscale Adv.* **2024**;6:3590–3601. doi:10.1039/D4NA00042K
53. Chen W, Liu L, Tang M, et al. Type I collagen-targeted liposome delivery of Serca2a modulates myocardium calcium homeostasis and reduces cardiac fibrosis induced by myocardial infarction. *Mater Today Bio.* **2024**;28.
54. Gao Z, Yan L, Meng J, et al. Targeting cardiac fibrosis with chimeric antigen receptor macrophages. *Cell Discov.* **2024**;10:1–4. doi:10.1038/s41421-024-00718-4

55. Aghajanian H, Kimura T, Rurik JG, et al. Targeting cardiac fibrosis with engineered T cells. *Nature*. 2019;573:430–433. doi:10.1038/s41586-019-1546-z
56. Wang Y, Jiang H, Chen Q, et al. Myofibroblast-targeting extracellular vesicles: a promising platform for cardiac fibrosis drug delivery. *Biomater Res*. 2025;29:0179. doi:10.34133/bmr.0179
57. Deng Z, Fan T, Xiao C, et al. TGF- $\beta$  signaling in health, disease, and therapeutics. *Signal Transduc Targeted Therap*. 2024;9.
58. Parichatikanond W, Luangmonkong T, Mangmool S, Kurose H. Therapeutic targets for the treatment of cardiac fibrosis and cancer: focusing on TGF- $\beta$  signaling. *Front Cardiovasc Med*. 2020;7. doi:10.3389/fcvm.2020.00034
59. Morfino P, Aimo A, Castiglione V, Gálvez-Montón C, Emdin M, Bayes-Genis A. Treatment of cardiac fibrosis: from neuro-hormonal inhibitors to CAR-T cell therapy. *Heart Failure Rev*. 2023;28:555–569. doi:10.1007/s10741-022-10279-x
60. Roată CE, Iacob Ș, Morărașu Ș, et al. Collagen-binding nanoparticles: a scoping review of methods and outcomes. *Crystals*. 2021;11:1396. doi:10.3390/cryst11111396
61. Rurik JG, Tombácz I, Yadegari A, et al. CAR T cells produced in vivo to treat cardiac injury. *Science*. 2022;375:91–96. doi:10.1126/science.abm0594
62. Jardin B, Epstein JA. Emerging mRNA therapies for cardiac fibrosis. *Am J Physiol Cell Physiol*. 2024;326:C107–11.
63. Rurik JG, Tombácz I, Yadegari A, et al. CAR T cells produced in vivo to treat cardiac injury. *Science*. 2022;375:91–96.
64. Nasrullah M, Meenakshi sundaram DN, Claerhout J, Ha K, Demirkaya E, Uludag H. Nanoparticles and cytokine response. *Front Bioeng Biotechnol*. 2023;11. doi:10.3389/fbioe.2023.1243651
65. Zhuang T, Chen MH, Wu RX, et al. ALKBH5-mediated m6A modification of IL-11 drives macrophage-to-myofibroblast transition and pathological cardiac fibrosis in mice. *Nat Commun*. 2024;15:1995. doi:10.1038/s41467-024-46357-x
66. Taniguchi R, Ohashi Y, Lee JS, et al. Endothelial cell TGF- $\beta$  (transforming growth factor-beta) signaling regulates venous adaptive remodeling to improve arteriovenous fistula patency. *Arterioscler Thromb Vasc Biol*. 2022;42:868–883. doi:10.1161/ATVBAHA.122.317676
67. Olson SR, Tang WHW, Liu CF. Non-coding ribonucleic acids as diagnostic and therapeutic targets in cardiac fibrosis. *Curr Heart Fail Rep*. 2024;21:262–275. doi:10.1007/s11897-024-00653-1
68. Zhang Y, Luo G, Zhang Y, et al. Critical effects of long non-coding RNA on fibrosis diseases. *Exp Mol Med*. 2018;50:e428–e428. doi:10.1038/emmm.2017.223
69. Yaghoobi A, Rezaee M, Behnush AH, et al. Role of long noncoding RNAs in pathological cardiac remodeling after myocardial infarction: an emerging insight into molecular mechanisms and therapeutic potential. *Biomed Pharmacother*. 2024;172:116248. doi:10.1016/j.biopha.2024.116248
70. Xu H, Li S, Liu YS. Nanoparticles in the diagnosis and treatment of vascular aging and related diseases. *Signal Trans Target Ther*. 2022;7(1):231.
71. Dong Y, Peng N, Dong L, Tan S, Zhang X. Non-coding RNAs: important participants in cardiac fibrosis. *Front Cardiovasc Med*. 2022;9. doi:10.3389/fcvm.2022.937995
72. Wang B, Zhang A, Wang H, et al. *miR-26a* limits muscle wasting and cardiac fibrosis through exosome-mediated microRNA transfer in chronic kidney disease. *Theranostics*. 2019;9:1864–1877. doi:10.7150/thno.29579
73. Creemers EE, van Rooij E. Function and therapeutic potential of noncoding RNAs in cardiac fibrosis. *Circ Res*. 2016;118:108–118. doi:10.1161/CIRCRESAHA.115.305242
74. Yue T, Xiong S, Zheng D, et al. Multifunctional biomaterial platforms for blocking the fibrosis process and promoting cellular restoring effects in myocardial fibrosis therapy. *Front Bioeng Biotechnol*. 2022;10. doi:10.3389/fbioe.2022.988683
75. Bancel S. Methods of tissue repair and regeneration. US2016022774A1. 2014.
76. Kang YJ. Methods of tissue repair and regeneration. US11077138B2. 2015.
77. Hamzah JB. Novel biomolecule conjugates and uses thereof. US2022143203A1. 2021.
78. Hagemeyer C. Agents and methods for the diagnosis and treatment of diseases associated with extracellular matrix turnover. US2019321496A1. 2017.
79. Cummings C. Compositions and methods for treating disease. US11376278B2. 2017.
80. Samir O. Methods and compositions for controlling cardiac fibrosis and remodeling. US2020197432A1. 2018.
81. Feng Z. CRISPR/CAS-adenine deaminase-based compositions, systems, and methods for targeted nucleic acid editing. US2021093667A1. 2018.
82. Moskowitz A. Platelets as delivery agents. US2020224164A1. 2019.
83. Schurpf T. LTBP complex-specific inhibitors of TGF $\beta$  and uses thereof. US11214614B2. 2020.
84. Glassbe M. Mesenchymal stem cell-derived extracellular vesicles and uses thereof for treating and diagnosing fibrotic diseases. US2022387510A1. 2022.
85. Elgebaly S. Cyclocreatine phosphate: a novel bioenergetic therapy to prevent and treat ischemia-induced and aging-related cardiovascular and neurodegenerative diseases. US11654134B2. 2020.
86. Elgebaly S. Nourin gene-based RNA molecular network: novel early diagnostic and prognostic biomarkers for coronary artery disease, unstable angina, STEMI/NSTEMI and heart failure. US11761966B2. 2020.
87. Hohman E. Boron-containing RHO kinase inhibitors. CN114173791A. 2020.
88. Sun D. Anti-gal3 antibodies and methods of use. US2023036181A1. 2021.
89. Glassberg M. Diagnostic and therapeutic uses of compositions comprising purified, enriched potent exosomes containing disease-based and therapy based signature cargo. US2022296645A1. 2022.
90. Jackson M. Anti-GDF15 antibodies, compositions and uses thereof. CA3228576A1. 2022.
91. Lingfeng L. Fusion protein targeting cell membrane receptor proteins and use thereof. WO2023227018A1. 2023.
92. Ping J. Application of MKP5 in the treatment of diabetic nephropathy. CN117398478A. 2023.
93. Bing Z. Mutant having casrx activity and use thereof. WO2024149283A1. 2024.
94. Goodman B. Circular RNA compositions and methods. WO2021113777A2. 2020.
95. Lee C. Natriuretic polypeptides. US2014005358A1. 2012.
96. Jinan L. Method and drug for preventing and treating abnormal blood pressure conditions. US2023139956A1. 2021.

97. Ocaranza M. Pharmaceutical composition containing angiotensin-(1-9) for cardiovascular, pulmonary and/or cerebral treatment. US20120172301A1. 2008.
98. Parhiz H. In vivo targeting of fibrosis by anti-CD5-targeted FAP-CAR T mRNA-LNP. US20120172301A1. 2023.
99. Datta A. TGFβ1 inhibitors and use thereof. US2021340238A1. 2019.
100. Datta A. High-affinity, isoform-selective TGFβ1 inhibitors and use thereof. US2021122814A1. 2019.
101. Karluri R. Exosome-based treatments for liver fibrosis and other fibrosis-related diseases. CN115297850A. 2020.
102. Longaker M. Use of inhibitors of the activator protein 1 (AP-1) for preventing adhesions. US2023241035A1. 2021.
103. Odell I. Methods and compositions for treating and preventing fibrosis. WO2023168087A1. 2023.
104. Yang C, Yang H, Wu J, et al. No overt structural or functional changes associated with PEG-coated gold nanoparticles accumulation with acute exposure in the mouse heart. *Toxicol Lett.* 2013;222:197–203.
105. Qiao Y, Zhu B, Tian A, Li Z. PEG-coated gold nanoparticles attenuate β-adrenergic receptor-mediated cardiac hypertrophy. *Int J Nanomed.* 2017;12:4709–4719. doi:10.2147/IJN.S130951
106. Coradeghini R, Gioria S, García CP, et al. Size-dependent toxicity and cell interaction mechanisms of gold nanoparticles on mouse fibroblasts. *Toxicol Lett.* 2013;217(3):205–216.
107. Wang Q, Song Y, Chen J, et al. Direct in vivo reprogramming with non-viral sequential targeting nanoparticles promotes cardiac regeneration. *Biomaterials.* 2021;276:121028. doi:10.1016/j.biomaterials.2021.121028
108. Guo T, Chen L, Li F, et al. Biomimetic nanoparticles loaded lutein functionalized by macrophage membrane for targeted amelioration pressure overload-induced cardiac fibrosis. *Biomed Pharmacother.* 2023;167:115579. doi:10.1016/j.biopha.2023.115579
109. Dong Y, Wang B, Liang T, et al. Melatonin-loaded cardiac homing peptide-functionalized gold nanoparticles for the care of anti-cardiac hypertrophy. *J Polym Environ.* 2022;30:3791–3801. doi:10.1007/s10924-022-02452-y
110. Ghann WE, Aras O, Fleiter T, Daniel MC. Syntheses and characterization of lisinopril-coated gold nanoparticles as highly stable targeted ct contrast agents in cardiovascular diseases. *Langmuir.* 2012;28:10398–10408. doi:10.1021/la301694q
111. Duncan B, Kim C, Rotello VM. Gold nanoparticle platforms as drug and biomacromolecule delivery systems. *J Contr Release.* 2010;148:122–127. doi:10.1016/j.jconrel.2010.06.004
112. Mitchell MJ, Billingsley MM, Haley RM, Wechsler ME, Peppas NA, Langer R. Engineering precision nanoparticles for drug delivery. *Nat Rev Drug Discov.* 2021;20:101–124. doi:10.1038/s41573-020-0090-8
113. Kumalasari MR, Alfanaar R, Andreani AS. Gold nanoparticles (AuNPs): a versatile material for biosensor applications. *Talanta Open.* 2024;9:100327. doi:10.1016/j.talo.2024.100327
114. Sarfraz N, Khan I. Plasmonic gold nanoparticles (AuNPs): properties, synthesis and their advanced energy, environmental and biomedical applications. *Chemistry.* 2021;16:720–742.
115. Bolaños K, Kogan MJ, Araya E. Capping gold nanoparticles with albumin to improve their biomedical properties. *Int J Nanomed.* 2019;14:6387–6406. doi:10.2147/IJN.S210992
116. Shah P, Chandra S. Review on the emergence of nanomaterial coatings in bio-engineered cardiovascular stents. *J Drug Deliv Sci Technol.* 2022;70:103224. doi:10.1016/j.jddst.2022.103224
117. Luo XM, Yan C, Feng YM. Nanomedicine for the treatment of diabetes-associated cardiovascular diseases and fibrosis. *Adv Drug Deliv Rev.* 2021;172:234–248. doi:10.1016/j.addr.2021.01.004
118. Yaqoob SB, Adnan R, Rameez Khan RM, Rashid M. Gold, silver, and palladium nanoparticles: a chemical tool for biomedical applications. *Front Chemis.* 2020;8. doi:10.3389/fchem.2020.00376
119. Sreenivasan Soumya R, Gopalan Raghu K. Recent advances on nanoparticle-based therapies for cardiovascular diseases. *J Cardiol.* 2023;81:10–18. doi:10.1016/j.jcc.2022.02.009
120. Yadav HKS, Almokdad AA, Shaluf SIM, Debe MS. Polymer-based nanomaterials for drug-delivery carriers. In: *Nanocarriers Drug Delivery.* Elsevier; 2019:531–556.
121. Perumal S. Polymer nanoparticles: synthesis and applications. *Polymers.* 2022;14:5449. doi:10.3390/polym14245449
122. Yue T, Xiong S, Zheng D, et al. Multifunctional biomaterial platforms for blocking the fibrosis process and promoting cellular restoring effects in myocardial fibrosis therapy. *Front Bioeng Biotechnol.* 2022;10.
123. Torrieri G, Ferreira MPA, Shahbazi MA, et al. In vitro evaluation of the therapeutic effects of dual-drug loaded spermine-acetalated dextran nanoparticles coated with tannic acid for cardiac applications. *Adv Funct Mater.* 2022;32(5). doi:10.1002/adfm.202109032.
124. Zhu JR, Tao YF, Lou S, Wu ZM. Protective effects of ginsenoside Rb3 on oxygen and glucose deprivation-induced ischemic injury in PC12 cells. *Acta Pharmacol Sin.* 2010;31:273–280. doi:10.1038/aps.2010.9
125. Zhang Y, Ji H, Qiao O, et al. Nanoparticle conjugation of ginsenoside Rb3 inhibits myocardial fibrosis by regulating PPARα pathway. *Biomed Pharmacother.* 2021;139.
126. Editorial. Let's talk about lipid nanoparticles. *Nat Rev Mater.* 2021;6:99. doi:10.1038/s41578-021-00281-4
127. Nsairat H, Khater D, Sayed U, Odeh F, Al Bawab A, Alshaer W. Liposomes: structure, composition, types, and clinical applications. *Heliyon.* 2022;8(5):e09394. doi:10.1016/j.heliyon.2022.e09394
128. Tenchov R, Bird R, Curtze AE, Zhou Q. Lipid nanoparticles from liposomes to mRNA vaccine delivery, a landscape of research diversity and advancement. *ACS Nano.* 2021;15:16982–17015. doi:10.1021/acsnano.1c04996
129. Alfutaimani AS, Alharbi NK, Alahmari AS, Alqabbani AA, Aldayel AM. Exploring the landscape of lipid nanoparticles (LNPs): a comprehensive review of LNPs types and biological sources of lipids. *Int J Pharm.* 2024;8:100305.
130. Li R, Liang H, Li J, et al. Paclitaxel liposome (Lipusu) based chemotherapy combined with immunotherapy for advanced non-small cell lung cancer: a multicenter, retrospective real-world study. *BMC Cancer.* 2024;24(1):1–8. doi:10.1186/s12885-023-11764-8
131. Hamad I, Harb AA, Bustanji Y. Liposome-based drug delivery systems in cancer research: an analysis of global landscape efforts and achievements. *Pharmaceutics.* 2024;16:400. doi:10.3390/pharmaceutics16030400
132. O'Brien MER, Wigler N, Inbar M, et al. Reduced cardiotoxicity and comparable efficacy in a Phase III trial of pegylated liposomal doxorubicin HCl (CAELYX™/Doxil®) versus conventional doxorubicin for first-line treatment of metastatic breast cancer. *Ann Oncol.* 2004;15:440–449. doi:10.1093/annonc/mdh097

133. Soroudi S, Jaafari MR, Arabi L. Lipid nanoparticle (LNP) mediated mRNA delivery in cardiovascular diseases: advances in genome editing and CAR T cell therapy. *J Contr Release*. 2024;372:113–140. doi:10.1016/j.jconrel.2024.06.023
134. Hou X, Zaks T, Langer R, Dong Y. Lipid nanoparticles for mRNA delivery. *Nat Rev Mater*. 2021;6:12. doi:10.1038/s41578-021-00358-0
135. Collén A, Bergenhem N, Carlsson L, et al. VEGFA mRNA for regenerative treatment of heart failure. *Nat Rev Drug Discov*. 2022;21:79–80. doi:10.1038/s41573-021-00355-6
136. Gaytan SL, Beaven E, Gadad SS, Nurunnabi M. Progress and prospect of nanotechnology for cardiac fibrosis treatment. *Interdiscip Med*. 2023;1:e20230018.
137. Shi Y, Shi M, Wang Y, You J. Progress and prospects of mRNA-based drugs in pre-clinical and clinical applications. *Signal Transduct Target Ther*. 2024;9(1).
138. Bejarano J, Rojas A, Ramírez-Sagredo A, et al. Light-induced release of the cardioprotective peptide angiotensin-(1–9) from thermosensitive liposomes with gold nanoclusters. *J Contr Release*. 2020;328:859–872. doi:10.1016/j.jconrel.2020.11.002
139. Liu M. Targeted gene silencing with small interfering RNA-loaded lipid nanoparticles to treat cardiac fibrosis. *Circ Res*. 2024;135(Suppl\_1): AWe101–AWe101. doi:10.1161/res.135.suppl\_1.We101
140. Dong C, Ma A, Shang L. Nanoparticles for postinfarct ventricular remodeling. *Nanomedicine*. 2018;13:3037–3050. doi:10.2217/nnm-2018-0264
141. Eck W, Craig G, Sigdel A, et al. PEGylated gold nanoparticles conjugated to monoclonal F19 antibodies as targeted labeling agents for human pancreatic carcinoma tissue. *ACS Nano*. 2008;2:2263–2272. doi:10.1021/nn800429d
142. Chen H, Shao L, Li Q, Wang J. Gold nanorods and their plasmonic properties. *Chem Soc Rev*. 2013;42:2679–2724. doi:10.1039/C2CS35367A
143. Her S, Jaffray DA, Allen C. Gold nanoparticles for applications in cancer radiotherapy: mechanisms and recent advancements. *Adv Drug Deliv Rev*. 2017;109:84–101. doi:10.1016/j.addr.2015.12.012
144. Bejarano J, Navarro-Marquez M, Morales-Zavala F, et al. Nanoparticles for diagnosis and therapy of atherosclerosis and myocardial infarction: evolution toward prospective theranostic approaches. *Theranostics*. 2018;8:4710–4732. doi:10.7150/thno.26284
145. Tian A, Yang C, Zhu B, et al. Polyethylene-glycol-coated gold nanoparticles improve cardiac function after myocardial infarction in mice. *Can J Physiol Pharmacol*. 2018;96:1318–1327. doi:10.1139/cjpp-2018-0227
146. Kawasaki T, Sakai C, Harimoto K, Yamano M, Miki S, Kamitani T. Usefulness of high-sensitivity cardiac troponin T and brain natriuretic peptide as biomarkers of myocardial fibrosis in patients with hypertrophic cardiomyopathy. *Am J Cardiol*. 2013;112:867–872. doi:10.1016/j.amjcard.2013.04.060
147. Mansouri S, Mezan SO, Altalbawy FM, et al. Recent advances in assembly strategies of new advanced materials-based analytical methods for the detection of cardiac biomarkers as a diagnosis tool. *Microchem J*. 2023;191:108827. doi:10.1016/j.microc.2023.108827
148. Sumbayev VV, Yasinska IM, Garcia CP, et al. Gold nanoparticles downregulate interleukin-1 $\beta$ -induced pro-inflammatory responses. *Small*. 2013;9:472–477. doi:10.1002/sml.201201528
149. Li Y, Yu J, Cheng C, et al. Platelet and erythrocyte membranes coassembled biomimetic nanoparticles for heart failure treatment. *ACS Nano*. 2024;18:26614–26630. doi:10.1021/acsnano.4c04814
150. Ding Y, Wang Y, Zhang W, et al. Roles of biomarkers in myocardial fibrosis. *Aging Dis*. 2020;11(5):1157. doi:10.14336/AD.2020.0604
151. Zhang Z, Lin M. Fast loading of PEG-SH on CTAB-protected gold nanorods. *RSC Adv*. 2014;4(34):17760–17767. doi:10.1039/c3ra48061e
152. Manson J, Kumar D, Meenan BJ, Dixon D. Polyethylene glycol functionalized gold nanoparticles: the influence of capping density on stability in various media. *Gold Bull*. 2011;44:99–105. doi:10.1007/s13404-011-0015-8
153. Jara-Guajardo P, Cabrera P, Celis F, et al. Gold nanoparticles mediate improved detection of  $\beta$ -amyloid aggregates by fluorescence. *Nanomaterials*. 2020;10(4). doi:10.3390/nano10040690.
154. Lara P, Palma-Florez S, Salas-Huenuleo E, et al. Gold nanoparticle-based double-labeling of melanoma extracellular vesicles to determine the specificity of uptake by cells and preferential accumulation in small metastatic lung tumors. *J Nanobiotechnol*. 2020;18(1). doi:10.1186/s12951-020-0573-0.
155. Lu J, Gao X, Wang S, et al. Advanced strategies to evade the mononuclear phagocyte system clearance of nanomaterials. *Exploration*. 2023;3(1). doi:10.1002/EXP.20220045.
156. Xu H, Li S, Liu YS. Nanoparticles in the diagnosis and treatment of vascular aging and related diseases. *Signal Transduct Target Ther*. 2022;7(1):231. doi:10.1038/s41392-022-01082-z
157. Riveros AL, Eggeling C, Riquelme S, et al. Improving cell penetration of gold nanorods by using an amphipathic arginine-rich peptide. *Int J Nanomed*. 2020;15:1837–1851. doi:10.2147/IJN.S237820
158. Yang C, Yang H, Wu J, et al. No overt structural or functional changes were associated with PEG-coated gold nanoparticles accumulation with acute exposure in the mouse heart. *Toxicol Lett*. 2013;222(2):197–203. doi:10.1016/j.toxlet.2013.07.018
159. Lara P, Palma-Florez S, Salas-Huenuleo E, et al. Gold nanoparticle based double-labeling of melanoma extracellular vesicles to determine the specificity of uptake by cells and preferential accumulation in small metastatic lung tumors. *J Nanobiotechnol*. 2020;18(1).
160. Zhang J, Xue Y, Ni Y, Ning F, Shang L, Ma A. Size dependent effects of gold nanoparticles in ISO-induced hyperthyroid rats. *Sci Rep*. 2018;8(1).
161. Sonavane G, Tomoda K, Makino K. Biodistribution of colloidal gold nanoparticles after intravenous administration: effect of particle size. *Colloids Surf B Biointerfaces*. 2008;66:274–280. doi:10.1016/j.colsurfb.2008.07.004
162. De Jong WH, Hagens WI, Krystek P, Burger MC, Sips AJAM, Geertsma RE. Particle size-dependent organ distribution of gold nanoparticles after intravenous administration. *Biomaterials*. 2008;29:1912–1919. doi:10.1016/j.biomaterials.2007.12.037
163. Chen KH, Lundy DJ, Toh EKW, et al. Nanoparticle distribution during systemic inflammation is size-dependent and organ-specific. *Nanoscale*. 2015;7:15863–15872. doi:10.1039/C5NR03626G
164. Fan Z, Guan J. Antifibrotic therapies to control cardiac fibrosis. *Biomater Res*. 2016;20(1). doi:10.1186/s40824-016-0060-8
165. Lundy DJ, Chen KH, Toh EKW, Hsieh PCH. Distribution of systemically administered nanoparticles reveals a size-dependent effect immediately following cardiac ischaemia-reperfusion injury. *Sci Rep*. 2016;6. doi:10.1038/srep25613
166. Yang C, Tian A, Li Z. Reversible cardiac hypertrophy induced by PEG-coated gold nanoparticles in mice. *Sci Rep*. 2016.
167. Ahmed SM, Abdelrahman SA, Salama AE. Efficacy of gold nanoparticles against isoproterenol-induced acute myocardial infarction in adult male albino rats. *Ultrastruct Pathol*. 2017;41(2):168–185. doi:10.1080/01913123.2017.1281367

168. Hillaireau H. Investigating interactions between nanoparticles and cells: internalization and intracellular trafficking. *Polym Nanopart Nanomed*. 2016;291–323.
169. Arathi, Megha KB, Joseph X, Mohanan PV. Biological safety and cellular interactions of nanoparticles. *Biomed Appl Toxicity Nanomater*. 2023;559–87.1.
170. Zhang YN, Poon W, Tavares AJ, McGilvray ID, Chan WCW. Nanoparticle–liver interactions: cellular uptake and hepatobiliary elimination. *J Contr Release*. 2016;240:332–348. doi:10.1016/j.jconrel.2016.01.020
171. Niznik L, Noga M, Kobylarz D, et al. Gold Nanoparticles (AuNPs)—toxicity, safety and green synthesis: a critical review. *Int J Mol Sci*. 2024;25(7):4057. doi:10.3390/ijms25074057
172. Carvalho GC, Marena GD, Karnopp JCF, et al. Cetyltrimethylammonium bromide in the synthesis of mesoporous silica nanoparticles: general aspects and in vitro toxicity. *Adv Colloid Interface Sci*. 2022;307:102746. doi:10.1016/j.cis.2022.102746
173. Andrianov K, Parvin N, Joo SW, Mandal TK. Biodegradable and stimuli-responsive nanomaterials for targeted drug delivery in autoimmune diseases. *J Funct Biomater*. 2025;16(1):24. doi:10.3390/jfb16010024
174. ClinicalTrials.gov [Internet]. 2025. Available from: <https://www.clinicaltrials.gov/>. Accessed July 31, 2025.
175. Dri DA, Rinaldi F, Carafa M, Marianecchi C. Nanomedicines and nanocarriers in clinical trials: surfing through regulatory requirements and physico-chemical critical quality attributes. *Drug Deliv Transl Res*. 2023;13:757–769. doi:10.1007/s13346-022-01262-y
176. Kumar S, Bharti B, Zha X, Ouyang F, Ren P. Recent development in industrial scale fabrication of nanoparticles and their applications. In: *Liquid and Crystal Nanomaterials for Water Pollutants Remediation*. Boca Raton: CRC Press; 2022:88–118.
177. Jindal AB editor. *Pharmaceutical Process Engineering and Scale-up Principles*. Vol. 13. Cham: Springer Nature Switzerland; 2023.
178. Gacem MA, Abd-Elsalam KA. Strategies for scaling up of green-synthesized nanomaterials: challenges and future trends. In: *Green Synthesis of Silver Nanomaterials*. Elsevier; 2022:669–698.
179. Eyube M, Enuesueke C, Alimikhena M. The future of nanomaterials in manufacturing. *ChemRxiv*. 2025.
180. Creutzenberg O. Nanoparticles and their regulation. In: Springer, editor. *Regulatory Toxicology*. Berlin, Heidelberg: Springer Berlin Heidelberg; 2021:1–17.
181. Bajpai M, Shafi H, Kumari S. Nanoparticles: importance and need for regulations. In: Springer, editor. *Nanoformulations in Human Health*. Cham: Springer International Publishing; 2020:93–107.
182. Kaur J, Singh H, Khatri M. Regulatory considerations for safety of nanomaterials. In: *Nanomedicine for Bioactives*. Singapore: Springer Singapore; 2020:431–450.
183. Wadhawan S, Wadhawan D, Jain A, Mehta SK. Toxic implication of nanoparticles: a review of factors, mechanisms, exposure, and control strategies. *Int J Environ Sci Technol*. 2025;22:1203–1224. doi:10.1007/s13762-024-05810-6
184. Kumarasamy RV, Manickam Natarajan P, Umapathy VR, Roy JR, Mironescu M, Palanisamy CP. Clinical applications and therapeutic potentials of advanced nanoparticles: a comprehensive review on completed human clinical trials. *Front Nanotechnol*. 2024. doi:10.3389/fnano.2024.1479993
185. Kharlamov AN, Feinstein JA, Cramer JA, Boothroyd JA, Shishkina EV, Shur V. Plasmonic photothermal therapy of atherosclerosis with nanoparticles: long-term outcomes and safety in NANOM-FIM Trial. *Future Cardiol*. 2017;13:345–363. doi:10.2217/fca-2017-0009
186. Hossain A, Rayhan M, Mobarak MH, et al. Advances and significances of gold nanoparticles in cancer treatment: a comprehensive review. *Results Chem*. 2024;8:101559. doi:10.1016/j.rechem.2024.101559
187. Damani M, Dhangar A, Momin M, Ningthoujam RS, Khan TA. Gold nanoparticles in nanomedicine: advances, prospects, and challenges. In: *Gold Nanoparticles, Nanomaterials and Nanocomposites*. Elsevier; 2025:687–722.
188. Mahmoud N, Sajadi M, Issaabadi Z. An Introduction to Nanotechnology. *Interface Sci Technol*. 2019;2019:1–27.
189. Díez-Pascual AM. Nanoparticle reinforced polymers. *Polymers*. 2019;11:625. doi:10.3390/polym11040625
190. Sarohi V, Chakraborty S, Basak T. Exploring the cardiac ECM during fibrosis: a new era with next-gen proteomics. *Front Mol Biosci*. 2022;9. doi:10.3389/fmolb.2022.1030226
191. Šimková A, Bušek P, Šedo A, Konvalinka J. Molecular recognition of fibroblast activation protein for diagnostic and therapeutic applications. *Biochim Biophys Acta Proteins Proteomics*. 2020;1868:140409. doi:10.1016/j.bbapap.2020.140409
192. Moita MR, Silva MM, Diniz C, et al. Transcriptome and proteome profiling of activated cardiac fibroblasts supports target prioritization in cardiac fibrosis. *Front Cardiovasc Med*. 2022;9. doi:10.3389/fcvm.2022.1015473
193. Nishiga M, Liu C, Qi LS, Wu JC. The use of new CRISPR tools in cardiovascular research and medicine. *Nat Rev Cardiol*. 2022;19:505–521. doi:10.1038/s41569-021-00669-3
194. Raziyeve K, Kim Y, Zharkinkbekov Z, Temirkhanova K, Saparov A. Novel therapies for the treatment of cardiac fibrosis following myocardial infarction. *Biomedicines*. 2022;10(9):2178. doi:10.3390/biomedicines10092178
195. Paratz ED, Mundisugih J, Rowe SJ, Kizana E, Semsarian C. Gene therapy in cardiology: is a cure for hypertrophic cardiomyopathy on the horizon? *Can J Cardiol*. 2024;40:777–788.
196. Liu N, Olson EN. CRISPR modeling and correction of cardiovascular disease. *Circ Res*. 2022;130:1827–1850. doi:10.1161/CIRCRESAHA.122.320496
197. Duan L, Ouyang K, Xu X, et al. Nanoparticle delivery of CRISPR/Cas9 for genome editing. *Front Genet*. 2021;12:673286.
198. Park H, Kim D, Cho B, et al. In vivo therapeutic genome editing via CRISPR/Cas9 magnetoplexes for myocardial infarction. *Biomaterials*. 2022;281:121327. doi:10.1016/j.biomaterials.2021.121327
199. Jiang L, Liang J, Huang W, et al. CRISPR activation of endogenous genes reprograms fibroblasts into cardiovascular progenitor cells for myocardial infarction therapy. *Mol Ther*. 2022;30:54–74. doi:10.1016/j.ymthe.2021.10.015
200. Ferrer-Curriu G, Soler-Botija C, Charvatova S, et al. Preclinical scenario targeting myocardial fibrosis with chimeric antigen receptor (CAR) immunotherapy. *Biomed Pharmacother*. 2023;158:114061. doi:10.1016/j.biopha.2022.114061
201. Vagnozzi RJ, Johansen AKZ, Molkentin JD. CARDiac immunotherapy: t cells engineered to treat the fibrotic heart. *Mol Ther*. 2019;27:1869–1871. doi:10.1016/j.ymthe.2019.09.021

## International Journal of Nanomedicine

**Dovepress**  
Taylor & Francis Group

### Publish your work in this journal

The International Journal of Nanomedicine is an international, peer-reviewed journal focusing on the application of nanotechnology in diagnostics, therapeutics, and drug delivery systems throughout the biomedical field. This journal is indexed on PubMed Central, MedLine, CAS, SciSearch®, Current Contents®/Clinical Medicine, Journal Citation Reports/Science Edition, EMBase, Scopus and the Elsevier Bibliographic databases. The manuscript management system is completely online and includes a very quick and fair peer-review system, which is all easy to use. Visit <http://www.dovepress.com/testimonials.php> to read real quotes from published authors.

Submit your manuscript here: <https://www.dovepress.com/international-journal-of-nanomedicine-journal>