

Engineering mRNA-lipid nanoparticle platforms for orthopedic regenerative medicine: From formulation design to biomaterial integration

Jianan Guo, Shiyi Yao, Gen Li, Yingying Wei, Sijia Li, Renhao Yang, Hanyu Wang, Lei Wang  , and Huitong Ruan  

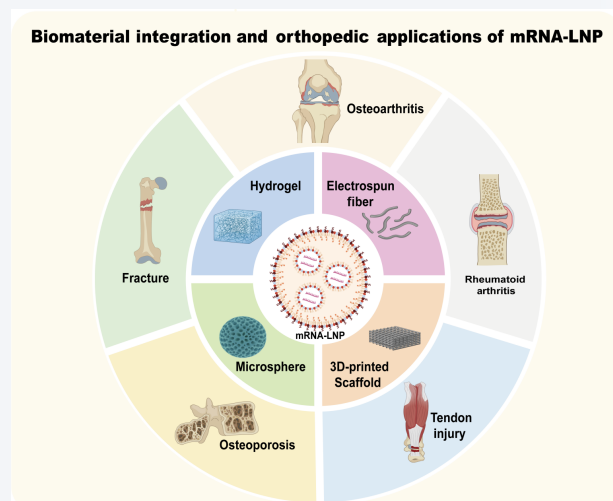
Department of Orthopaedics, Shanghai Institute of Traumatology and Orthopaedics, Shanghai Key Laboratory for Prevention and Treatment of Bone and Joint Diseases, Ruijin Hospital, College of Health Science and Technology, Shanghai Jiao Tong University School of Medicine, Shanghai 200025, China



Cite this article: Nano Research, 2026, 19, 94909012. <https://doi.org/10.26599/NR.2026.94909012>

ABSTRACT: Messenger RNA (mRNA) therapeutics have emerged as a transformative treatment by directing in vivo therapeutic protein synthesis. Lipid nanoparticles (LNPs) currently serve as the predominant delivery vectors for mRNA. Recently, the application of mRNA-LNPs in orthopedic diseases has presented an innovative approach to musculoskeletal tissue regeneration. This review thus focuses on the pioneering design strategies and applications of mRNA-LNP in the treatment of orthopedic diseases. The review first outlines the structural compositions of LNPs, subsequently discussing design strategies for local administration of LNPs and the engineering of synergistic therapeutic platforms that integrate LNPs within functional biomaterial scaffolds, including microspheres, hydrogels, 3D-printed scaffolds, and nanofibers. Furthermore, recent advancements utilizing mRNA-LNPs to treat osteoarthritis, bone fractures, osteoporosis, rheumatoid arthritis, tendon injuries, and other orthopedic diseases are systematically summarized. This review also critically examines the current translational bottlenecks of mRNA-LNPs in orthopedic medicine and proposes future research directions.

KEYWORDS: messenger RNA, lipid nanoparticles, orthopedic diseases, biomedical materials



1 Introduction

The current scope of orthopedic research encompasses pathologies affecting the entire musculoskeletal system, including bones, joints, and skeletal muscles. Conventional treatment modalities comprise surgical intervention, rehabilitation, and pharmacological, biological, or biomaterial-based therapies. However, the field encounters critical challenges that necessitate further innovation. Owing to the anatomical complexity of the musculoskeletal system,

tissues such as cartilage, tendons, and bone-tendon junctions exhibit limited regenerative potential following injuries, contrasting with the robust regenerative capacity of bone. Consequently, existing therapeutic approaches fail to adequately address the distinct repair requirements of heterogeneous injury sites. Furthermore, there is a lack of localized therapeutic approaches for the musculoskeletal system that can focus on lesion sites and provide targeted intervention at the cellular level. Current pharmacological and biological agents are hampered by low delivery efficiency, poor targeting during systemic administration, rapid clearance upon local application, and the inability to maintain sustained therapeutic effects via single-dose administration [1].

Messenger RNA (mRNA) therapy represents a transformative technology that has garnered immense interest, particularly in the development of infectious disease vaccines, protein replacement therapies, gene editing, and tumor immunotherapy. By conveying

Received: May 18, 2026; Revised: July 3, 2026

Accepted: July 8, 2026

✉ Address correspondence to Huitong Ruan, ruanhuitong@163.com; Lei Wang, ray_wangs@hotmail.com

protein-coding information, mRNA directs intracellular protein synthesis to regulate cellular activities or elicit specific immune responses. Lipid nanoparticles (LNPs) currently serve as the most prevalent delivery vectors for mRNA. The regulatory approval of two LNP-formulated COVID-19 vaccines marked a critical milestone in the clinical translation of mRNA-LNP therapeutics [2]. Within orthopedic researches, mRNA-LNP systems are being extensively investigated for the management of osteoarthritis, fractures, osteoporosis, rheumatoid arthritis, and tendon injuries. The deployment of these platforms offers a pioneering paradigm to overcome the limitations inherent to conventional orthopedic

treatments. Unlike the administration of exogenous drugs or growth factors, LNPs efficiently transfect target cells within bone, cartilage, and tendinous tissues, thereby directing the synthesis of endogenous therapeutic proteins. LNPs achieve the precise modulation of cellular metabolism, activity, and fate, ultimately augmenting tissue regeneration and improving long-term prognoses.

This review comprehensively delineates emerging applications of mRNA-LNP therapies for orthopedic disorders (Fig. 1). Initially, the review outlines the compositions of mRNA-LNP, detailing both the mRNA and the lipid components. Subsequently, material

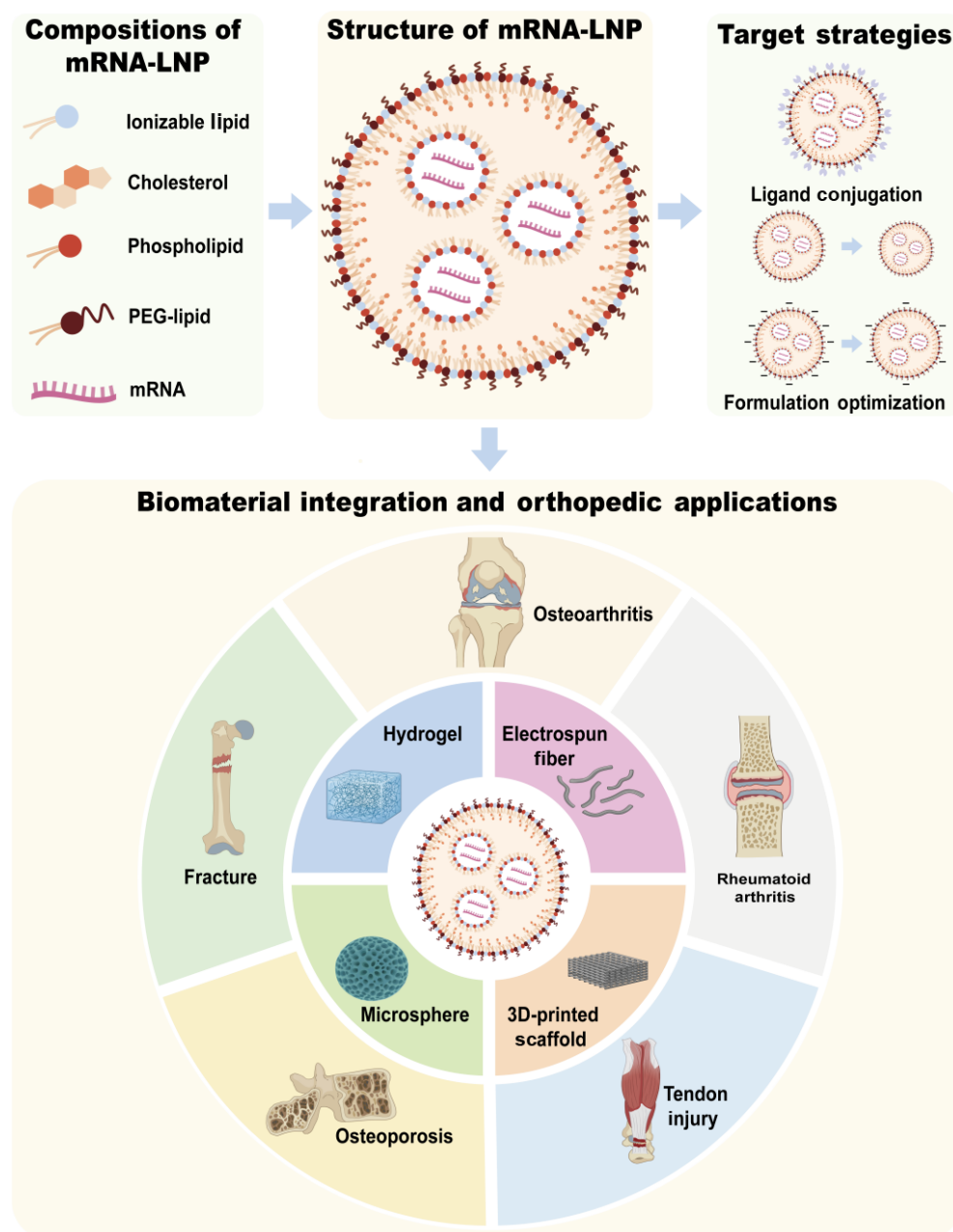


Figure 1 Schematic illustration of compositions, structure, target strategies, biomaterial integration, and orthopedic applications of mRNA-LNPs. Typical mRNA-LNPs contain mRNA and different lipids comprising ionizable lipids, cholesterol, phospholipids, and polyethylene glycol (PEG)-lipids. Ionizable lipids bind to the mRNA via electrostatic interactions, encapsulating the mRNA within the nanoparticle core. Ligand conjugation and formulation optimization represent two primary strategies for achieving the targeted delivery of mRNA-LNPs in orthopedic diseases. Furthermore, LNPs can be integrated within functional biomaterial scaffolds including microspheres, hydrogels, 3D-printed scaffolds, and nanofibers to engineer synergistic therapeutic platforms. Currently, mRNA-LNPs have been extensively investigated for the treatment of osteoarthritis, bone fractures, osteoporosis, rheumatoid arthritis, tendon injuries, and other orthopedic diseases.

strategies for local administration of mRNA-LNPs to overcome delivery barriers are examined. Furthermore, the review discusses innovative approaches for integrating LNPs with functional biomaterial scaffolds. Following this, the therapeutic applications of mRNA-LNPs across various orthopedic pathologies including osteoarthritis, bone fractures, osteoporosis, rheumatoid arthritis, tendon injuries, spinal cord injuries, and femoral head necrosis are elucidated in detail. Finally, the review assesses the primary challenges faced by mRNA-LNPs in orthopedic medical applications and proposes directions for future studies. This review aims to guide the development of mRNA-LNPs toward clinical translation and inspire the exploration of next-generation mRNA-LNP platforms for orthopedic disease.

2 Structure, function and therapeutic mechanisms of mRNA

mRNA typically comprises five structural elements: a 5'-cap, a 5'-untranslated region (UTR), a protein-coding sequence, a 3'-UTR, and a 3'-poly(A) tail [3]. These structural elements dictate the initiation and termination of translation, as well as the post-transcriptional stability and eventual degradation of the transcript. The rational engineering of these components can significantly enhance mRNA stability and translational efficiency [4–7]. However, the direct administration of unmodified mRNA frequently elicits robust inflammatory and immune responses, thereby compromising therapeutic efficacy and raising safety concerns [8]. To overcome these barriers during the synthesis of therapeutic mRNA, multiple structural modifications are employed. Modifying the 5'-cap and 3'-poly(A) tail modulates the mRNA half-life and translational efficacy. Codon optimization and UTR engineering synergistically prolong mRNA stability and maximize protein expression. Crucially, the incorporation of chemically modified nucleosides mitigates innate immunogenicity and promotes translation. Furthermore, engineering the transcript as circular RNA (circRNA) substantially prolongs protein expression [9].

In contrast to DNA-based therapeutics, mRNA delivery circumvents the need for nuclear translocation, as translation occurs directly within the cytoplasm. Besides, mRNA eliminates the

risk of genomic integration, thereby conferring superior safety and efficiency. However, the high molecular weight and polyanionic nature of naked mRNA severely restrict its cellular internalization. Additionally, naked mRNA is susceptible to nuclease-mediated degradation and exhibits innate immunogenicity. Together, these physicochemical and biological barriers necessitate the deployment of highly efficient delivery vectors.

Currently serving as the most commonly used vehicle for mRNA delivery, LNPs encapsulate the mRNA, thereby shielding it from ribonuclease degradation. Following cellular uptake via endocytosis, the acidic endosomal environment triggers the protonation of ionizable lipids, which induces electrostatic interactions with the negatively charged endosomal membrane, resulting in membrane destabilization or fusion. Consequently, this process facilitates endosomal escape and the release of the mRNA into the cytoplasm to initiate protein translation.

3 LNP compositions

The conventional LNP comprises four fundamental components: ionizable lipids, phospholipids, cholesterol, and PEG-lipids (Fig. 2).

3.1 Ionizable lipids

Ionizable lipids represent the critical component of LNP formulations. During the assembly of mRNA-LNPs, these lipids become protonated and positively charged, binding to the negatively charged mRNA via electrostatic interactions to drive encapsulation. Compared to traditional cationic lipids, ionizable lipids maintain a minimal net positive charge or exhibit electroneutrality at physiological pH, thereby significantly mitigating immune responses [10, 11]. Structurally, ionizable lipids comprise three parts: an ionizable hydrophilic head, a central linker, and a hydrophobic tail [12]. The head typically contains one or more hydrophilic groups, such as amino groups, guanidino groups, and heterocyclic groups. These head groups fundamentally affect mRNA encapsulation efficiency, LNP stability, cell membrane interactions, and endosomal escape capabilities. Linkers are generally classified into two categories: non-biodegradable (e.g., ether and urethane bonds) and biodegradable (e.g., ester, amide, and thiol bonds). While lipids with non-degradable linkers exhibit high stability and transfection efficiency, they often induce greater

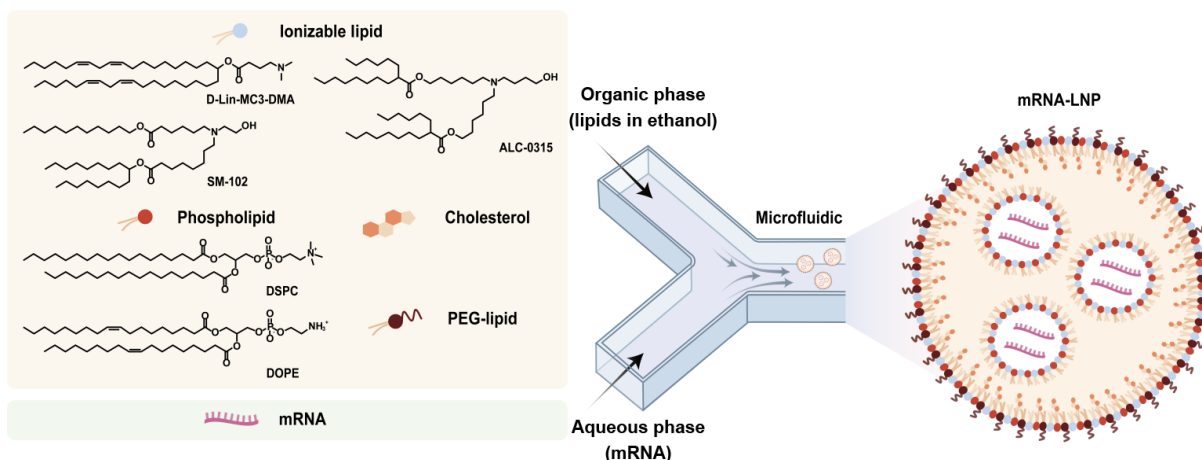


Figure 2 Representative components, structure, and fabrication of mRNA-LNPs. LNP typically consists of four components—ionizable lipids, phospholipids, cholesterol, and PEG-lipids. MC3, SM-102, and ALC-0315 are three representative ionizable lipids that have been approved for clinical use. The most commonly used phospholipids are DSPC and DOPE. During mRNA-LNP formulation, lipids and mRNA are respectively dissolved in ethanol and an appropriate aqueous buffer. The organic and aqueous phases are then introduced into the microfluidic device to drive the self-assembly of mRNA-LNPs.

cytotoxicity. Consequently, degradable linkers are currently preferred owing to their more rapid *in vivo* clearance, reduced potential for adverse effects, and suitability for repeated administration [13]. The hydrophobic tail, typically consisting of saturated, unsaturated, or branched hydrocarbon chains, modulates lipid fluidity and subsequent cellular uptake of ionizable lipids. Lipids with highly saturated tails promote the endocytosis of LNPs but impair endosomal escape, ultimately compromising overall delivery efficiency [14]. Based on the number of amine groups, ionizable lipids are categorized as either monoamine or polyamine ionizable lipids. To date, three monoamine ionizable lipids—D-Lin-MC3-DMA (MC3), SM-102, and ALC-0315—have obtained U.S. Food and Drug Administration (FDA) approval for clinical mRNA delivery [15, 16].

3.2 Cholesterol

Cholesterol constitutes the second most abundant lipid component within LNPs, accounting for approximately 40 mol% of the formulation [17]. The proportion of cholesterol profoundly influences the stability, biodistribution, and transfection efficiency of the LNPs. Mechanistically, cholesterol enhances the structural rigidity of the lipid shell of the LNP, thereby preventing premature payload leakage from the nanoparticle core. Furthermore, it significantly attenuates the adsorption of serum proteins onto the LNP surface within the bloodstream, consequently prolonging the circulation half-life [18]. But an excessive cholesterol fraction can induce crystallization, which subsequently diminishes the adjuvant activity of the LNPs [19]. Conversely, reducing cholesterol levels alters LNP biodistribution, facilitating targeted delivery to extrahepatic tissues such as the spleen and lungs [20]. Since cholesterol comprises a substantial molar fraction of the LNP, the chemical conjugation of targeting ligands to cholesterol molecules empowers the LNPs to actively target distinct cell populations. For instance, Kim et al. formulated LNPs incorporating mannose-functionalized cholesterol, which markedly enhanced LNPs uptake by hepatic sinusoidal endothelial cells and Kupffer cells—both of which highly express mannose receptors [21]. Beyond native cholesterol, incorporating various structural analogues of cholesterol into LNP formulations can fundamentally transform the surface composition and structural properties of the vectors, thereby modulating their intracellular trafficking. Notably, replacing native cholesterol with phytosterol has been demonstrated to substantially augment mRNA delivery efficacy [22].

3.3 Phospholipids

Phospholipids serve as essential helper lipids within LNP formulations. A typical phospholipid molecule comprises a hydrophilic phosphate head group and a hydrophobic tail linked via ester bonds. Studies indicate that phospholipids facilitate the fusion of LNPs with biological membranes, thereby promoting endosomal escape [23]. Currently, 1,2-distearoyl-*sn*-glycero-3-phosphocholine (DSPC) and 1,2-dioleoyl-*sn*-glycero-3-phosphoethanolamine (DOPE) represent the most widely utilized phospholipids. Compared to DSPC, DOPE exhibits a superior capacity to mediate endosomal escape, ultimately yielding higher mRNA delivery efficiency. Additionally, the structural differences between them significantly influence the biodistribution of the nanoparticles. Specifically, LNPs containing DOPE demonstrate a heightened affinity for apolipoprotein E (ApoE) and preferentially accumulate in the liver, whereas DSPC-containing formulations show a greater propensity for splenic accumulation [24].

3.4 PEG-lipids

Although PEG-lipids constitute a relatively minor molar fraction of LNPs—typically only around 1.5 mol%—they remain indispensable for LNPs formulation. These lipids significantly influence the size, stability, and polydispersity of the LNPs. Indeed, formulations devoid of PEG-lipids typically exhibit particle sizes exceeding 200 nm and demonstrate profound instability [25]. The surface distribution of PEG-lipids generates a pronounced steric hindrance effect. This steric shield markedly attenuates the formation of the protein corona, impedes phagocytic clearance by monocytes and macrophages, and consequently prolongs the *in vivo* circulation half-life. Conversely, the steric hindrance effect invariably impedes the interaction between LNPs and target cell membranes, thereby compromising mRNA delivery efficiency [26]. Furthermore, while PEG-lipids are canonically employed to evade immune recognition, studies reveal that the polymer itself is intrinsically immunogenic and can elicit the production of anti-PEG antibodies in humans [27]. These antibodies mediate the accelerated systemic clearance of PEG-containing LNPs and risk triggering severe hypersensitivity reactions.

4 mRNA-LNP: Delivering challenges and targeting strategies in orthopedic diseases

The precise delivery of mRNA-LNPs to specific target organs and cells constitutes a critical determinant of their therapeutic efficacy. Following systemic intravenous administration, the surfaces of LNPs readily adsorb endogenous ApoE. This adsorption facilitates high-affinity interactions with low-density lipoprotein (LDL) receptors abundantly expressed on hepatocytes, thereby driving preferential off-target accumulation in the liver. In the context of orthopedic diseases, the localized application of mRNA-LNPs emerges as a superior therapeutic strategy. Localized application of LNPs—including direct intra-articular injection or integration within biomaterials—can substantially attenuate the accumulation of LNPs in non-target organs. However, these localized approaches do not completely abrogate the risk of nanoparticle dissemination into the peripheral circulation. Consequently, given the inherent constraints of current administration routes, it is imperative to systematically optimize LNP formulations tailored to the distinct pathophysiological microenvironments of the target tissues.

The relatively limited blood supply and highly dense cortical structure of bone pose significant challenges for achieving targeted delivery to the bone microenvironment, potentially compromising the bioavailability of pharmacological or biological agents within bone tissue [28]. Conjugating affinity ligands to LNPs represents a promising strategy for the targeted delivery of mRNA-LNPs. Notably, targeting modification methods developed to enhance the delivery efficacy of systemically administered LNPs are highly translatable and hold significant value for the design of localized delivery platforms. For example, capitalizing on the principle that bisphosphonates (BPs) exhibit a strong affinity for hydroxyapatite in bone, Xue et al. developed a series of bisphosphonate-conjugated ionizable lipids and formulated a library of BP-LNPs for the systemic delivery of mRNA to the bone microenvironment. Compared to unmodified LNPs, BP-LNPs—exemplified by 490BP-C14—demonstrate significantly enhanced accumulation in the bone microenvironment following systemic administration, resulting in markedly improved transfection efficiency (Fig. 3(a)) [29]. However, the long-term or excessive administration of BPs

frequently precipitates adverse effects, including gastrointestinal and renal complications, as well as compromised skeletal perfusion that increases fracture risk [30]. To enhance the stability of BP-modified ionizable lipids and mitigate these potential side effects, Yin et al. designed piperazine-based BP ionizable lipids by conjugating a robust piperazine (PIP) ring structure to a BP group, thereby further improving the transfection efficiency of the LNPs (Fig. 3(b)) [31]. In addition, to specifically target chondrocytes *in vivo*, Wang et al. conjugated chondrocyte-affinity peptides (CAPs) to LNPs

encapsulating palmitoyltransferase *Zdhhc11* mRNA, effectively delaying the progression of osteoarthritis (Fig. 3(c)) [32].

Beyond ligand conjugation, optimizing the molar ratios of foundational LNP components can independently augment bone delivery efficiency while circumventing the potential immunogenicity introduced by exogenous ligands. Mu et al. screened LNP formulations across varying N/P ratios and PEG proportions, demonstrating that an architecture comprising MC3/DSPC/cholesterol/DMG-PEG2000 exhibited optimal skeletal

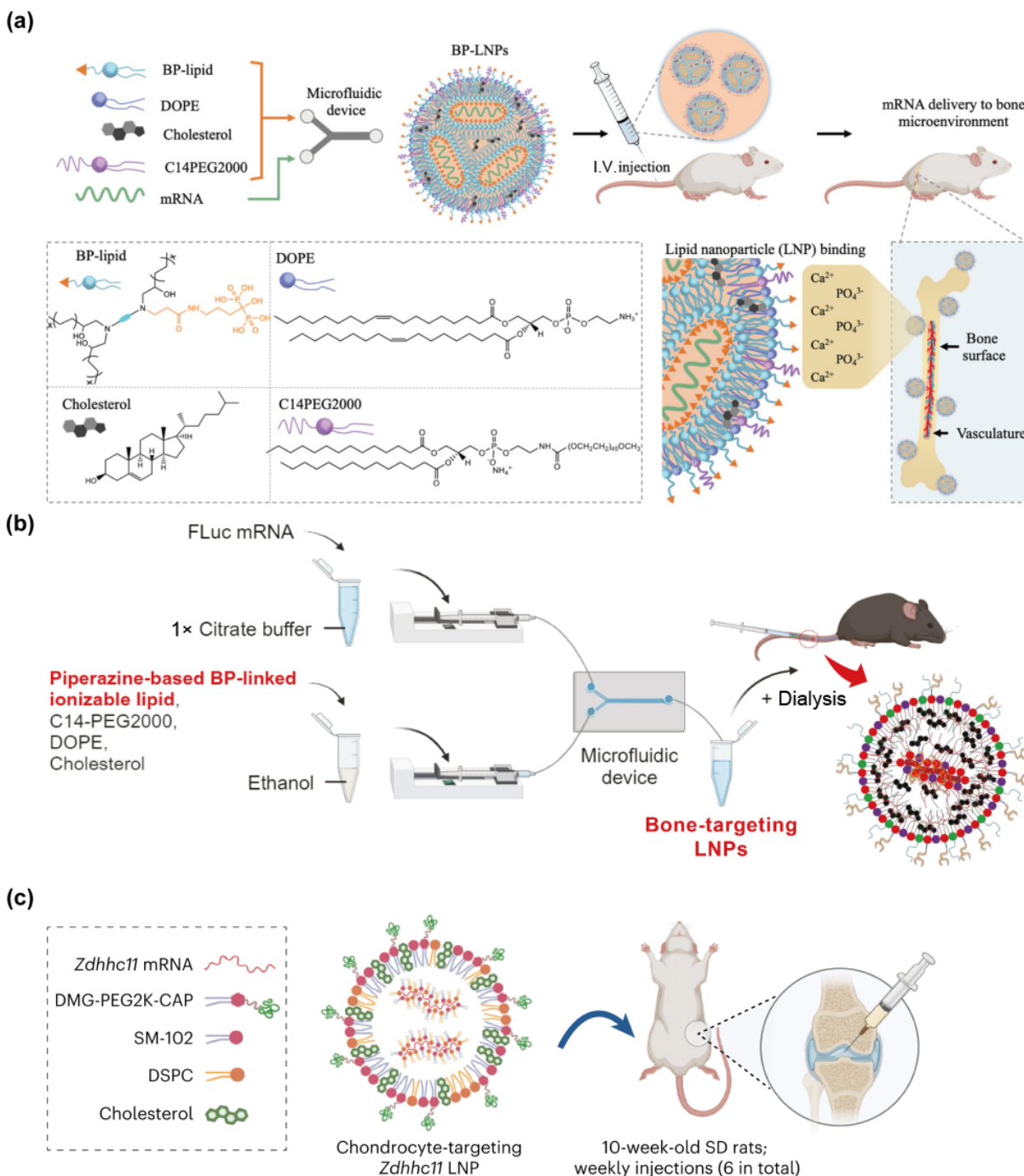


Figure 3 LNPs conjugated with affinity ligands for targeting. (a) LNPs prepared by components including bisphosphonate (BP)-conjugated ionizable lipids promote mRNA delivery to the bone microenvironment. Reproduced with permission from Ref. [29], © American Chemical Society 2022. (b) Piperazine-based bisphosphonate-linked LNPs enhance efficient mRNA delivery efficiency in the bone microenvironment. Adapted with permission from Ref. [31], © Yoon, I. C. et al. *Angewandte Chemie International Edition* published by Wiley-VCH GmbH 2024. (c) *Zdhhc11* mRNA-LNPs based on CAPs functionalized PEG-lipids specifically target chondrocytes. Reproduced with permission from Ref. [32], © Wang, K. et al. 2025.

delivery performance at an N/P ratio of 8:1 and a PEG fraction of 2.5% [33]. Furthermore, as a direct physicochemical consequence of the formulation, the size of LNP constitutes a critical determinant of targeting efficiency. Martin et al. elucidated that bone marrow vessels display high permeability primarily for nanoparticles smaller than 30 nm [34]. Consequently, the further miniaturization of LNPs may improve their enrichment within bone tissue after intravenous administration. Conversely, in the context of intra-articular injection, the rapid leakage and clearance of LNPs present substantial challenges. While smaller particles are highly susceptible to clearance via lymphatic drainage and systemic circulation, increasing the diameter of nanoparticles promotes the *in situ* retention of the mRNA-LNPs following injection into the joint cavity. Sun et al. reported that increasing the aqueous-to-lipid (W/O) ratio, reducing PEG content, neutralizing the LNP surface charge, and enlarging particle size can significantly enhance LNP accumulation and mRNA expression within the articular space, thereby minimizing systemic exposure [35].

5 Strategy for combination of LNPs and functional biomaterial scaffolds

By integrating various biomedical materials as carriers of LNPs according to their characteristics, multifunctional drug delivery platforms capable of precise spatiotemporal control over release can be formed.

5.1 Hydrogel

Hydrogels represent a class of three-dimensional polymeric networks characterized by high porosity and substantial water content. Based on their source materials, hydrogels are classified as natural or synthetic [36]. Natural hydrogels encompass networks derived from collagen, gelatin, silk fibroin, hyaluronic acid (HA), alginate, chitosan, DNA, and extracellular matrix (ECM).

Synthetic hydrogels are typically fabricated from polymers such as polyacrylamide (PAM), polyvinyl alcohol (PVA), and PEG. Capable of mimicking the native extracellular matrix, hydrogels exhibit remarkable biocompatibility. By altering their chemical composition, crosslinking density, and pore architecture, the degradation kinetics, mechanical strength, and other physicochemical properties of hydrogels can be precisely modulated to meet diverse application requirements [37]. The incorporation of LNPs within hydrogel scaffolds significantly prolongs nanoparticle retention at pathological sites and facilitates microenvironment-responsive payload release. For instance, Li et al. fabricated a stimuli-responsive drug delivery platform, circ-srIκBα@LNP-SHC, by encapsulating LNPs carrying circular RNA (circRNA) srIκBα into a silk fibroin composite hydrogel (SHC) crosslinked via matrix metalloproteinase (MMP)-cleavable peptides (Fig. 4(a)) [38]. Upon exposure to the elevated MMP concentrations characteristic of the osteoarthritis microenvironment, this system undergoes localized degradation to trigger the on-demand release of circ-srIκBα@LNPs. This targeted intervention successfully suppressed local inflammation and downregulated the production of matrix-degrading enzymes, thereby effectively ameliorating osteoarthritis pathology.

Due to their excellent biocompatibility, low immunogenicity and tunable degradation rate, composite hydrogels can serve as long-term reservoirs of LNPs. Clinically, the precise modulation of physicochemical properties enables minimally invasive

interventions, such as injectable formulations, and facilitates on-demand payload release adapted to specific pathological conditions.

5.2 Microsphere

Microspheres are spherical particles with micrometer-scale diameters, typically synthesized from natural or synthetic polymers via techniques including emulsion polymerization and microfluidics [39–41]. These discrete carriers exhibit high porosity, a large specific surface area, and excellent injectability. During the manufacturing process, microspheres' particle size, chemical composition, pore architecture, and surface properties can be precisely engineered to accommodate specific tissue microenvironments and drug delivery requirements [42–44].

Microspheres can incorporate LNPs via physical adsorption or chemical conjugation, enabling the sustained and controlled release of mRNA-LNPs. Besides, these systems can be engineered to respond to the microenvironment. For instance, Zheng et al. developed MMP-responsive gelatin methacryloyl (GelMA) hydrogel microspheres encapsulating ubiquitin-specific protease 26 (USP26) mRNA-loaded LNPs [45]. Within the osteoporotic bone defect microenvironment, these microspheres underwent specific degradation, releasing the therapeutic LNPs, thereby precisely modulating the crosstalk between osteoblasts and osteoclasts and promoting intervertebral fusion in a rat model. Moreover, microsphere platforms offer robust multifunctional integration capabilities. For example, Gong et al. developed two-component microspheres comprising GelMA and fucoidan, and utilized chemical grafting to immobilize *Brachyury* (Bry) mRNA-loaded LNPs onto the microsphere surface (Fig. 4(b)) [46]. While the LNPs delivered mRNA to promote extracellular matrix synthesis, the sustained release of fucoidan from the microspheres exerted a concomitant anti-inflammatory effect, thereby achieving the synergistic treatment of intervertebral disc degeneration.

Compared to hydrogels, microspheres exhibit highly uniform sizes and enhanced porosity. Clinically, their inherent lubricating properties can significantly alleviate joint symptoms by reducing friction between damaged articular cartilage surfaces. Furthermore, precise surface modifications including tuning the surface charge, functionalizing with antibodies or peptides, enable superior penetration into the ECM. Notably, engineering dual-targeting systems by simultaneously modifying both the microspheres and the encapsulated LNPs holds immense clinical potential for maximizing therapeutic efficacy at specific lesion sites.

5.3 3D-printed scaffold

3D-printed scaffolds represent an advanced class of materials precisely fabricated via additive manufacturing technologies to feature customized macroscopic architectures and interconnected pore networks. Besides, their spatial organization, mechanical properties, and degradation kinetics are intricately governed by the selected raw materials and printing parameters, thereby enabling highly precise structural design [47]. The integration of LNPs within 3D-printed scaffolds simultaneously addresses the complex requirements of localized therapy by providing both essential mechanical support and targeted drug delivery. It is important to note that, at present, the specific integration of mRNA-loaded LNPs with 3D-printed scaffolds remains an unexplored gap in orthopedic applications. However, the successful integration of conventional LNPs provides a critical technical reference. For instance, Dahiya et al. loaded carvacrol-containing LNPs onto a 3D-printed β-

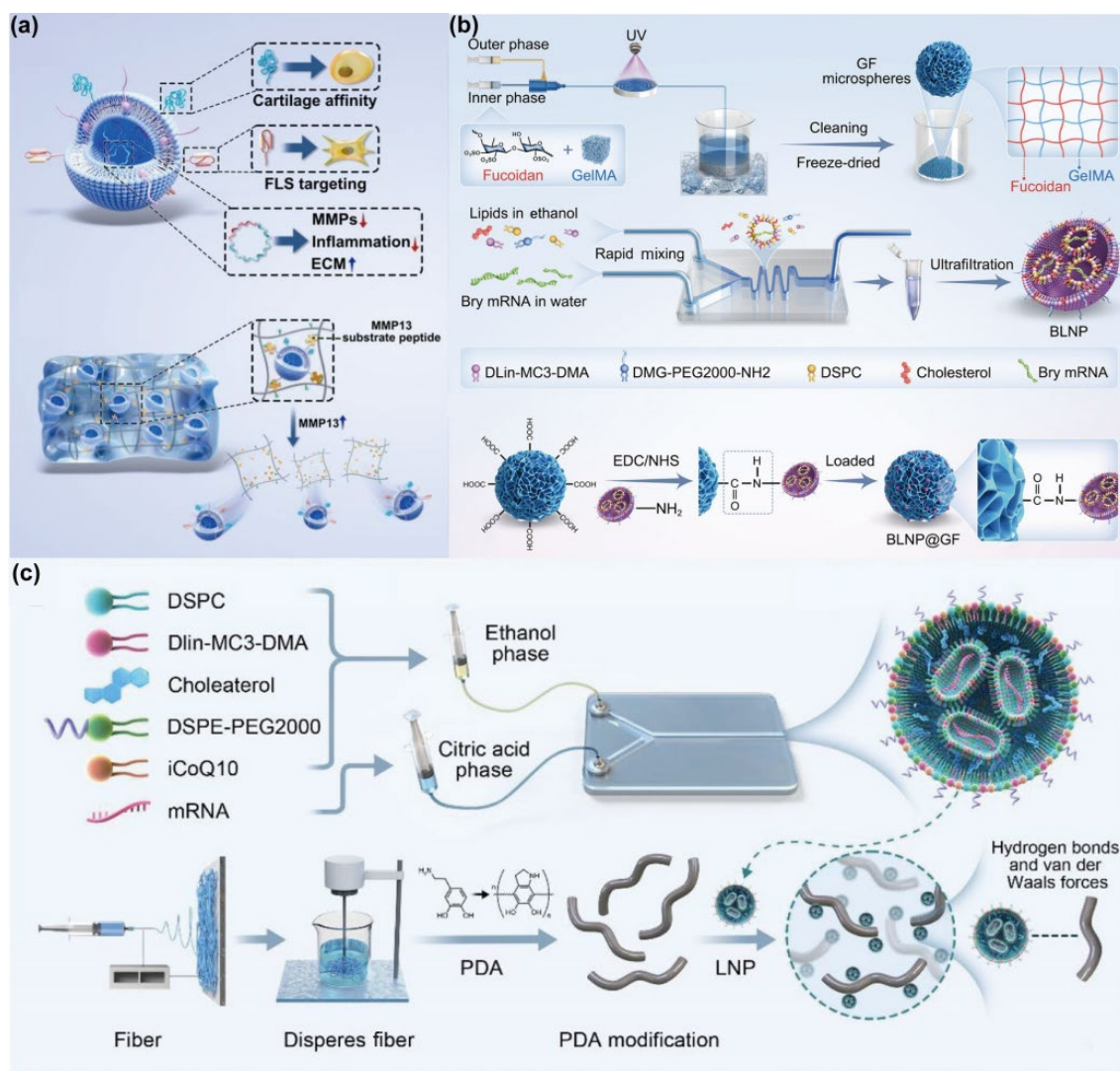


Figure 4 Representative biomaterial scaffolds loaded with LNPs. (a) SHC cross-linked with circ-srIxBa@LNPs by MMP13-specific substrate peptides. Reproduced with permission from Ref. [38], © Li, M. et al. Published by Elsevier B.V. 2025. (b) GelMA/fucoidan (GF) microspheres integrated with *Brachyury* (Bry) mRNA LNPs via amide bonds. Reproduced with permission from Ref. [46], © Gong, Y. et al. Advanced Science published by Wiley-VCH GmbH 2025. (c) PDA-modified short fibers assembled with iCoQ10 functionalized LNPs to construct iCLNP@SF. Reproduced with permission from Ref. [50], © Wiley-VCH GmbH 2025.

tricalcium phosphate (β -TCP) scaffold doped with SiO_2 , achieving the pH-responsive sustained release of carvacrol for up to 35 days [48]. This composite system demonstrated potent antibacterial activity and pro-osteogenic properties, highlighting its significant potential for the treatment of complex bone defects.

Ultimately, 3D-printed scaffolds can recapitulate both the mechanical and biological functions of native tissues, especially bone. By mimicking the properties of the ECM and offering high mechanical strength, these layer-manufactured constructs provide a robust foundation for tissue regeneration. Clinically, the utilization of medical imaging data and computer-aided design allows for the creation of scaffolds with patient-specific anatomical conformity [49]. The fabrication strategies and release mechanisms of these personalized platforms hold immense translational value for the future design of mRNA-LNP-loaded 3D-printed scaffolds.

5.4 Nanofiber/electrospun fiber

Electrospun fibrous membranes represent an advanced class of fibrous scaffolds fabricated via electrospinning technology, currently among the most widely utilized biomedical materials for

drug delivery, exhibiting advantages that include a high surface-area-to-volume ratio, extensive porosity, and tunable fiber alignment, diameter, mechanical properties, and degradation kinetics. Modifying their surface topography enables the precise modulation of cellular activities—such as adhesion, proliferation, and migration—subsequently influencing the interactions between LNPs and target cells. Furthermore, electrospun fibrous membranes protect encapsulated LNPs from premature release and enzymatic degradation *in vivo*, thereby substantially enhancing their stability. Based on these properties, Ling et al. designed a novel ionizable coenzyme Q10 (CoQ10)-engineered lipid/fiber microplexes system (iCLNP@SF) to achieve the efficient co-delivery of CoQ10 and mRNA. They synthesized a new ionizable CoQ10 (iCoQ10) to partially replace traditional ionizable lipids and prepare functionalized iCLNPs [50]. The injectable iCLNP@SF, prepared by assembling iCLNPs with polydopamine (PDA)-modified short fibers, markedly prolonged the stability and local retention time of the encapsulated mRNA (Fig. 4(c)). The system was subsequently utilized to deliver *Runx2* mRNA in femoral condyle defect models of aged mice, significantly improving bone repair.

The structural diversity of electrospun fibers offers tailored clinical options depending on the anatomical characteristics of the injury sites. Injectable short fibers demonstrate minimally invasive applicability, while electrospun fibers can serve as conformable structural coverings that mimic mechanical properties and ECM-like microstructure of native tissues to support regeneration.

6 mRNA-LNP applications in orthopedic diseases

mRNA-LNP platforms are being extensively investigated for the treatment of osteoarthritis, fractures, osteoporosis, rheumatoid arthritis, tendon injuries and other orthopedic disorders.

6.1 Osteoarthritis (OA)

Osteoarthritis is a degenerative joint disease characterized by the progressive destruction of articular anatomical structures and subsequent functional impairment. Its primary pathological hallmarks include osteochondral degeneration and subchondral bone sclerosis [51, 52]. Rapid clearance and limited tissue penetration depth represent the main physiological barriers to effective cartilage-targeted drug delivery. However, compared to the direct administration of therapeutic proteins, LNPs demonstrate a superior capacity to deliver mRNA into the deeper layers of articular cartilage. At the molecular level, OA pathogenesis is driven by multiple inflammatory mediators (e.g., interleukin-1 β (IL-1 β), IL-6, and tumor necrosis factor- α (TNF- α)) and the aberrant activation of signaling pathways, including mitogen-activated protein kinase (MAPK), nuclear factor- κ B (NF- κ B), and Janus kinase/signal transducer and activator of transcription (JAK/STAT). It has been revealed that the expression levels of autophagy-related genes and fibroblast growth factor 18 (FGF-18) are significantly downregulated in the chondrocytes of aged mice. Physiologically, autophagy promotes cell survival under stress, whereas FGF-18 mitigates the degradation of the chondrocyte ECM and stimulates matrix synthesis. Capitalizing on these mechanisms, Kong et al. designed an LNP formulation to deliver FGF-18 mRNA for OA therapy. Following intra-articular injection in an OA model of mouse, LNP-FGF18 effectively upregulated chondrocyte autophagy, suppressed inflammation and cellular senescence, and promoted chondrocyte proliferation (Fig. 5(a)) [53].

The *ex vivo* genetic engineering of mesenchymal stem cells (MSCs) utilizing mRNA-LNPs, followed by their transplantation, represents a promising therapeutic strategy for OA management. Huang et al. synthesized a novel ionizable glycerol ester, TG6A, to formulate LNPs capable of delivering circRNA encoding FGF-18 for the genetic engineering of MSCs. Compared to commercial LNPs formulated with MC3 or ALC-0315, TG6A-LNPs demonstrated significantly augmented transfection efficiency. Following injection into the articular cavities of OA rats, these engineered MSCs effectively promoted cartilage repair [54].

Despite these advances, the intra-articular injection of mRNA-LNPs inherently carries a risk of systemic leakage, which can precipitate off-target mRNA expression in the liver. To achieve the selective retention of LNPs within the articular cavity, Sun et al. systematically optimized their LNP formulation. By utilizing the ionizable lipid PL14 and optimizing both the PEG fraction and W/O ratio, they developed the WG-PL14 LNP for delivering recombinant human FGF-18 mRNA (Fig. 5(b)). Exhibiting a larger particle diameter (100–200 nm) and carrying a net negative surface

charge, this optimized vector successfully achieved prolonged intra-articular enrichment and elicited higher FGF-18 mRNA expression levels compared to standard MC3-based LNPs, subsequently remodeling subchondral bone homeostasis [35].

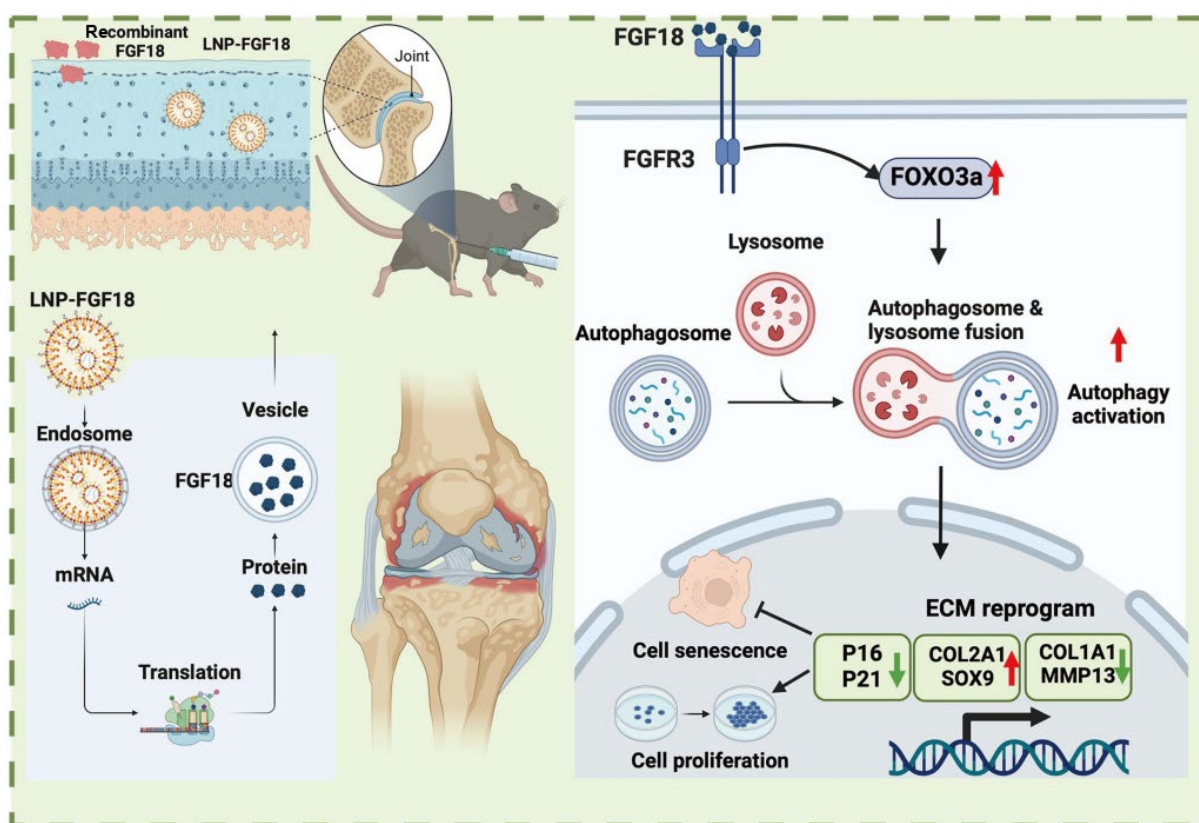
6.2 Fracture

Fractures are characterized by the disruption of bone integrity or continuity. Among all clinical cases, approximately 5% to 10% of patients experience nonunion [55]. Obesity constitutes a significant risk factor that impairs fracture healing. It is associated with elevated levels of pro-inflammatory cytokines, which precipitate chronic inflammation and subsequently delay bone regeneration [56]. Adiponectin (APN) activates APPL1-mediated Akt1 signaling to inhibit osteoclastogenesis, while simultaneously stimulating Runx and bone morphogenetic protein 2 (BMP2) pathways to promote osteoblast differentiation [57–59]. Notably, circulating APN levels are typically downregulated in obese individuals [60]. To address this, Xie et al. synthesized an LNP formulation for delivering APN mRNA, which successfully upregulated the expression of osteogenic genes in MC3T3-E1 osteoblast precursor cells. Utilizing a diet-induced obese mouse model of femoral fracture, the researchers demonstrated that the intravenous injection of APN-LNPs significantly accelerated bone healing (Fig. 6(a)) [61]. Besides, this treatment attenuated body weight and blood glucose levels in the obese mice, concomitant with an increase in the levels of the anti-inflammatory cytokine IL-10 within white adipose tissue. The callus formation phase of fracture healing encompasses two processes: endochondral ossification and intraosseous ossification. Studies indicate that β -catenin regulates the transdifferentiation of chondrocytes into osteoblasts, thereby driving endochondral ossification. Anna et al. developed an LNP based on the ionizable lipid SM-102 for delivering β -catenin mRNA. This formulation demonstrated superior mRNA transfection efficiency compared to LNPs fabricated with the ionizable lipid MC3 [62]. In a mouse tibial fracture model, the SM-102-LNP-mediated delivery of β -catenin mRNA activated the canonical Wnt pathway. It promoted the transdifferentiation of chondrocytes into osteoblasts and effectively facilitated bone formation, thereby providing a novel strategy for improving bone healing after fractures (Fig. 6(b)).

6.3 Osteoporosis

Osteoporosis is a prevalent chronic skeletal disorder arising from an imbalance between bone resorption and bone formation. The precise modulation of osteoblast and osteoclast metabolism, aimed at restoring bone tissue homeostasis, has proven effective in mitigating bone loss [63]. USP26 plays a pivotal role in maintaining bone tissue homeostasis. By stabilizing β -catenin via deubiquitination, USP26 promotes the osteogenic differentiation of bone marrow-derived mesenchymal stem cells (BMSCs). Additionally, USP26 inhibits the NF- κ B signaling pathway, thereby attenuating osteoclastogenesis. According to these mechanisms, Zheng et al. encapsulated USP26 mRNA-LNPs within MMP-responsive GelMA hydrogel microspheres to engineer bone homeostasis repair carriers (BHRCs). These microcarriers effectively stimulated the osteogenic differentiation of BMSCs while suppressing the osteoclastogenic differentiation of bone marrow-derived macrophages. The dual action successfully modulated osteoblast-osteoclast crosstalk and facilitated intervertebral fusion in an osteoporotic rat model (Fig. 7(a)) [45]. The differentiation mechanism of BMSCs is complex. Liu et al. demonstrated that the depletion of methyltransferase 1 (METTL1) in BMSCs significantly

(a)



(b)

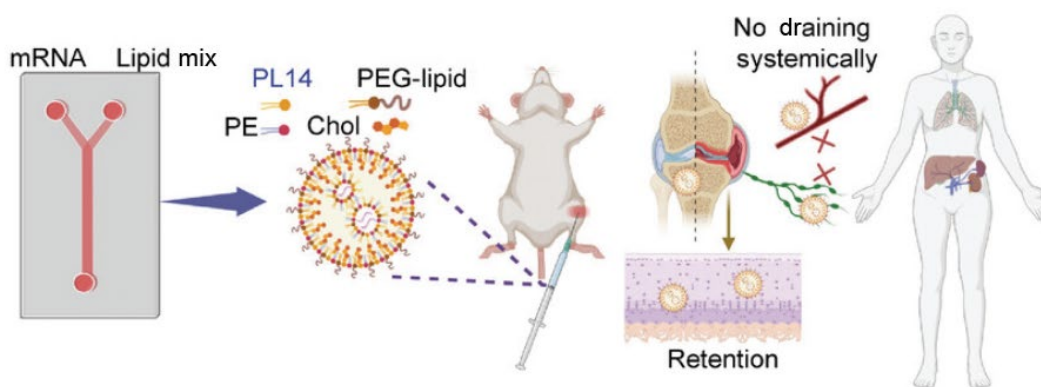


Figure 5 mRNA-LNPs improved osteoarthritis. (a) FGF18 mRNA-LNPs significantly alleviated chondrocyte senescence and degeneration in senile OA models. Reproduced with permission from Ref. [53], © Kong, K. et al. 2025. (b) WG-PL14 LNPs achieve specific accumulation in the articular cavity while minimizing expression in the liver. Reproduced with permission from Ref. [35], © Sun, M. et al. Advanced Healthcare Materials published by Wiley-VCH GmbH 2024.

diminished the m⁷G modification levels of Runx2 mRNA, thereby impeding BMSC osteoblast differentiation and driving age-related osteoporosis [64]. To improve osteogenesis, Liu et al. engineered LNPs to deliver m⁷G-methylated *Runx2* mRNA. By conjugating zoledronic acid to the PEG-lipids of these nanoparticles to enable skeletal targeting, the formulation significantly accelerated bone repair and formation in a murine model of age-related osteoporosis (Fig. 7(b)).

6.4 Rheumatoid arthritis

Rheumatoid arthritis (RA) is a common chronic autoimmune disease characterized by persistent joint inflammation, synovial

hyperplasia, and progressive bone destruction. Heat shock protein 10 (HSP10) represents a critical therapeutic target for RA. It drives the maturation of immunosuppressive regulatory T (Treg) cells and modulates systemic immune responses [65, 66], while simultaneously inhibiting the activity of invasive synovial fibroblasts responsible for cartilage degradation. Liu et al. engineered heparin-protamine-modified LNPs to deliver HSP10 mRNA specifically to RA lesion sites, capitalizing on their upregulated expression of heparinase (Fig. 8(a)). In a mouse RA model, this LNP formulation demonstrated superior therapeutic efficacy compared to methotrexate, a first-line clinical drug [67]. Furthermore, because antibody glycosylation patterns profoundly dictate their

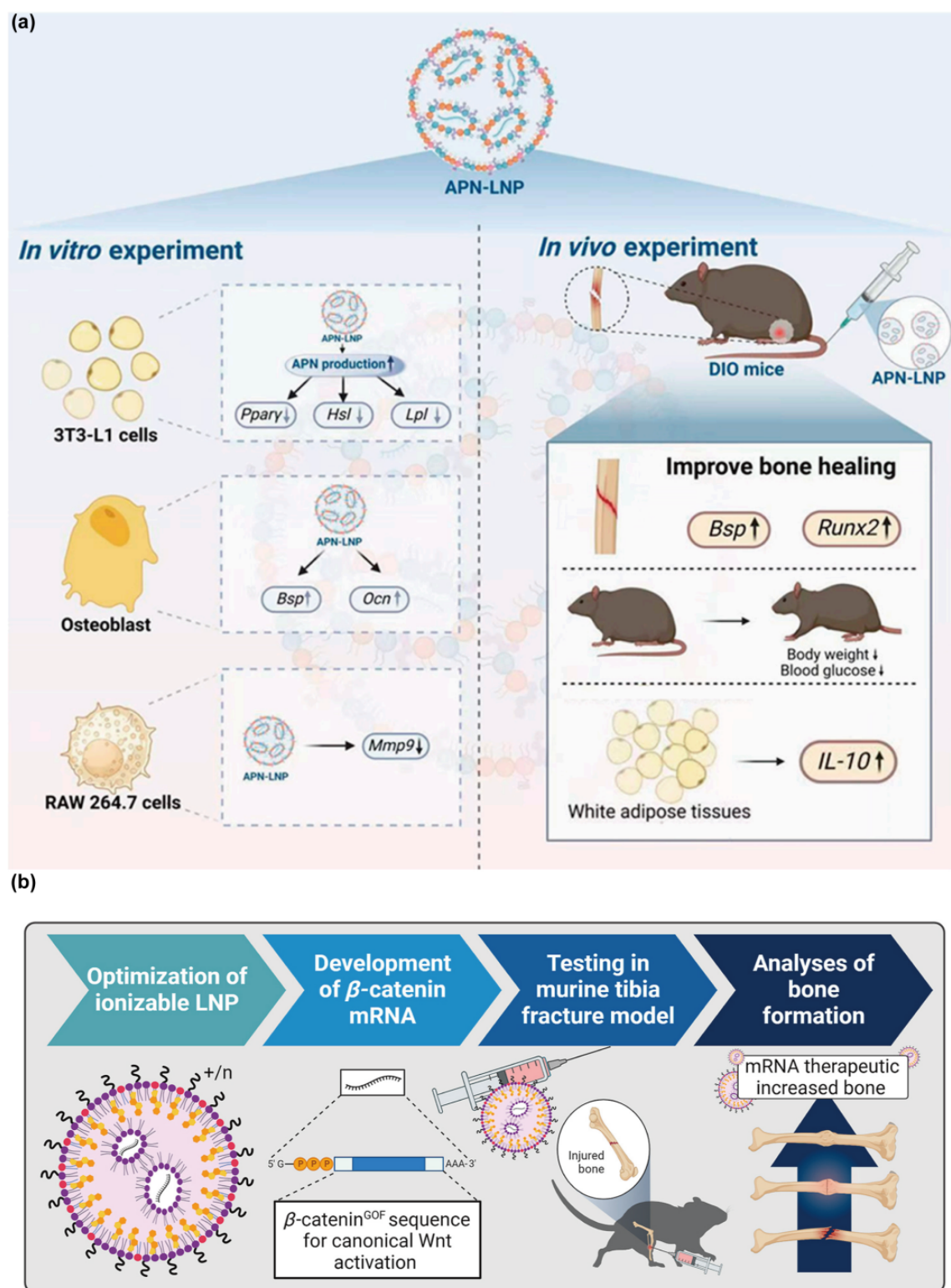


Figure 6 mRNA-LNPs promote bone healing. (a) APN-LNP treatment enhances bone healing in a femoral fracture model of male diet-induced obesity mice. Reproduced with permission from Ref. [61], © Xie, Y. et al. Licensee MDPI, Basel, Switzerland 2025. (b) SM-102-β-catenin^{GOF} mRNA promotes bone formation in a murine tibia fracture model. Reproduced with permission from Ref. [62], © Nelson, A. L. et al. Publishing services by Elsevier B.V. on behalf of KeAi Communications Co. Ltd. 2024.

immunological function, modulating these post-translational modifications represents a promising strategy for treating RA. Studies reveal that diminished sialylation and galactosylation of the immunoglobulin G (IgG) Fc region strongly correlate with pro-

inflammatory responses. To address this, Peng et al. utilized LNPs for the simultaneous co-delivery of mRNA encoding the glycosyltransferases beta-1,4-galactosyltransferase (B4GALT) and beta-galactoside alpha-2,6-sialyltransferase 1 (ST6GAL1) (Fig. 8(b)).

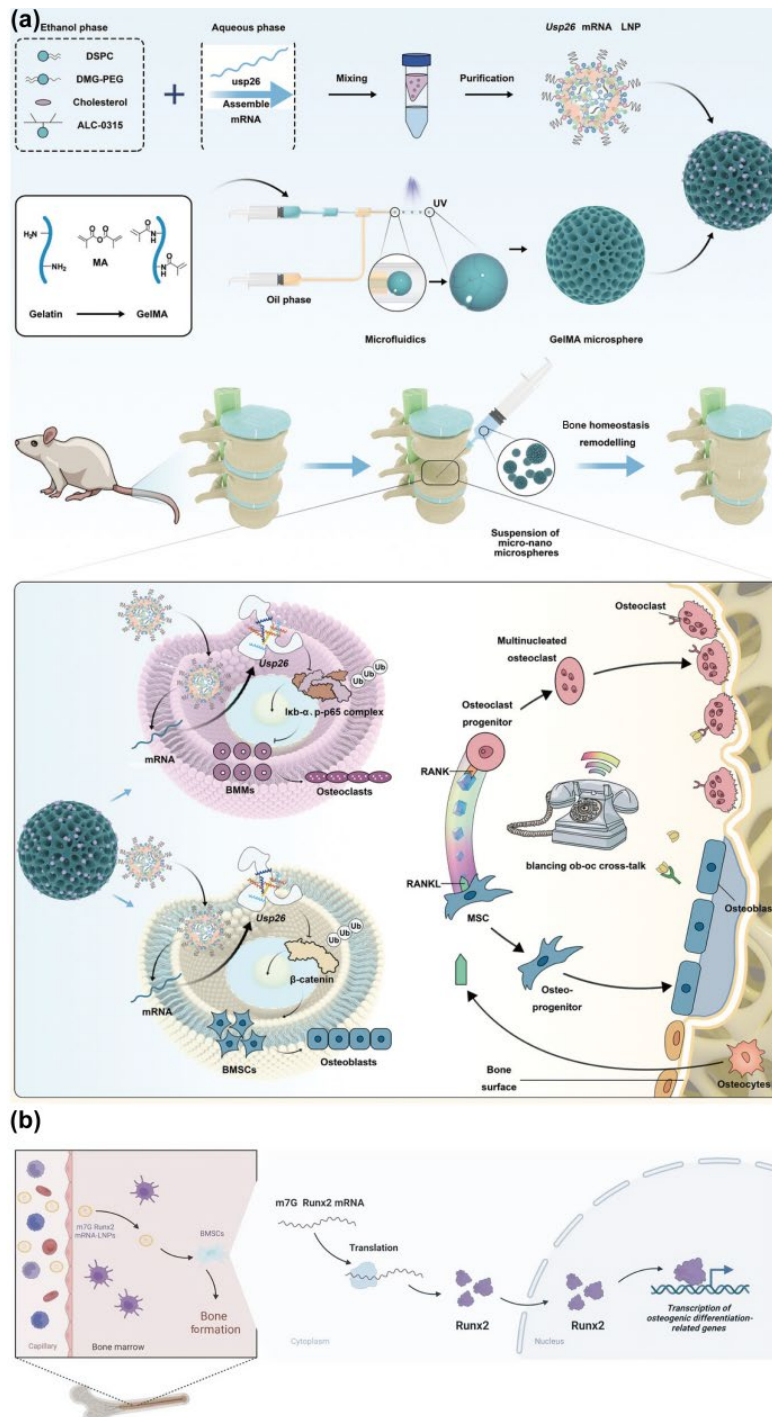


Figure 7 mRNA-LNPs alleviate osteoporosis. (a) The fabrication procedure and therapeutic mechanism of GelMA hydrogel microspheres encapsulating USP26 mRNA. Reproduced with permission from Ref. [45], © Wiley-VCH GmbH 2024. (b) Zoledronic acid modified LNPs deliver m7G *Runx2* mRNA to accelerate bone formation. Reproduced with permission from Ref. [64], © Elsevier Ltd. All rights reserved 2023.

Following intra-articular injection, this approach successfully enhanced IgG Fc glycosylation and significantly alleviated RA pathology [68].

6.5 Tendon injury

Tendon injuries are broadly classified into acute ruptures and chronic tendinopathies. In contrast to normal tissue, chronic tendinopathy is characterized by an elevated ratio of type III collagen (Col III) to type I collagen (Col I) synthesis. This

pathological shift results in disorganized collagen alignment and compromised mechanical properties. To address this, Ruben et al. engineered hybrid lipid nanoparticles by fusing MSC-derived extracellular vesicles (EVs) with synthetic LNPs to deliver *col1a1* mRNA into tendon stem/progenitor cells (TSPCs), thereby upregulating Col I expression within the tendinopathic microenvironment [69]. By integrating the inherent biocompatibility of EVs with the high transfection capacity of LNPs, this platform successfully promoted *in vitro* Col I synthesis in

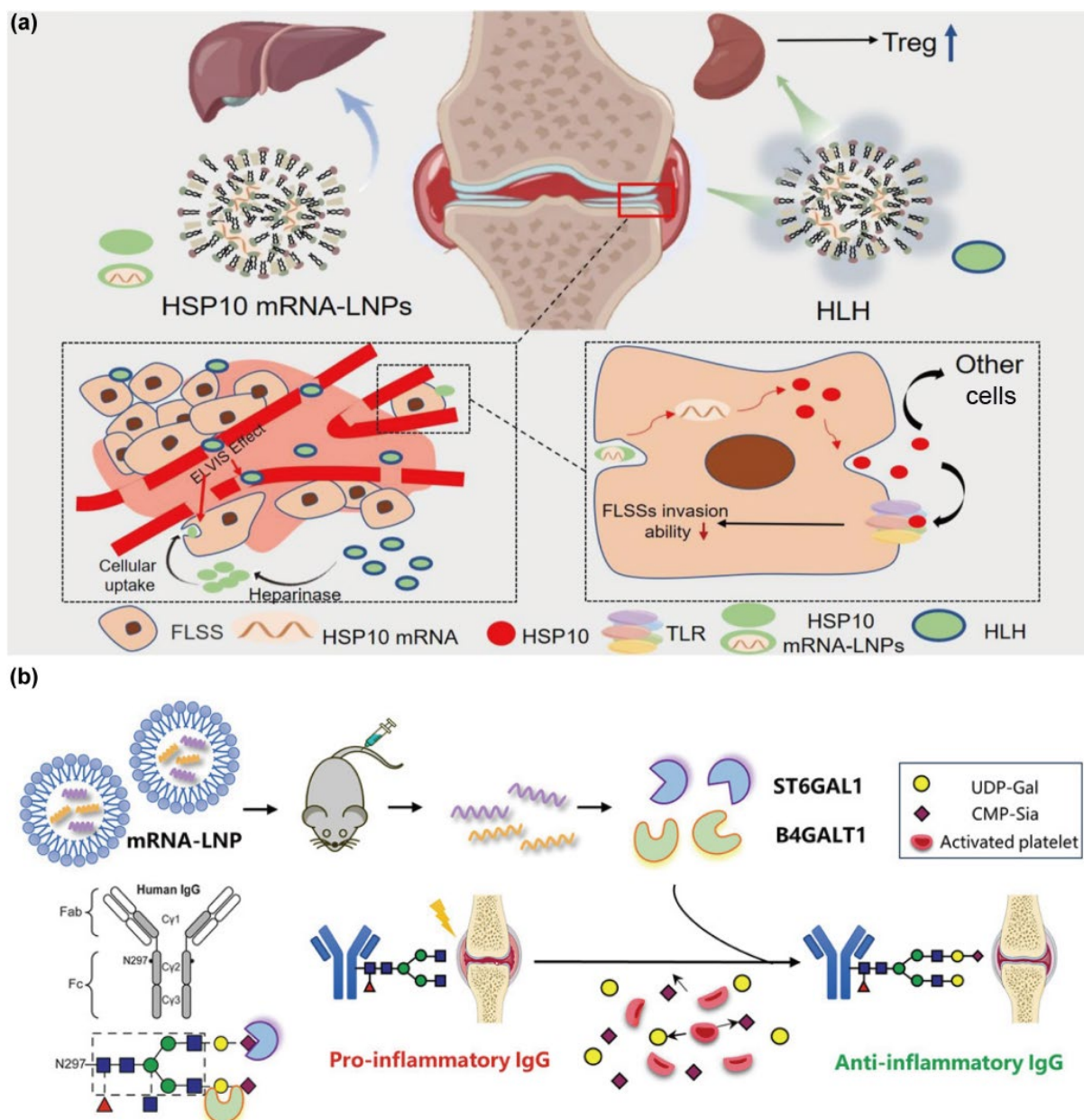


Figure 8 The application of mRNA-LNPs in rheumatoid arthritis treatment. (a) Heparin particles modified HSP10 mRNA-LNPs are able to reach RA sites after intravenous injection. Reproduced with permission from Ref. [67], © American Pharmacists Association. Published by Elsevier Inc. All rights are reserved, including those for text and data mining, AI training, and similar technologies 2024. (b) Glycosyltransferase-encoded mRNA-LNP modulate IgG glycosylation, shifting immune responses from pro-inflammatory to anti-inflammatory states. Reproduced with permission from Ref. [68], © The American Society of Gene and Cell Therapy. Published by Elsevier Inc. All rights are reserved, including those for text and data mining, AI training, and similar technologies 2025.

TSPCs at levels comparable to conventional LNPs.

The repair process following acute tendon injury progresses through three overlapping phases: inflammation, proliferation, and remodeling. During this progression, the ECM undergoes dynamic reorganization, shifting from the initial deposition of provisional Col III to the synthesis of mature Col I. Tendon fibrosis constitutes a prevalent complication following acute injury, which restricts tendon gliding and significantly elevates the risk of re-rupture. The transforming growth factor- β (TGF- β)/SMAD3 signaling pathway drives the development of peritendinous fibrotic adhesions; consequently, the targeted knockdown of SMAD3 expression has been shown to promote scarless tendon healing [70]. To achieve the dual therapeutic objectives of accelerating tendon regeneration and

preventing adhesions, Sandra et al. engineered LNPs for the simultaneous co-delivery of SMAD3 small interfering RNA (siRNA) and *col1a1* mRNA. This combinatorial nanotherapeutic effectively silenced SMAD3 expression while concomitantly upregulating collagen type I alpha 1 chain (COL1A1) production in tenocytes (Fig. 9(a)) [71].

6.6 Other orthopedic diseases

Following spinal cord injury, profound inflammatory responses, cell death, and scar formation within the injury microenvironment significantly impede neural regeneration and functional recovery. Gál et al. encapsulated nucleoside-modified mRNA encoding human IL-10 into LNPs and administered these vectors directly

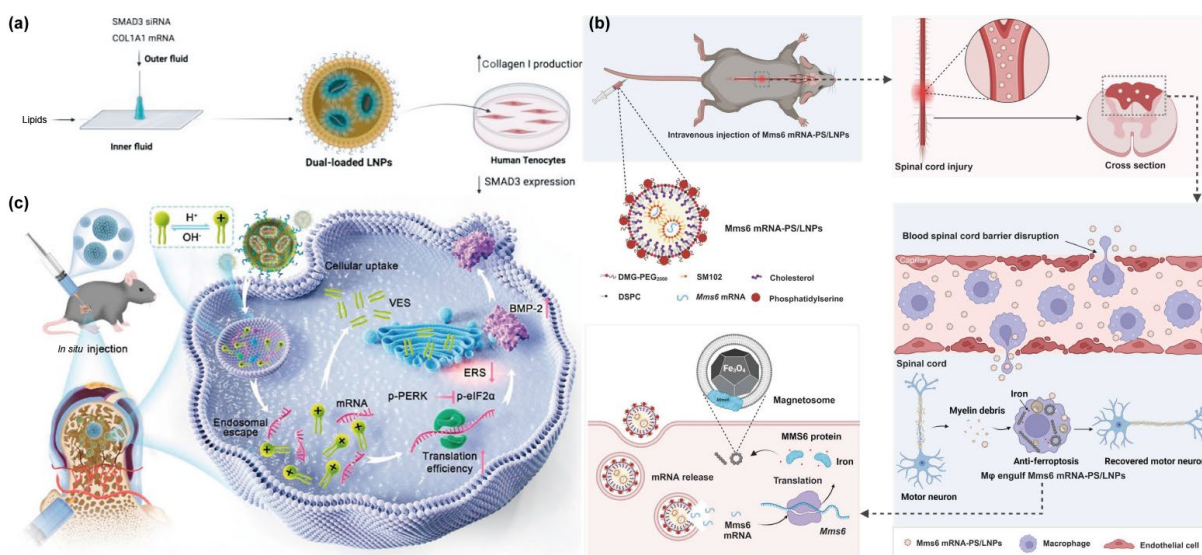


Figure 9 mRNA-LNPs promote the regeneration of tendon, spinal cord, and femoral head. (a) LNPs loaded simultaneously with SMAD3 siRNA and collagen I mRNA enhance collagen I production and inhibit SMAD3 in tenocytes. Reproduced with permission from Ref. [71], © López-Cerdá, S. et al. Published by American Chemical Society 2024. (b) Intravenous injection of Mms6 mRNA-LNPs promote locomotor function recovery and morphological repair in traumatic spinal cord injury mice. Reproduced with permission from Ref. [73], © Fu, C. et al. some rights reserved; exclusive licensee American Association for the Advancement of Science 2025. (c) Injectable iLMPs relieve SANFH through mitigating ERS and reducing eukaryotic initiation factor 2 α (eIF2 α) phosphorylation. Reproduced with permission from Ref. [75], © Chen, Y. et al. Publishing services by Elsevier B.V. on behalf of KeAi Communications Co. Ltd. 2026.

into the lesion site of a rat spinal cord injury model one week post-injury [72]. This delayed therapeutic intervention significantly attenuated the microglial and macrophage activation within the perilesional zone, upregulated the expression of neuroprotective factors such as tissue inhibitor of metalloproteinases 1 (TIMP-1) and ciliary neurotrophic factor (CNTF), and ultimately facilitated both tissue preservation and locomotor recovery. To achieve targeted cellular modulation, Fu et al. engineered phosphatidylserine-modified LNPs encapsulating Mms6 mRNA (Mms6 mRNA-PS/LNPs) designed to specifically home to macrophages [73]. Following intravenous administration, these functionalized LNPs efficiently localized within macrophages at the injury epicenter, augmenting their resistance to ferroptosis (Fig. 9(b)). This intracellular regulation effectively mitigated secondary tissue damage, promoted neuronal survival and axonal regeneration, and significantly enhanced the locomotor function of the murine subjects.

Steroid-associated necrosis of the femoral head (SANFH) is a refractory orthopedic disorder primarily induced by long-term use of glucocorticoids (GCs) [74]. GCs lead to elevated reactive oxygen species (ROS) levels and activation of endoplasmic reticulum stress (ERS) signaling pathways. Given that the increase of ERS significantly reduced mRNA translation in ribosomes, targeted intervention in ERS signaling may mitigate bone destruction and delay the progression of SANFH. To address this, Chen et al. synthesized a novel vitamin E-derived ionizable lipid (iVES) and employed it to construct lipid-hydrogel microplexes (iLMPs) capable of simultaneously delivering mRNA and attenuating ERS (Fig. 9(c)) [75]. After loading BMP-2 mRNA, the *in situ* injection of these iLMPs into the femoral heads of rat SANFH models demonstrated a marked reduction in necrotic severity alongside a significant improvement of bone structure.

To provide a clear overview of the current research landscape, the aforementioned mRNA-LNP platforms applied in various orthopedic diseases are systematically summarized in Table 1.

7 Conclusions and perspectives

Propelled by recent breakthroughs in mRNA technology and LNP delivery systems, mRNA-LNP platforms have demonstrated immense therapeutic promise in the management of orthopedic diseases. Their application across diverse disorders—including osteoarthritis, fractures, osteoporosis, rheumatoid arthritis, and tendon injuries—underscores their profound utility in restoring or augmenting intracellular proteins and cytokines, modulating cellular fate, and orchestrating tissue repair. Nevertheless, translating these preclinical findings into routine clinical practice remains hindered by several formidable challenges.

Foremost among these challenges is the inherent *in vivo* instability and rapid systemic clearance of mRNA-LNPs, which substantially limit their duration of action and long-term efficacy. To overcome these translatability barriers, promising strategies include rigorous optimization of lipid formulation, alongside employment of localized administration routes, and integration of LNPs into advanced biomaterial scaffolds. Moving forward, artificial intelligence (AI) is poised to serve as transformative tools to accelerate LNP formulation design and component screening [76]. Furthermore, the current storage and logistical distribution of mRNA-LNPs predominantly rely on ultra-low temperature cryopreservation or freeze-drying methods, demanding urgent innovations in both formulation chemistry and the development of next-generation dosage forms to extend shelf life.

Beyond stability issues, mRNA-LNPs possess the potential to trigger hypersensitivity reactions via complement system activation and mediating the production of anti-PEG antibodies. Consequently, future research must prioritize the rational design of ionizable lipids and the development of viable alternatives to PEG-lipids to enhance delivery efficiency while minimizing the inherent immunogenicity, thereby mitigating the incidence of adverse reactions associated with prolonged, repeated administrations.

To address the multifaceted pathogenic mechanisms and highly heterogeneous microenvironments of orthopedic disorders,

Table 1 mRNA-LNP platforms for different orthopedic diseases

Name	mRNA	Delivery strategy	Disease	Ref.
Zdhhc11 mRNA-LNP-CAP complex	Zdhhc11 mRNA	Ligand modification	Osteoarthritis	[32]
circ-srIxB α @LNP-SHC	circ-srIxB α	Ligand modification, hydrogel encapsulation	Osteoarthritis	[38]
LNP-FGF18 mRNA system	FGF18 mRNA	Localized injection	Osteoarthritis	[53]
LNP-rhFGF18 mRNA complex	RhFGF18 mRNA	Localized injection, formulation optimization	Osteoarthritis	[35]
APN-LNP	APN mRNA	\	Fracture	[61]
SM-102- β -cateninGOF mRNA	β -cateninGOF mRNA	Localized injection	Fracture	[62]
Bone homeostasis repair microcarrier	USP26 mRNA	Microsphere encapsulation, localized injection	Osteoporosis	[45]
ZA-m7G Runx2 mRNA-LNP	m7G methylated Runx2 mRNA	Ligand modification	Osteoporosis	[64]
H-mRNA-LNPs@heparin/Protamine	HSP10 mRNA	Ligand modification	Rheumatoid arthritis	[67]
Glycosyltransferase-encoded mRNA-LNP	B4GALT1 and ST6GAL1 mRNAs	\	Rheumatoid arthritis	[68]
MSC-Hyb NP	Collagen type I mRNA	\	Tendon injury	[69]
LNPs loaded simultaneously with SMAD3 siRNA and collagen I mRNA	Collagen type I mRNA	\	Tendon injury	[71]
Mms6 mRNA-PS/LNP	Mms6 mRNA	Ligand modification	Spinal cord injury	[73]
iLMP	BMP-2 mRNA	Microsphere encapsulation, localized injection	Steroid-associated necrosis of the femoral head	[75]
BLNP@GF	Brachyury mRNA	Microsphere encapsulation, localized injection	Intervertebral disc degeneration	[46]
iCLNP@SF	Runx2 mRNA	Nanofiber encapsulation, localized injection	Bone defect	[50]

advanced carrier design and combinatorial delivery strategies are essential. The co-delivery of multiplexed mRNA or the combinatorial delivery of mRNA alongside other drugs or biological agents via LNPs holds profound translational potential. Furthermore, integrating LNPs with advanced biomaterials necessitates disease-specific rational design to achieve stimuli-responsive, spatiotemporally controlled release.

To date, no mRNA-LNP therapeutics have received approval for clinical applications for orthopedic disorders. The successful clinical translation of mRNA-LNP from preclinical models demands the resolution of critical manufacturing and logistical bottlenecks. Current large-scale production of mRNA-LNPs relies predominantly on microfluidic mixing technologies, demanding continuous refinement of analytical methodologies to ensure formulation uniformity, nucleic acid encapsulation efficiency, and batch-to-batch consistency. These quality control measures are particularly vital for antibody-conjugated or ligand-conjugated LNPs, where minute batch-to-batch variations can profoundly compromise efficacy and safety. After surmounting these scale-up challenges, executing rigorous, large-scale clinical trials is imperative to definitively establish both the long-term safety and efficacy of mRNA-LNP interventions in human cohorts.

Data availability

Not applicable.

Acknowledgements

This work is supported by Key Research and Development Plan of the Ministry of Science and Technology (No. 2023YFC2410705, L.

W.), Shanghai Oriental Talents Program Youth Project (H. T. R.), the National Natural Science Foundation of China (No. 82472419, L. W.), "Two Hundred Talent" Program supported by Shanghai Jiao Tong University School of Medicine (No. 20240701, H. T. R.).

Declaration of competing interest

All the contributing authors report no conflict of interests in this work.

Author contribution statement

L. W. and H. T. R. supervised and revised the manuscript. J. N. G. led the writing of the manuscript. S. Y. Y., G. L., Y. Y. W., S. J. L., R. H. Y. and H. Y. W. contributed to the writing and revisions of the manuscript. All authors approved the submitted version.

Informed consent

Not applicable.

Ethics statement

Not applicable.

Use of AI statement

None.

References

- [1] Du, M. Z.; Gu, Y. L.; Xu, T. H.; Wu, T.; Wang, Y. W.; Liu, J. J.; Fu, W. L.; Lin, S. E.; Jiang, D.; Li, G. Healing beyond tradition:

- Advanced regenerative therapies for joint-related sports injuries. *Innov. Med.* **2025**, *3*, 100132.
- [2] Wilson, B.; Geetha, K. M. Lipid nanoparticles in the development of mRNA vaccines for COVID-19. *J. Drug Deliv. Sci. Technol.* **2022**, *74*, 103553.
 - [3] Baptista, B.; Carapito, R.; Laroui, N.; Pichon, C.; Sousa, F. mRNA, a revolution in biomedicine. *Pharmaceutics* **2021**, *13*, 2090.
 - [4] Sahin, U.; Karikó, K.; Türeci, Ö. mRNA-based therapeutics-developing a new class of drugs. *Nat. Rev. Drug Discov.* **2014**, *13*, 759–780.
 - [5] Pardi, N.; Hogan, M. J.; Porter, F. W.; Weissman, D. mRNA vaccines—a new era in vaccinology. *Nat. Rev. Drug Discov.* **2018**, *17*, 261–279.
 - [6] Hajj, K. A.; Whitehead, K. A. Tools for translation: Non-viral materials for therapeutic mRNA delivery. *Nat. Rev. Mater.* **2017**, *2*, 17056.
 - [7] Uchida, S.; Perche, F.; Pichon, C.; Cabral, H. Nanomedicine-based approaches for mRNA delivery. *Mol. Pharm.* **2020**, *17*, 3654–3684.
 - [8] Pandey, M.; Ojha, D.; Bansal, S.; Rode, A. B.; Chawla, G. From bench side to clinic: Potential and challenges of RNA vaccines and therapeutics in infectious diseases. *Mol. Aspects Med.* **2021**, *81*, 101003.
 - [9] Liu, H. Y.; Yao, X. L.; Zhou, Y.; Chen, L. CircRNA-based therapeutics: Current opinions and clinical potential. *Innov. Med.* **2024**, *2*, 100081.
 - [10] Han, X. X.; Zhang, H. W.; Butowska, K.; Swingle, K. L.; Alameh, M. G.; Weissman, D.; Mitchell, M. J. An ionizable lipid toolbox for RNA delivery. *Nat. Commun.* **2021**, *12*, 7233.
 - [11] Xu, Y.; Golubovic, A.; Xu, S. F.; Pan, A. N.; Li, B. W. Rational design and combinatorial chemistry of ionizable lipids for RNA delivery. *J. Mater. Chem. B* **2023**, *11*, 6527–6539.
 - [12] Eygeris, Y.; Gupta, M.; Kim, J.; Sahay, G. Chemistry of lipid nanoparticles for RNA delivery. *Acc. Chem. Res.* **2022**, *55*, 2–12.
 - [13] Yang, L.; Gong, L. M.; Wang, P.; Zhao, X. H.; Zhao, F.; Zhang, Z. J.; Li, Y. F.; Huang, W. Recent advances in lipid nanoparticles for delivery of mRNA. *Pharmaceutics* **2022**, *14*, 2682.
 - [14] Heyes, J.; Palmer, L.; Bremner, K.; MacLachlan, I. Cationic lipid saturation influences intracellular delivery of encapsulated nucleic acids. *J. Controlled Release* **2005**, *107*, 276–287.
 - [15] Samaridou, E.; Heyes, J.; Lutwyche, P. Lipid nanoparticles for nucleic acid delivery: Current perspectives. *Adv. Drug Deliv. Rev.* **2020**, *154–155*, 37–63.
 - [16] Kim, J.; Eygeris, Y.; Gupta, M.; Sahay, G. Self-assembled mRNA vaccines. *Adv. Drug Deliv. Rev.* **2021**, *170*, 83–112.
 - [17] Vasileva, O.; Zaborova, O.; Shmykov, B.; Ivanov, R.; Reshetnikov, V. Composition of lipid nanoparticles for targeted delivery: Application to mRNA therapeutics. *Front. Pharmacol.* **2024**, *15*, 1466337.
 - [18] Semple, S. C.; Chonn, A.; Cullis, P. R. Influence of cholesterol on the association of plasma proteins with liposomes. *Biochemistry* **1996**, *35*, 2521–2525.
 - [19] Anindita, J.; Tanaka, H.; Yamakawa, T.; Sato, Y.; Matsumoto, C.; Ishizaki, K.; Oyama, T.; Suzuki, S.; Ueda, K.; Higashi, K. et al. The effect of cholesterol content on the adjuvant activity of nucleic-acid-free lipid nanoparticles. *Pharmaceutics* **2024**, *16*, 181.
 - [20] Su, K. X.; Shi, L.; Sheng, T.; Yan, X. X.; Lin, L. X.; Meng, C. Y.; Wu, S. Q.; Chen, Y. X.; Zhang, Y.; Wang, C. R. et al. Reformulating lipid nanoparticles for organ-targeted mRNA accumulation and translation. *Nat. Commun.* **2024**, *15*, 5659.
 - [21] Kim, Y.; Chatterjee, S.; Robertson, A. L.; Guerrero, E. D.; Vaidya, A.; Wang, X.; Lee, S. M.; Wei, J. W.; Miller, W. E.; Farbiak, L. et al. Mannose-conjugated cholesterol containing lipid nanoparticles for active targeted mRNA delivery to liver sinusoidal endothelial and kupffer cells. *Bioconjug. Chem.* **2025**, *36*, 2181–2196.
 - [22] Patel, S.; Ashwanikumar, N.; Robinson, E.; Xia, Y.; Mihai, C.; Griffith, J. P.; Hou, S. G.; Esposito, A. A.; Ketova, T.; Welsher, K. et al. Naturally-occurring cholesterol analogues in lipid nanoparticles induce polymorphic shape and enhance intracellular delivery of mRNA. *Nat. Commun.* **2020**, *11*, 983.
 - [23] Álvarez-Benedicto, E.; Farbiak, L.; Ramirez, M. M.; Wang, X.; Johnson, L. T.; Mian, O.; Guerrero, E. D.; Siegwart, D. J. Optimization of phospholipid chemistry for improved lipid nanoparticle (LNP) delivery of messenger RNA (mRNA). *Biomater. Sci.* **2022**, *10*, 549–559.
 - [24] Zhang, R.; El-Mayta, R.; Murdoch, T. J.; Warzecha, C. C.; Billingsley, M. M.; Shepherd, S. J.; Gong, N. Q.; Wang, L. L.; Wilson, J. M.; Lee, D. et al. Helper lipid structure influences protein adsorption and delivery of lipid nanoparticles to spleen and liver. *Biomater. Sci.* **2021**, *9*, 1449–1463.
 - [25] Lokugamage, M. P.; Vanover, D.; Beyersdorf, J.; Hatit, M. Z. C.; Rotolo, L.; Echeverri, E. S.; Peck, H. E.; Ni, H. Z.; Yoon, J. K.; Kim, Y. et al. Optimization of lipid nanoparticles for the delivery of nebulized therapeutic mRNA to the lungs. *Nat. Biomed. Eng.* **2021**, *5*, 1059–1068.
 - [26] Saber, N.; Senti, M. E.; Schifflers, R. M. Lipid nanoparticles for nucleic acid delivery beyond the liver. *Hum. Gene Ther.* **2024**, *35*, 617–627.
 - [27] Shi, D.; Beasock, D.; Fessler, A.; Szebeni, J.; Ljubimova, J. Y.; Afonin, K. A.; Dobrovolskaia, M. A. To PEGylate or not to PEGylate: Immunological properties of nanomedicine's most popular component, polyethylene glycol and its alternatives. *Adv. Drug Deliv. Rev.* **2022**, *180*, 114079.
 - [28] McCarthy, I. The physiology of bone blood flow: A review. *J. Bone Joint Surg. Am.* **2006**, *88*, 4–9.
 - [29] Xue, L. L.; Gong, N. Q.; Shepherd, S. J.; Xiong, X. H.; Liao, X. Y.; Han, X. X.; Zhao, G.; Song, C.; Huang, X. S.; Zhang, H. W. et al. Rational design of bisphosphonate lipid-like materials for mRNA delivery to the bone microenvironment. *J. Am. Chem. Soc.* **2022**, *144*, 9926–9937.
 - [30] Pazianat, M.; Cooper, C.; Ebetino, F. H.; Russell, R. G. G. Long-term treatment with bisphosphonates and their safety in postmenopausal osteoporosis. *Ther. Clin. Risk Manag.* **2010**, *6*, 325.
 - [31] Yoon, I. C.; Xue, L. L.; Chen, Q. Y.; Liu, J. Y.; Xu, J. C.; Siddiqui, Z.; Kim, D.; Chen, B. L.; Shi, Q. Q.; Laura Han, E. et al. Piperazine-derived bisphosphonate-based ionizable lipid nanoparticles enhance mRNA delivery to the bone microenvironment. *Angew. Chem., Int. Ed.* **2025**, *64*, e202415389.
 - [32] Wang, K. F.; He, W.; Gong, Z.; Gao, J.; Gao, T. Y.; Pan, N.; Wu, D. Z.; Yang, Y.; Li, Z.; Zhao, X. et al. ZDHC11-mediated palmitoylation alleviates chondrocyte senescence and serves as a therapeutic target for osteoarthritis. *Nat. Aging* **2025**, *5*, 2228–2246.
 - [33] Mu, X. M.; Du, X. Y.; Han, H. T.; Liu, F.; Zheng, Z. F.; Hao, J.; Liu, L. J.; Liu, S.; Wei, Z.; Huang, C. F. et al. Intravenous administration of sRNA nanoparticles for treatment of osteoporosis in mice. *Pharmaceutics* **2025**, *17*, 789.
 - [34] Martin, J. D.; Toh, K.; Martin, M. R.; Chen, P. W.; Wang, C. Y.; Igarashi, K.; Mpekris, F.; Stylianopoulos, T.; Stuber, M. D.; Kataoka, K. et al. Bone marrow vessels are hyperpermeable to macromolecules and nanoscale medicine in a size-dependent manner. *J. Controlled Release* **2025**, *382*, 113669.
 - [35] Sun, M. Z.; Ma, B.; Pan, Z. H.; Zhao, Y.; Tian, L. L.; Fan, Y. F.; Kong, W. J.; Wang, J. Y.; Xu, B. Y.; Ao, Y. F. et al. Targeted therapy of osteoarthritis via intra-articular delivery of lipid-nanoparticle-encapsulated recombinant human FGF18 mRNA. *Adv. Healthc. Mater.* **2024**, *13*, 2400804.
 - [36] Fan, M. H.; Pi, J. K.; Zou, C. Y.; Jiang, Y. L.; Li, Q. J.; Zhang, X. Z.; Xing, F.; Nie, R.; Han, C.; Xie, H. Q. Hydrogel-exosome system in tissue engineering: A promising therapeutic strategy. *Bioact. Mater.* **2024**, *38*, 1–30.
 - [37] Wu, W.; Cheng, R. Y.; das Neves, J.; Tang, J. C.; Xiao, J. Y.; Ni, Q.; Liu, X. N.; Pan, G. Q.; Li, D. C.; Cui, W. G. et al. Advances in biomaterials for preventing tissue adhesion. *J. Controlled Release* **2017**, *261*, 318–336.
 - [38] Li, M.; Jia, X. M.; Lai, P. H.; Ma, Y. Q.; Li, F.; Yang, L. Y.; Zhang, S.

- Y.; Yang, C. F.; Luo, Y.; Wang, Q. H. Injectable microenvironment-responsive hydrogels encapsulating engineered NF- κ B-targeting circular RNA for osteoarthritis therapy. *J. Controlled Release* **2025**, 385, 114039.
- [39] Chen, W.; Zheng, D. D.; Chen, H.; Ye, T. J.; Liu, Z. H.; Qi, J.; Shen, H. X.; Ruan, H. T.; Cui, W. G.; Deng, L. F. Circadian clock regulation via biomaterials for nucleus pulposus. *Adv. Mater.* **2023**, 35, 2301037.
- [40] Gan, L.; Xu, T. T.; Zheng, D. D.; Zhu, N.; Li, S. J.; Deng, S. Y.; Lian, Q. Y.; Wang, J. X.; Li, W. L.; Zhang, Z. J. et al. Immuno-training remodeled vascular microenvironment via peptide-functionalized hydrogel microspheres for neural repair. *Adv. Funct. Mater.* **2026**, 36, e19806.
- [41] Xu, T. T.; Gan, L.; Chen, W.; Zheng, D. D.; Li, H. L.; Deng, S. Y.; Qian, D. L.; Gu, T. T.; Lian, Q. Y.; Shen, G. et al. Bridging immune-neurovascular crosstalk via the immunomodulatory microspheres for promoting neural repair. *Bioact. Mater.* **2025**, 44, 558–571.
- [42] Zhu, N.; Wang, F.; Han, Z. Y.; Ling, S. F.; Wong, D. W. C.; Ye, S. L.; Liu, M. Y.; Chen, Y. Y.; Shen, G.; Ni, M. et al. Noninvasive method for achieving the regeneration of damaged nerves via ultrasonic nasal drops. *Bioact. Mater.* **2025**, 49, 342–361.
- [43] Zheng, D. D.; Chen, W.; Chen, T. T.; Chen, X. Y.; Liang, J.; Chen, H.; Shen, H. X.; Deng, L. F.; Ruan, H. T.; Cui, W. G. Hydrogen ion capturing hydrogel microspheres for reversing inflammaging. *Adv. Mater.* **2024**, 36, 2306105.
- [44] Ruan, H. T.; Li, Y. F.; Zheng, D. D.; Deng, L. F.; Chen, G.; Zhang, X. C.; Tang, Y. H.; Cui, W. G. Engineered extracellular vesicles for ischemic stroke treatment. *Innovation* **2023**, 4, 100394.
- [45] Zheng, J. C.; Li, X. Y.; Zhang, F. K.; Li, C. W.; Zhang, X. K.; Wang, F.; Qi, J.; Cui, W. G.; Deng, L. F. Targeting osteoblast-osteoclast cross-talk bone homeostasis repair microcarriers promotes intervertebral fusion in osteoporotic rats. *Adv. Healthc. Mater.* **2024**, 13, 2402117.
- [46] Gong, Y. H.; Shi, W. X.; Liu, X. Z.; Yu, H.; Wu, Y. H.; Xia, Y. Z.; Yue, C. C.; Yang, C. K.; Shen, C.; Pei, R. J. et al. Brachyury-activated fucoidan hydrogel microspheres rejuvenate degenerative intervertebral discs microenvironment. *Adv. Sci.* **2025**, 12, e04195.
- [47] Li, J. T.; Liang, D. N.; Chen, X.; Sun, W. J.; Shen, X. Applications of 3D printing in tumor treatment. *Biomed. Technol.* **2024**, 5, 1–13.
- [48] Dahiya, A.; Chaudhari, V. S.; Kushram, P.; Bose, S. 3D Printed SiO₂-tricalcium phosphate scaffolds loaded with carvacrol nanoparticles for bone tissue engineering application. *J. Med. Chem.* **2024**, 67, 2745–2757.
- [49] Wang, J. W.; Zhao, P. Z.; Li, M. M.; Li, J.; Lin, Y. F. Remedying infectious bone defects via 3D printing technology. *Chin. Chem. Lett.* **2025**, 36, 110686.
- [50] Ling, S. F.; Zhang, Y. X.; Chen, Y. Y.; He, W.; Wu, T. Q.; Wang, J.; Wei, Y. Y.; Lu, M.; Zhao, W. L.; Li, Q. F. et al. Ionizable coenzyme-engineered lipid/fiber microplexes boost ribosomal translation to improve mRNA therapy for degenerative diseases. *Adv. Mater.* **2026**, 38, e13720.
- [51] Molnar, V.; Matišić, V.; Kodvanj, I.; Bjelica, R.; Jeleč, Ž.; Hudetz, D.; Rod, E.; Čukelj, F.; Vrdoljak, T.; Vidović, D. et al. Cytokines and chemokines involved in osteoarthritis pathogenesis. *Int. J. Mol. Sci.* **2021**, 22, 9208.
- [52] Katz, J. N.; Arant, K. R.; Loeser, R. F. Diagnosis and treatment of hip and knee osteoarthritis: A review. *JAMA* **2021**, 325, 568.
- [53] Kong, K. Y.; Li, B. X.; Chang, Y. Y.; Zhao, C.; Qiao, H.; Jin, M. H.; Wu, X. R.; Fan, W. X.; Wang, L.; Qi, Y. S. et al. Delivery of FGF18 using mRNA-LNP protects the cartilage against degeneration via alleviating chondrocyte senescence. *J. Nanobiotechnology* **2025**, 23, 34.
- [54] Huang, K.; Liu, X. Y.; Qin, H. T.; Li, Y. W.; Zhu, J. F.; Yin, B.; Zheng, Q. J.; Zuo, C. J.; Cao, H.; Tong, Z. B. et al. FGF18 encoding circular mRNA-LNP based on glycerolipid engineering of mesenchymal stem cells for efficient amelioration of osteoarthritis. *Biomater. Sci.* **2024**, 12, 4427–4439.
- [55] Zura, R.; Xiong, Z.; Einhorn, T.; Watson, J. T.; Ostrum, R. F.; Prayson, M. J.; Della Rocca, G. J.; Mehta, S.; McKinley, T.; Wang, Z. et al. Epidemiology of fracture nonunion in 18 human bones. *JAMA Surg.* **2016**, 151, e162775.
- [56] Rohm, T. V.; Meier, D. T.; Olefsky, J. M.; Donath, M. Y. Inflammation in obesity, diabetes, and related disorders. *Immunity* **2022**, 55, 31–55.
- [57] Tu, Q. S.; Zhang, J.; Dong, L. Q.; Saunders, E.; Luo, E.; Tang, J.; Chen, J. Adiponectin inhibits osteoclastogenesis and bone resorption via APPL1-mediated suppression of akt1. *J. Biol. Chem.* **2011**, 286, 12542–12553.
- [58] Oshima, K.; Nampei, A.; Matsuda, M.; Iwaki, M.; Fukuhara, A.; Hashimoto, J.; Yoshikawa, H.; Shimomura, I. Adiponectin increases bone mass by suppressing osteoclast and activating osteoblast. *Biochem. Biophys. Res. Commun.* **2005**, 331, 520–526.
- [59] Williams, G. A.; Wang, Y.; Callon, K. E.; Watson, M.; Lin, J. M.; Lam, J. B. B.; Costa, J. L.; Orpe, A.; Broom, N.; Naot, D. et al. *In vitro* and *in vivo* effects of adiponectin on bone. *Endocrinology* **2009**, 150, 3603–3610.
- [60] Yosae, S.; Khodadost, M.; Esteghamati, A.; Speakman, J. R.; Djafarian, K.; Bitarafan, V.; Shidfar, F. Adiponectin: An indicator for metabolic syndrome. *Iran. J. Public Health* **2019**, 48, 1106–1115.
- [61] Xie, Y.; Ma, Q.; Wang, J. H.; Zhu, Z. X.; El-Araby, R. E.; Tu, M.; Li, Z. Y.; Xu, X. Y.; Tu, Q. S.; Chen, J. Effects of modified messenger RNA of adiponectin delivered by lipid nanoparticles on adipogenesis and bone metabolism *in vitro* and *in vivo*. *Cells* **2025**, 14, 891.
- [62] Nelson, A. L.; Mancino, C.; Gao, X. Q.; Choe, J. A.; Chubb, L.; Williams, K.; Czachor, M.; Marcucio, R.; Taraballi, F.; Cooke, J. P. et al. β -catenin mRNA encapsulated in SM-102 lipid nanoparticles enhances bone formation in a murine tibia fracture repair model. *Bioact. Mater.* **2024**, 39, 273–286.
- [63] Luo, X.; Xiao, J. W.; Yang, Q. M.; Lu, X. L.; Huang, Q. J.; Ai, X. J.; Li, B.; Sun, L.; Chen, L. Biomaterials for surgical repair of osteoporotic bone defects. *Chin. Chem. Lett.* **2025**, 36, 109684.
- [64] Liu, J. L.; Zhang, Y. W.; Wu, Y.; Li, G. F.; Ji, N.; Han, R. N.; Tang, H.; Liu, X. R.; Liu, H.; Wang, C. J. et al. Delivery of m7G methylated Runx2 mRNA by bone-targeted lipid nanoparticle promotes osteoblastic bone formation in senile osteoporosis. *Nano Today* **2024**, 54, 102074.
- [65] van Eden, W.; Jansen, M. A. A.; Ludwig, I. S.; Leufkens, P.; van der Goes, M. C.; van Laar, J. M.; Broere, F. Heat shock proteins can be surrogate autoantigens for induction of antigen specific therapeutic tolerance in rheumatoid arthritis. *Front. Immunol.* **2019**, 10, 279.
- [66] Vanags, D.; Williams, B.; Johnson, B.; Hall, S.; Nash, P.; Taylor, A.; Weiss, J.; Feeney, D. Therapeutic efficacy and safety of chaperonin 10 in patients with rheumatoid arthritis: A double-blind randomised trial. *The Lancet* **2006**, 368, 855–863.
- [67] Liu, Y. Y.; Shen, H.; Zang, H. C.; Shi, Y. B.; Qiu, S. N.; Fu, X. L.; Zhang, Y. K.; Chen, X. Q.; Zhou, J. M.; Wan, X. Y. et al. A study on the treatment of rheumatoid arthritis with lipid nanoparticles containing mRNA encoding heat shock protein 10. *J. Pharm. Sci.* **2025**, 114, 1455–1463.
- [68] Peng, X. Y.; Li, Y. Y.; Sun, X.; Ren, G. R.; Li, H. J.; Wang, X. C.; Wang, P. G.; Wang, Q. W.; Ji, Y. Therapeutic effect of galactosyltransferase- and sialyltransferase-encoding mRNA in rheumatoid arthritis. *Mol. Ther.* **2025**, 33, 4143–4155.
- [69] Tello, R. P.; Pavel Lamparelli, E.; Camilla Ciardulli, M.; Hirvonen, J.; Barreto, G.; Mafulli, N.; Porta, G. D.; Santos, H. A. Hybrid lipid nanoparticles derived from human mesenchymal stem cell extracellular vesicles by microfluidic sonication for collagen I mRNA delivery to human tendon progenitor stem cells. *Biomater. Sci.* **2025**, 13, 2066–2081.

- [70] Katzel, E. B.; Wolenski, M.; Loisel, A. E.; Basile, P.; Flick, L. M.; Langstein, H. N.; Hilton, M. J.; Awad, H. A.; Hammert, W. C.; O'Keefe, R. J. Impact of Smad3 loss of function on scarring and adhesion formation during tendon healing. *J. Orthop. Res.* **2011**, *29*, 684–693.
- [71] López-Cerdá, S.; Molinaro, G.; Tello, R. P.; Correia, A.; Waris, E.; Hirvonen, J.; Barreto, G.; Santos, H. A. Antifibrotic and pro-regenerative effects of SMAD3 siRNA and collagen I mRNA-loaded lipid nanoparticles in human tenocytes. *ACS Appl. Nano Mater.* **2024**, *7*, 17736–17747.
- [72] Gál, L.; Bellák, T.; Marton, A.; Fekécs, Z.; Weissman, D.; Török, D.; Biju, R.; Vizler, C.; Kristóf, R.; Beattie, M. B. et al. Restoration of motor function through delayed intraspinal delivery of human IL-10-encoding nucleoside-modified mRNA after spinal cord injury. *Research* **2023**, *6*, 0056.
- [73] Fu, C. Y.; Jin, X. Q.; Ji, K. F.; Lan, K.; Mao, X. J.; Huang, Z. B.; Chen, J.; Zhao, F. D.; Li, P. F.; Hu, X. F. et al. Macrophage-targeted *Mms6* mRNA-lipid nanoparticles promote locomotor functional recovery after traumatic spinal cord injury in mice. *Sci. Adv.* **2025**, *11*, eads2295.
- [74] Zhang, J.; Cao, J. Z.; Liu, Y. F.; Zhao, H. Y. Advances in the pathogenesis of steroid-associated osteonecrosis of the femoral head. *Biomolecules* **2024**, *14*, 667.
- [75] Chen, Y. Y.; He, W.; Ling, S. F.; Zhou, Y.; Chen, J.; Du, Y. W.; Mo, R.; Cui, W. G. Boosting ribosomal translation via ionizable lipid-hydrogel microplexes for localized mRNA therapy. *Bioact. Mater.* **2026**, *59*, 678–696.
- [76] Witten, J.; Raji, I.; Manan, R. S.; Beyer, E.; Bartlett, S.; Tang, Y. H.; Ebadi, M.; Lei, J. Y.; Nguyen, D.; Oladimeji, F. et al. Artificial intelligence-guided design of lipid nanoparticles for pulmonary gene therapy. *Nat. Biotechnol.* **2025**, *43*, 1790–1799.



This is an open access article under the terms of the Creative Commons Attribution 4.0 International License (CC BY 4.0, <https://creativecommons.org/licenses/by/4.0/>).

© The Author(s) 2026. Published by Tsinghua University Press.



清华大学出版社
Tsinghua University Press

SciOpen

94909012 (17 of 17)

Nano Research, 2026, 19, 94909012