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Emerging trends in nanomedicine: The role of RNAi-based therapies and onpattro's clinical journey

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ABSTRACT

Nanomedicine has transformed therapeutic strategies by enabling precise delivery of nucleic acid-based drugs, including small interfering RNA (siRNA), messenger RNA (mRNA), and antisense oligonucleotides. A landmark achievement is Onpattro (patisiran), the first FDA-approved RNAi therapy, which employs lipid nanoparticles (LNPs) to silence transthyretin in hereditary amyloidosis. Its approval validates RNAi as a viable therapeutic modality and underscores the central role of nanocarriers in clinical translation. Despite this success, barriers such as nanoparticle stability, targeted delivery, immunogenicity, and manufacturing scalability remain. Recent advances in mRNA vaccines, CRISPR-based gene editing, and stimuli-responsive nanoparticles are addressing these challenges, supported by growing clinical case studies and real-world data. This review highlights Onpattro's clinical development, compares delivery platforms, discusses translational challenges, and examines emerging technologies that will guide the next generation of RNAi nanomedicines in personalized therapy.

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Introduction

Nanomedicine applies nanotechnology to disease treatment, offering new hope for previously intractable conditions (1). Among the most promising applications of nanomedicine is the delivery of nucleic acid-based therapeutics, such as small interfering RNA (siRNA), messenger RNA (mRNA), and antisense oligonucleotides (ASOs), which offer unprecedented precision in targeting the molecular underpinnings of diseases (1, 2). The advent of these therapies has unlocked the potential to treat genetic disorders, cancers, and viral infections by modulating the expression of specific genes at the RNA level, providing a powerful tool to address previously untreatable conditions (2). Despite their promise, nucleic acid-based nanomedicines face major barriers. These include poor stability, limited delivery efficiency, immune activation, and manufacturing difficulties (3).

A key breakthrough in the clinical development of RNA-based therapeutics has been the approval of Onpattro (patisiran), the first FDA-approved RNA interference (RNAi) therapy. Onpattro, a lipid nanoparticle (LNP)-formulated siRNA, targets the genetic mutation responsible for hereditary transthyretin-mediated amyloidosis (hATTR), a life-threatening disease caused by the accumulation of amyloid fibrils in tissues and organs (4). The approval of Onpattro has set a precedent for RNAi therapies, demonstrating that nucleic acid-based treatments can not only be developed but also successfully commercialized for the treatment of genetic diseases (5). The ability of Onpattro to reduce transthyretin (TTR) protein levels in patients and improve their clinical symptoms represents a significant milestone in the use of RNAi for therapeutic purposes and has sparked further interest in the development of RNA-based nanomedicines for other genetic and chronic diseases (5, 6).

Despite the success of Onpattro, the journey of translating RNAi therapies from preclinical studies to clinical practice is fraught with obstacles. One of the major hurdles is the development of efficient and safe delivery systems for nucleic acids (7). Nucleic acids are highly unstable in the bloodstream and are quickly degraded by nucleases, necessitating the use of advanced drug delivery platforms, such as nanoparticles, to protect these fragile molecules and facilitate their delivery to the target tissues (8).

The field of nanomedicine continues to evolve, with emerging technologies such as CRISPR-Cas9 gene editing, mRNA vaccines, and stimuli-responsive nanoparticles showing promise in overcoming some of these challenges (9). The combination of nanotechnology with gene editing and RNA-based therapies is poised to revolutionize the treatment of genetic disorders, cancers, and other complex

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diseases by providing highly targeted and personalized treatment options (9, 10). As research advances, it is expected that innovations in nanoparticle design, delivery methods, and combination therapies will address current limitations, paving the way for more effective and widely accessible nucleic acid-based treatments. This review specifically focuses on the clinical development of RNAi-based nanomedicines, with Onpattro as a case study, and highlights emerging technologies that may overcome current barriers and guide the future of personalized medicine

Methodology

The literature for this review was identified through a structured search of PubMed, Scopus, and Web of Science databases, covering the period 2010-2024. Keywords included *RNAi, siRNA, lipid nanoparticles, RNAi clinical trials, Onpattro, CRISPR gene editing, nanomedicine delivery*. Priority was given to peer-reviewed articles, clinical trial reports, and high-impact reviews. Inclusion criteria were: (a) studies reporting preclinical or clinical evaluation of RNAi or nucleic acid-based nanomedicines; (b) publications describing clinical development of Onpattro or related RNAi therapeutics; (c) emerging nanotechnologies with translational potential (e.g., mRNA, CRISPR, stimuli-responsive NPs, AI in nanomedicine).

Exclusion criteria were: (a) non-English language papers; (b) conference abstracts without peer-reviewed full texts; (c) studies lacking clear mechanistic or clinical relevance. This approach ensured inclusion of both seminal works and the most recent (2022-2024) literature to maintain relevance and translational focus.

Mechanism of action of nucleic acid-based drugs

Nucleic acid-based drugs, such as RNAi, antisense oligonucleotides, and gene editing technologies, represent a transformative approach in the treatment of genetic and acquired diseases (10, 11). The mechanism of action for these drugs is intricately tied to their ability to modulate gene expression at the RNA or DNA level. RNAi is a key therapeutic strategy that involves that mediate the degradation or inhibition of specific target mRNA molecules, thereby preventing protein synthesis (11). This mechanism plays a critical role in silencing genes responsible for diseases like genetic disorders, cancers, and viral infections (12, 13). Figure 1 illustrates the process of RNAi, highlighting how siRNA molecules mediate the degradation or inhibition of target mRNA to prevent protein synthesis. This mechanism plays a critical role in silencing genes responsible for diseases like genetic disorders, cancers, and viral infections (13). A landmark example of RNAi-based therapy is Onpattro, which targets the mutant transthyretin (TTR) mRNA to treat hereditary TTR-mediated amyloidosis, demonstrating the clinical potential of RNAi-based therapeutics (14). Table 1 provides an overview of significant studies on RNAi therapies, including the conditions treated, the corresponding treatments, and the outcomes achieved (14). At the molecular level, siRNA duplexes are incorporated into the RNA-induced silencing complex (RISC). The guide strand directs Argonaute-2 (Ago2) to the complementary mRNA, leading to endonucleolytic cleavage and degradation (14). This process not only suppresses translation but also recruits exonucleases that ensure irreversible silencing.

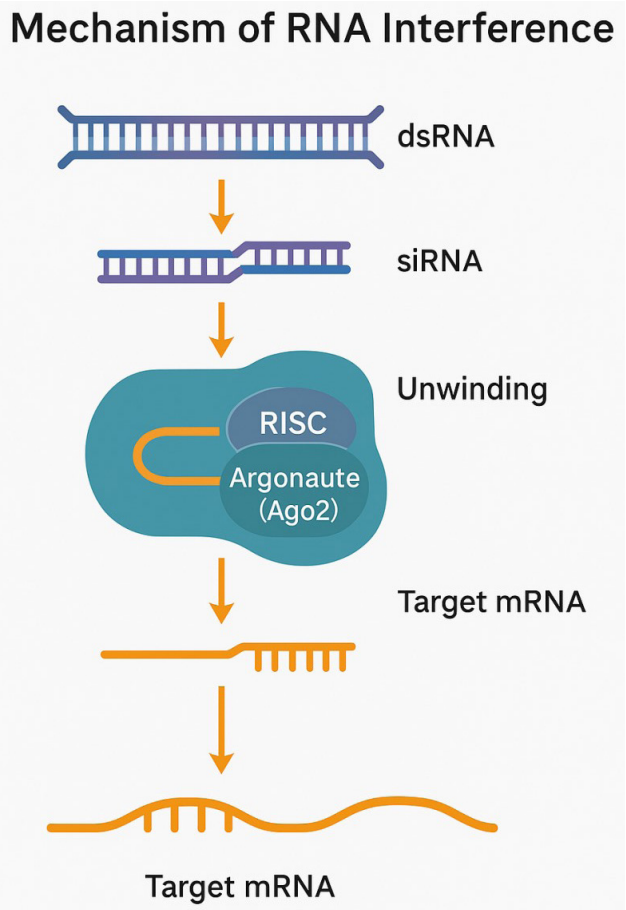

Figure 1. Mechanism of action of RNA interference (RNAi) in nucleic acid-based nanomedicine

Gene editing technologies, particularly CRISPR–Cas9, offer profound opportunities by introducing targeted double-stranded breaks in genomic DNA, which are then repaired through either non-homologous end joining (NHEJ) or homology-directed repair (HDR)(15). This enables correction of pathogenic mutations at the molecular level with unprecedented precision. Recent clinical trials have demonstrated the therapeutic potential of CRISPR-based therapies, such as ex vivo editing of hematopoietic stem cells for sickle cell disease and β-thalassemia, as well as ongoing investigations in Duchenne muscular dystrophy and TTR amyloidosis (15). While these technologies remain under clinical exploration, their capacity to correct disease-causing alleles highlights a paradigm shift toward curative genomic medicine (16). CRISPR-Cas9 editing relies on a ribonucleoprotein complex where the Cas9 endonuclease is guided by sgRNA to the protospacer adjacent motif (PAM) site. Upon DNA cleavage, endogenous repair pathways such as NHEJ or HDR determine editing outcomes. The efficiency of this system depends heavily on intracellular delivery and nuclear localization of the complex (17). These technologies are still under clinical exploration but hold the promise for curing genetic disorders like sickle cell anemia and muscular dystrophy (18). The application of these nucleic acid-based drugs, while revolutionary, also comes with several challenges, such as delivery efficiency, specificity, and potential off-target effects (18). The development of nanoparticle-based delivery systems, as seen with Onpattro, has proven critical in improving the bioavailability and

Table 1. Summary of key FDA-approved RNAi therapeutics (as of August 2025)

Drug (brand name)	Platform	Target Gene	Indication	Approval (FDA/EMA)	Sponsor	Key Clinical Findings	Research Gaps	Ref
Patisiran (Onpattro)	LNP-siRNA	TTR	Hereditary ATTR amyloidosis	FDA 2018	Alnylam	Improved mNIS+7 score, reduced TTR protein levels	Long-term safety, immunogenicity	(19)
Givosiran (Givlaari)	GalNAc-siRNA	ALAS1	Acute hepatic porphyria	FDA 2019	Alnylam	Reduced annual attack rate, improved quality of life	Rare hepatotoxicity events	(20)
Lumasiran (Oxlumo)	GalNAc-siRNA	HAO1	Primary hyperoxaluria type 1	FDA/EMA 2020	Alnylam	Lowered urinary oxalate	Pediatric, long-term follow-up	(21)
Inclisiran (Leqvio)	GalNAc-siRNA	PCSK9	Hypercholesterolemia	FDA 2021; EMA 2020	Novartis/Alnylam	LDL-C reduction (~50%) sustained with biannual dosing	Hard outcomes data pending	(22)
Vutrisiran (Amvuttra)	GalNAc-siRNA	TTR	ATTR amyloidosis (polyneuropathy, cardiomyopathy)	FDA 2022	Alnylam	Effective SC dosing, favorable safety vs patisiran	Comparative long-term efficacy	(23)
Nedosiran	GalNAc-siRNA	LDHA	Primary hyperoxaluria (type 1 and 2)	FDA 2024	Dicerna/Eli Lilly	Reduced urinary oxalate excretion	Broader PH population efficacy	(23)
Ongoing: Zilebesiran	GalNAc-siRNA	AGT	Hypertension	Phase 3	Alnylam	Sustained BP reduction	CV outcomes, large-population safety	(24)

targeted delivery of these delicate molecules to tissues where they exert their therapeutic effects (19).

Nanomedicine in drug delivery

Nanomedicine has revolutionized the field of drug delivery, particularly in the context of nucleic acid-based therapies, by addressing critical challenges related to the stability, bioavailability, and targeted delivery of therapeutic agents (25). These nanoparticles are designed to encapsulate or conjugate nucleic acids like siRNA, mRNA, or antisense oligonucleotides, protecting them from degradation by nucleases and enhancing their delivery to target cells (26, 27). The ability to precisely control the size, surface charge, and functionalization of nanoparticles allows for the optimization of their interaction with specific cellular targets, ensuring that the nucleic acid drugs reach the intended site of action with minimal systemic exposure (28). For instance, liposomal formulations, such as those used in Onpattro, utilize lipid nanoparticles to deliver RNAi molecules to liver cells, where they reduce the production of toxic TTR protein (29). Effective delivery requires nanoparticles to cross the cellular membrane, typically via clathrin-mediated endocytosis or macropinocytosis. Once internalized, a major bottleneck is endosomal escape. Ionizable lipids in LNPs become protonated in acidic endosomes, disrupting the membrane and releasing siRNA into the cytoplasm (30). Failure to escape results in lysosomal degradation, reducing therapeutic efficacy. Additionally, intracellular trafficking determines whether nucleic acids reach cytosolic RNAi machinery or the nucleus for CRISPR activity, underscoring the importance of nanoparticle composition in dictating pharmacodynamics (30, 31) Figure 2 demonstrates the structure and function of lipid nanoparticles (LNPs) used in the delivery of nucleic acid-based drugs, showcasing how they encapsulate RNA molecules to enhance stability and facilitate targeted delivery to cells (32). LNPs encapsulate nucleic acids via microfluidic mixing of lipids (ionizable lipid, DSPC, cholesterol, PEG-lipid) with siRNA in acidic buffer, forming stable nanostructures that undergo PEG shedding, systemic circulation, cellular uptake, and endosomal escape. Recent reports have provided detailed mechanistic insights into this process, including the role of ionizable lipid protonation, helper lipid stabilization, and endosomal membrane disruption (32). Research has shown that these nanoparticles enhance the uptake of therapeutic agents into target cells by exploiting cellular endocytosis mechanisms, thereby increasing their intracellular accumulation (33). In addition to enhancing stability and targeting, nanomedicines also offer the advantage of controlled and sustained release, which can improve the therapeutic index by maintaining

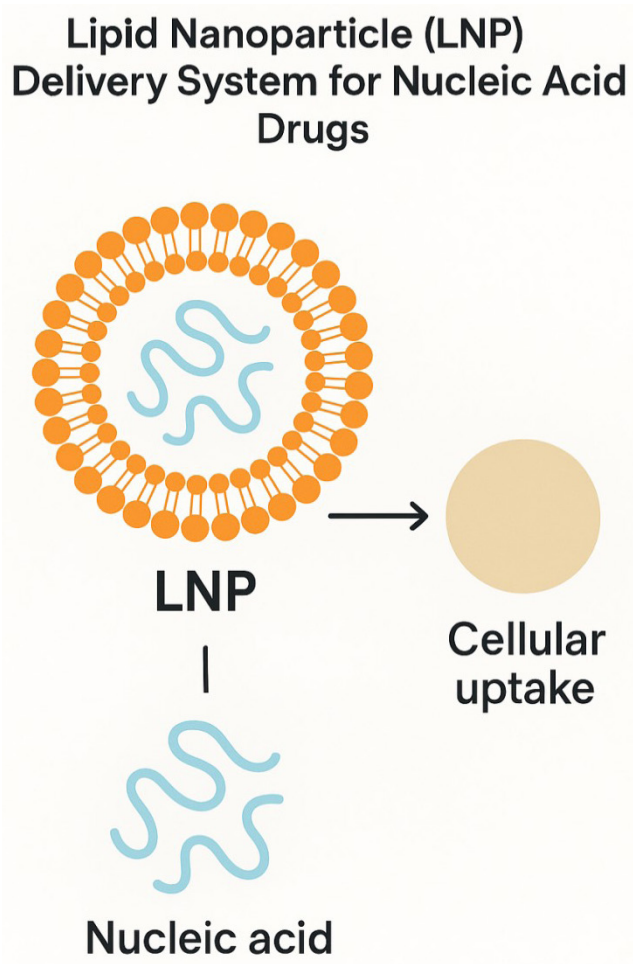

Figure 2. Depiction of lipid nanoparticle (LNP) encapsulation and delivery of nucleic acids

Table 2. Comparison of nanoparticle delivery systems for nucleic acid drugs

Nanoparticle Type	Advantages	Quantitative Parameters	Example	Research Gaps
Liposomes	Biocompatible, versatile, well-established	Size: 70-100 nm; Encapsulation efficiency: >90%; Circulation half-life: ~3 days	Onpattro (Patisiran)	Scalable manufacturing methods remain underexplored
Polymeric Nanoparticles	Controlled release, customizable surface	Size: 100-200 nm; Loading efficiency: 60 80%; Release: sustained up to 72 h	siRNA delivery systems	Batch-to-batch reproducibility and regulatory standardization needed
Dendrimers	High drug loading, targeted delivery	Size: 5-20 nm; Drug payload capacity: up to 10% w/w; Rapid clearance unless modified	Dendrimer-based siRNA carriers	Limited *in vivo* safety data
Solid Lipid Nanoparticles (SLN)	Stable, long shelf life, high payload	Size: 50-200 nm; Loading efficiency: 40-60%; Release: controlled over 24-48 hr	mRNA delivery platforms	Optimization for hydrophilic payload delivery is lacking
Gold Nanoparticles	High stability, easy surface modification	Size: 10-50 nm; High surface functionalization density (~100 200 ligands/particle)	Gene silencing therapies	Long-term biocompatibility still unclear

optimal drug concentrations over extended periods (34). However, challenges remain in overcoming biological barriers such as the blood-brain barrier and improving the specificity of drug delivery to avoid off-target effects (35). Researchers are actively exploring innovative nanocarriers, including stimuli-responsive nanoparticles that release their payload in response to specific environmental cues, such as pH changes or the presence of certain enzymes (35). The clinical success of Onpattro in treating hereditary TTR amyloidosis underscores the potential of nanomedicine in revolutionizing nucleic acid-based drug delivery, paving the way for future advancements in the treatment of genetic disorders, cancers, and viral infections (36). As research progresses, the combination of nanotechnology with nucleic acid-based therapies promises to deliver more effective, safer, and personalized treatments for a wide range of diseases. Table 2 compares different types of nanoparticles used for delivering nucleic acid drugs, highlighting their advantages, disadvantages, and examples of applications in RNAi-based therapies.

While first-generation LNPs (used in patisiran and mRNA vaccines) consist of ionizable cationic lipids, phospholipids, cholesterol, and PEG-lipids, advances in formulation chemistry have yielded next-generation LNPs with enhanced pharmacokinetics and targeting precision (37). One innovation is the development of rapidly eliminated lipid nanoparticles (reLNPs), designed to minimize prolonged circulation and reduce off-target accumulation in non-hepatic tissues. reLNPs accelerate clearance post-delivery, thereby improving safety for repeated dosing regimens (37). Another important advancement is targeted LNPs (tLNPs), which incorporate surface ligands (e.g., antibodies, peptides, aptamers, GalNAc analogs) to achieve organ- or cell-specific delivery beyond the liver. These targeted systems address one of the major limitations of conventional LNPs, namely their hepatic tropism (38). Together, reLNPs and tLNPs represent the second wave of LNP innovation, expanding the clinical potential of RNAi and mRNA therapeutics for extrahepatic indications such as oncology, neurology, and immunology.

Clinical development of onpattro (Patisiran)

The clinical development of Onpattro, the first FDA-approved RNAi therapeutic, represents a significant milestone in the translation of nanomedicine-based nucleic acid therapies from the lab to the clinic (39). Onpattro was developed to treat hATTR, a rare and debilitating genetic disorder characterized by the accumulation of amyloid deposits, which are primarily composed of misfolded TTR proteins (40). The drug utilizes lipid nanoparticle technology to deliver siRNA that silences the mutant TTR gene, preventing the production of the abnormal TTR protein and thereby halting the progression of amyloidosis (41). This targeted approach highlights the efficacy of RNAi therapeutics in treating diseases caused by specific genetic mutations. The clinical journey of Onpattro began with preclinical studies that demonstrated its ability to reduce TTR levels in animal models, paving the way for human trials (42). Table 3 summarizes key clinical trials evaluating Onpattro for the treatment of hATTR, including trial names, conditions, phases, and key outcomes. The pivotal Phase 3 APOLLO study, which evaluated the safety and efficacy of Onpattro in patients with hATTR, showed a significant reduction in serum TTR levels and improvement in neuropathy symptoms, establishing the therapeutic potential of RNAi in treating genetic diseases (43). Beyond the pivotal

Table 3. Key clinical trials of onpattro

Trial Name	Condition	Treatment	Phase	Key Outcome	Research Gaps	Ref
APOLLO Trial (2018)	Hereditary transthyretin-mediated amyloidosis (hATTR)	Onpattro	Phase 3	Significant reduction in TTR protein, improvement in neuropathy	Limited trial data in diverse populations	(50)
OLE Study (2019)	HATTR	Onpattro	Open-label Extension	Long-term safety and efficacy, sustained reduction in TTR	Need for extended follow-up beyond 5 years	(51)
Study 2015	Familial amyloid polyneuropathy (FAP)	Onpattro	Phase 1/2	Improved nerve function and quality of life	Early phase, small cohorts limit generalization	(52)
Study 2017	Familial amyloid polyneuropathy (FAP)	Onpattro	Phase 2	Safe, well-tolerated, significant biomarker reduction	Ethnic and geographic diversity of trial participants underexplored	(53)

APOLLO trial, real-world case studies and extended clinical follow-up have reinforced the clinical relevance of RNAi therapeutics. For instance, post-marketing surveillance of Onpattro has demonstrated sustained reductions in serum TTR and improvements in neuropathy scores across diverse patient groups, with over 36 months of follow-up (44). Furthermore, the success of patisiran has spurred additional RNAi approvals: givosiran (Givlaari®, 2019) for acute hepatic porphyria, lumasiran (Oxlumo®, 2020) for primary hyperoxaluria type 1, and vutrisiran (Amvuttra®, 2022) as a next-generation subcutaneous RNAi therapeutic for hATTR amyloidosis. Together, these approvals provide concrete clinical validation of RNAi platforms, complementing controlled trials with growing real-world evidence (45).

Furthermore, the success of Onpattro has demonstrated the viability of using LNPs as a delivery platform for RNAi therapeutics, opening new possibilities for treating other genetic disorders and diseases with a known genetic basis (46). Post-marketing surveillance and long-term studies continue to evaluate the safety of Onpattro, with ongoing research focusing on its effects in diverse patient populations and its potential use in combination with other treatments (47). However, despite its success, there are challenges that remain, including the high cost of treatment, the need for regular intravenous infusions, and the requirement for individualized dosing based on patient response (48). As the clinical landscape evolves, the development of next-generation RNAi therapies that require less frequent dosing, improved patient accessibility, and better cost-effectiveness remains a priority (49). The clinical approval of Onpattro serves as a case study in the potential of RNAi-based therapies and highlights the important role of nanomedicine in delivering nucleic acid drugs that can address the underlying causes of genetic diseases.

Comparison with other RNAi therapies

The approval of Onpattro in 2018 marked the first clinical validation of RNAi therapy. However, several subsequent approvals have expanded the therapeutic scope of RNAi. Clinically, all demonstrate robust biomarker knockdown (>80% TTR reduction for patisiran and vutrisiran; significant urinary oxalate reduction for lumasiran; decreased porphyria attacks for givosiran), with improved patient convenience and generally favorable safety profiles (54). While Onpattro established proof-of-concept for RNAi in humans, these analogues represent iterative innovation in delivery routes, dosing schedules, and safety optimization, illustrating the rapid maturation of RNAi-based nanomedicines. Givosiran (Givlaari®, 2019) was approved for acute hepatic porphyria, representing the first siRNA drug delivered subcutaneously, reducing the burden of intravenous infusions required with patisiran (55). Lumasiran (Oxlumo®, 2020) extended RNAi applications to primary hyperoxaluria type 1, while vutrisiran (Amvuttra®, 2022) emerged as a next-generation TTR silencer with improved dosing convenience (quarterly subcutaneous injection) compared to patisiran's biweekly intravenous regimen. Compared to Onpattro, these newer RNAi drugs highlight the field's shift toward enhanced patient convenience, expanded indications, and improved safety profiles (56). For example, vutrisiran achieves comparable TTR knockdown to patisiran but with reduced infusion-related reactions and less frequent administration. Similarly, givosiran and lumasiran demonstrate the versatility of RNAi platforms beyond amyloidosis, targeting metabolic disorders with high unmet needs (57). These differences underscore two key points: (1) Onpattro's success was pivotal but technologically limited by the requirement for intravenous delivery; (2) subsequent RNAi drugs demonstrate the importance of iterative innovation in delivery systems, dosing schedules, and therapeutic targeting (58). Together, these therapies reflect the rapid evolution of RNAi nanomedicines and provide valuable context for future design of personalized nucleic acid drugs (Table 4).

While Onpattro represents a landmark in RNAi therapeutics, its high cost poses significant challenges for widespread patient access. The estimated annual treatment cost exceeds USD 450,000, making it one of the most expensive drugs currently available (59). This economic burden raises issues of healthcare affordability, insurance coverage, and equity of access, particularly in low- and middle-income countries. Moreover, the need for biweekly intravenous infusions adds indirect costs related to clinical administration, patient travel, and time lost from work. Compared to newer RNAi drugs such as vutrisiran, which allows quarterly subcutaneous dosing, Onpattro is less convenient and imposes higher cumulative costs (60). These socioeconomic barriers highlight the urgent need for cost-effective manufacturing strategies, competitive pricing, and policy interventions to ensure that RNAi therapies, despite their transformative potential, are accessible to the patients who need them most.

Challenges in clinical translation of nucleic acid-based nanomedicines

The clinical translation of nucleic acid-based nanomedicines, including RNAi therapeutics like Onpattro, is an exciting frontier in modern medicine, yet it is fraught

Table 4. Comparison of FDA-approved RNAi therapeutics (2018-2022)

Drug (Year)	Indication	Molecular Target	Delivery System	Route	Dosing Frequency	Key Outcomes	Safety Considerations
Onpattro (2018)	hATTR amyloidosis	TTR mRNA	Lipid Nanoparticle (LNP)	IV	Every 3 weeks	~80% TTR knockdown; improved neuropathy scores	Infusion-related reactions, requires premedication
Givosiran (2019)	Acute hepatic porphyria	ALAS1 mRNA	GalNAc-siRNA conjugate	SC	Monthly	Reduces ALA/PBG accumulation; lowers porphyria attacks	Possible hepatic and renal adverse events
Lumasiran (2020)	Primary hyperoxaluria type 1	HAO1 (glycolate oxidase) mRNA	GalNAc-siRNA conjugate	SC	Monthly	Significant reduction in urinary oxalate levels	Favorable safety profile
Vutrisiran (2022)	hATTR amyloidosis	TTR mRNA	GalNAc-siRNA conjugate	SC	Quarterly	Non-inferior to patisiran; sustained TTR knockdown	Lower infusion burden; fewer treatment-related reacti

with several significant challenges (61). One of the foremost obstacles is the issue of immunogenicity and toxicity. The inherent nature of nanoparticles, such as their size, shape, and surface chemistry, can influence how the immune system recognizes and responds to them (62). For example, certain types of nanoparticles may be recognized as foreign by the body's immune system, which could result in the activation of immune pathways, thereby limiting the effectiveness and safety of the treatment (63). Additionally, the long-term toxicity of nanoparticles, particularly their potential accumulation in organs like the liver or spleen, is still not fully understood, and this remains a major concern for patient safety (64).

Another critical challenge lies in the stability and pharmacokinetics of nucleic acid-based nanomedicines. Nucleic acids, by their very nature, are susceptible to degradation by nucleases, enzymes that break down RNA and DNA molecules (65). As such, ensuring that nucleic acid drugs remain stable during delivery to the target site and avoid rapid degradation in the bloodstream is a major hurdle (66). LNPs, such as those used in Onpattro, have shown promise in protecting these fragile molecules, but the efficiency of these systems can vary depending on their design and the specific therapeutic agent (67). Furthermore, the pharmacokinetics—how the drug is absorbed, distributed, metabolized, and eliminated by the body—can be highly variable and may not always support optimal therapeutic effects (68). Table 5 presents the major challenges in translating RNAi-based nanomedicines into clinical practice and potential solutions to overcome these barriers (61-76). While LNPs have shown remarkable clinical success, particularly in the approval of Onpattro and mRNA COVID-19 vaccines, their performance is highly context-dependent. For instance, their efficiency in targeting hepatocytes is largely due to the natural tropism of ionizable lipids toward the liver, but this same property limits applications in non-hepatic diseases (69). In contrast, polymeric nanoparticles demonstrate superior control over release kinetics and surface modification but often fail clinically due to aggregation tendencies and complex manufacturing requirements (69). Similarly, dendrimers offer high drug-loading capacity and multivalency for targeting ligands, yet their toxicity profile has prevented translation (70). This comparative trend indicates that the success or failure of delivery systems hinges not only on biological performance but also on manufacturability, scalability, and safety in chronic dosing scenarios.

The challenge of targeted delivery and cellular uptake also remains a major barrier. While nanoparticles can be engineered to preferentially accumulate in certain tissues, such as the liver in the case of Onpattro, the ability to ensure that these particles are taken up by the right cells and tissues at the right time is complex (71). Efficient cellular uptake is dependent on various factors, including the nanoparticle's size, surface charge, and functionalization (72). Furthermore, the biological barriers, such as the blood-brain barrier, pose additional difficulties for the delivery of nucleic acid-based drugs to certain tissues, such as the central nervous system (73). Overcoming these barriers requires advanced engineering strategies and better understanding of the interactions between nanoparticles and biological systems.

Finally, there are manufacturing and scalability issues that hinder the widespread use of nucleic acid-based nanomedicines (74). The production of these sophisticated drug delivery systems at a scale suitable for commercial use remains challenging. High costs associated with the synthesis of nanoparticles, as well as the purification and formulation processes, can drive up the price of treatments, making them less accessible to patients (75). Moreover, the complexity of scaling up the manufacturing of RNA-based therapeutics, while maintaining quality control and reproducibility, adds to the difficulties faced by the industry (76). Despite these hurdles, the continued progress in the field of nanomedicine, along with ongoing research into overcoming these challenges, holds great promise for the future of nucleic acid-based therapeutics (77).

Emerging trends in nucleic acid-based nanomedicines

One of the most notable advancements is the use of mRNA vaccines and therapeutics, which gained significant attention during the COVID-19 pandemic (78). mRNA vaccines, such as the Pfizer-BioNTech and Moderna vaccines, demonstrated the power of mRNA in generating an immune response and preventing viral infections (79). This technology, which had been in development for years, showed that mRNA could be safely delivered via LNPs to stimulate protein production in cells. This success has sparked a broader exploration of mRNA-based therapeutics for treating a variety of diseases beyond infectious ones, including cancers, autoimmune diseases, and genetic disorders (80). Another exciting area of growth is the use

Table 5. Challenges and solutions in clinical translation of RNAi-based nanomedicines

Challenge	Impact	Potential Solutions
Immunogenicity and toxicity	Immune system activation, inflammation, potential organ damage	Modify surface charge, use PEGylation, optimize nanoparticle size and composition
Stability and degradation	Nucleic acids degrade in circulation, limiting effectiveness	Use lipid nanoparticles (LNPs) for stability, employ polymeric coatings
Targeted delivery and cellular uptake	Difficulty in directing therapeutics to specific cells or tissues	Functionalize nanoparticles with targeting ligands, utilize cell-specific receptors
Blood-brain barrier (BBB) penetration	Difficulty delivering therapeutics to brain tissues	Use nanoparticle-based transporters, design nanoparticles for BBB penetration
Manufacturing and scalability	High production costs, difficulty scaling up for mass production	Develop cost-effective, scalable manufacturing processes, enhance nanoparticle stability for storage

of CRISPR-Cas9 and other gene-editing technologies in combination with nanomedicine. CRISPR-Cas9 allows for the precise modification of genes within living organisms, offering the potential to correct genetic mutations that cause a wide range of diseases (81). However, the delivery of CRISPR components to the appropriate cells in vivo has been a major challenge due to the size and complexity of the gene-editing machinery (81). Nanomedicine holds the key to overcoming this barrier, as nanoparticles can be engineered to carry CRISPR components—such as the Cas9 enzyme and guide RNA—directly into cells (82). This could enable precise, in situ gene editing for conditions like sickle cell anemia, muscular dystrophy, and cystic fibrosis, and offers hope for curative treatments for genetic disorders (82). The most prominent trend is the clinical expansion of mRNA vaccines and therapeutics. Following the success of COVID-19 mRNA vaccines, LNPs are now being explored for oncology vaccines and protein replacement therapies (83). Several phase I/II trials are evaluating mRNA-based cancer vaccines (e.g., BioNTech's individualized neoantigen vaccines) with encouraging immunogenicity results. In the realm of cancer therapy, there is growing interest in using nanomedicine to enhance the delivery of nucleic acid-based drugs to tumors (84). Nanoparticles can be designed to selectively target cancer cells based on specific biomarkers, such as receptors overexpressed in tumor tissues (84). This precision targeting, coupled with the ability of nanoparticles to carry RNAi molecules, antisense oligonucleotides, or mRNA therapeutics, opens up new possibilities for treating cancers that are resistant to traditional therapies (85). Researchers are exploring the use of nucleic acid-based nanomedicines to silence oncogenes, restore tumor suppressor genes, or activate immune responses against tumors. Several preclinical studies have shown promise in using these strategies, and clinical trials are underway to evaluate the safety and efficacy of these novel treatments (85). Figure 3 provides an overview of cutting-edge technologies, such as mRNA-based therapeutics, CRISPR-Cas9 gene editing, and AI-driven nanomedicine, and their potential applications in improving the delivery and efficacy of nucleic acid-based therapies. Another emerging trend is the development of stimuli-responsive nanoparticles, which can release their payload in response to specific environmental conditions within the body (86). For instance, nanoparticles can be designed to release their therapeutic cargo in response to changes in pH, temperature, or the presence of certain enzymes, which is particularly useful for targeting disease sites such as tumors or inflamed tissues (87).

Additionally, the integration of artificial intelligence (AI) and machine learning (ML) with nanomedicine is becoming increasingly prevalent. AI is being utilized to design nanoparticles with optimized properties, predict the behavior of nanomaterials in biological systems, and analyze vast datasets to identify potential therapeutic targets for nucleic acid-based drugs (88). AI can also be employed to accelerate the discovery of new drug candidates and improve the efficiency of clinical trials by identifying patient populations that are most likely to benefit from specific treatments (89). As these technologies continue to mature, they are expected to play a significant role in the future development and clinical application of nucleic acid-based nanomedicines (90).

Beyond LNPs and GalNAc-siRNA conjugates, several innovative delivery platforms have entered clinical translation. ESC+ technology (Enhanced Stabilization Chemistry Plus), pioneered by Alnylam, incorporates advanced chemical modifications to improve siRNA stability and reduce dosing frequency (91). Its most prominent candidate is Zilebesiran, currently in Phase III trials for hypertension. Similarly, mRNAi GOLD™ technology by Silence Therapeutics enables precise siRNA design and conjugation for cardiovascular disease targets, exemplified by Zerlasiran (SLN360). Another breakthrough is lipid chain conjugation strategies, such as the addition of C16 lipid moieties, which improve biodistribution and enable siRNA penetration across the blood–brain barrier—a strategy under development for Mivelsiran, targeting Alzheimer's disease (92). Collectively, these technologies represent the next wave of nucleic acid delivery innovations, broadening therapeutic applications to extrahepatic organs and complex disorders previously inaccessible to standard platforms. Figure 4 illustrates representative next-generation delivery platforms beyond conventional LNP and GalNAc systems, while Table 6 summarizes broader emerging technologies in nanomedicine for nucleic acid-based drug delivery (78-92).

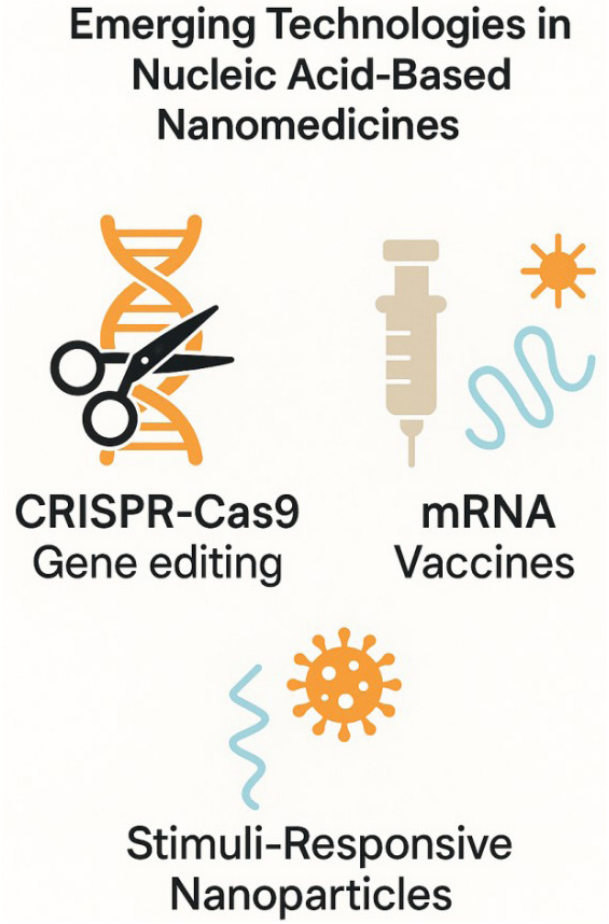

Figure 3. Emerging technologies in nucleic acid-based nanomedicines

Future perspectives and potential applications

The future of nucleic acid-based nanomedicines holds immense promise, particularly as advancements in nanotechnology, gene therapy, and drug delivery systems continue to evolve. One of the most exciting prospects is the integration of personalized medicine with nanomedicine

Table 6. Emerging technologies in nanomedicine for nucleic acid-based drug delivery

Technology	Description	Potential Application	Example
mRNA-Based Therapeutics	Delivery of mRNA molecules to stimulate protein production within cells	Vaccines, cancer therapy, genetic diseases	Pfizer-BioNTech, Moderna vaccines
CRISPR-Cas9 Gene Editing	Targeted gene editing to correct genetic mutations	Genetic disorders (e.g., sickle cell, cystic fibrosis)	CRISPR-based therapies
Stimuli-Responsive Nanoparticles	Nanoparticles that release therapeutic agents in response to environmental stimuli (e.g., pH or enzymes)	Targeted drug delivery, cancer therapy	Stimuli-responsive polymer systems
Nanogels	Hydrophilic nanoparticles that respond to changes in environmental conditions (e.g., pH or temperature)	Controlled drug release, tissue-specific delivery	Nanogels for RNAi delivery
Artificial Intelligence in Nanomedicine	AI-driven design of nanoparticles for optimized drug delivery and monitoring	Personalized medicine, optimized nanoparticle design	AI in nanoparticle development

(93). Personalized medicine aims to tailor treatments to individual patients based on their genetic makeup, lifestyle, and specific disease characteristics. Nanomedicines, particularly those that deliver nucleic acid-based drugs, offer a unique opportunity to develop treatments that are highly specific to each patient's genetic profile (94). With the growing availability of genetic sequencing and advances in AI-driven data analysis, it is becoming increasingly feasible to design custom therapies that can target specific mutations or molecular pathways in diseases such as cancer, genetic disorders, and cardiovascular diseases (94). This personalized approach promises to improve therapeutic outcomes, reduce side effects, and optimize treatment efficacy by selecting the most appropriate nanomedicine for each individual.

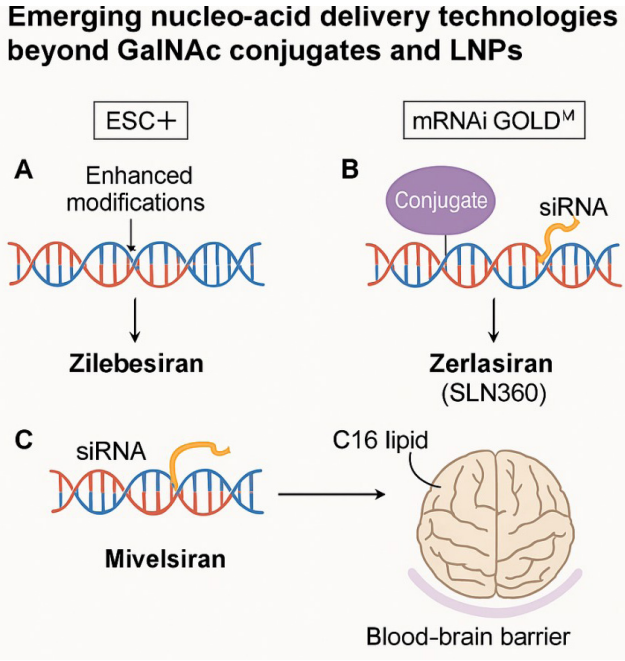

Figure 4. Emerging nucleic acid delivery platforms beyond conventional GalNAc and LNP systems
(A) ESC+ (Enhanced Stabilization Chemistry Plus) platform developed by Alnylam, featuring chemically stabilized siRNA to enhance durability and reduce dosing, exemplified by Zilebesiran (Phase III for hypertension). (B) mRNAi GOLD™ technology from Silence Therapeutics, enabling precise siRNA design and conjugation for targeted delivery, illustrated by Zerlasiran (SLN360) for cardiovascular disease. (C) C16 lipid conjugation strategy, in which lipid-modified siRNA crosses the blood–brain barrier to enable extrahepatic targeting, exemplified by Mivelsiran for Alzheimer's disease

Overcoming current barriers in nucleic acid delivery is a key area of focus for future research (95). Challenges remain in terms of the effective and precise delivery of nucleic acids to the right tissues, particularly for diseases affecting tissues like the brain or lungs are critical issues (95). Overcoming biological barriers, such as the blood-brain barrier or extracellular matrix in tumors, remains a significant challenge (96). To address these barriers, novel nanoparticle designs are being explored, including the development of biodegradable nanoparticles, targeted ligands, and stimuli-responsive systems that can release their therapeutic payload only when they reach the specific site of disease (96). Such advancements could enable the treatment of conditions like Alzheimer's disease, neurological disorders, and pulmonary diseases, where effective drug delivery has been historically challenging.

In addition to overcoming delivery challenges, enhanced drug release profiles will play a crucial role in improving the therapeutic index of nucleic acid-based drugs (97). Researchers are increasingly focused on developing controlled release systems, which allow for the sustained and gradual release of drugs over time, improving patient compliance and ensuring that therapeutic levels are maintained in the body for longer durations (98). This is particularly important for treatments that require continuous delivery, such as those targeting chronic diseases or genetic disorders that require long-term management (98). By using advanced formulations, such as nanogels, liposomes, or dendrimers, it is possible to achieve targeted, sustained, and site-specific release of drugs, minimizing side effects and enhancing therapeutic efficacy.

Another promising application is in combination therapies, where nucleic acid-based nanomedicines are combined with other treatment modalities, such as chemotherapy, immunotherapy, or radiotherapy (99). The combination of RNA-based therapeutics with traditional therapies could lead to synergistic effects that improve treatment outcomes in complex diseases like cancer (100). For instance, RNAi could be used to silence cancer-promoting genes, while chemotherapy targets rapidly dividing cancer cells (68). In combination with immunotherapy, which enhances the body's immune system to recognize and destroy cancer cells, this approach could significantly increase the therapeutic efficacy of these treatments (101). Similarly, gene editing technologies like CRISPR could be used to enhance the effects of chemotherapy or immunotherapy by repairing mutated

genes or enhancing immune cell function, creating a more powerful and multifaceted approach to cancer treatment (101).

The advancement of gene editing technologies in clinical applications also holds tremendous potential. Although technologies like CRISPR-Cas9 are still largely in the experimental phase, their combination with nanomedicine could lead to breakthroughs in curing genetic disorders (102). By directly editing the genome of diseased cells, CRISPR-based therapies could correct mutations at the DNA level, offering a potential cure for previously untreatable genetic diseases. The use of nanoparticles to deliver CRISPR components (Cas9 enzyme and guide RNA) directly to specific cells is a critical area of development, and ongoing research aims to improve the precision, efficiency, and safety of these gene-editing strategies (103). Once perfected, CRISPR-based gene therapies could potentially revolutionize the treatment of diseases such as sickle cell anemia, cystic fibrosis, and Duchenne muscular dystrophy (104).

Furthermore, the integration of AI and ML into the design, development, and clinical implementation of nucleic acid-based nanomedicines will accelerate the pace of innovation in this field (104). AI can be used to predict the interactions between nanoparticles and biological systems, optimize the design of nanoparticles for specific drug delivery applications, and enhance the analysis of clinical trial data (104). By analyzing large datasets, AI can identify novel therapeutic targets, suggest the most promising drug combinations, and predict patient responses to treatment. These technologies will help researchers and clinicians make data-driven decisions, personalize treatments, and improve patient outcomes in real-time (105).

Conclusion

The clinical success of Onpattro represents a groundbreaking achievement in the field of nanomedicine, validating the potential of RNAi therapies for treating genetic disorders such as hATTR. By utilizing LNPs to deliver siRNA to target cells, Onpattro has demonstrated the ability to reduce the toxic protein buildup that causes the disease, significantly improving patient outcomes. Despite this success, the clinical translation of RNAi-based nanomedicines is not without challenges. Issues such as stability, targeted delivery, immunogenicity, toxicity, and scalability of manufacturing processes remain significant barriers that must be addressed to expand the use of these therapies to other diseases. Advances in nanotechnology, including the development of more efficient delivery systems and the integration of gene editing technologies like CRISPR-Cas9, offer promising solutions to these challenges, potentially enhancing the precision and efficacy of RNA-based therapeutics. Furthermore, the growing field of personalized medicine, driven by advances in genomic sequencing and AI, is expected to play a key role in optimizing treatment outcomes for patients receiving nucleic acid-based therapies. As research progresses, it is likely that the field of RNA-based nanomedicines will continue to evolve, offering innovative treatments for a wide range of genetic and chronic diseases. The continued development of RNAi-based therapies, including the successful clinical translation of Onpattro, offers significant hope for the future of precision medicine and the treatment of previously untreatable conditions.

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Authors' Contributions

D S conceived the study and performed the literature survey. D S, A T, and N T performed data validation and analysis for tables and figures. SS proof read the article. All authors approved the final draft for publication.

Conflicts of Interest

Not applicable.

Declaration

We have not used any AI tools or technologies to prepare this manuscript.

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