# The expanding role of m-RNA – Based therapeutics in modern medicine

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**Abstract**

*Messenger RNA (mRNA) therapeutics have rapidly evolved into a powerful platform following the success of COVID-19 vaccines, demonstrating high efficacy and rapid development potential (Pardi et al., 2018). Unlike traditional therapies, mRNA enables transient protein expression through cellular translation machinery, offering flexibility across vaccines, cancer immunotherapy, and genetic disease treatment. A defining feature of mRNA technology is its amplification effect, where a single molecule can produce $10^3$–$10^6$ protein copies depending on cellular and molecular conditions (Vogel et al., 2010). While this property enhances therapeutic efficiency, it complicates dose control and long-term protein regulation. Current lipid nanoparticle (LNP) delivery systems exhibit predictable kinetics, including rapid onset, peak expression within 24–48 hours, and decline over 7–14 days (Hou et al., 2021). Although highly effective in vaccines and immunotherapy, these kinetics present limitations for chronic disease treatment requiring sustained protein levels. This review explores the principles, applications, challenges, and future prospects of mRNA therapeutics, emphasizing their role in protein replacement, regenerative medicine, and gene editing (Sahin et al., 2020).*

**Keywords:** mRNA therapeutics, protein replacement therapy, lipid nanoparticle (LNP) delivery, mRNA pharmacokinetics, gene expression kinetics.

## 1. INTRODUCTION

The emergence of mRNA-based vaccines during the COVID-19 pandemic marked a paradigm shift in biotechnology and molecular medicine (Sahin et al., 2020). Vaccines such as BNT162b2 and mRNA-1273 demonstrated efficacy rates above 90%, validating decades of foundational research (Polack et al., 2020; Baden et al., 2021). These vaccines highlighted the advantages of mRNA platforms, including rapid development, scalability, and strong immune activation. Despite this success, transitioning mRNA technology from preventive vaccination to therapeutic applications introduces new complexities. Unlike vaccines, where transient protein expression is sufficient, therapeutic interventions require precise dosage control and sustained protein production (Kowalski et al., 2019). mRNA molecules are inherently unstable and prone to enzymatic degradation, necessitating chemical modifications such as pseudouridine incorporation to improve stability and reduce immunogenicity (Karikó et al., 2008).

Another critical challenge lies in delivery. mRNA is a negatively charged macromolecule that cannot easily cross

cellular membranes, making delivery systems such as lipid nanoparticles essential (Hou et al., 2021). Even with advanced delivery systems, only a small fraction of administered mRNA reaches the cytoplasm for translation, limiting efficiency (Verbeke et al., 2019). These limitations highlight the need for improved design strategies to fully realize the therapeutic potential of mRNA. The rapid development and global deployment of messenger RNA (mRNA)-based vaccines during the COVID-19 pandemic marked a transformative milestone in biotechnology and modern medicine (Sahin et al., 2020; Pardi et al., 2018). Vaccines such as BNT162b2 and mRNA-1273 demonstrated remarkably high efficacy rates of approximately 95% and 94.1%, respectively, validating decades of foundational research in RNA biology, delivery systems, and immunology (Polack et al., 2020; Baden et al., 2021). This success not only established mRNA as a reliable platform for infectious disease prevention but also accelerated interest in its broader therapeutic applications, including cancer immunotherapy, protein replacement, and gene editing (Dolgin et al., 2021).

Unlike traditional vaccine technologies that rely on weakened pathogens or protein subunits, mRNA vaccines function by delivering synthetic genetic instructions into host cells, enabling the production of antigenic proteins that stimulate immune responses (Pardi et al., 2018). This mechanism offers several advantages, including rapid design, scalable manufacturing, and the ability to encode multiple targets simultaneously (Sahin et al., 2014). However, while transient protein expression is sufficient for vaccination purposes, therapeutic applications demand a much higher level of precision in dosage, timing, and protein regulation (Kowalski et al., 2019). This shift from preventive to therapeutic use introduces new scientific and clinical challenges that must be carefully addressed.

One of the defining characteristics of mRNA technology is its intrinsic amplification effect. A single mRNA molecule can be translated multiple times by ribosomes, producing thousands to millions of protein copies depending on cellular context and molecular optimization (Vogel et al., 2010; Liu et al., 2022). While this amplification enhances efficiency and reduces the amount of mRNA required, it complicates dose-response relationships, making it difficult to achieve predictable and controlled protein levels in therapeutic settings (Walsh et al., 2020). In applications such as enzyme replacement or hormone therapy, where precise protein concentrations are critical, this variability presents a significant limitation. Another major challenge is the inherent instability of mRNA molecules. Due to their single-

stranded structure and susceptibility to ribonuclease (RNase) degradation, mRNA molecules are rapidly broken down in biological environments (Karikó et al., 2008). To overcome this limitation, various chemical modifications, such as the incorporation of pseudo uridine and optimized 5′ capping structures, have been developed to enhance stability, reduce immunogenicity, and improve translation efficiency (Sahin et al., 2014). Although these modifications have significantly improved mRNA performance, they also increase production complexity and cost. Efficient delivery of mRNA into target cells remains another critical barrier. As a large, negatively charged molecule, mRNA cannot easily cross the lipid bilayer of cell membranes (Verbeke et al., 2019). Lipid nanoparticle (LNP) systems have emerged as the most effective delivery vehicles, protecting mRNA from degradation and facilitating cellular uptake (Hou et al., 2021). Despite these advances, delivery efficiency is still limited, with only a small fraction of administered mRNA successfully reaching the cytoplasm for translation (Kowalski et al., 2019). Furthermore, current LNP systems lack precise spatial and temporal control, leading to non-specific distribution and transient expression patterns. In addition to delivery and stability challenges, mRNA therapeutics can trigger unintended immune responses. The innate immune system can recognize exogenous RNA molecules, leading to inflammation and reduced protein translation (Verbeke et al., 2019). Although chemical modifications and optimized formulations have mitigated these effects, immunogenicity remains an important consideration in therapeutic design. Despite these limitations, the potential applications of mRNA therapeutics are vast and rapidly expanding. From personalized cancer vaccines to treatments for rare genetic disorders, mRNA offers a flexible and powerful platform capable of addressing previously untreatable conditions (Dolgin et al., 2021). The continued advancement of delivery technologies, molecular engineering, and clinical strategies will be essential to overcome current barriers and fully realize the promise of mRNA-based therapies in modern healthcare (Sahin et al., 2014).

## 2. The role of mRNA therapeutics in replacement of protein and monogenic diseases

mRNA therapeutics have shown promising applications in treating monogenic diseases by enabling the production of functional proteins directly within target cells. Unlike conventional protein replacement therapies, which face challenges related to delivery efficiency and high production costs,

mRNA offers a more flexible and scalable approach (Sahin et al., 2014).

In metabolic disorders such as methylmalonic acidemia (MMA), mRNA encoding methylmalonyl-CoA mutase has demonstrated significant therapeutic potential by reducing toxic metabolite accumulation in preclinical models (Chakraborty et al., 2021). Similarly, in Fabry disease, mRNA-based delivery of α-galactosidase A has shown effective restoration of enzyme activity in multiple organs (Zangi et al., 2013). Respiratory diseases like cystic fibrosis also benefit from mRNA approaches, where functional CFTR protein expression restores chloride transport in epithelial cells (Robinson et al., 2018). In genetic bleeding disorders such as haemophilia, mRNA therapies encoding clotting factors have demonstrated rapid and sustained protein expression sufficient to restore haemostatic function (Rosenblum et al., 2020). Beyond protein replacement, mRNA plays a critical role in gene editing technologies. Delivery of CRISPR-Cas9 components via mRNA allows transient expression of nucleases, reducing off-target effects and improving safety profiles compared to DNA-based methods (Lino et al., 2018). This has opened new possibilities for precision medicine and targeted genetic correction.

### 2.1 Methylmalonic acidemia

Methylmalonic acidemia (MMA), a rare metabolic disorder caused by a deficiency in the methylmalonyl-CoA mutase enzyme or its associated cofactors, represents a significant target for mRNA-based therapeutic intervention (Chandler et al., 2019). This condition leads to severe metabolic instability and the accumulation of methylmalonic acid in the body, resulting in multi-organ complications. An mRNA-based therapy for MMA, designed to encode the functional methylmalonyl-CoA mutase (MMU) enzyme, has been actively developed by Moderna as a novel treatment strategy (An et al., 2017). Preclinical studies have demonstrated that intravenous administration of this mRNA therapy, delivered twice weekly in MMU-deficient animal models, significantly reduced plasma methylmalonic acid levels by approximately 60% to 90% (An et al., 2017). These findings highlight the therapeutic potential of mRNA technology in restoring enzyme function and managing metabolic disorders such as MMA in human patients (Manoli et al., 2016).

### 2.2 Fabry disease

Fabry disease is a metabolic illness caused by an accumulation of glycosphingolipids as a result of an enzyme α-galactosidase A deficiency (Jiang et al., 2020). The buildup may result in serious organ malfunction that affects the kidneys, heart, and brain system.

For Fabry disease, Moderna is developing an mRNA-based treatment that encodes α-galactosidase A. Intravenous administration of α-galactosidase A mRNA found in LNPs effectively restored the activity of the enzyme in important organs like the liver, spleen, heart, and kidneys in preclinical tests, indicating that the treatment may be able to stop or reverse the development of Fabry disease (Moderna et al., 2021).

### 2.3 Cystic fibrosis

Mutations in the cystic fibrosis transmembrane conductance regulator (CFTR) gene are responsible for cystic fibrosis (CF), a genetic disorder characterized by impaired chloride transport and the accumulation of thick mucus in the lungs and digestive system (Elborn et al., 2016). To address this defect, mRNA-based therapies aim to deliver functional CFTR mRNA into epithelial cells of the respiratory and gastrointestinal tracts, thereby restoring normal ion transport and cellular function (Robinson et al., 2018). Preclinical studies on Translate Bio's mRNA-based therapy, MRT5005, have demonstrated encouraging results. In CFTR knockout mouse models, administration of CFTR mRNA restored chloride channel activity to near-normal levels, with effects sustained for up to 14 days (Robinson et al., 2018). Furthermore, early-phase clinical trials evaluating nebulized delivery of MRT5005 reported favourable safety and tolerability profiles, with no serious adverse events observed in patients (Rowe et al., 2019). These findings highlight the potential of mRNA therapeutics as a promising strategy for treating cystic fibrosis.

### 2.4 Hemophilia

Factor VIII (Hemophilia A) or factor IX (Hemophilia B) mutations cause hemophilia, a genetic bleeding condition that impairs blood coagulation (Rowe et al., 2019). In order to restore normal clotting function, mRNA treatments are being developed to encode these clotting proteins. Factor VIII mRNA given to hemophiliacs During preclinical research, a mouse model with circulating factor VIII levels above therapeutic levels for longer than 72 hours was able to cure the hemostatic deficiency (Ratjen et al., 2015). Factor IX mRNA given as LNPs caused a rapid spike in factor IX expression in hemophilia B models by 4–6 hours after injection, with longer-term effects lasting up to 6 days (Boyle et al., 2020).

### 2.5 Muscular dystrophy

Mutations in the dystrophin gene are responsible for Duchenne muscular dystrophy (DMD), a severe genetic disorder characterized by progressive

muscle degeneration and weakness due to the absence of functional dystrophin protein (Hoffman et al., 1987). To address this deficiency, mRNA-based therapeutic strategies aim to deliver functional dystrophin-encoding mRNA into muscle cells, enabling the transient production of dystrophin protein necessary for maintaining muscle integrity (Kaczmarek et al., 2017). This approach offers a promising alternative to conventional gene therapy, as it does not involve permanent modification of the host genome, thereby reducing the risk of insertional mutagenesis and allowing repeated dosing for sustained therapeutic benefit (Pardi et al., 2018). Consequently, mRNA therapy is considered a safer and more flexible strategy for restoring dystrophin function in patients with Duchenne muscular dystrophy (Mercuri et al., 2019).

## 3. Regenerative medicine

The adaptability of mRNA therapies extends to regenerative medicine, where they can help promote tissue regeneration and repair, offering novel treatments for illnesses and injuries with limited therapeutic options (Dowdy et al., 2022). mRNA can improve tissue repair and healing processes by encoding growth factors, cytokines, or tissue-specific proteins (Sahin et al., 2014).

### 3.1 cardiovascular disease

Ischemic tissues often experience severe hypoxia following medical emergencies such as myocardial infarction, leading to significant cell death and impaired cardiac function (Ibanez et al., 2015). In such conditions, mRNA-based therapies can be utilized to encode proteins like vascular endothelial growth factor (VEGF), which plays a crucial role in promoting angiogenesis, the formation of new blood vessels (Zangi et al., 2013; Pardi et al., 2018). Preclinical studies have demonstrated that delivery of VEGF-encoding mRNA into ischemic tissues enhances neovascularization and improves cardiac function in animal models (Zangi et al., 2013; Carlsson et al., 2018). These findings highlight the therapeutic potential of mRNA-mediated VEGF expression as a promising approach for treating ischemic heart disease and other cardiovascular disorders associated with reduced blood supply.

### 3.2 Cartilage and bone regeneration

In orthopedic therapy, mRNA has shown great promise in promoting bone and tissue repair. mRNA encoding for cartilage-specific proteins, like collagen II or aggrecan, can be used to restore damaged cartilage in conditions like osteoarthritis. Similarly, in fracture or bone degenerative

illnesses, mRNA encoding for bone morphogenetic proteins can promote bone development and repair (Hausenloy et al., 2017). Preclinical studies have demonstrated that mRNA-based regenerative medicine can support tissue regeneration and repair, despite the fact that research in this area is still in its infancy. Heart disease, musculoskeletal issues, and chronic wounds are just a few of the degenerative diseases and injuries for which there are currently few therapeutic alternatives. This novel technique gives hope for treating these conditions (Pardi et al., 2018).

### 3.3 Pompe disease

Acid alpha-glucosidase (GAA) deficiency is the primary cause of Pompe disease, a lysosomal storage disorder characterized by the accumulation of glycogen in muscle tissues, leading to progressive muscle weakness and respiratory complications (van der Ploeg et al., 2008; Hirschhorn et al., 2016). Currently, enzyme replacement therapy (ERT) is the standard treatment; however, it has limitations, including variable efficacy and challenges in delivering the enzyme efficiently to target tissues (Kishnani et al., 2017). In contrast, mRNA-based therapy introduces genetic instructions for the production of GAA directly within muscle cells, enabling endogenous synthesis of the functional enzyme (Pardi et al., 2018; Sabnis et al., 2018). This approach has the potential to enhance therapeutic outcomes by improving muscle function and reducing disease severity through more efficient and sustained protein expression (Sabnis et al., 2018; Kowalski et al., 2019).

## 4. Gene editing therapy

mRNA has emerged as a crucial tool for delivering CRISPR-Cas9 and other gene-editing components into cells, enabling efficient and controlled genome modification (Lino et al., 2018). By encoding the Cas9 nuclease within an mRNA molecule, transient expression of the enzyme can be achieved, reducing long-term exposure and minimizing off-target effects (Kim et al., 2014). Precise genome editing occurs when the Cas9 enzyme, guided by a specific single-guide RNA (sgRNA), introduces targeted double-strand breaks in the DNA sequence, allowing for the correction of mutations or the insertion of new genetic material (Jinek et al., 2012). This approach highlights the versatility and safety advantages of mRNA-mediated delivery systems in gene editing applications.

## 5. CRISPR-Cas9 mRNA

Compared to zinc finger nucleases (ZFNs)192 and transcription activator-like effector nucleases (TALENs), CRISPR-

Cas9 is the most often used programmable nuclease because of its ease of use and versatility.38, 39 Its potential in therapeutic gene editing and precision medicine is highlighted by its notable success in mRNA-mediated delivery, which allows for precise targeted insertions and deletions (Yin et al., 2017).

With combined knockouts of human leukocyte antigen class I, T cell receptor (TCR), and programmed cell-death protein 1 (PD-1) increasing anticancer activity in allogeneic CAR T cells in both in vitro and in vivo experiments, CRISPR-Cas9 mRNA seems especially promising for T cell engineering Furthermore, new lentiviral vectors containing hybrid ΔU3-sgRNAs have simplified TRAC locus editing, resulting in TCR-negative CAR19 T cells that exhibit potent antileukemic effects in preclinical animals (Doudna et al., 2014).

6. **Significances**: The significance of mRNA therapeutics lies in their versatility and ability to address a wide range of medical conditions. One of the most important features is the intrinsic amplification mechanism, which allows a single mRNA molecule to generate a large number of protein copies, significantly enhancing therapeutic efficiency (Vogel et al., 2010). In cancer immunotherapy, mRNA-based vaccines have demonstrated the ability to induce strong immune responses against tumor-specific antigens. For instance, the mRNA-4157 vaccine, when combined with immune checkpoint inhibitors, has shown a substantial reduction in cancer recurrence risk (Luke et al., 2023). This highlights the potential of mRNA in personalized medicine. Additionally, mRNA therapeutics offer advantages in regenerative medicine. By encoding growth factors such as vascular endothelial growth factor (VEGF), mRNA can promote tissue repair and angiogenesis in ischemic conditions (Zangi et al., 2013). Similarly, applications in bone and cartilage regeneration demonstrate the ability of mRNA to stimulate tissue-specific protein synthesis. Another key advantage is safety. Unlike DNA-based therapies, mRNA does not integrate into the host genome, reducing the risk of insertional mutagenesis (Pardi et al., 2018). Furthermore, mRNA-based treatments can be rapidly designed and manufactured, making them highly adaptable for emerging diseases. The choice of clinical applications must be guided by the unique benefits and drawbacks of mRNA therapies, a revolutionary technological platform. These applications' steady-state needs are inherently at odds with the characteristic expression kinetics (fast initiation, peak at 24–48 hours, drop over 7–14 days).

**6. Future prospectives** current mRNA technologies face inherent limitations in applications requiring precise and sustained protein levels, such as hormone replacement, enzyme replacement therapy, and chronic protein deficiency disorders (Kowalski et al., 2019; Sahin et al., 2014). These limitations arise because the steady-state requirements of such therapies conflict with the characteristic expression kinetics of mRNA, which involve rapid initiation, peak expression within 24–48 hours, and a decline over 7–14 days (Hou et al., 2021; Ey geris et al., 2022).Recent industry analyses indicate that over 190 organizations are currently working on approximately 310 mRNA-based vaccines and therapeutic candidates across various disease areas (Dolgin et al., 2021; Nature Reviews Drug Discovery, 2023). Of these, around 65% are focused on vaccines, while 35% are dedicated to therapeutic applications, highlighting significant opportunities for drug discovery and innovation (Dolgin et al., 2021). Given its flexibility and scalability, mRNA technology is expected to play a dominant role in preventive medicine over the next decade (Pardi et al., 2018; Sahin et al., 2020). According to industry experts such as Patterson (Thermo Fisher Scientific), RNA-based therapeutics—including mRNA, antisense oligonucleotides (ASO), RNA aptamers, and RNA interference (RNAi)—are rapidly advancing as next-generation treatment modalities (Patterson et al., 2022; Roberts et al., 2020). Currently, nearly 90 mRNA-based therapeutics are undergoing clinical trials, alongside several other RNA-based drugs in development (Roberts et al., 2020; Nature Reviews Drug Discovery, 2023). The accelerated clinical development timelines observed during the COVID-19 pandemic have further demonstrated the potential of RNA-based medicines to reach the market faster than traditional therapeutic approaches (Sahin et al., 2020; Dolgin et al., 2021).

**7 Conclusion:** mRNA therapeutics represent a groundbreaking advancement in modern medicine, offering a flexible and powerful platform for treating a wide range of diseases. The success of COVID-19 vaccines has accelerated research and demonstrated the feasibility of large-scale mRNA applications. However, the transition from vaccines to therapeutic protein replacement introduces challenges related to dosage control, delivery efficiency, and expression stability. While mRNA therapies are highly effective in applications requiring transient protein expression, such as vaccines and cancer immunotherapy, their current limitations restrict their use in conditions requiring long-term protein regulation. Advances in delivery systems, molecular modifications,

and controlled expression technologies are essential to overcome these barriers. Overall, mRNA therapeutics hold immense potential to transform healthcare, particularly in precision medicine, gene editing, and regenerative therapies. Continued research and innovation will be crucial in unlocking their full capabilities and expanding their clinical applications in the coming years.

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