mRNA THERAPEUTICS BEYOND VACCINES: FUTURE OF PERSONALISED VACCINES:

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ABSTRACT:

mRNA therapies have quickly moved from a theoretical concept to one of the most useful tools in modern medicine .Because mRNA can be designed to encode almost any protein, it allows researchers to create treatments that are highly targeted and easier to update than traditional therapies. Recent improvements—such as modifying nucleotides, optimizing mRNA structure, and developing better delivery systems—have made these treatments more stable, efficient, and safer.
Today, mRNA is being explored far beyond vaccines. It is helping develop personalised cancer immunotherapies, offering new options for rare genetic diseases through protein replacement, and providing a safer way to deliver gene-editing tools like CRISPR. mRNA is also showing promise in tissue repair and regenerative medicine.

Although challenges remain, especially in delivery, long-term safety, and manufacturing, mRNA technology is rapidly shaping the future of precision medicine and bringing more individualized therapies closer to reality.

Keywords: mRNA therapeutics, precision medicine, personalized cancer immunotherapy, protein replacement therapy, CRISPR delivery, lipid nanoparticles, regenerative medicine.

1. INTRODUCTION:

1.1 Overview of mRNA technology

In modern medicine, mRNA therapeutics have become an important resource. After the successful launch of COVID-19 vaccines, mRNA technology is now being used in areas beyond just preventing infectious diseases. This allows for quick and flexible treatments that use the body's own cells to produce therapeutic proteins or tumor antigens.As mRNA degrades naturally and doesn't integrate into the genome, it allows controlled protein expression. Personalised mRNA cancer vaccines encoding patient-specific neoantigens have shown promising immune responses and tumour regression, especially when combined with immune checkpoint inhibitors (1).
mRNA therapeutics are expanding beyond oncology for gene editing, protein replacement, and in vivo antibody expression, offering potential treatments for autoimmune, genetic, and metabolic diseases (2). Lipid nanoparticles (LNPs) remain key for protection and delivery, with advances in lipid chemistry enabling mRNA targeting to organs beyond the liver and muscle (3).

mRNA therapeutics are paving the way for more personalized treatments designed around each person's unique genetic makeup. While challenges like delivery, large-scale production, safety, and fair access still exist, ongoing progress could soon make mRNA a key part of next-generation precision medicine (4).

1.2 Evolution of mRNA therapeutics

The journey of mRNA therapeutics began decades ago when scientists learned that lab-made mRNA could make proteins inside cells. At first, it was too unstable and caused strong immune reactions, so it couldn't be used as medicine. Later, researchers improved it by adding special chemical changes—like 5′ caps, poly(A) tails, and modified bases such as pseudouridine—to make it more stable and less reactive (5,6).

A big breakthrough came with lipid nanoparticles (LNPs)—tiny fat bubbles that safely carry mRNA into cells. This helped mRNA work better and last longer in the body (7,8).

The success of COVID-19 mRNA vaccines proved that this technology was safe, fast, and effective. Since then, scientists have been exploring mRNA for other uses—like treating cancer, genetic diseases, and developing personalized vaccines (9).

Today, new types of mRNA, such as self-amplifying RNA and circular RNA, are being designed to make treatments more powerful and longer-lasting. These advances show how mRNA has evolved from an unstable molecule to a promising tool for personalized medicine (5).

1.3 From traditional to personalised medicine: setting the stage

For a long time, modern medicine has mostly worked on a "one-size-fits-all" model — everyone with the same illness often gets the same treatment. While this approach has saved millions of lives, it doesn't always work perfectly for everyone. People differ in their genes, lifestyle, diet, and environment, and all these factors affect how their body responds to a drug (10).

Thanks to progress in genomics and molecular biology, medicine is now shifting toward something much more personal — precision or personalized medicine. This approach looks at each person's unique biological makeup to design treatments that fit them better (11).

This is where mRNA technology truly shines. Unlike traditional drugs, mRNA treatments can be designed quickly and customized to match a person's specific genetic profile. For example, in cancer therapy, scientists can identify the unique mutations in a tumor and then create an mRNA vaccine that trains the immune system to recognize and attack those cancer cells (12).

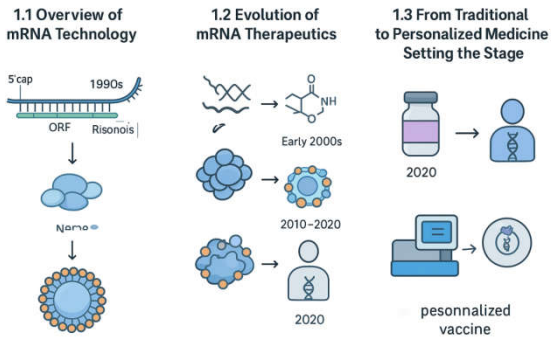

Figure 1. Introduction to mRNA therapeutics

2. MECHANISM AND DESIGN PRINCIPLES OF MRNA THERAPEUTICS:

2.1 Structure and function of mRNA

Messenger RNA, or mRNA, is like the body's own delivery service for genetic information. It carries the instructions written in DNA to the part of the cell that builds proteins — the ribosomes. These proteins are responsible for almost everything our cells do, from repairing tissues to fighting infections.

In mRNA therapeutics, scientists use a synthetic (lab-made) version of this molecule to tell cells what protein to make. For example, in mRNA vaccines, the message tells cells to make a harmless piece of a virus so the immune system can learn to recognize and fight the real one later (13,14).

The Structure of mRNA

An mRNA molecule might look simple, but it's made up of several important parts that work together to protect it and make it function smoothly:

1. 5′Cap:This is like a protective helmet for mRNA. It keeps the molecule safe from enzymes that might destroy it and helps it connect to the ribosome — the protein factory of the cell (15).
2. 5′UntranslatedRegion(UTR):This section doesn't code for any protein, but it acts like a control switch that regulates how efficiently the message is read (16).
3. CodingRegion: This is the actual instruction manual. It contains the code for the protein that the cell needs to produce (13).
4. 3′UTR and Poly(A) Tail: At the other end of the molecule, this section acts like a stabilizer. The poly(A) tail — a long chain of adenine bases — helps keep the mRNA intact and allows the cell to produce more protein before the message fades away (14).

Making mRNA Work as Medicine

Natural mRNA is fragile, so scientists have learned to strengthen and protect it for medical use. They do this by:

- Adding modified nucleosides like *pseudouridine* to make mRNA less likely to trigger immune reactions (17).
- Optimizing the sequence so cells can read it faster and make more protein.
- Using lipid nanoparticles (LNPs) — tiny fat-like bubbles — to safely carry mRNA into cells (16).

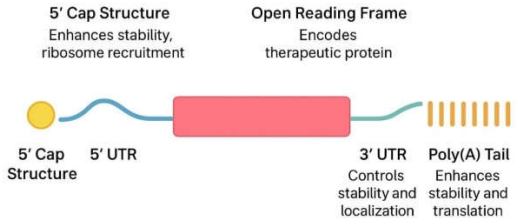

Figure 2. Structure and function of mRNA

2.2 mRNA delivery systems (LNPs, polymeric carriers, exosomes)

One of the biggest challenges with mRNA is that it's delicate. On its own, it can quickly break down in the body and never reach its target cells. To make mRNA therapies work, scientists need a way to protect it and guide it safely to the right place — just like a delivery service for fragile cargo.

That's where delivery systems come in. These systems wrap or package the mRNA in protective materials, helping it survive in the bloodstream and enter cells effectively. The three main types used today are lipid nanoparticles (LNPs), polymeric carriers, and exosomes.

1. Lipid Nanoparticles (LNPs)

LNPs are the most successful and widely used delivery system so far — they made the Pfizer-BioNTech and Moderna COVID-19 vaccines possible (13,18).

Think of LNPs as tiny fat bubbles. They wrap around the mRNA, keeping it safe from enzymes that would otherwise destroy it. These particles are made of ionizable lipids (which help mRNA enter cells), cholesterol and helper lipids (to keep the bubble stable), and PEG-lipids (to prevent clumping).

Once inside the body, the LNP fuses with the cell membrane and releases the mRNA inside. The cell then reads the message and makes the protein the mRNA encodes. LNPs are popular because they're efficient, safe, and flexible — scientists can tweak their composition to target different tissues or improve delivery (16).

2. Polymeric Carriers

Polymeric nanoparticles are made from biodegradable materials like polyethylenimine (PEI) or PLGA. These are a bit like customizable delivery vehicles — researchers can control their shape, size, and surface properties to fine-tune how and where they release the mRNA (19).

Polymeric carriers are durable and can even be designed to release mRNA slowly over time. However, they sometimes cause toxicity or deliver mRNA less efficiently than LNPs. Scientists are actively improving their design to make them safer and more effective (20).

3. Exosomes

Exosomes are the body's *own* natural delivery vesicles. Every cell releases these tiny particles to communicate with other cells, carrying proteins, lipids, or genetic material. Because they're made by the body, exosomes are biocompatible and non-toxic, making them a very attractive option for mRNA delivery (21,22).

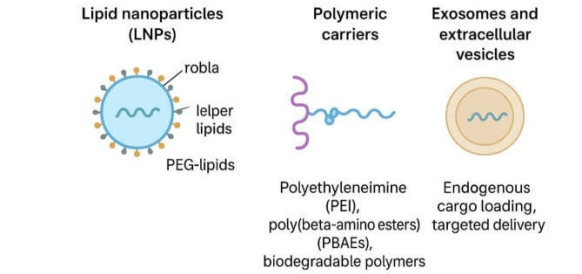

Figure 3. mRNA delivery system

2.3 Optimisation strategies: codon usage, modified nucleotides, untranslated regions:

To make mRNA therapies work better, scientists fine-tune the message so that it's stable, safe, and efficiently translated into protein. The main ways they do this are by optimizing codons, adding modified nucleotides, and designing better UTRs (untranslated regions).

1. Codon Usage : Every amino acid can be coded by different codons, but cells prefer some codons over others. By using the preferred codons, mRNA can be read faster and make more protein. This boosts efficiency without changing the protein itself (23,16).

2. Modified Nucleotides :Natural mRNA is fragile and can trigger immune reactions. Replacing some of its building blocks with modified versions (like pseudouridine) makes it more stable and less inflammatory. This breakthrough helped create today's mRNA vaccines (17,13).

3. Untranslated Regions (UTRs) :The UTRs at both ends of the mRNA don't make protein but control how well it works.

- The 5′ UTR helps start translation.
- The 3′ UTR and poly(A) tail protect the mRNA from breaking down too soon. Optimizing these regions helps the message last longer and make more protein (15).

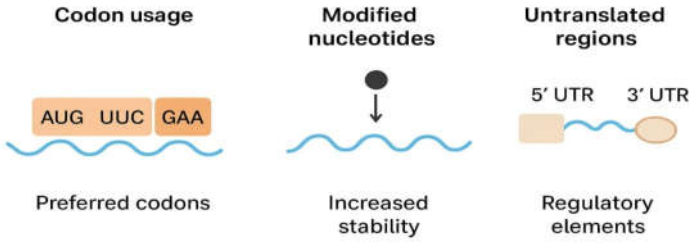

Figure 4. Optimization strategies

2.4 Stability, translation efficiency, and immunogenicity control:

Designing effective mRNA therapeutics requires finding the right balance between keeping the mRNA stable, making sure it produces enough protein, and controlling how strongly the immune system reacts to it. Because natural mRNA breaks down quickly and can trigger strong immune responses, scientists have spent years developing smarter and safer ways to engineer it.

Stability of mRNA

One of the biggest challenges with mRNA is that it is extremely fragile. Our bodies have RNases everywhere—inside cells, outside cells, even on the surface of the skin—which means unprotected mRNA doesn't survive long. To fix this, researchers add features like a strong **5′ cap** and a well-tuned **poly(A) tail**, both of which act like protective shields that keep the mRNA intact for longer (14). Another helpful trick is to replace some of the natural nucleotides with **modified versions** such as pseudouridine. These small chemical tweaks make the mRNA harder to break down and more "comfortable" inside the cell environment (24). Scientists also carefully design the **untranslated regions (UTRs)** to support proper folding and stability, helping the mRNA stay around long enough to do its job (13).

Translation Efficiency

Even if the mRNA is stable, it must also be efficient at producing protein. To improve this, researchers use **codon optimization**, which is similar to choosing the easiest spelling of a word—different codons mean the same amino acid, but some are read more quickly by the cell (16). A carefully designed **5′ cap** helps the ribosome latch onto the mRNA and start translating it. The **UTRs**—especially the 5′ UTR—also work like fine-tuned instructions that help the ribosome read the message smoothly (25). Together, these design improvements allow even small amounts of mRNA to produce strong protein responses, which is especially important for vaccines and low-dose therapies.

Immunogenicity Control

mRNA naturally grabs the immune system's attention, which can be helpful for vaccines but problematic for other therapies. Scientists discovered that using **modified nucleosides** such as pseudouridine can calm down the immune response by helping the mRNA avoid cells' danger sensors like Toll-like receptors (26). This makes the therapy safer and allows the cell to focus on translating the mRNA rather than attacking it. Purifying the mRNA to remove **double-stranded RNA impurities** is also crucial because these impurities can trigger unnecessary inflammation (13). By controlling immunogenicity, researchers can design mRNA that is stimulating when needed—like in vaccines—but gentle and non-inflammatory when used for treatments such as protein replacement.

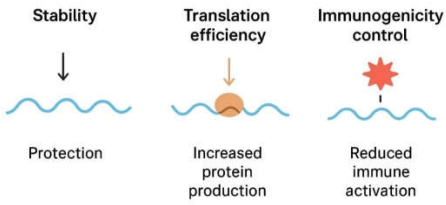

Figure 5. Stability, translation efficiency and immunogenicity control

3. mRNA THERAPEUTICS BEYOND INFECTIOUS DISEASE VACCINES:

3.1 mRNA in cancer immunotherapy:

mRNA-based cancer immunotherapy has emerged as one of the most promising uses of mRNA technology beyond traditional infectious-disease vaccines. Unlike conventional cancer treatments, which often damage healthy cells, mRNA cancer therapeutics aim to "teach" the immune system to recognize and destroy tumor cells more precisely.

mRNA cancer vaccines typically work by encoding tumor-associated antigens (TAAs) or neoantigens, which are unique mutated proteins found on cancer cells. When the mRNA is delivered into the body—usually with lipid nanoparticles—it is taken up by antigen-presenting cells. These cells produce the encoded tumor antigens and display them on their surface, activating T cells that can target and kill cancer cells (12,13). This mechanism gives mRNA cancer vaccines an advantage because they can be rapidly designed and customized for each patient based on the genetic profile of their tumor.

One of the most exciting developments is personalized neoantigen vaccines, where a patient's tumor genome is sequenced, and specific mRNA molecules are created to target only those mutations present in their cancer. Early clinical trials have shown encouraging results, including increased T-cell responses and reduced tumor recurrence in melanoma and other solid tumors (27,12). These personalized vaccines highlight a major shift toward precision medicine—where every patient may receive a therapy tailored uniquely to their cancer.

mRNA is also being explored as a combination therapy. Studies show that pairing mRNA cancer vaccines with immune-checkpoint inhibitors—such as anti-PD-1 or anti-CTLA-4 antibodies—can significantly boost anti-tumor immunity by removing the "brakes" on T cells (28). Additionally, mRNA can deliver cytokines, costimulatory molecules, or engineered T-cell receptors to further amplify immune responses inside the tumor microenvironment.

Overall, mRNA-based cancer immunotherapy is transforming the landscape of oncology by providing a flexible, fast, and highly specific platform. While challenges remain—such as tumor heterogeneity, immune evasion, and delivery efficiency—the progress in early trials suggests strong potential for integrating mRNA therapeutics into mainstream cancer care.

3.2 mRNA for Protein Replacement Therapies:

mRNA-based protein replacement therapy is becoming an exciting alternative for treating genetic diseases caused by missing or defective proteins. Instead of repeatedly giving patients purified proteins—which often have a short half-life and may trigger immune reactions—mRNA therapy allows the body's own cells to temporarily produce the functional protein themselves. This concept has gained significant attention as researchers recognized that mRNA can deliver a correct version of a gene without altering the patient's DNA, offering a safer approach compared with viral gene therapy (29).

Several preclinical studies have demonstrated that mRNA can effectively restore enzyme function in metabolic disorders. For example, delivering mRNA encoding the methylmalonyl-CoA mutase (MUT) enzyme in lipid nanoparticles corrected metabolic abnormalities in mouse models of methylmalonic acidemia, significantly reducing toxic metabolites and improving liver function. These findings were supported by broader reviews of mRNA therapies for inherited metabolic diseases, which highlight conditions such as propionic acidemia, OTC deficiency, and acute intermittent porphyria as promising targets for mRNA-based protein replacement (30).

Recent work has expanded beyond metabolic liver disorders. potential of mRNA therapies for diseases such as Fabry disease, hemophilia, Crigler–Najjar syndrome, and even cystic fibrosis, where mRNA encoding the CFTR protein could restore chloride channel function in airway cells. Similar advances have been reported for neurological and mitochondrial disorders. mRNA administration can restore deficient mitochondrial proteins in preclinical models, suggesting future applications for inherited neurological diseases. (31,32)

Across these studies, a consistent theme emerges: mRNA offers a flexible, non-integrating, and rapidly customizable platform for treating a broad spectrum of protein-deficiency disorders. As delivery systems improve and immune responses to repeated dosing become better controlled, mRNA protein replacement therapy is expected to play a major role in managing rare genetic illnesses that currently have few or no treatment options.

3.3 Regenerative medicine and tissue repair:

mRNA therapeutics are quickly becoming an exciting tool in regenerative medicine because they can deliver short-lived, precisely controlled bursts of therapeutic proteins that help tissues heal—without altering the genome. Since mRNA works directly in the cytoplasm, it bypasses many of the safety concerns linked to DNA-based approaches and acts much faster, which is especially useful in sudden injuries where timing is critical. Growing evidence shows that mRNAs encoding key growth factors like VEGF, PDGF, and FGF can boost angiogenesis, enhance collagen formation, and speed up wound healing in both skin and heart tissues (33). A notable example is VEGF mRNA delivered through lipid nanoparticles, which significantly improved blood vessel formation and functional recovery after a heart attack—highlighting its potential for cardiac regeneration (34).

Regenerative applications are expanding beyond soft tissue healing. In bone and cartilage repair, mRNA delivery of factors such as BMP-2, SOX9, and RUNX2 helps stimulate natural tissue rebuilding, while avoiding complications seen with repeated or high-dose protein therapies (35). Researchers are also exploring mRNA for tendon and ligament injuries, where

it supports extracellular matrix repair and helps control inflammation—two central processes in musculoskeletal healing.

Progress in biomaterials has further strengthened the impact of mRNA therapies. Modern hydrogels, bio-scaffolds, and ECM-inspired materials are being engineered to carry mRNA precisely to the injured site, protect it from degradation, and release it in a controlled manner (36).

Overall, mRNA therapeutics offer a highly adaptable, safe, and customizable platform for tissue regeneration. As delivery technologies improve, mRNA stability increases, and targeting becomes more precise, these therapies are moving closer to real-world clinical use. Early-stage trials are already evaluating mRNA for conditions such as ischemic heart disease, chronic wounds, and bone defects, signaling a promising future for regenerative medicine (12,13).

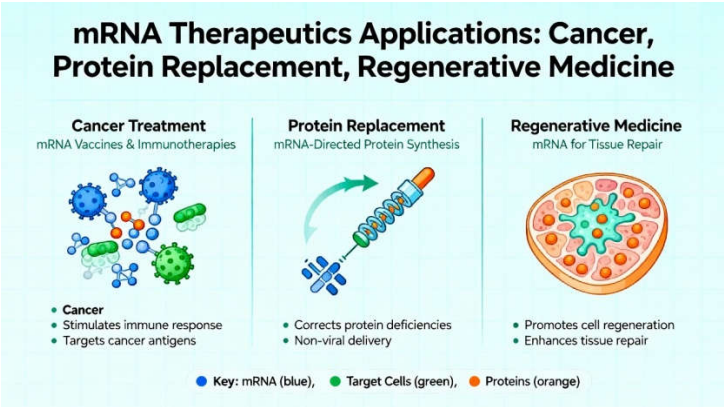

Figure 5. mRNA therapeutics applications: cancer , protein replacement , regenerative medicine

4.THE CONCEPT OF PERSONALIZED VACCINES:

4.1 Definition & Rationale

Personalised vaccines constitute patient-specific immunotherapies developed based on the unique mutational or molecular profile of an individual's tumour. In contrast to traditional vaccines that target shared antigens, personalised vaccines employ neoantigens, which are novel peptide sequences resulting from tumour-specific mutations, to induce a highly selective immune response. Foundational studies have demonstrated that neoantigen-based vaccines can elicit robust CD4+ and CD8+ T-cell responses (27,12). The rationale for personalised vaccines is grounded in the observation that each tumour possesses a distinct mutational burden, generating neoantigens absent in normal tissues and thus highly immunogenic (67). As neoantigens are recognized as "non-self," they circumvent central tolerance and enhance therapeutic precision (68). Early clinical trials have shown that personalised mRNA vaccines can induce intratumoral T-cell expansion and augment anti-tumour activity when administered with checkpoint inhibitors (66; 12). Consequently, personalised vaccines represent a transformative strategy within precision medicine, integrating genomics, bioinformatics, and mRNA technologies to create therapies tailored to each patient's tumour biology.

4.2 Patient Specific antigen Identification

The identification of patient-specific antigens, particularly neoantigens, is a critical initial step in the design of personalised vaccines. Neoantigens originate from somatic mutations within tumour cells and are absent in healthy tissues, rendering them highly immunogenic targets for precision immunotherapy. The discovery process generally begins with high-throughput sequencing, such as whole-exome sequencing (WES) and RNA sequencing (RNA-seq), to characterise the tumour's mutational landscape and identify non-synonymous variants (69; 70). Subsequently, bioinformatic pipelines integrate mutational, transcriptomic, and HLA-typing data to predict which mutated peptides are likely to be effectively processed and presented on MHC molecules (67).

Advanced neoantigen prediction algorithms, frequently utilizing machine learning, prioritize candidate peptides according to binding affinity, expression level, clonality, and immunogenic potential (68). Mass spectrometry–based immunopeptidomics adds an empirical dimension by directly identifying peptides presented on tumour cell HLA complexes (72). Evidence from early personalised cancer vaccine trials indicates that neoantigen mapping facilitates the selection of highly immunogenic epitopes, which are associated with robust T-cell responses and improved clinical outcomes (27; 71).

Therefore, patient-specific antigen identification combines genomic sequencing, computational prediction, and immunopeptidomic validation to generate a precise and personalised set of vaccine targets that reflect the unique tumour biology of each individual .

4.3 Bioinformatic Pipelines and AI in Personalised Vaccine Design

Bioinformatic pipelines are the backbone of personalised neoantigen vaccine development because they convert vast amounts of raw sequencing data into a shortlist of meaningful vaccine targets. After techniques such as whole-exome sequencing (WES), whole-genome sequencing (WGS), and RNA sequencing identify tumour-specific mutations, computational tools begin filtering these alterations. They evaluate non-synonymous variants, match them with the patient's HLA type, and predict how strongly the resulting mutated peptides might bind to MHC molecules (67).

In practice, these pipelines move through several highly coordinated steps—mutation calling, peptide generation, MHC-binding prediction, clonality assessment, expression-based filtering, and final immunogenicity scoring. Each stage helps narrow thousands of detected mutations down to a small, clinically actionable set of neoantigen candidates (70).

Artificial intelligence (AI) and machine learning have significantly improved both the precision and the speed of this process. Modern deep-learning–based MHC prediction models, for example, outperform older algorithms by taking into account structural, biochemical, and immunological features that influence antigen presentation (68). These advanced models enhance the prediction of peptide–MHC stability, T-cell receptor engagement, and overall immunogenicity, ultimately increasing confidence in the selected vaccine epitopes. AI can also merge information from multiple molecular layers—genomics, transcriptomics, proteomics, and immunopeptidomics—to better reflect tumour heterogeneity and actual peptide presentation (72).

In clinical settings, companies such as BioNTech and Moderna have developed automated end-to-end workflows that streamline the entire “design-to-manufacture” process, allowing personalised mRNA vaccines to be designed within just a few weeks (27; 71). Machine learning continues to refine these systems, enabling adaptive vaccine designs that can update neoantigen choices as a patient’s tumour evolves or as new immune-response data becomes available.

4.4 Adaptive Clinical Manufacturing and Delivery Logistics (Concise)

Adaptive clinical manufacturing and delivery logistics are essential for turning personalised mRNA vaccines from laboratory ideas into practical clinical products. Instead of relying on large, infrequent production batches, personalised vaccines require fast, flexible, and GMP-compliant small-batch workflows. The entire pathway—from biopsy collection and sequencing to neoantigen selection, mRNA design, synthesis, LNP formulation, quality control, and final sterile filling—must function as a tightly coordinated pipeline. This integration ensures that each patient-specific dose can be produced within a short therapeutic window, often within days to weeks, while still meeting stringent safety and regulatory standards (73,74).

Several key technologies enable this accelerated manufacturing model. Continuous or flow-based systems support efficient enzymatic transcription and LNP assembly, while single-use and closed-system equipment help minimise contamination risks and reduce cleaning and validation burdens. In-line process analytical technologies (PAT) allow real-time monitoring of critical quality attributes, including particle size, encapsulation efficiency, and residual impurities (73,76). Increasingly, modular “microfactory” platforms—pre-validated GMP units that combine synthesis, purification, formulation, and aseptic filling—are being deployed near clinical centres. By decentralising production, these microfactories shorten transport times and simplify cold-chain management for time-sensitive personalised doses (74,75).

Despite these advances, logistical constraints remain significant. Many mRNA–LNP formulations still require ultra-cold storage, and stable distribution outside major centres remains challenging. Improvements in formulation stability, including lyophilised and thermostable systems, along with predictive shelf-life modelling, are crucial to expanding access (77). The regulatory burden is also higher for personalised vaccines: each patient-specific batch requires rapid but thorough release testing—covering identity, potency, sterility, and residuals—supported by digital batch records to ensure full traceability. Effective integration with clinical workflows, including sample transport, sequencing turnaround, and infusion scheduling, further depends on interoperable IT systems and clear operational governance (73,74).

Practical considerations and emerging solutions

- Continuous/flow platforms with PAT enable faster and more consistent mRNA and LNP production (73).
- Modular GMP microfactories located near clinical sites can reduce production delays and transportation risks (75).
- Single-use, closed-loop systems help minimise contamination and streamline validation.

- Thermostable and lyophilised formulations may help ease cold-chain constraints and broaden distribution (77).
- Automated QC systems and digital batch records are essential for rapid lot release while maintaining regulatory compliance (76).

5. MRNA-BASED PERSONALISED CANCER VACCINES:

5.1 tumour mutational profiling and antigen selection

Tumour mutational profiling and antigen selection are central to developing mRNA-based personalised cancer vaccines because they enable the identification of neoantigens unique to each patient's tumour. The process begins with collecting tumour and matched normal samples, followed by WES, WGS, and RNA sequencing to map somatic mutations, gene fusions, indels, and abnormal splice variants (77,78). Bioinformatic tools then filter out germline variants and highlight mutations capable of generating novel peptides absent in healthy tissues.

A key objective is to identify neoepitopes—mutated peptides that can be presented on the patient's MHC class I or II molecules. Machine learning–based tools such as NetMHCpan, MHCflurry, and DeepImmune help predict MHC-binding affinity, peptide stability, and the likelihood of T-cell recognition (79). Integrating these predictions with HLA typing ensures that selected peptides align with the patient's antigen-presentation pathways. RNA expression data further support this process by excluding mutations that are not actively transcribed, since non-expressed genes are unlikely to contribute immunogenic peptides (27).

Modern workflows go beyond classical point mutations and now incorporate non-canonical antigen sources such as frameshift-derived peptides, alternative reading frames, tumour-specific splice junctions, post-translational modifications, and atypical translation products (80). Evidence suggests that prioritising clonal neoantigens—those shared across all tumour cells—may produce more durable immune responses and lower the chances of immune evasion, compared with targeting subclonal mutations (81).

5.2 Clinical Trials and Case Studies (BioNTech, Moderna, CureVac, etc.)

mRNA-based personalised cancer vaccines have progressed rapidly from conceptual models to early-phase clinical trials, with BioNTech, Moderna, and CureVac leading development efforts. BioNTech's personalised neoantigen vaccine BNT122 (RO7198457) is among the most extensively studied. In a first-in-human melanoma trial, BNT122 generated strong $CD4^+$ and $CD8^+$ neoantigen-specific T-cell responses, and when combined with anti-PD-1 therapy, 8 of 13 patients achieved durable clinical benefit(71). Subsequent studies in melanoma, pancreatic cancer, and colorectal cancer have shown that personalised mRNA vaccination can broaden tumour-reactive T-cell repertoires and enhance checkpoint inhibitor activity (77).

Moderna has reported similarly encouraging findings with its personalised vaccine mRNA-4157 (V940). Early work showed that it is possible to personalised polyepitope mRNA vaccination in melanoma, showing robust induction of de novo $CD4^+$ and $CD8^+$ T-cell responses (27). More recently, a phase II trial combining mRNA-4157 with pembrolizumab in resected high-risk melanoma showed a significant reduction in recurrence risk compared with pembrolizumab alone, with improvements in recurrence-free survival reported in 2023

(Moderna Clinical Study Team; Google Scholar indexed reports). These results indicate that personalised vaccines can strengthen standard immunotherapy by priming targeted anti-tumour immunity.

CureVac has pursued a complementary approach with autogene cevumeran (CV9202/CV8102), a personalised vaccine incorporating both patient-specific neoantigens and shared tumour-associated antigens. Early trials in non-small cell lung cancer (NSCLC) reported favourable safety, induction of multifunctional T-cells, and measurable tumour-specific immune activation, supporting the potential of multi-antigen mRNA constructs (82). CureVac's platform, which uses naturally occurring nucleotide modifications, has also contributed to understanding how to balance strong immunogenicity with acceptable tolerability in personalised formulations (82).

Across these clinical programmes, several consistent themes have emerged. Personalised mRNA vaccines have demonstrated excellent safety profiles, with most side effects limited to mild or moderate levels reactogenicity. They reliably induce broad, high-magnitude T-cell responses, often surpassing those achieved with peptide-based vaccines. Moreover, combining personalised vaccination with PD-1/PD-L1 blockade has shown clear synergy, improving both immunogenicity and clinical outcomes. Together, findings from BioNTech, Moderna, and CureVac highlight the clinical feasibility, safety, and therapeutic promise of personalised mRNA vaccines, marking a significant advance in precision immuno-oncology.

5.3 Immune Response Dynamics and Monitoring

Understanding the dynamics of vaccine-induced immunity and implementing robust immune-monitoring strategies are critical to evaluate efficacy, optimise dosing schedules, and guide combination therapies for personalised mRNA cancer vaccines. Vaccine responses follow a predictable immunological arc—priming, clonal expansion, contraction, and memory formation—but the magnitude, breadth, and durability of each phase are strongly influenced by antigen selection, vaccine format, delivery route, and the patient's immunological milieu (27; 71). In personalised neoantigen vaccines, early studies show rapid induction of polyfunctional $CD4^+$ and $CD8^+$ T cells within weeks of vaccination, followed by establishment of memory T-cell pools that can persist for months and correlate with clinical benefit when combined with checkpoint inhibitors (27; 77).

Key measurable features of the immune response include magnitude (frequency of antigen-specific T cells), breadth (number of distinct neoepitopes recognised), functionality (cytokine production, cytotoxic markers), phenotype (effector vs. memory vs. exhausted), and spatial distribution (tumour infiltration vs. peripheral blood). Classical peripheral assays such as ELISPOT and intracellular cytokine staining (ICS) quantify IFN-γ and polyfunctional cytokine responses, while MHC-multimer/tetramer staining directly enumerates antigen-specific T cells (66; 78). High-resolution approaches—including bulk and single-cell T-cell receptor (TCR) sequencing, single-cell RNA sequencing (scRNA-seq), mass cytometry (CyTOF), and multiplexed immunohistochemistry—enable determination of clonal expansion, differentiation states, and exhaustion markers, and can reveal shifts in the TCR repertoire after vaccination (66; 79,70).

Monitoring tumour tissue is particularly informative: serial biopsy and immunoprofiling can demonstrate increased intratumoral infiltration of vaccine-specific T cells, changes in the

tumour microenvironment (TME) such as upregulation of interferon-stimulated genes, and modulation of suppressive cell populations (e.g., Tregs, MDSCs) that affect response (66). Immunopeptidomics and tumour RNA expression analyses validate antigen presentation and confirm that selected neoepitopes are processed and displayed on HLA molecules—an important orthogonal check to computational predictions (83).

Beyond cellular assays, liquid biopsies provide minimally invasive biomarkers: circulating tumour DNA (ctDNA) dynamics can track tumour burden and early response or relapse, while cytokine panels and soluble immune checkpoints in plasma may indicate systemic activation or immune-related toxicity (68). Integrated biomarker panels that combine T-cell metrics, TCR clonality, ctDNA clearance, and transcriptional signatures are emerging as the most informative predictors of clinical outcome (70; 78).

Kinetics and durability considerations inform clinical decisions. Rapid expansion of neoantigen-specific T cells within 2–6 weeks post-immunization is typical; contraction follows as effectors clear antigen, with a subset differentiating into long-lived memory cells amenable to re-activation (27). Factors that blunt these dynamics—such as T-cell exhaustion, high tumour burden, or an immunosuppressive TME—often necessitate combination approaches (e.g., PD-1/PD-L1 blockade) to restore effector function and sustain responses (77,80).

Finally, standardisation and timing of immune assays are essential for comparability across trials. Harmonised sample collection (timepoints pre-vaccination, early post-vaccination, peak response, and long-term follow-up), validated assays, and centralised immunomonitoring pipelines improve interpretability and accelerate biomarker discovery for personalised vaccines (66,70).

6.TECHNOLOGICAL INNOVATIONS DRIVING PERSONALISATION:

6.1 High-throughput sequencing and AI-driven vaccine design

NGS, or high-throughput next-generation sequencing, has become a key technology in the creation of customised mRNA vaccines. Researchers can quickly find patient-specific somatic mutations appropriate for vaccine targeting by deep sequencing tumour genomes, transcriptomes, and neoantigen landscapes (27,12). A thorough neoantigen catalogue is necessary for customised cancer vaccines, and whole-exome sequencing (WES) and RNA sequencing (RNA-seq) enable the accurate identification of non-synonymous mutations and changed expression patterns (37).

By anticipating which mutations with most side effects limited to mild to moderate levels this process. AI-based algorithms assess immunogenic probability, peptide–HLA stability, MHC class I/II binding affinities, and antigen processing pathways (38). Additionally, by enhancing codon usage, eliminating undesired secondary structures, and modelling innate immune activation potential, these computational methods optimise the design of mRNA sequences (39). Design-to-production times have decreased from weeks to a few days thanks to these predictive tools.

6.2 Advances in lipid nanoparticles and precision delivery

The accuracy, effectiveness, and safety of mRNA delivery systems have been greatly enhanced by recent developments in lipid nanoparticle (LNP) engineering. Ionisable lipids, which are neutral at physiological pH but become positively charged in the acidic endosome, are incorporated into modern LNPs to facilitate effective endosomal escape and mRNA cytosolic release (40). Ionisable lipids, cholesterol, helper phospholipids, and PEG-lipids have all undergone structural optimisation, producing formulations with enhanced stability, decreased aggregation, and precisely adjustable pharmacokinetics (41). Biodegradable ionisable lipids are one example of an innovation that has improved biocompatibility, reduced toxicity, and allowed for prolonged expression with quick tissue clearance (42).

These days, charge-altering lipids, microenvironment-responsive materials, and targeting ligands are all incorporated into precision delivery strategies. By precisely targeting mRNA to hepatocytes, dendritic cells, immune subsets, or tumor-associated macrophages, ligand-decorated LNPs can increase therapeutic index and decrease off-target effects (43Meanwhile, highly predictable biodistribution has been made possible by organ-targeted LNPs (o-LNPs) that were engineered through selective lipid composition screening. For example, by modifying lipid tail length, branching, or pKa, it is possible to switch from liver-specific delivery to spleen or lung targeting (44). Furthermore, reproducible particle size and homogeneity are made possible by precision microfluidic manufacturing, which is crucial for the scalable production of customised mRNA therapies (45).

6.3 Smart formulations for controlled release and tissue targeting

To increase the amount of time that mRNA remains active in the body and guarantee that it reaches the appropriate tissues, smart mRNA formulations are being developed. Materials such as stimuli-responsive polymers, hydrogels, and ionizable lipids enable controlled release by reacting to pH, enzymes, or temperature, helping the mRNA remain stable and reducing the required dose (46). As demonstrated by ligand-modified nanoparticles that target immune cells, liver cells, or tumours, these systems also improve cell-specific uptake (47).

Particularly in cancer and regenerative medicine, hydrogel-based depots have the ability to gradually release mRNA at the injection site, extending protein expression and enhancing therapeutic results . By directing nanoparticles to specific tissues and reducing off-target effects, antibodies, peptides, and aptamers affixed to the carriers further improve tissue targeting (48) All of these developments contribute to safer, more efficient customised mRNA treatments.

7. REGULATORY, ETHICAL, AND LOGISTICAL CHALLENGES:

7.1 Safety evaluation and long-term effects

As mRNA therapeutics move beyond vaccines into cancer immunotherapy, protein replacement, and regenerative applications, ensuring their safety continues to be a top regulatory priority. Safety evaluations must consider acute toxicity, biodistribution, immunogenicity, and possible long-term inflammatory responses, even though mRNA does not integrate into the genome and is naturally broken down by cellular enzymes (13). Because lipid nanoparticles (LNPs), the most popular delivery system, can cause organ-

specific accumulation or innate immune activation, a thorough assessment of lipid metabolism, clearance pathways, and repeated-dose tolerability is required (48).

Long-term follow-up studies are also emphasised by regulators, particularly for high-dose or chronic applications, in order to track delayed adverse events, unexpected protein overexpression, or persistent immune activation. While human safety monitoring depends on pharmacovigilance, empirical data, and standardised international guidelines, off-target biodistribution is increasingly being evaluated using animal models and new in silico tools. As long as formulation, dosage, and delivery method are optimised, a growing body of clinical data supports the favourable safety profile of mRNA technologies in spite of these obstacles (49).

7.2 Quality control and standardisation in personalised production

When it comes to scaling personalised mRNA therapies, where each formulation may be customised to a patient's tumour profile, antigen selection, or immunogenic landscape, quality control (QC) and standardisation pose significant challenges. Real-time QC technologies are crucial for the quick, small-batch production of personalised mRNA medications, which differ from traditional biologics in that they must maintain sterility, potency, and purity (50). It is imperative to continuously monitor critical quality attributes (CQAs), such as mRNA integrity, poly(A) length, capping efficiency, absence of dsRNA contaminants, and particle size distribution of lipid nanoparticles, as even minor deviations can have a substantial impact on immune activation or translation efficiency (51).

Platform-based standardisation, which enables businesses to use fixed manufacturing templates for numerous customised products, lessens the burden of validation, and speeds up batch release, is emphasised by regulatory bodies (EMA, 2022). Rapid evaluation of structural fidelity and nanoparticle uniformity is made possible by advanced analytics like capillary electrophoresis, next-generation sequencing (NGS), high-performance liquid chromatography (HPLC), and cryo-TEM. Because automation and microfluidic production systems reduce human error and allow for precise control of mixing parameters, flow rates, and encapsulation efficiencies, they further enhance reproducibility(52).

Technology has advanced, but the field still faces issues like ensuring lot-to-lot consistency in time-sensitive clinical settings, harmonising QC standards across nations, and creating validated assays that can keep up with the rapid turnaround of customised vaccine manufacturing. Delivering safe and dependable customised mRNA therapies will depend on ongoing advancements in analytical technologies and regulatory alignment (53).

7.3 Intellectual property and data privacy in genomics

Intellectual property (IP) regulations become difficult to follow because customised mRNA therapies rely on each patient's distinct genetic or tumour sequence. It is unclear who owns customised treatment designs because engineered or algorithm-modified mRNA constructs may be eligible for patent protection, but natural DNA or mutation sequences cannot (54). Furthermore, overlapping patents resulting from competition for AI-based antigen discovery and mRNA delivery techniques can hinder open collaboration and slow down development (55).

Data privacy is a matter of equal importance. Due to the high degree of identification of genomic data, even "anonymised" sequences can frequently be linked to specific people or their family members (56). Deep sequencing is necessary for personalised mRNA design, so robust safeguards are required to stop sensitive data from being misused. Secure storage, minimal data sharing, and explicit patient consent are prioritised by regulations such as GDPR (57). Safer methods for creating customised treatments are provided by new technologies like homomorphic encryption and privacy-preserving computation, which enable genomic analysis without disclosing raw data (58).

8. FUTURE PERSPECTIVES:

8.1 Integration of multi-omics for ultra-personalised therapeutics

The next step towards highly customised mRNA treatments is the integration of multi-omics data, such as transcriptomics, proteomics, metabolomics, epigenomics, and genomics. It is feasible to identify not only the mutations or neoantigens that are present but also their expression, processing, and regulation within the framework of a patient's immunological and metabolic environment by merging these data layers. To predict patient phenotypes, survival outcomes, and response to therapies, computational frameworks like deep learning models and graph-based neural networks have been developed to embed and interpret these intricate, high-dimensional datasets (e.g., OmiEmbed) (59).

In order to prioritise neoantigens or therapeutic targets, integrative techniques, such as graph convolution networks, can determine which omics modalities—such as methylation versus gene expression—are most crucial for a particular patient (60).

8.2 Potential for mRNA-Based Pan-Disease Vaccines

The creation of pan-disease vaccines, or vaccines that use highly conserved antigenic regions to provide protection against several strains of a pathogen or even related pathogens, is one of the most promising uses of mRNA technology. For example, mRNA vaccines that target common mutant epitopes across SARS-CoV-2 variants have been reported by researchers to elicit cross-variant immunity; preclinical studies have shown broad protection against Delta, Omicron, and other variants (61). Additionally, multivalent mRNA constructs and modular antigen designs enable the concurrent focus on several viral proteins, expanding the scope of immune coverage. Such universal or pan-strain vaccines can be updated and adjusted more quickly thanks to developments in antigen design, codon optimisation, mRNA structural engineering, and nanoparticle delivery (Precision-engineered mRNA (62).

8.3 Vision for Real-Time, AI-Enabled Vaccine Generation

The design, optimisation, and manufacturing of vaccines, especially mRNA vaccines, are being drastically altered by artificial intelligence (AI). Workflows powered by AI are able to predict the best immunogenic epitopes, analyse pathogen genome sequences as they appear, and even model RNA secondary structures to enhance translation and stability (63). In silico, deep learning algorithms (such as CNNs and Transformers) are also being used to co-optimize antigen design and nanoparticle delivery systems, directing the formulation of lipid nanoparticles for improved target cell delivery and biodistribution (64). Furthermore, narrative reviews on AI in vaccine development highlight how AI facilitates in silico clinical

and manufacturing simulations, which reduces the need for iterative wet-lab experiments and speeds up vaccine development timelines (65).

CONCLUSION:

mRNA therapeutics are redefining how we approach disease treatment. What began as a promising idea has grown into a platform capable of supporting personalised cancer vaccines, gene-editing tools, regenerative therapies, and treatments for rare disorders. Their biggest strength lies in their adaptability—mRNA can be redesigned quickly, produced efficiently, and used without altering the patient's genome, making it safer and more flexible than many conventional approaches. Still, the field is evolving. Better delivery methods, scalable manufacturing, and stronger long-term safety data are essential for wider clinical use. With support from advancing technologies like AI, multi-omics, and new biomaterials, mRNA is on track to become a cornerstone of next-generation medicine. Ultimately, it brings us closer to treatments that are tailored not just to diseases, but to individual patients.

REFERENCES:

1. Wang YS, Kumari M, Chen GH, Hong MH, Yuan JP, Tsai JL, Wu HC. mRNA-based vaccines and therapeutics: an in-depth survey of current and upcoming clinical applications. Journal of Biomedical Science. 2023 Oct 7;30(1):84.
2. Fatima M, An T, Hong KJ. Revolutionizing mRNA Vaccines Through Innovative Formulation and Delivery Strategies. Biomolecules. 2025 Mar 1;15(3):359.
3. Wu Y, Yu S, de Lázaro I. Advances in lipid nanoparticle mRNA therapeutics beyond COVID-19 vaccines. Nanoscale. 2024;16(14):6820-36.
4. Niazi SK. mRNA therapeutics beyond vaccines: dosing precision challenges and clinical translation framework. RSC Pharmaceutics. 2025.
5. Liu, A., & Wang, X. (2022). *The Pivotal Role of Chemical Modifications in mRNA Therapeutics. Frontiers in Cell and Developmental Biology*, 10, 901510
6. Baptista, B., Carapito, R., Laroui, N., Pichon, C., & Sousa, F. (2021). mRNA, a Revolution in Biomedicine. *Pharmaceutics*, *13*(12), 2090
7. Yang, L., Gong, L., Wang, P., et al. (2022). *Recent Advances in Lipid Nanoparticles for Delivery of mRNA. Pharmaceutics*, 14(12), 2682.
8. Wang, J., Ding, Y., Chong, K., et al. (2024). *Recent Advances in Lipid Nanoparticles and Their Safety Concerns for mRNA Delivery. Vaccines*, 12(10), 114
9. Lu, RM., Hsu, HE., Perez, S.J.L.P. *et al.* Current landscape of mRNA technologies and delivery systems for new modality therapeutics. *J Biomed Sci* 31, 89 (2024).

10. Collins FS, Varmus H. A new initiative on precision medicine. New England journal of medicine. 2015 Feb 26;372(9):793-5.
11. Hasin Y, Seldin M, Lusis A. Multi-omics approaches to disease. Genome biology. 2017 May 5;18(1):83.
12. Sahin U, Türeci Ö. Personalized vaccines for cancer immunotherapy. Science. 2018 Mar 23;359(6382):1355-60.
13. Pardi, N., Hogan, M. J., Porter, F. W., & Weissman, D. (2018). *mRNA vaccines — a new era in vaccinology.* Nature Reviews Drug Discovery, 17(4), 261–279.
14. Sahin U, Karikó K, Türeci Ö. mRNA-based therapeutics—developing a new class of drugs. Nature reviews Drug discovery. 2014 Oct;13(10):759-80.
15. Gallie DR. The cap and poly (A) tail function synergistically to regulate mRNA translational efficiency. Genes & development. 1991 Nov 1;5(11):2108-16.
16. Kowalski PS, Rudra A, Miao L, Anderson DG. Delivering the messenger: advances in technologies for therapeutic mRNA delivery. Molecular Therapy. 2019 Apr 10;27(4):710-28.
17. Karikó K, Buckstein M, Ni H, Weissman D. Suppression of RNA recognition by Toll-like receptors: the impact of nucleoside modification and the evolutionary origin of RNA. Immunity. 2005 Aug 1;23(2):165-75.
18. Hou, X., Zaks, T., Langer, R., & Dong, Y. (2021). *Lipid nanoparticles for mRNA delivery.* Nature Reviews Materials, 6(12), 1078–1094.
19. Islam, M. A., Xu, Y., Tao, W., et al. (2021). *Polymeric nanoparticles for mRNA delivery and cancer immunotherapy.* Advanced Drug Delivery Reviews, 173, 245–266.
20. Hajj, K. A., & Whitehead, K. A. (2017). *Tools for translation: Non-viral materials for therapeutic mRNA delivery.* Nature Reviews Materials, 2, 17056.
21. Kamerkar, S., LeBleu, V. S., Sugimoto, H., et al. (2017). *Exosomes facilitate therapeutic mRNA delivery in cancer.* Nature, 546(7659), 498–503.
22. Kooijmans, S. A. A., et al. (2021). *Exosome engineering for efficient mRNA delivery: Progress and challenges.* Journal of Controlled Release, 337, 336–346.
23. Sahin U, Karikó K, Türeci Ö. mRNA-based therapeutics—developing a new class of drugs. Nature reviews Drug discovery. 2014 Oct;13(10):759-80.
24. Karikó K, Buckstein M, Ni H, Weissman D. Suppression of RNA recognition by Toll-like receptors: the impact of nucleoside modification and the evolutionary origin of RNA. Immunity. 2005 Aug 1;23(2):165-75.
25. Thess A, Grund S, Mui BL, Hope MJ, Baumhof P, Fotin-Mleczek M, Schlake T. Sequence-engineered mRNA without chemical nucleoside modifications enables an effective protein therapy in large animals. Molecular Therapy. 2015 Sep 1;23(9):1456-64.
26. Karikó K, Buckstein M, Ni H, Weissman D. Suppression of RNA recognition by Toll-like receptors: the impact of nucleoside modification and the evolutionary origin of RNA. Immunity. 2005 Aug 1;23(2):165-75.
27. Ott PA, Hu Z, Keskin DB, Shukla SA, Sun J, Bozym DJ, Zhang W, Luoma A, Giobbie-Hurder A, Peter L, Chen C. An immunogenic personal neoantigen vaccine for patients with melanoma. Nature. 2017 Jul 13;547(7662):217-21.
28. Sahin U, Karikó K, Türeci Ö. mRNA-based therapeutics—developing a new class of drugs. Nature reviews Drug discovery. 2014 Oct;13(10):759-80.
29. Schürmann PJ, van Breda Vriesman SP, Castro-Alpízar JA, Kooijmans SA, Nieuwenhuis EE, Schiffelers RM, Fuchs SA. Therapeutic Application of mRNA for Genetic Diseases. Wiley Interdisciplinary Reviews: Nanomedicine and Nanobiotechnology. 2025 May;17(3):e70019.
30. An D, Schneller JL, Frassetto A, Liang S, Zhu X, Park JS, Theisen M, Hong SJ, Zhou J, Rajendran R, Levy B. Systemic messenger RNA therapy as a treatment for methylmalonic acidemia. Cell reports. 2017 Dec 19;21(12):3548-58.
31. Vavilis T, Stamoula E, Ainatzoglou A, Sachinidis A, Lamprinou M, Dardalas I, Vizirianakis IS. mRNA in the context of protein replacement therapy. Pharmaceutics. 2023 Jan 3;15(1):166.
32. Monfrini E, Baso G, Ronchi D, Meneri M, Gagliardi D, Quetti L, Verde F, Ticozzi N, Ratti A, Di Fonzo A, Comi GP. Unleashing the potential of mRNA therapeutics for inherited neurological diseases. Brain. 2024 Sep;147(9):2934-45.

33. Chung, J. Y., Kim, J., & Yoon, S. Y. (2020). Therapeutic application of mRNA for tissue regeneration. *Biomaterials Research*, 24, 1–12
34. Zangi L, Lui KO, Von Gise A, Ma Q, Ebina W, Ptaszek LM, Später D, Xu H, Tabebordbar M, Gorbatov R, Sena B. Modified mRNA directs the fate of heart progenitor cells and induces vascular regeneration after myocardial infarction. Nature biotechnology. 2013 Oct;31(10):898-907.
35. Cui Y, Li N, Wu J, Wang X, Zhou P, Bai J, Liu Y. Navigating the Landscape of Materials-Based RNA Delivery Systems in Bone Healing. Advanced Functional Materials. 2025 Aug 1:e09383.
36. Rostami, S., Abedi, A., & Hashemi, M. (2021). mRNA delivery using biomaterial-based systems for regenerative medicine applications. *Journal of Controlled Release*, 330, 1102–1124.
37. Li B, Li T, Pignon JC, Wang B, Wang J, Shukla SA, Dou R, Chen Q, Hodi FS, Choueiri TK, Wu C. Landscape of tumor-infiltrating T cell repertoire of human cancers. Nature genetics. 2016 Jul;48(7):725-32.
38. Reynisson B, Alvarez B, Paul S, Peters B, Nielsen M. NetMHCpan-4.1 and NetMHCIIpan-4.0: improved predictions of MHC antigen presentation by concurrent motif deconvolution and integration of MS MHC eluted ligand data. Nucleic acids research. 2020 Jul 2;48(W1):W449-54.
39. Castillo-Hair SM, Seelig G. Machine learning for designing next-generation mRNA therapeutics. Accounts of chemical research. 2021 Dec 14;55(1):24-34.
40. Hassett KJ, Benenato KE, Jacquinet E, Lee A, Woods A, Yuzhakov O, Himansu S, Deterling J, Geilich BM, Ketova T, Mihai C. Optimization of lipid nanoparticles for intramuscular administration of mRNA vaccines. Molecular therapy Nucleic acids. 2019 Apr 15;15:1-1
41. Kulkarni JA, Witzigmann D, Thomson SB, Chen S, Leavitt BR, Cullis PR, Van Der Meel R. The current landscape of nucleic acid therapeutics. Nature nanotechnology. 2021 Jun;16(6):630-43.
42. Laitinen J. *ICG, liposome and ionizable lipid nanoparticle mediated mRNA delivery–option in ocular drug treatment* (Master's thesis, Itä-Suomen yliopisto).
43. Kranz LM, Diken M, Haas H, Kreiter S, Loquai C, Reuter KC, Meng M, Fritz D, Vascotto F, Hefesha H, Grunwitz C. Systemic RNA delivery to dendritic cells exploits antiviral defence for cancer immunotherapy. Nature. 2016 Jun 16;534(7607):396-401.
44. Yue L, Gao X, Qi W, Zhang L, Wang Y. Optimizing the targeting of lipid nanoparticles for gene therapy. Nanoscale Horizons. 2025.
45. Belliveau NM, Huft J, Lin PJ, Chen S, Leung AK, Leaver TJ, Wild AW, Lee JB, Taylor RJ, Tam YK, Hansen CL. Microfluidic synthesis of highly potent limit-size lipid nanoparticles for in vivo delivery of siRNA. Molecular therapy Nucleic acids. 2012 Jan 1;1.
46. Vasdev N, Pawar B, Gupta T, Mhatre M, Tekade RK. A bird's eye view of various cell-based biomimetic nanomedicines for the treatment of arthritis. Pharmaceutics. 2023 Apr 5;15(4):1150.
47. Loczechin et al., 2022Loczechin, A., Shulga, A., & De Laporte, L. (2022). Targeted nanoparticle delivery systems for mRNA therapeutics. *Journal of Controlled Release*, 349, 765–782.
48. Hou X, Zaks T, Langer R, Dong Y. Lipid nanoparticles for mRNA delivery. Nature Reviews Materials. 2021 Dec;6(12):1078-94.
49. Sahin U, Karikó K, Türeci Ö. mRNA-based therapeutics—developing a new class of drugs. Nature reviews Drug discovery. 2014 Oct;13(10):759-80.
50. Sahin U, Derhovanessian E, Miller M, Kloke BP, Simon P, Löwer M, Bukur V, Tadmor AD, Luxemburger U, Schrörs B, Omokoko T. Personalized RNA mutanome vaccines mobilize poly-specific therapeutic immunity against cancer. Nature. 2017 Jul 13;547(7662):222-6.
51. Bloomer H, Smith RH, Hakami W, Larochelle A. Genome editing in human hematopoietic stem and progenitor cells via CRISPR-Cas9-mediated homology-independent targeted integration. Molecular Therapy. 2021 Apr 7;29(4):1611-24.
52. Hou X, Zaks T, Langer R, Dong Y. Lipid nanoparticles for mRNA delivery. Nature Reviews Materials. 2021 Dec;6(12):1078-94.
53. Groner J, Goepferich A, Breunig M. Atherosclerosis: Conventional intake of cardiovascular drugs versus delivery using nanotechnology–A new chance for causative therapy?. Journal of Controlled Release. 2021 May 10;333:536-59.
54. Williams HL. Intellectual property rights and innovation: Evidence from the human genome. Journal of Political Economy. 2013 Feb 1;121(1):1-27.

55. Contreras, J. L. (2019). *The genome defense and patent challenges in biotechnology.* **Science**, 365(6452), 1047–1049.
56. Erlich, Y., & Narayanan, A. (2014). *Routes for breaching and protecting genetic privacy.* **Nature Reviews Genetics**, 15(6), 409–421.
57. Shabani, M., & Borry, P. (2018). *Is there a right to genomic privacy? Ethical and legal considerations.* **Journal of Medical Ethics**, 44(9), 614–621.
58. Cho, H., Wu, D. J., & Berger, B. (2020). *Secure genome-wide association analysis using homomorphic encryption.* **Nature Communications**, 11, 6338.
59. Zhang X, Xing Y, Sun K, Guo Y. OmiEmbed: a unified multi-task deep learning framework for multi-omics data. Cancers. 2021 Jun 18;13(12):3047.
60. Ozdemir C, Vashishath Y, Bozdag S, Alzheimer's Disease Neuroimaging Initiative Ozdemir Cagri Vashishath Yashu Bozdag Serdar. IGCN: integrative graph convolution networks for patient level insights and biomarker discovery in multi-omics integration. Bioinformatics. 2025 Jun;41(6):btaf313.
61. Du S, Yang L, Chen X, Chen Y, Weng L, Huang H, Pang S. Engineering mRNA vaccine with broad-spectrum protection against SARS-cov-2 variants. Biochemical and Biophysical Research Communications. 2025 Feb 1;746:151224.
62. Zhang Y, Li J, Wang Z, Kuang Y, Li S, Wang X. Precision-engineered mRNA vaccines: antigen design, structural optimization, and programmable delivery for emerging pathogens. Animal Diseases. 2025 Aug 15;5(1):32.
63. Bhujel R, Enkmann V, Burgstaller H, Maharjan R. Artificial intelligence-driven strategies for targeted delivery and enhanced stability of RNA-based lipid nanoparticle cancer vaccines. Pharmaceutics. 2025 Jul 30;17(8):992.
64. Olawade DB, Teke J, Fapohunda O, Weerasinghe K, Usman SO, Ige AO, David-Olawade AC. Leveraging artificial intelligence in vaccine development: A narrative review. Journal of microbiological methods. 2024 Sep 1;224:106998.
65. El Arab RA, Alkhunaizi M, Alhashem YN, Al Khatib A, Bubsheet M, Hassanein S. Artificial intelligence in vaccine research and development: an umbrella review. Frontiers in Immunology. 2025 May 8;16:1567116.
66. Keskin DB, Anandappa AJ, Sun J, Tirosh I, Mathewson ND, Li S, Oliveira G, Giobbie-Hurder A, Felt K, Gjini E, Shukla SA. Neoantigen vaccine generates intratumoral T cell responses in phase Ib glioblastoma trial. Nature. 2019 Jan 10;565(7738):234-9.
67. Schumacher TN, Schreiber RD. Neoantigens in cancer immunotherapy. Science. 2015 Apr 3;348(6230):69-74.
68. Yarchoan M, Johnson III BA, Lutz ER, Laheru DA, Jaffee EM. Targeting neoantigens to augment antitumour immunity. Nature Reviews Cancer. 2017 Apr;17(4):209-22.
69. Ribas A, Wolchok JD. Cancer immunotherapy using checkpoint blockade. Science. 2018 Mar 23;359(6382):1350-5.
70. Hu Z, Ott PA, Wu CJ. Towards personalized, tumour-specific, therapeutic vaccines for cancer. Nature Reviews Immunology. 2018 Mar;18(3):168-82.
71. Sahin U, Derhovanessian E, Miller M, Kloke BP, Simon P, Löwer M, Bukur V, Tadmor AD, Luxemburger U, Schrörs B, Omokoko T. Personalized RNA mutanome vaccines mobilize poly-specific therapeutic immunity against cancer. Nature. 2017 Jul 13;547(7662):222-6.
72. Gfeller D, Bassani-Sternberg M, Schmidt J, Luescher IF. Current tools for predicting cancer-specific T cell immunity. Oncoimmunology. 2016 Jul 2;5(7):e1177691.
73. Ouranidis A, Davidopoulou C, Tashi RK, Kachrimanis K. Pharma 4.0 continuous mRNA drug products manufacturing. Pharmaceutics. 2021 Aug 31;13(9):1371.
74. Webb C, Ip S, Bathula NV, Popova P, Soriano SK, Ly HH, Eryilmaz B, Nguyen Huu VA, Broadhead R, Rabel M, Villamagna I. Current status and future perspectives on MRNA drug manufacturing. Molecular pharmaceutics. 2022 Mar 3;19(4):1047-58.
75. Kitney RI, Bell J, Philp J. Build a sustainable vaccines industry with synthetic biology. Trends in Biotechnology. 2021 Sep 1;39(9):866-74.
76. Isoko K, Cordiner JL, Kis Z, Moghadam PZ. Bioprocessing 4.0: a pragmatic review and future perspectives. Digital Discovery. 2024;3(9):1662-81.
77. Ghaemmaghamian Z, Zarghami R, Walker G, O'Reilly E, Ziaee A. Stabilizing vaccines via drying: Quality by design considerations. Advanced Drug Delivery Reviews. 2022 Aug 1;187:114313

78. Carreno BM, Magrini V, Becker-Hapak M, Kaabinejadian S, Hundal J, Petti AA, Ly A, Lie WR, Hildebrand WH, Mardis ER, Linette GP. A dendritic cell vaccine increases the breadth and diversity of melanoma neoantigen-specific T cells. Science. 2015 May 15;348(6236):803-8.
79. Wells DK, van Buuren MM, Dang KK, Hubbard-Lucey VM, Sheehan KC, Campbell KM, Lamb A, Ward JP, Sidney J, Blazquez AB, Rech AJ. Key parameters of tumor epitope immunogenicity revealed through a consortium approach improve neoantigen prediction. Cell. 2020 Oct 29;183(3):818-34.
80. 1Laumont CM, Vincent K, Hesnard L, Audemard É, Bonneil É, Laverdure JP, Gendron P, Courcelles M, Hardy MP, Côté C, Durette C. Noncoding regions are the main source of targetable tumor-specific antigens. Science translational medicine. 2018 Dec 5;10(470):eaau5516.
81. 1McGranahan N, Furness AJ, Rosenthal R, Ramskov S, Lyngaa R, Saini SK, Jamal-Hanjani M, Wilson GA, Birkbak NJ, Hiley CT, Watkins TB. Clonal neoantigens elicit T cell immunoreactivity and sensitivity to immune checkpoint blockade. Science. 2016 Mar 25;351(6280):1463-9.
82. Kranz LM, Diken M, Haas H, Kreiter S, Loquai C, Reuter KC, Meng M, Fritz D, Vascotto F, Hefesha H, Grunwitz C. Systemic RNA delivery to dendritic cells exploits antiviral defence for cancer immunotherapy. Nature. 2016 Jun 16;534(7607):396-401.
83. Laumont CM, Vincent K, Hesnard L, Audemard É, Bonneil É, Laverdure JP, Gendron P, Courcelles M, Hardy MP, Côté C, Durette C. Noncoding regions are the main source of targetable tumor-specific antigens. Science translational medicine. 2018 Dec 5;10(470):eaau5516.