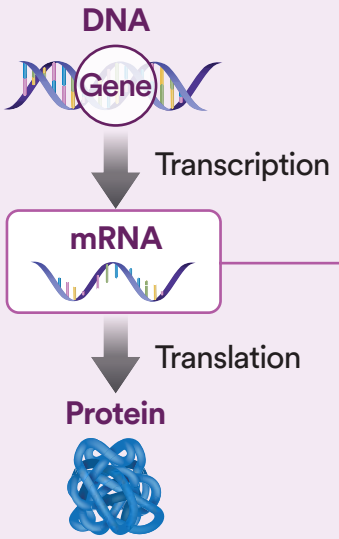
The Science Behind RNA Targeting: Small Interfering RNAs Explained

Why target messenger RNA (mRNA) in the gene expression pathway?

Various diseases arise from dysregulation of one or multiple points within the process of gene expression[1,2]



Targeting mRNA represents a specific upstream approach to treating diseases[1,3]

Key Advantages of Targeting mRNA

Sequence-specific precision
Because each mRNA has a unique nucleotide sequence, it can be selectively recognized and targeted without altering the gene[4]

Non-permanent
Unlike the DNA blueprint, mRNA is a transient molecule, so alterations to it allow for reversible changes in genetic expression[1,5]

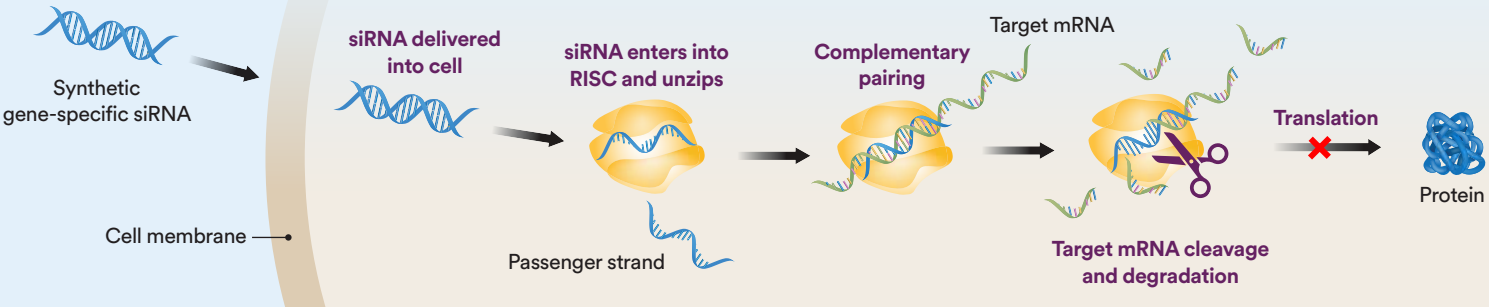
Regulation of pathogenic gene products
By blocking or degrading mRNA, build-up of mRNA and the production of proteins associated with disease can be inhibited[4,6]

What is RNA interference?

RNA interference (RNAi) is a natural mechanism for silencing mRNA.[6] This process has been adapted therapeutically through small interfering RNAs (siRNAs), which are short double-stranded RNA molecules that are designed to specifically match the target mRNA, mediating its degradation[4,6]

siRNAs guide the RISC to precisely silence an mRNA of interest[6]

Following delivery to the target cell, siRNAs are loaded into the RNA-induced silencing complex (RISC), which uses one siRNA strand as a guide to find and cleave matching mRNA sequences. This action specifically blocks expression of the gene of interest and, subsequently, reduces production of the associated protein[6]



Clinical rationale for siRNA therapeutics

- siRNA therapies aim to enable **precise, sequence-specific silencing of the target mRNA**, and have the potential to modulate disease pathways[6]
- Potential long-lasting gene-silencing effects allow for **infrequent dosing**[a]

RNAi has been studied for its therapeutic potential for >25 years, and several approved siRNA-based therapies have well-characterized long-term safety profiles[7-11]

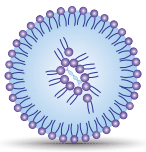
See reverse side for more information

siRNAs as a Therapeutic Modality

siRNA therapies rely on precise sequence targeting and tissue-specific delivery for therapeutic effect.[10] Some key considerations for development are as follows:

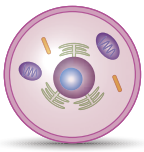
Target selection and specificity

Selecting a clinically-validated mRNA target and optimizing sequence specificity helps ensure that the siRNA achieves its intended biological effect while minimizing potential off-target effects[10]



Delivery approaches

Approaches include lipid nanoparticles and conjugates, with careful consideration of intended target cells, the potential immunogenicity of each delivery system, and systemic vs local administration[4,11]



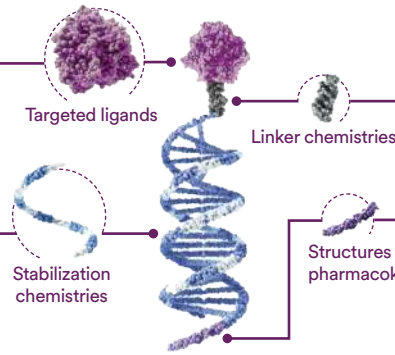
Intracellular challenges

siRNAs are subject to degradation and entrapment in endosomes.[6,11] Strategies to overcome these challenges are important for therapeutic efficacy[11]

siRNAs can be chemically and structurally modified with the goal of improving stability and enabling targeted delivery[6]

Conjugates and ligands[6,11]
Tissue-specific conjugates or ligands (eg, peptides, small molecules, or antibodies) can be added to help guide siRNAs to target tissues, including the liver, lungs, brain, muscle, or heart

Stabilization chemistry[6]
Chemical modifications can be made to help optimize potency and duration, and target specificity



Linkers[6,11]
Linkers enhance siRNA stability and are selected to support delivery to the intended tissue, helping to minimize off-target effects

Pharmacokinetic structures[6]
Novel structures may be added to help extend the half-life of the siRNA (eg, PEGylated lipid and hydroxy-fatty acid)

The Targeted RNAi Molecule (TRiM™) uses an siRNA paired with targeting ligands, linkers, and structural elements designed to enhance pharmacokinetics and support tissue-specific delivery, enabling selective gene silencing while aiming to reduce in vivo toxicity[6,12]

The versatility of siRNA platforms allows for the potential to treat various genetic and acquired diseases[10,13,b]

Muscular
Facioscapulohumeral muscular dystrophy,[c] myotonic dystrophy[c]

Cardiac
Transthyretin amyloidosis,[d] primary hypercholesterolemia,[d] atherosclerotic cardiovascular disease[c]

Pulmonary
Idiopathic pulmonary fibrosis[c]

Hepatic
Acute hepatic porphyria,[d] Hepatitis B[c]

Neurodegenerative
Amyotrophic lateral sclerosis,[c] spinocerebellar ataxia,[c] Huntington's disease[c]

Advances in delivery methods and the modular design of siRNAs are expanding their potential to address diseases across diverse and challenging target tissues[11]

[a]Administered every 3 weeks to 6 months.[7,8] [b]List of approved and investigational therapeutic areas is not exhaustive. [c]siRNA-based therapy is under investigation for the treatment of this disease.[10,13] [d]siRNA-based therapy is approved for the treatment of this disease.[10]

Abbreviations: DNA, deoxyribonucleic acid; mRNA, messenger RNA; PEG, polyethylene glycol; RISC, RNA-induced silencing complex; RNA, ribonucleic acid; RNAi, RNA interference; siRNA, small interfering RNA.

References: 1. Alberts B, et al. *Essential Cell Biology*. Garland Science. 2014; 4th Edition:1-864. 2. Manning KS, Cooper TA. *Nat Rev Mol Cell Biol*. 2017;18(2):102-114. 3. Ige MA, et al. *Mol Ther Nucleic Acids*. 2025;36(4):102721. 4. Zhu Y, et al. *Cell Death Dis*. 2022;13(7):644. 5. Ranasinghe P, et al. *Br J Pharmacol*. 2023;180(21):2697-2720. 6. Hu B, et al. *Signal Transduct Target Ther*. 2020;5(1):101. 7. Adams D, et al. *JAMA Neurol*. 2025;82(3):228-236. 8. Wright RS, et al. *J Am Coll Cardiol*. 2023;82(24):2251-2261. 9. Lieske JC, et al. *Kidney Int Rep*. 2025;10(6):1993-2002. 10. Xiao B, et al. *Mol Ther Nucleic Acids*. 2025;36(1):102437. 11. Tang Q, Khvorova A. *Nat Rev Drug Discov*. 2024;23(5):341-364. 12. Wooddell CI, et al. *JCI Insight*. 2020;5(12):e135348. 13. Sarepta Therapeutics (2025). Accessed January 6, 2026. https://www.sarepta.com/products-pipeline/pipeline

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