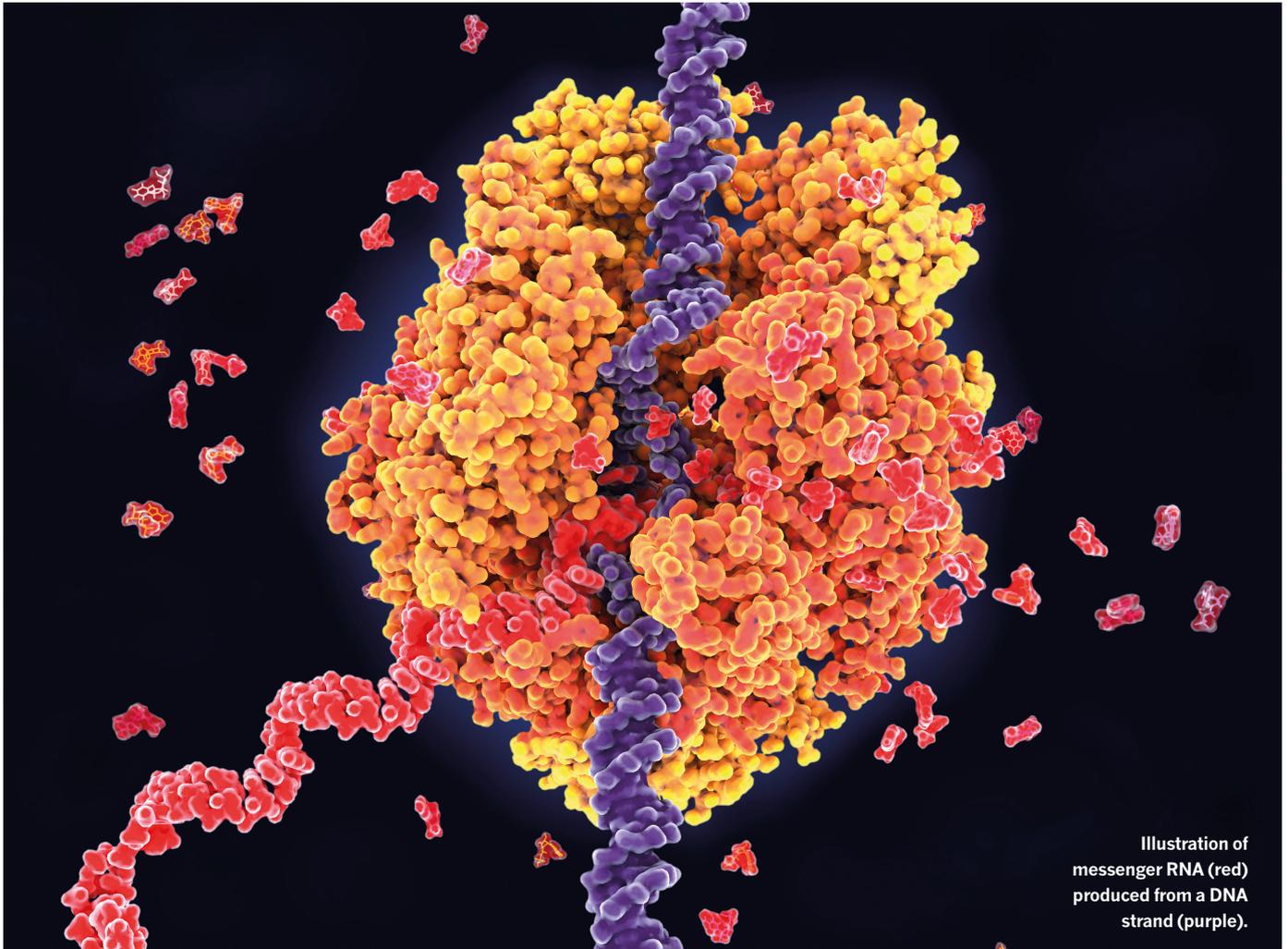


RNA THERAPIES EXPLAINED

Treatments that target RNA or deliver it to cells fall into three broad categories, with hybrid approaches also emerging.

BY SARAH DEWEERDT



JUAN GAERTNER/SPL

Illustration of messenger RNA (red) produced from a DNA strand (purple).

PATH TO THE CLINIC

Messenger RNA was discovered in 1961, but RNA therapies only took off in the 1990s.

1990	1993	1998	1998	2001	2002
A study in mice shows that injecting mRNA into skeletal muscle leads to production of the protein encoded by the RNA ¹ . This result lays the groundwork for treatments based on mRNA.	Injection of mRNA from the influenza virus induces an immune response in mice ² , providing a proof of concept for RNA-based vaccines.	A team led by Andrew Fire at the Carnegie Institution for Science, Washington DC, and Craig Mello at the University of Massachusetts, Worcester, show that short interfering RNAs (siRNA) can suppress gene activity in <i>Caenorhabditis elegans</i> ³ .	The first RNA therapy, fomivirsen, is approved by the US Food and Drug Administration (FDA). The antisense oligonucleotide (ASO) drug tackles cytomegalovirus retinitis, inflammation of the retina.	Researchers demonstrate that RNA interference (RNAi), a mechanism for gene silencing underpinned by siRNA, occurs not only in plants and invertebrates, but also in mammalian cells ⁴ , suggesting its potential for targeting harmful genes.	Researchers led by Mark Kay at Stanford University, California, use RNAi to target a sequence in the hepatitis C virus for destruction in mice ⁵ , highlighting the technology's therapeutic potential.

The conventional view of RNA casts the molecule in a supporting role — the intermediary between DNA and protein, and a passive conduit for information.

And that's how the most familiar form of RNA, messenger RNA, works. But only a small fraction of RNA molecules in cells are mRNAs.

As well as carrying instructions for making proteins, RNAs help to turn genes on and off, aid chemical reactions, slice and dice other RNAs, and even build proteins — by transporting amino acids and linking them together.

“They’re much more active players in cell biology than we initially thought,” says Bruce Sullenger, who investigates RNA therapies at Duke University School of Medicine in Durham, North Carolina.

In turn, these diverse roles have inspired a host of ideas about how to harness RNA for use in medicine. These developments culminated in the 2018 approval, in both the United States and Europe, of two RNA-based therapies for hereditary ATTR amyloidosis — a progressive and potentially fatal disorder in which abnormal proteins build up in nerves and organs such as the heart. Many more RNA therapies are in the development pipeline, with around a dozen already being tested in clinical trials.

Most RNA therapies can be sorted into one of three broad categories: those that target nucleic acids (either DNA or RNA), those that target proteins, and those that encode proteins. Hybrid approaches that combine several RNA-based mechanisms into a single package are also emerging.

There are two distinct types of therapies that target nucleic acids: single-stranded antisense oligonucleotides (ASOs), and double-stranded molecules that operate through a cellular pathway known as RNA interference (RNAi).

ASOs are short stretches of modified DNA made up of about 13–25 building blocks, or nucleotides. These molecules prevent mRNA from being translated into protein by several mechanisms, including blocking the start of translation or tagging the mRNA for degradation. One of the amyloidosis drugs approved last year, inotersen, is an ASO.

ASOs can also alter splicing, the process that sculpts a precursor messenger RNA into its mature form. Two of this type of ASO received FDA approval in 2016: nusinersen, which targets a fatal inherited condition called spinal muscular atrophy, and eteplirsen, a treatment for Duchenne muscular dystrophy. The latter is an example of an ‘exon’ skipping drug, which uses an ASO to block only the mutated portion of a gene from being expressed. The result is a protein that is functional, but that lacks the mutated portion that causes a pathology (see page S16).

Because RNAi makes use of double-stranded molecules, these therapies are tougher to get into cells than ASOs. But fewer molecules are needed for the therapy to be effective. RNAi involves small interfering RNAs (siRNAs), 21–23 nucleotides long, or similar molecules such as microRNAs, to degrade mRNA and prevent it from being translated into protein. The other amyloidosis drug approved in 2018, patisiran, is an siRNA therapy.

RNA therapies that target proteins use a type of molecule known as an RNA aptamer. The molecule is designed to bind to a specific site on



MARIO TAMAW/GETTY IMAGES

A mother and her baby who has microcephaly, which is linked to Zika virus.

a specific protein to modulate its function. Pegaptanib, a treatment for a form of age-related macular degeneration in which blood vessels penetrate the retina and cause vision to deteriorate, is an example of such a drug. Pegaptanib binds to and blocks the function of the protein vascular endothelial growth factor, leading to a reduction in the growth and permeability of blood vessels in the eye. RNA aptamers might be useful in surgery and emergency medicine, in which their rapid action and reversibility could aid anaesthesia and modulate blood clotting.

RNA therapies that use mRNAs are being used to develop personalized cancer vaccines (S10), as well as vaccines for infectious diseases such as Zika virus, which has been linked with the condition microcephaly. Researchers are also exploring whether these type of treatments can be used as protein-replacement therapies for rare conditions such as the blood-clotting disorder haemophilia.

Some researchers are pursuing hybrid strategies that draw on the three main forms of RNA therapy. Apic Bio in Cambridge, Massachusetts, uses an approach that it calls silence and replace, which uses an RNAi component to silence a harmful gene and an mRNA component that encodes a normal version of the corresponding mutated protein. Both are delivered by a single viral vector. The company is doing pre-clinical work on hybrid drugs aimed at α 1-antitrypsin deficiency, an inherited lung and liver disease, and hereditary amyotrophic lateral sclerosis, a degenerative neurological condition.

The biggest barrier to RNA therapy has long been delivering RNA to the correct place in the correct cells. The past several years have seen a flurry of advances that have improved researchers’ ability to get such drugs into liver cells — an important development because so many proteins implicated in diseases are made in the liver. ■

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2003

Judith Lieberman and her colleagues at Harvard Medical School in Boston, Massachusetts, demonstrate that RNAi can suppress HIV replication in macrophages¹.

2004

Pegaptanib becomes the first RNA aptamer that targets proteins to be approved by the FDA.

2006

Fire and Mello receive the Nobel Prize in Physiology or Medicine for their work on RNAi. Meanwhile, Kay and his team report that long-term siRNA expression can cause liver damage and even death in mice², temporarily halting progress on RNAi-based therapies.

2010

The first use of RNAi-mediated gene silencing in humans is reported in a phase I trial in people with the skin cancer melanoma³. The therapy reduced expression of a gene needed for tumour cells to multiply.

2018

Approval to market patisiran and inotersen for hereditary ATTR amyloidosis is granted in the United States and Europe.

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