News & views

Translation

RNA structure underpins a dynamic regulatory switch

Yizhu Lin & Stephen N. Floor

During translation, messenger RNA guides protein production, and certain conditions can favour particular proteins. Helicase enzymes and mRNA structure control translation during defence responses in plants. **See p.423**

Organisms can respond to stressful conditions by changing which proteins they make. On page 423, Xiang *et al.*[1] reveal how plants harness structural aspects of RNA sequences to determine which proteins are made during a defence response and show that this mechanism can occur in human cells.

Most genes encode the instructions needed to make protein products. The initiation of this protein-production process, called translation, is generally the rate-limiting step in protein synthesis from messenger RNA.

The classic model of translation initiation involves several steps. First, a subunit of the protein-production machinery (the small ribosomal subunit) and many other factors assemble on one end (termed the 5′ end) of an mRNA molecule. This menagerie of proteins and associated factors then scans along the mRNA in search of a nucleotide sequence known as an AUG start codon. When the start codon is recognized, another key component (the large ribosomal subunit, which is needed to assemble a complete ribosome) joins, and protein synthesis begins.

However, not all AUG codons are equal, and some are recognized as start codons and initiate protein synthesis more efficiently than others. The preferentially used main start codon (mAUG) is typically surrounded by a nucleotide sequence, called the Kozak sequence, that promotes translation. Most mRNAs contain nucleotide sequences upstream of the mAUG, in a part of the mRNA called the 5′ untranslated region (5′ UTR). The 5′ UTR often harbours regulatory elements including other potential upstream start codons (uAUGs) or related sequences and double-stranded RNA (dsRNA) structures amid the single-stranded RNA.

The presence of an uAUG generally inhibits recognition of the downstream mAUG, which inhibits protein synthesis from the mAUG[2]. An uAUG might prevent expression of a particular protein in a variety of ways, for example, by driving premature termination of translation (if the nucleotides following the uAUG are in a 'reading frame' that contains a premature 'stop' sequence), although there are also cases in which an uAUG can enhance translation[3]. A ribosome scanning along mRNA needs the molecule to be unfolded, therefore stable dsRNA regions inhibit protein synthesis[4].

Things become yet more complicated when both an uAUG and dsRNA are present in an mRNA. More than 30 years ago, it was demonstrated that inserting dsRNA downstream of an uAUG increases the chance that the uAUG is selected as a start codon, but only when the dsRNA is separated by a ribosome-sized distance from the uAUG[5]. This can be referred to as uAUG–dsRNA cooperation.

A probable explanation for such cooperation is that start-codon recognition and ribosome scanning are in a race, and dsRNA structures slow down scanning, providing more time for start-codon recognition and therefore recognition of the uAUG. Indeed, uAUG–dsRNA cooperation can be observed

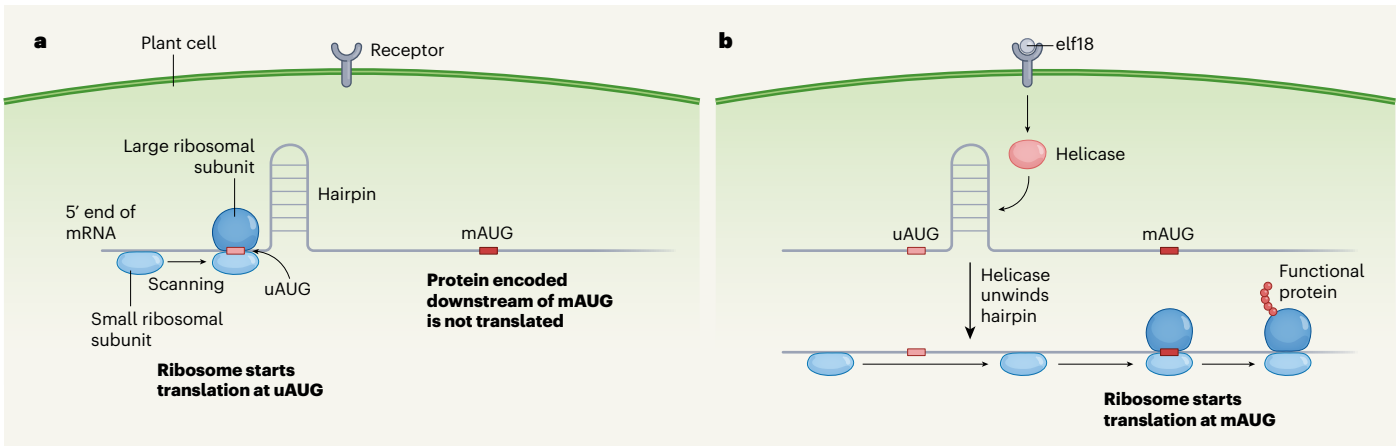

Figure 1 | Messenger RNA structure can influence protein production. Xiang *et al.*[1] examined protein production from mRNA during the process of translation in plants. **a**, The small subunit of the protein-producing ribosome scans along the mRNA, starting at what is termed its 5′ end. This subunit recognizes a sequence at which protein production can start when the large ribosomal subunit joins to initiate translation. Such start sites, called AUG start codons, can be an upstream (uAUG) or a main (mAUG) site. The site chosen determines whether a functional protein is made. The protein encoded downstream of an mAUG is usually the functional protein encoded by the mRNA. A hairpin structure can affect scanning and start-site selection, for example, by slowing scanning and enabling translation to start at the uAUG site. **b**, The authors report that a defence response, triggered by a fragment of the bacterial protein EF-Tu termed elf18, induces expression of a type of enzyme called an RNA helicase. Such enzymes unwind RNA hairpins and thereby favour mAUG over uAUG start sites.

at the level of a single molecule of mRNA[6]. It is also known that enzymes called RNA helicases can unwind RNA structures adjacent to AUG start codons[7]. However, it was unclear whether cells use uAUG–dsRNA cooperation to dynamically regulate protein synthesis.

To identify functional uAUGs in the plant *Arabidopsis thaliana*, Xiang and colleagues used the approach of sequencing mRNA associated with ribosomes in seedlings. These seedlings were undergoing a defence response triggered by the presence of a fragment, termed elf18, of a bacterial protein called EF-Tu. The authors report that mRNAs that increased their association with ribosomes after treatment with elf18 were enriched in uAUGs that could be used as start sites by ribosomes. Moreover, the authors found that the ribosome density at active uAUGs decreased in response to elf18 compared with the situation during normal growth conditions. These findings suggest that translation preferentially begins at the mAUG through a defence-induced release of uAUG-mediated inhibition.

The authors investigated why some mRNAs might experience a decrease in upstream start codon preference during a defence response mediated by the immune system. Xiang and colleagues identified dsRNA structures near uAUGs, and such structures in mRNAs increased the ribosome occupancy at these start codons under normal growth conditions. Notably, the authors observed consistent dsRNA structures both *in vivo* and *in vitro*, suggesting that they are independent of potential interactions with RNA-binding proteins.

Xiang *et al.* tested whether widespread (global) translational reprogramming was regulated by changes in uAUG-associated structures during the immune response. Indeed, the authors found that dsRNA structures were preferentially unfolded in mRNAs that had increased ribosome occupancy during the immune response. A machine-learning classifier tool helped the authors to identify dsRNA near regulated uAUGs. Cooperation between dsRNA and uAUGs was also observed for the 5′ UTRs of the genes *ATF4* and *BRCA* in human cells, which suggests that translation regulation through uAUG structures is an evolutionarily conserved mechanism found in humans, too.

The authors searched for regulatory factors that unwind dsRNA during the immune response. Xiang and colleagues focused on increased amounts of RNA helicases after treatment with elf18, because these enzymes might act as binding partners, called chaperones, for RNA[8]. The authors found that *A. thaliana* helicases, called RH37-like helicases, which are similar to the yeast protein Ded1p and human protein DDX3X (ref. 9), regulate dsRNA structures near uAUGs during the immune response. Expressing higher than normal levels of RH37-like helicases increased translation from the mAUG for many mRNAs. In plant strains lacking RH37-like helicases, these dsRNA structures persisted during the immune response.

Xiang and colleagues' work has uncovered a previously unknown pathway through which cells can regulate protein synthesis in response to stress conditions by inducing RNA-remodelling enzymes, which remove dsRNA structures to shift from uAUG to mAUG start-codon selection (Fig. 1). This finding was made possible by integrating systematic analyses of translation and of RNA structures.

This research also sheds light on how it is possible to achieve dynamic translation regulation through the arrangement of several regulatory elements in the linear sequence of an mRNA. Furthermore, with mRNA becoming a topic of increasing interest for therapeutic purposes, understanding the regulatory mechanisms that govern translation should provide valuable information for the rational sequence design of mRNA-based treatments.

Given that independent regulatory functions of uAUG, dsRNA and RNA helicases have been reported across different species, the authors propose that the uAUG–dsRNA helicase regulatory module might be widely present in multicellular organisms called eukaryotes. This is partially supported by the authors' experiments examining human cells. However, future studies will be necessary to determine the extent of this phenomenon across different species and conditions.

Moreover, changes to transcription[10] or RNA processing[11] can also change start-codon use. Are those mechanisms used in combination with uAUG–dsRNA cooperation? The authors report that three RH37-like RNA helicases are involved in unwinding dsRNA structures associated with uAUG start codons. This finding raises fresh questions. Are the functions of these helicases the same, or do they have different specificities? How are they regulated? Luckily, Xiang and colleagues showcase what approaches could be used to answer these questions.

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1. Xiang, Y. *et al. Nature* **621**, 423–430 (2023).
2. Calvo, S. E., Pagliarini, D. J. & Mootha, V. K. *Proc. Natl Acad. Sci. USA* **106**, 7507–7512 (2009).
3. Dever, T. E., Ivanov, I. P. & Hinnebusch, A. G. *Genes Dev.* **37**, 474–489 (2023).
4. Hinnebusch, A. G. *Annu. Rev. Biochem.* **83**, 779–812 (2014).
5. Kozak, M. *Proc. Natl Acad. Sci. USA* **87**, 8301–8305 (1990).
6. Wang, J. *et al. Cell* **185**, 4474–4487 (2022).
7. Guenther, U.-P. *et al. Nature* **559**, 130–134 (2018).
8. Bohnsack, K. E., Yi, S., Venus, S., Jankowsky, E. & Bohnsack, M. T. *Nature Rev. Mol. Cell Biol.* https://doi.org/10.1038/s41580-023-00628-5 (2023).
9. Ryan, C. S. & Schröder, M. *Front. Cell Dev. Biol.* **10**, 1033684 (2022).
10. Cheng, Z. *et al. Cell* **172**, 910–923 (2018).
11. Hossain, M. A. *et al. Mol. Cell* **62**, 346–358 (2016).

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Astrophysics

Interstellar dust revealed by light from cosmic dawn

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The obscuration of light from a distant galaxy has raised the possibility that a type of carbon dust existed in the earliest epochs of the Universe — challenging the idea that stars had not yet evolved enough to make such material. **See p.267**

The space between stars is full of fine solid particles that range in size from several ångströms to a few micrometres. This 'interstellar dust' is a key component of galaxies[1] both near and far, and it dims the ultraviolet and optical light that arrives at Earth from these galaxies. The way that the dimming varies with wavelength is captured in 'extinction curves' that contain clues about the dust's composition and size. Curves for the Milky Way and other local galaxies exhibit a prominent bump at 2,175 Å, but some of these galaxies do not have this bump, and the same is often assumed of galaxies far away from Earth[2]. It therefore comes as a surprise that there is a pronounced extinction bump for a galaxy that lies in the distant reaches of the Universe, as Witstok *et al.*[3] report on page 267.

First discovered in the Milky Way nearly six decades ago[4], the 2,175 Å extinction bump is generally thought to be caused by nanoparticles containing aromatic carbon[5] — carbon atoms arranged in flat hexagonal rings. Graphite is the most stable form of pure aromatic carbon, and extraterrestrial graphite grains formed in the outflows of old, evolved stars and in the ejecta from supernovae have