10-YEAR ANNIVERSARY

Great expectations of small RNAs

During the 1990s, two small RNAs in *Caenorhabditis elegans*, lin-4 and let-7, were found to regulate developmental timing through a unique mechanism, by annealing to a target mRNA and preventing its translation. This unusual behaviour later proved to be the first glimpse of a novel tier of gene expression control that is conserved from plants to mammals and is governed by small or 'micro' RNAs.

The first indication that microRNAs (miRNAs) were not a peculiarity of worms came in 2000, when Pasquinelli *et al.* showed that let-7 is highly conserved across a wide range of species — from flies to humans.

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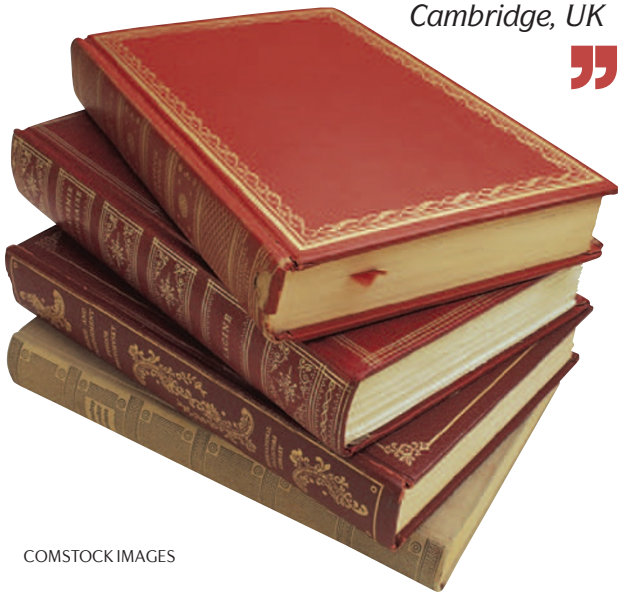

COMSTOCK IMAGES

Importantly, let-7 sequence length was conserved at ~21 nucleotides and, in some cases, its target gene was also retained. On the basis of this, and its temporal regulation, the author's proposed a conserved role of let-7 in regulating gene expression during development, and they also predicted that there may be other such miRNAs. Indeed, a year later, the Ambros, Bartel and Tuschl laboratories reported that there are numerous, diverse miRNAs in human cells, flies and worms.

The next challenge was to identify the potential targets of these miRNAs. Computational approaches were initially used for this, but in 2004 Doench and Sharp went back to basics and experimentally investigated the pairing rules for a miRNA–mRNA interaction using a mutational analysis. Surprisingly, they found that the first eight nucleotides in the 5′ region of a miRNA contribute the most to target specificity and activity, forming a 'seed' that drives the interaction with a target mRNA. This principle formed the basis of modern miRNA target prediction.

Computational predictions also suggested that miRNAs might have several targets. In 2005, Lim *et al.* made the experimental case for this. Using microarray analysis, they showed that a miRNA could drive the downregulation of several target mRNAs in human cells. In addition to reducing protein levels of these targets, the transcripts themselves were also reduced. "This idea, that miRNAs have many targets and act to destabilize mRNA, was heretic at the time but turned out to be exactly right", says Eric Miska (University of Cambridge, UK).

The field has continued to gain momentum since these studies and numerous insights have been gained into the diverse functions of miRNAs both during development and in the adult, for example during mammalian tumorigenesis. The endogenous pathways that drive their production and processing are also being unravelled. This, together with the therapeutic potential of miRNAs that is now emerging, suggests that we may be right to have such high hopes for these small RNAs.

Alison Schuldt

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