To address this, Eulalio *et al.* conducted an unbiased search for microRNAs (miRNAs) that can induce re-entry of postnatal cardiomyocytes into the cell cycle. MicroRNAs are natural, single-stranded RNAs that are around 22 nucleotides long and bind directly to and suppress messenger RNA targets through imprecise base-pairing. Active miRNAs thus suppress expression of cellular proteins from mRNAs, and so their identification can provide insight into the protein regulators that keep cells in a quiescent state.

The authors' screen yielded 204 miRNAs that more than doubled the rate of division of postnatal rat cardiomyocytes (and more than 300 miRNAs that suppressed division). Of the 204, a minority (40) also functioned in mice, and Eulalio *et al.* selected two of these (dubbed hsa-miR-590-3p and hsa-miR-199a-3p) for further analysis. In the follow-up studies, they gathered impressive microscopy images to document mitotic cell division and even its final step, cytokinesis.

The researchers also tested the effect of the two miRNAs *in vivo*. When delivered directly to the rat heart — by means of a viral vector that is used in gene therapy — these miRNAs increased cardiomyocyte proliferation. Moreover, the two miRNAs boosted the normally ineffective process of heart repair after infarction in mice. Specifically, several parameters of cardiac function and structure improved over time, despite the initial loss of cardiac muscle due to the infarction.

These findings are notable because they not only add to evidence for the replicative capacity of cardiomyocytes, but also show that miRNAs can modulate cell regeneration in the heart. Furthermore, by overriding the cellular processes that maintain the quiescent state, they open the door to a broader, systems-level understanding of what prevents cardiomyocytes from re-entering the cell cycle and dividing. This information might enable the design of drugs to target some of the proteins whose expression is affected by these miRNAs, enhancing regeneration.

The miRNAs that Eulalio *et al.* tested did not affect the division of another class of cell in the heart, cardiac fibroblasts, suggesting that these regulatory sequences tap into fundamental mechanism(s) of cardiomyocyte differentiation and quiescence. It remains to be discovered whether the same miRNAs also mediate the regrowth of blood vessels in ischaemic tissue and/or blunt adverse changes that occur upon prolonged stress, such as a detrimental shift in energy utilization or a decline in contractility of the heart muscle. Moreover, it should be investigated whether boosting cardiomyocyte proliferation using miRNAs has undesirable consequences that might compromise heart function in the long term.

In my opinion, the main strength of this study lies in its unbiased search of the miRNAs, and thus of the total cellular pool of proteins (the proteome) that they could have affected, in cardiomyocytes. Functional screening using synthetic miRNAs has also been performed previously to study heart development[10] and disease-related increase in cardiomyocyte size (hypertrophy)[11]. Given that the 1,500 or so known human miRNAs control an estimated 60% of the proteome[12], this approach is emerging as an effective means of gaining comprehensive insight into complex biology. Eulalio *et al.* used a library of about 900 synthetic miRNAs, thus probing a large swathe of the proteome. The miRNA hits that they got might not be the natural regulators of cardiomyocyte quiescence (using miRNAs with an antisense sequence in a similar screening is more likely to lead to miRNAs that sustain quiescence), but are nevertheless valuable because of the proteins they downregulate.

Unfortunately, identifying the target proteins of miRNAs is not easy. The binding of an miRNA to its cognate mRNA is not just a matter of base-pair recognition: sequences surrounding the recognition site and RNA-binding cofactors also influence site accessibility[12]. This makes computational approaches too error-prone to use as the sole means of target prediction. Eulalio *et al.* therefore profiled changes in the levels of mRNA gene transcripts in cardiomyocytes treated with hsa-miR-590-3p and hsa-miR-199a-3p. They found about 700 gene transcripts that showed decreased expression and 1,000 transcripts with enhanced expression; there was little overlap between the mRNAs affected by the two miRNAs.

"The main strength of this study lies in its unbiased search of the miRNAs, and thus of the total cellular pool of proteins that they could have affected."

The authors attempted to determine the importance of particular transcripts by reducing their expression using specific short interfering RNAs (siRNAs). Individual siRNAs only partially reproduced the miRNA effects, in agreement with the expectation that the miRNAs act on multiple cellular mRNA targets and, thus, that multiple proteins cooperate to maintain quiescence. Clearly, systems-level analysis of changes to the proteome is needed to further the understanding of cardiomyocyte quiescence.

Knowledge of the mechanisms that keep cardiomyocytes in a differentiated, non-dividing state will greatly aid the development of regenerative drugs. Ideally, such a drug would target an intracellular signalling pathway coordinating several processes that enable a cardiomyocyte to divide successfully, including cell-cycle entry, faithful DNA synthesis to avoid triggering apoptotic cell death, and disassembly and rebuilding of the contractile apparatus. This is a tall order. The present study is important because it takes a step

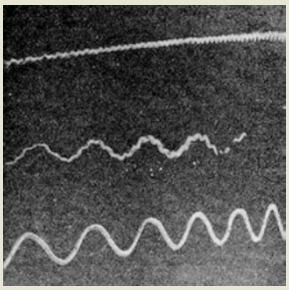
50 Years Ago

I wish to appeal to my fellow geologists and workers in the rapidly growing subject of geochronology to discontinue a habit or fashion which is both unnecessary and misleading. I refer to the habit of calling radiometric ages 'absolute' ages. An age does not become 'absolute' by virtue of being expressed in units of time such as a year … The term is not only redundant and both philosophically and scientifically without meaning: it is also misleading in its psychological suggestion of a higher degree of accuracy than can be justified … If any of the culprits claim that they are in good company with Newton and Kelvin, who both wrote of 'absolute time', they have only to remember Einstein and the coming of relativity.

From *Nature* 22 December 1962

100 Years Ago

'Smoke trace of compound vibrations of tuning-fork' — The accompanying print is from one corner of a smoke trace used by me at a popular lecture in 1901. One curve shows the fundamental (128 per second), another the first upper



partial, while the centre curve of the three shows the form of vibration executed when the first upper partial is sounding, together with the prime. The three sounds may be heard by the audience, and the smoke traces of each obtained in their presence.

From *Nature* 19 December 1912