Circles in the desert

This is the first in a series of editorials I plan to write over the coming 12 months, looking back at papers that caught my attention in bygone years, but which represent still unresolved puzzles or topics recently returned to widespread attention. And some of them are just cool.

I suppose another of my purposes here is to clobber the idea that the value of a piece of published work is robustly measured by the number of times it is cited in the years immediately following publication. I am far from immune to this trend. My own funding, reputation and even the viability of the journal you are now reading depend on my ability to conjure an always rising trend, even if I have to resort to differential equations or chaos theory to stake my claim. In terms of real intellectual progress, I know it's largely meaningless or little more than an ephemeral indicator of fashion. Sometimes observations that were barely noticed and never pursued turn out to be the most important.

I'm going to open my box of memories with a paper that was a side-track on the road to a Nobel Prize, even if it has only rarely been cited.

The late 1970s and early 1980s saw the first fruits of DNA sequencing technology, among them the detailed molecular characterization of eukaryotic viruses. One weekend in this period I drove out to the Californian desert with a clutch of such papers on the backseat of my Beetle, to educate myself a little in this burgeoning field. One of those papers, already a little 'dated' (1978), was the analysis of unintegrated proviral DNA in cells infected with the avian sarcoma virus, from the Bishop/Varmus lab [1].

I camped at Borrego Springs in the arid hinterland of San Diego. As I discovered, American campgrounds are an odd combination: hookups for luxury RVs, plus a few patches of rough gravel for those foolish enough to sleep in a tent; less comfortable than just picking a random spot in the wilderness, where at least there are no

people peeking fearfully out of their plexiglass windows, expecting you to pounce on them at any moment to steal a scrap of toilet paper. As I recall, the only other people under canvas or polyester were a couple of off-duty Marines on some mission to defeat wild rodents with overwhelming fire-power. On reflection, I was surely safer inside the campground than out among the rodents.

In the daytime I hiked up to the oasis, bathed in a cool rock pool, then slouched under a palm tree to read about the mysteries of retroviral replication.

The authors were describing a snapshot in the life cycle of an extrachromosomal genome. It was early days in my love affair with mitochondrial DNA, but the parallels were striking. Shank *et al* had found circular molecules, which they interpreted as an intermediate on the way from the RNA of the infectious virion to the fully fledged, integrated provirus. Or possibly a dead-end recombination product, or even some kind of artefact, though as the major species of unintegrated DNA in infected cells, this seemed unlikely. A subpopulation of these molecules contained two copies of the viral long-terminal repeat (LTR), as if the provirus had simply circularized instead of integrating. However, the major species had only a single LTR, as if the repeats of the linear provirus had undergone intramolecular recombination.

Such circles do not quite fit the paradigm of the retroviral lifestyle. But a circular DNA containing a powerful promoter, information for RNA 3' end formation, and encoding reverse transcriptase, RNase H and a promiscuous integrase, not to mention a signalling molecule that drives the cell into endless proliferation, seems like the mother of all selfish elements.

As far as we know, mtDNA is not closely related to retro-elements. Its polymerases share their evolutionary origins with those of the T-odd phages, although the presumed replicase, DNA polymerase γ , does

possess RNA-dependent DNA polymerase activity, which confounded some early attempts to purify and characterize the reverse transcriptases encoded by retroviruses. Furthermore, mtDNA is transcribed into a full genome-length RNA, which is then processed into different classes of functional transcripts. In some fungi and even some animals, all the genes are encoded on the same strand. Intriguingly, mammalian mtDNA replicates via an RNA/DNA hybrid intermediate on the way to a dsDNA circle containing a single copy of the regulatory information for transcription and replication. Like the LTRs of retroelements, this 'control' region contains a collection of both evolutionarily conserved and hypervariable segments. The idea that replication proceeds around the circle and through a second copy of the control region to create a linear product with terminal repeats that must be resolved by recombination to generate the fully replicated circle, does not seem so far fetched, even if evidence supporting such a model has never been published. It would represent just another solution to the classic 'end' problem in DNA replication: how replicons protect themselves from the attritional loss of terminal information.

There is no point labouring this analogy further. In many small details it breaks down, although retro-plasmids found in fungal mitochondria do combine some of the features of mtDNA and conventional retro-elements. We remain ignorant of how replication terminates in mtDNA, just as we remain ignorant of the meaning of single-LTR circles in retrovirally infected cells. And in my memory these ideas finally dissolve into the circles of the stars traced in a luminous desert sky.

REFERENCE

1. Shank PR *et al* (1978) *Cell* **15**: 1383–1395

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