TUMORIGENESIS

Shoddy repair leads to a bad end

Gross chromosomal rearrangements (GCRs), as well as gains and losses of chromosomes, are a common feature of cancers, but we still have an incomplete understanding of their generation. Now, Wang and colleagues have shown that subunit 1 of replication protein A (RPA1) is essential for maintaining chromosomal stability and tumour suppression in mice.

The trimeric RPA complex is crucial for DNA repair and recombination, and its largest subunit, RPA1, is conserved throughout eukaryotes. Interestingly, previous results from yeast showed that the L221P mutation in the yeast RPA1 homologue resulted in GCRs similar to those found in human cancers. The authors therefore investigated the effect of producing the equivalent RPA1 mutation (*Rpa1* 689T→C) in mice.

Homozygous $Rpa1^{689C/689C}$ embryos died at embryonic day 3.5, but heterozygous $Rpa1^{689C/+}$ mice were obtained that successfully produced the full-length L230P mutant RPA protein. Furthermore, the heterozygous mice were less healthy than their wild-type counterparts. They developed lymphoid hyperplasia and displayed altered haematopoesis, and most mice developed lymphomas, which were highly aggressive in some cases.

Analysis of the tumours showed that neither the wild-type nor the mutant *Rpa1* allele was lost, consistent with its essential role in DNA metabolism. However, all the tumours showed genomic alterations; notably, gains of the whole of chromosomes 6 and 15, and segmental gains and losses of other chromosomes. However, these rearrangements did not involve the loss of any tumour-suppressor genes.

Was this observed genomic instability a direct result of the *Rpa1* 689T→C mutation, or was it a secondary event that arose during tumour progression? Abnormal karyotypes and chromosomal breaks were far more common in $Rpa1^{689C/+}$ mouse embryonic fibroblast (MEF) cells than they were in wild-type MEFs. Moreover, it seems likely that this is due to a loss of DNA double-stand break (DSB) repair that is caused by defective RPA1. One of the markers of DSBs, the phosphorylated histone γ-H2AX, was highly induced in the $Rpa1^{689C/+}$ MEFs, and persisted after the transient inhibition of DNA replication.

The authors returned to yeast to carry out a series of genetic-interaction experiments using the yeast equivalent of the $Rpa1^{689C}$ mutant protein. A significant increase in the incidence of GCR was seen when this mutant protein was expressed in yeast strains that were deficient in homologous-recombination components or various checkpoint proteins. The authors conclude that defects in these processes might therefore underlie lymphoma formation in $Rpa1^{689C/+}$mice.

These results have implications for human tumour formation, as loss of human chromosome 17p13.3, on which the human *RPA1* gene lies, is evident in a wide range of malignancies.

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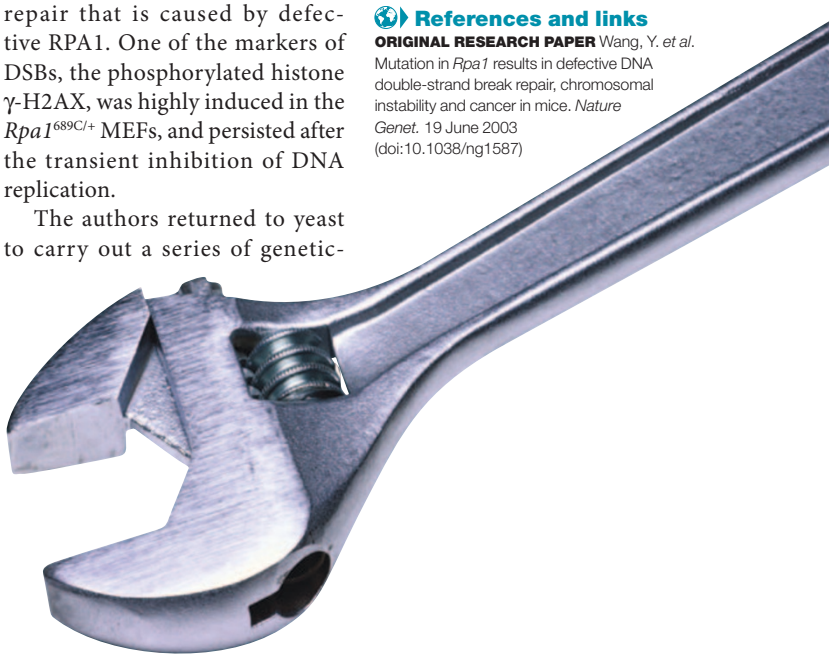

References and links

ORIGINAL RESEARCH PAPER Wang, Y. *et al.* Mutation in *Rpa1* results in defective DNA double-strand break repair, chromosomal instability and cancer in mice. *Nature Genet.* 19 June 2003 (doi:10.1038/ng1587)