expression was selectively increased after treatment with DC101 and that inhibition of the angiopoietin-1 receptor, TIE2, blocked recruitment of the pericytes. In addition, the authors found that the abnormally thick basement membrane often seen in tumour vessels was temporarily reduced. This was not a function of pericyte recruitment, but one of increased matrix-metalloproteinase activity.

Collectively, these results show that blockade of VEGFR2 transiently 'normalizes' the vascular network in tumours and decreases hypoxia, making radiotherapy more effective. The identification of a normalization window in patients might lead to more successful treatments for glioblastoma multiforme and other solid tumours.

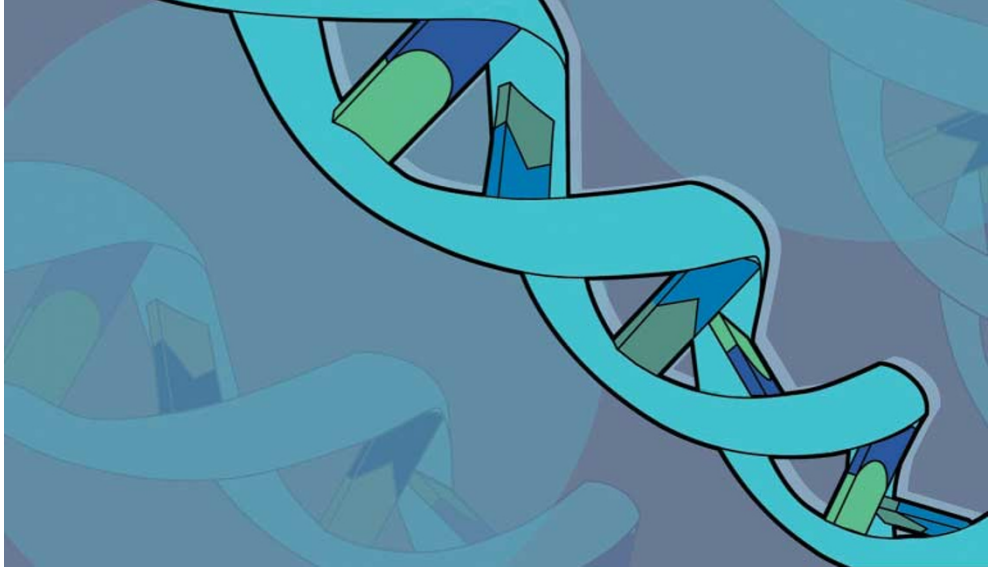
Nicola McCarthy

References and links

ORIGINAL RESEARCH PAPER Winkler, F. *et al.* Kinetics of vascular normalization by VEGFR2 blockade governs brain tumour response to radiation: role of oxygenation, angiopoietin-1 and matrix metalloproteinases. *Cancer Cell* 20 Dec 2004 (doi:10.1016/j.ccr.2004.10.011)

WEB SITE

Rakesh K. Jain's lab: http://steele.mgh.harvard.edu



TUMOUR SUPPRESSORS

A new SNP

The *TP53* tumour suppressor is mutated in at least half of all cancers, and other components of the pathway, such as *MDM2* and *ARF*, are mutated in some of the remaining cancers, which emphasizes the importance of the pathway in tumour development. In the latest issue of *Cell*, Arnold Levine and colleagues investigate whether naturally occurring polymorphisms in these components might influence an individual's susceptibility to cancer, and discover that this is, indeed, correct.

The authors focused on a 300 base pair region of the first intron of the intronic promoter of *MDM2* and looked for sequence variation in 50 healthy individuals. They identified a single-nucleotide polymorphism (SNP) — SNP309, a T to G change — that was present at a relatively high frequency. SNP309 lies within an area that contains several putative binding sites for the SP1 transcription factor, and the nucleotide change was thought to extend one of the sites. This might increase the affinity of SP1 binding. The authors analysed this using electromobility shift assays and found that purified human SP1 does bind with greater affinity to oligonucleotides containing SNP309 than to wild-type oligonucleotides. This binding was confirmed *in vivo* using chromatin immunoprecipitation assays.

What is the effect of increased SP1 binding? *Drosophila* SL2 cells, which are deficient in SP proteins, were transfected with SP1 and an expression vector driven by the *MDM2* promoter containing either the wild-type or G/G homozygous variant. The luciferase reporter was expressed at a higher level with the SNP309 variant than with the wild-type promoter. So, it seems that SNP309 does result in higher levels of transcription from the *MDM2* promoter. Tumour-derived cell lines that contain SNP309 were also found to have eightfold higher *MDM2* mRNA expression and fourfold higher protein expression than those with a wild-type *MDM2* promoter. SP1 was shown to be responsible for this increase, as inhibiting its activity with either RNA interference or the antibiotic mithramycin A markedly reduced MDM2 levels.

But how might this SNP affect the p53 pathway? MDM2 is a negative regulator of p53, so its increased expression should attenuate the p53 pathway. This was found to be true, as the p53 response to the chemotherapeutic drug etoposide was different in cell lines that contained SNP309. In cell lines with the wild-type promoter, 20–35% of cells died following etoposide treatment, but in those homozgous for SNP309, only 2–3% of cells died. This low death rate was found to be due to poor induction of the p53 transcriptional programme.

But does this affect the risk of tumour formation? The authors first investigated this in individuals with Li–Fraumeni syndrome — who carry only one germline copy of *TP53*. Those who also possessed SNP309 had higher levels of MDM2 and a weak DNA-damage response. Furthermore, these individuals developed tumours at a younger age and were more likely to develop multiple primary tumours. The SNP309 variant also affects the risk of sporadic cancers, as soft-tissue sarcomas were diagnosed on average 12 years earlier in individuals with the SNP309 variant.

So, naturally occurring genetic variants can influence susceptibility to tumour development. It will be interesting to look for more of these variants in the population.

Emma Greenwood

NPG Executive Editor, Cell Death and Differentiation

References and links

ORIGINAL RESEARCH PAPER Bond, G. L. *et al.* A single nucleotide polymorphism in the *MDM2* promoter attenuates the p53 tumor suppressor pathway and accelerates tumor formation in humans. *Cell* **119**, 591–602 (2004)