COLORECTAL CANCER

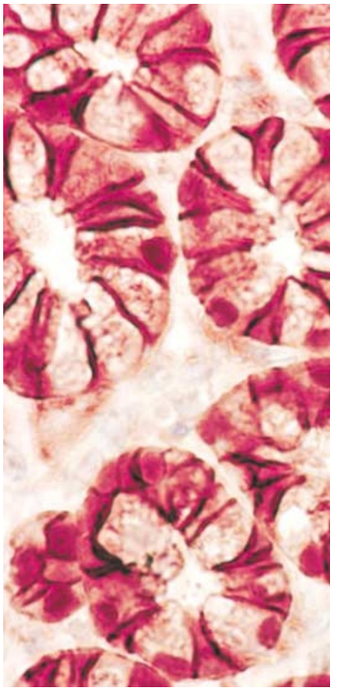
Know your place

Activating mutations in the WNT pathway are the only known genetic alterations that cause intestinal epithelial cells to develop premalignant lesions (polyps). Back-to-back reports in *Cell* by Clevers and colleagues now provide insight into how dysregulation of the WNT pathway might cause colorectal cancer (CRC).

Activation of the WNT pathway induces the translocation of β-catenin to the nucleus to interact with TCF transcription factors. To understand the role of β-catenin–TCF complexes in CRC, the authors used dominant-negative TCF4 (dnTCF4) to inhibit TCF transactivation in CRC cell lines; this caused cell-cycle arrest in G1. Analysis of complementary DNA from the dnTCF4-expressing CRC cells showed small subsets of genes that were upregulated and downregulated. Those that were downregulated were normally expressed in the proliferative compartment of colon crypts, whereas genes that were markedly upregulated localized to the top of the crypts (where differentiated cells are usually found), or were absent when polyps arose.

Of the genes upregulated by dnTCF4, *CDKN1A* — which encodes the cyclin-dependent kinase inhibitor WAF1 — was the only cell-cycle regulator. When the authors induced *CDKN1A* in CRC cells, G1 arrest and differentiation occurred. Conversely, c-*MYC* was the only dnTCF4-downregulated gene that overrode the growth arrest induced by dnTCF4 or WAF1, by binding to and repressing the promoter of *CDKN1A*. So levels of *CDKN1A* are key in regulating differentiation or proliferation.

In the second paper, the role of Eph–ephrin signalling in mediating cell positioning in the small intestine was studied. EphB2 and EphB3 were downregulated in response to TCF4 inhibition. In wild-type embryos, both genes are expressed in the proliferative intervillus pockets, whereas the ephrin-B1 ligand is expressed on adjacent differentiated villus cells. The authors propose that the interaction between cells at the boundary of these two compartments restricts cell intermingling in newborns, which is consistent with established roles for Eph–ephrin signalling. In adults, the pattern of EphB–ephrin-B1 expression is more complex. And in polyps, high levels of EphB2 and EphB3, but not ephrin-B ligands, were expressed. Ephrin-B1-expressing normal cells surrounded, but didn't mix with, EphB-expressing polyps. So, an attractive model put forward by the authors is that "β-catenin–TCF signalling couples cell positioning with cell proliferation, cell-cycle arrest and differentiation."



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References and links

ORIGINAL RESEARCH PAPERS van de Wetering, M. *et al.* The β-catenin/TCF-4 complex imposes a crypt progenitor phenotype on colorectal cancer cells. *Cell* **111**, 241–250 (2002) | Batlle, E. *et al.* β-catenin and TCF mediate cell positioning in the intestinal epithelium by controlling the expression of EphB/ephrinB. *Cell* **111**, 251–263 (2002)

FURTHER READING van Noort, M. & Clevers. H. TCF transcription factors, mediators of Wnt-signaling in development and cancer. *Dev. Biol.* **244**, 1–8 (2002)

WEB SITES

Encyclopedia of Life Sciences:
http://www.els.net
signal transduction pathways in development: Wnts and their receptors | ephrins

Hans Clevers' laboratory:
www.BiomedicalGenetics.nl

The Wnt gene home page:
http://www.stanford.edu/~rnusse/wntwindow.html

TRIAL WATCH

Attack of the clones

Although many studies have shown that tumour-specific T cells can slow tumour growth in mice, there has been little evidence that T-cell-based immunotherapy is effective in human cancer patients. A Phase I clinical trial involving the adoptive transfer of melanoma-specific T-cell clones into patients with therapy-resistant metastatic melanoma has provided new evidence that T cells can be induced to target tumours.

Yee *et al*. isolated cytotoxic T lymphocytes (CTLs) that were specific for two well-defined melanoma/melanocyte antigens — MART1 and gp100 — from ten stage-IV melanoma patients. They primed these T cells *in vitro* using peptide-loaded dendritic cells, and then selected those that specifically lysed MART1- or gp100-expressing cells. These CTL clones were expanded in culture, and transferred back into patients in four separate infusions. After the first infusion, the cells were initially observed to have a short survival time (6.7 days), but when interleukin (IL)-2 was co-administered with subsequent infusions, the average CTL survival time increased to almost 17 days.

Biopsies taken 3 days post-infusion revealed that the tumour-specific CTLs preferentially localized to the tumour. In one patient, the tumour-antigen-specific CTLs were found to make up 37% of the total tumour-infiltrating CTL population, whereas these cells made up less than 1% of the total CTLs in the peripheral blood. Melanoma-antigen-specific T cells were found to make up 0.5–2.2% of all CTLs, compared with the 0.0–0.3% of tumour-specific CTLs detected in previous studies of patients who received vaccine-based therapies.

The adoptive T-cell therapy resulted in disease stabilization in five of ten patients, and minor or mixed responses in an additional three patients for up to 21 months. The average survival time of patients was 11 months, and some patients survived for as long as 21 months. Although the number of patients in this study is small, this is a large improvement over the median survival time of 4 months for patients with refractory metastatic disease. No serious toxicity was observed in any patients after adoptive therapy.

In an accompanying editorial, Drew Pardoll points out that the patients did not have any partial or complete clinical responses to the therapy. This doesn't mean, however, that the transferred CTLs were incapable of antitumour activity. Based on analysis of tumour biopsies, tumour-cell expression of the targeted antigens was lost in three of the five patients examined. This indicates that antigen-expressing tumour cells were eliminated by the CTLs.

These findings support the emerging view that tumour-reactive T cells are present in the peripheral blood of individuals with cancer, and that these can be activated and traffic to metastatic tumour deposits, where they eliminate tumour cells that express target antigen. Further studies to determine the specific signals that regulate T-cell proliferation, as well as ways to increase T-cell activation, localization to tumours and affinity for their antigenic target are necessary to improve this immunotherapeutic approach.

ORIGINAL RESEARCH PAPER Yee, C. *et al*. Adoptive T cell therapy using antigen-specific $CD8^+$ T cell clones for the treatment of patients with metastatic melanoma: *in vivo* persistence, migration, and anti-tumor effect of transferred T cells. *Proc. Natl Acad. Sci. USA* 11 Nov 2002 (doi:10.1073/pnas.242600099)