WEB WATCH

Career choices

- *Science*'s Next Wave: http://nextwave.sciencemag.org/

It's not only cells of the immune system that are faced with decisions, and for those decisions relating to career choice, *Science*'s Next Wave web site could be a good place to start. This career-development resource, which is updated weekly, aims to enable you to find out about the range of options that are open to you at various stages of your career, including research and non-research jobs, whether in academia, industry or elsewhere. Articles provide expert advice from individuals who have followed particular career paths, as well as the latest news from the science job market.

The site is helpfully organized so that recent articles can be found through the country homepages, which bring together news from each region (although homepages for some countries are not available), or through special-focus portals. These portals include the 'Career Development Center', which contains news and feature articles, and a search facility, which allows visitors to search for jobs in academic research or for grants, as well as 'The Grant Doctor', where questions about grants and fellowships are answered. The 'Postdoc Network' includes articles covering many issues that are of importance to postdocs, as well as career-development news, and it is a place where postdocs can raise concerns about their career needs or find the answers to their questions.

Next Wave's Career Resources Library is a useful archive of previously published articles divided into sections, including those specifically for graduate students and for individuals who are interested in career transitions or science policy.

Jenny Buckland

AUTOIMMUNITY

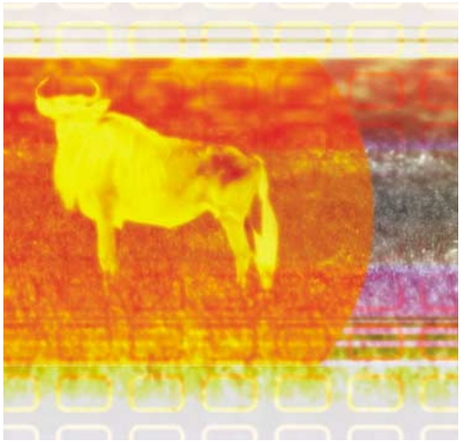
Come and get me!

Few prey specifically try to attract their predator. However, the β cells of the islets of Langerhans in the pancreas do just that, by secreting chemokines to attract islet-specific effector T cells, which then destroy them, according to a paper now published in *Nature Medicine*.

Type 1 diabetes results from the T-cell-mediated autoimmune destruction of insulin-secreting β cells. Early in disease pathogenesis, macrophages and T cells infiltrate the islets and, in combination with pro-inflammatory cytokines, including interleukin-1β, tumour-necrosis factor and interferon-γ, cause insulitis. T cells are not a normal constituent of islets, so what controls the recruitment of these effector cells to these sites?

In this study, Frigerio and colleagues investigated the involvement of chemokines in the recruitment of T cells to the islets. First, they cultured mouse islets and a β-cell line with pro-inflammatory cytokines, and tested whether this resulted in changes in the chemokine expression profiles of these cells. Cytokine treatment caused the upregulated expression of many chemokines, including CXCL9 and CXCL10 transcripts. So, in the presence of pro-inflammatory cytokines, islets and β cells express higher levels of chemokines that are known to attract activated T cells and macrophages.

The authors then used a transgenic mouse model of type 1 diabetes to test the *in vivo* role of chemokines in disease pathogenesis. RIP-GP transgenic mice express the glycoprotein (GP) of lymphocytic choriomeningitis virus (LCMV) under the control of the rat insulin promoter (RIP), which leads to the expression of GP on pancreatic cells. Infection of these mice with LCMV leads to severe insulitis and T-cell-mediated β-cell destruction. After infection, increased levels of pro-inflammatory cytokines, chemokines (including CXCL9 and CXCL10) and chemokine receptors (including CXCR3, the receptor for CXCL9 and CXCL10) were detected in the islets of these mice. Immunohistology of



THYMIC DEVELOPMENT

FoxN1 gene regulation in the thymus

T-cell development and selection in the thymus depend on distinct populations of thymic epithelial cells (TECs). The forkhead transcription factor FoxN1 (also known as Whn), which is lacking in athymic *nude* mice, is required for the growth and differentiation of TECs. However, the signalling pathways that control expression of FoxN1 are not known. In *Nature Immunology*, Balciunaite and colleagues now report that Wnt signalling pathways regulate the expression of FoxN1 and are crucial for TEC development.

Signals from Wnt glycoproteins are transduced through three intracellular pathways, including the Wnt–β-catenin pathway, which is central to many cell-fate decisions during development. Wnt proteins bind cell-surface receptors composed of Frizzled proteins and low-density lipoprotein receptor-related proteins 5 and 6 (Lrp5 and Lrp6), and downstream signalling inhibits the phosphorylation of β-catenin. Dephosphorylated β-catenin interacts with the high-mobility group (HMG) proteins T-cell factor 1 (Tcf1), Tcf3 and Tcf4, and with lymphoid enhancer binding factor 1 (Lef1), enabling these proteins to activate the transcription of downstream target genes.

Mice that are deficient in Wnt1 and Wnt4 have reduced thymic cellularity, and following experiments that showed that Wnt glycoproteins are expressed by TECs, the authors investigated whether TECs respond to Wnt signalling. TEC lines were transfected with a reporter construct (known as TOP) that contains the luciferase gene under the control of Tcf and Lef1 proteins. These cells were then co-transfected to overexpress various Wnt proteins. Wnt1 and Wnt4 were shown to activate Tcf- and Lef1-dependent transcription of the TOP construct, which indicates that TECs do respond to Wnt signals.

Next, the authors asked whether *FoxN1* gene transcription is regulated by Wnt signalling. The level of *FoxN1* messenger RNA was increased in TECs cultured with cells that overexpressed Wnt4 or Wnt5. In addition, *FoxN1* transcription was blocked in the presence of soluble Frizzled proteins (which inhibit Wnt signalling). The putative promoter region of *FoxN1* was shown to be responsive to Wnt4 when it was cloned into a promoter-less luciferase construct.

These results show that Wnt signalling pathways control the transcription of *FoxN1* in TECs and that they are implicated in control of the genetic programme of TECs that is required for their development and thymic function.

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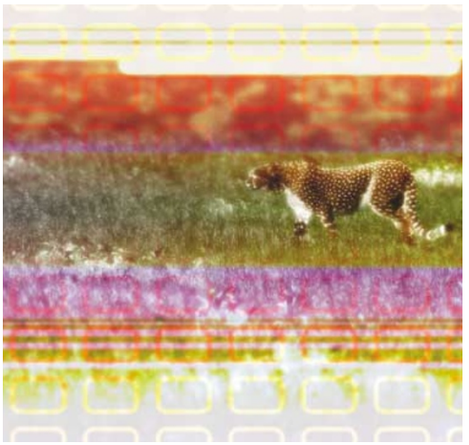

pancreatic tissue indicated that β cells were the main source of CXCL10 during insulitis.

So, chemokines are expressed in islets during insulitis, but are T cells attracted to these chemokines? *In vitro* and *in vivo* studies showed that LCMV-activated T cells are attracted to the chemokines that are present in inflamed islets. To investigate which chemokine receptors are involved, Frigerio *et al.* exposed T cells from LCMV-infected mice to CXCL10 (to desensitize CXCR3 on these cells) before they were cultured with the supernatant from a β-cell line. CXCL10 treatment led to a reduced migratory capacity of T cells towards the supernatants of stimulated β cells, indicating that β-cell chemokines preferentially attract T cells through CXCR3. These observations were confirmed *in vivo* by studies of RIP-GP mice deficient for CXCR3. In the absence of CXCR3, insulitis, diabetes and hyperglycaemia were delayed.

Therefore, in type 1 diabetes, β cells contribute to their own destruction by secreting CXCL9 and CXCL10, which specifically attract $CXCR3^+$ effector T cells to the islets. The authors conclude that CXCR3 might be a new target for therapeutic intervention early in disease.

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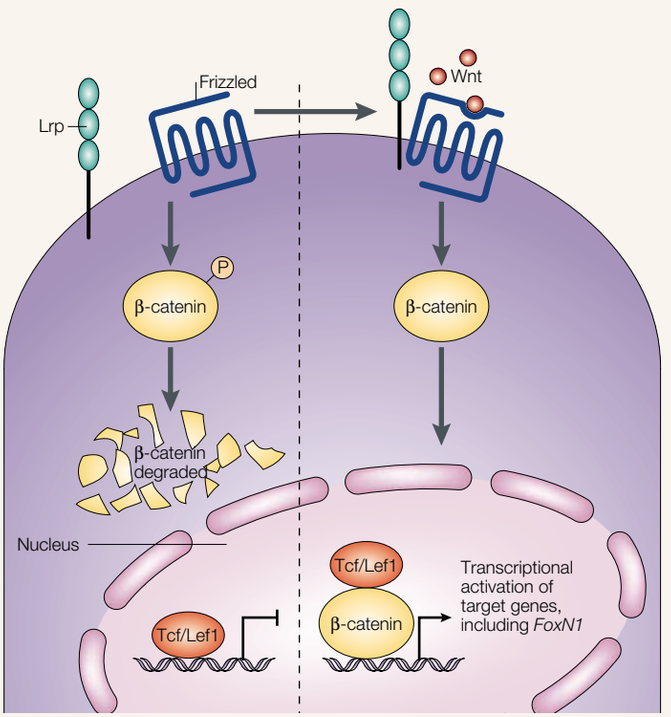
References and links

ORIGINAL RESEARCH PAPER Frigerio, S. *et al.* β cells are responsible for CXCR3-mediated T-cell infiltration in insulitis. *Nature Med.* 4 November 2002 (DOI: 10.1038/nm792)



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ORIGINAL RESEARCH PAPER Balciunaite, G. *et al.* Wnt glycoproteins regulate the expression of *FoxN1*, the gene defective in *nude* mice. *Nature Immunol.* 15 October 2002 (DOI: 10.1038/ni850)

FURTHER READING Anderson, G. & Jenkinson, E. J. Lymphostromal interactions in thymic development and function. *Nature Rev. Immunol.* **1**, 31–40 (2002)

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-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 none of the patients experienced significant tumour regression. 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.

Kristine Novak, Senior Editor, Nature Reviews Cancer

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 November 2002 (DOI:10.1073/pnas.242600099)