CELL BIOLOGY

Splitting hairs

A dual-transgenic mouse with localized expression of two different fluorescent markers is the foundation for an inventive strategy for dissecting hair follicles and isolating their component cell populations.

Baldness jokes aside, hair growth is a surprisingly complicated process; follicle formation relies on an intricate, synchronized process of cell migration, and postnatal follicular function requires maintenance of the stem cell population that drives hair formation and growth. The mesenchymal cells of the dermal papilla (DP), residing at the base of the follicle, have long been recognized as being responsible for niche maintenance, although the specifics of this process are poorly understood. Surgical isolation of DP cells is difficult, owing to the follicle's small size and the variety of other cell types closely surrounding the DP, and even successfully isolated DP cells rapidly lose their *in vivo* properties in culture. As a result, current gene expression data for DP cells is scant—and potentially suspect.

Elaine Fuchs' laboratory at Rockefeller University has long been interested in the processes of hair and skin formation and maintenance, and recently developed an inventive solution to the DP problem. They began by crossing two transgenic mouse lines—one expressing green fluorescent protein (GFP) exclusively in skin epithelium, and the other specifically expressing red fluorescent protein (RFP) in the DP and melanocytes—to generate a strain expressing both fluorescent tags (**Fig. 1**). Starting with a follicular homogenate derived from mouse backskins, the researchers were thus able to efficiently purify individual cell types via fluorescence-activated cell sorting (FACS), using surface antigens and GFP or RFP expression as indicators.

Fuchs' group confirmed the identity and purity of their cell populations and demonstrated that grafts containing keratinocytes and their purified DP cells were capable of generating haired skin on the backs of *nude* mouse recipients. They then proceeded to conduct an exhaustive series of gene expression profiling experiments on each of their purified cell populations, data that they analyzed closely in hopes of getting new insights into follicle function.

"What was really nice about this whole system was that there are closely interacting cell types right next to each other," explains lead author Michael Rendl, "but getting all of them out in a pure form to define what is very specific to each of them and define what is common to them—that was not done before."

They established two categories of genes: 'backbone' genes common to each purified cell type, and 'signature' genes that are dramatically upregulated only in individual cell types. These signatures offered a treasure trove of cell type–specific expression data for a wide variety of signaling factors and genes previously linked with hair formation, as well as several previously unknown genes, and Rendl believes that this information could reveal the communication processes by which the DP collaborates with other cells to maintain the follicle. "There's something very unique about the dermal papilla, because it can induce hair formation," he says. "The dermal papilla are considered to be mesenchymal cells, like fibroblasts. The question is, what makes the dermal

PROTEOMICS

PROTEIN DETECTION GOES DOWN TO THE WIRE

Nanowire-based arrays offer a novel approach for the multiplexed detection of marker proteins in biological samples—and may prove useful for a wide range of other diagnostic applications.

Microarrays are old news—or so one might think. But Harvard University investigator Charles Lieber has given chips a new flavor, developing unusual electronic arrays for sensitive protein detection. The principle of these arrays is simple; silicon nanowires are deposited at specific locations on a specially prepared chip, after which the wires are coated with a receptor molecule selected for the detection of a target of interest. If the target is a charged molecule, then successful binding will manifest as a shift in the electrical conductance of the receptor-conjugated nanowire, with the extent of the shift proportional to the concentration of bound ligand.

Initial trials using prostate-specific antigen (PSA) as a target demonstrated successful detection even at low-femtomolar concentrations. In subsequent experiments, the researchers tested the multiplexing potential of their system by designing a chip with three sets of nanowires linked to antibodies against three different targets and were pleased to observe that each wire showed appropriate antigen specificity with the same remarkable femtomolar sensitivity. The chip design allows considerable multiplexing capabilities, with the potential for simultaneously detecting tens or even hundreds of targets, making it a potentially valuable tool for quantifying biomarkers for human disease from serum or tissue samples. To this end, Lieber's team used their system to quantify PSA from undiluted serum samples, and they were pleased to observe that even with complex natural mixtures, the approach remains effective. "The fact that we could measure a marker at subpicomolar concentration in the presence of all the serum proteins to me was a little surprising," says Lieber, "and it tells you about not only the sensitivity, but also the selectivity."

Of course, the specificity of this system is only as good as its receptors, and Lieber recognizes the importance of keeping an open mind. "The beauty of it is that… there's a lot of different receptors that one could envision using," he says, citing the

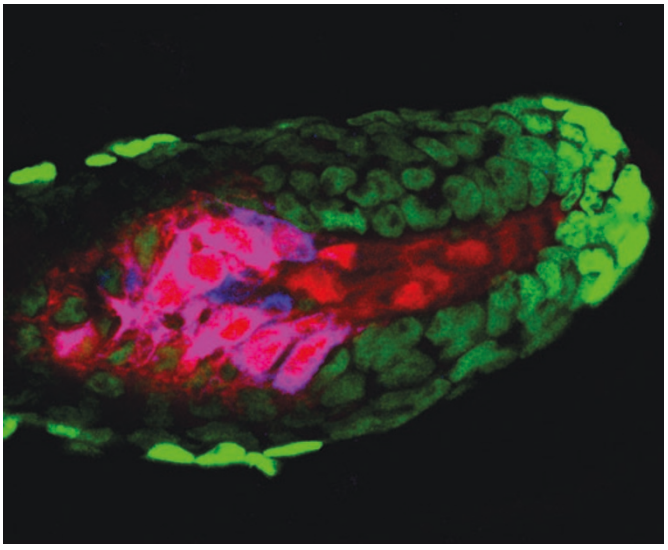

Figure 1 | Section of a hair follicle. This section was taken from a GFP$^+$ (green; epithelial cells) and RFP$^+$ (red; dermal papilla cells and melanocytes) double transgenic mouse after counterstaining with tyrosinase antibodies (blue; melanocytes). Image courtesy of Michael Rendl and *PLoS Biology*.

papilla cells special so that they can do that, compared to regular fibroblasts in the skin or other tissues… we basically want to tackle the whole idea of why it's inductive."
Michael Eisenstein

RESEARCH PAPERS
Rendl, M. *et al.* Molecular dissection of mesenchymal-epithelial interactions in the hair follicle. *PloS Biology* **3**, e331 (2005).

possibility of applying specialized aptamers, or cell-surface molecules such as glycosides. For such molecules, he adds, "that could work even better if you have a membrane-like layer as your receptor, with these essentially cell-like receptor ligands popping out of this membrane over your nanowire, because now you have the very low dielectric constant of this membrane, and you don't have this open aqueous network, which somewhat reduces your sensitivity." Lieber's group has also demonstrated more unconventional applications, such as detecting telomerase activity by monitoring the extension of oligonucleotide-modified substrates.

Lieber suggests that devices like this could represent a new generation of solutions for complex biological problems. "In the broader sense, this is kind of the natural interface between nanoelectronics and biological systems," he says. "It's one of the things I'm most excited about right now."
Michael Eisenstein

RESEARCH PAPERS
Zheng, G. *et al.* Multiplexed electrical detection of cancer markers with nanowire sensor arrays. *Nat. Biotechnol.*; published online 18 September 2005.

NEWS IN BRIEF

GENE TRANSFER

Tumor-targeted, systematic delivery of therapeutic viral vectors using hitchhiking on antigen-specific T cells

Noting that retroviral particles adsorb nonspecifically to the surface of T cells, Cole *et al.* have developed a gene delivery strategy wherein retroviruses are pre-incubated with tumor antigen-specific primary T cells. Back in the host, the virus 'hitchhikes' with these T cells to tumor sites throughout the body for systemic delivery of therapeutic genes.
Cole, C. *et al. Nat. Med.*; published online 18 September 2005.

CHEMICAL BIOLOGY

In vitro selection of RNA aptamers against a composite small molecule–protein surface

Plummer *et al.* describe the *in vitro* engineering of an orthogonal RNA–small molecule–protein complex, in which the RNA and target protein can only bind each other via ternary association with the small molecule. None of these components interact with natural cellular components, and the authors suggest this strategy could prove useful for future gene expression and regulation studies.
Plummer, K.A. *et al*. *Nucleic Acids Res.* **33**, 5602–5610 (2005).

PROTEIN BIOCHEMISTRY

An active enzyme constructed from a nine-amino-acid alphabet

Studies have demonstrated that dramatically reduced amino acid alphabets can be used to encode polypeptides that will assume native-like folds. Walter *et al.* take this process a step further, modifying a bacterial metabolic enzyme so that it is encoded by only nine different amino acids. This 'reduced' enzyme is slightly less stable, but folds properly and retains full catalytic activity.
Walter, K.U. *et al. J. Biol. Chem.*; published online 6 September 2005.

STEM CELLS

Genomic alterations in cultured human embryonic stem cells

Maitra *et al.* offer a cautionary tale for researchers working with embryonic stem cells, noting that, as with other cell lines, long-term passaging increases the risk of genomic changes. They find that after many rounds of passaging, stem cell lines are likely to exhibit alterations like those typically seen in cancers, such as aberrant chromosomal copy number or promoter methylation.
Maitra, A. *et al. Nat. Genet.* **37**, 1099–1103 (2005)

ANIMAL MODELS

An aneuploid mouse strain carrying human chromosome 21 with Down syndrome phenotypes

Down syndrome (DS) is difficult to model in mice, as trisomy of the chromosome syntenic with most of human chromosome 21 (Hsa21) leads to embryonic lethality. O'Doherty *et al.* have developed a method for generating mouse embryonic stem cell lines that contain Hsa21, generating mice whose phenotype partly mirrors DS, and offering hope for modeling other human aneuploidies.
O'Doherty, A *et al. Science* **309**, 2033–2037 (2005).