Another ugly fact.

A few months ago, I headed a blog article (*An ugly fact*) with T H Huxley's quotation about the pain of having a beautiful idea about how the body works refuted by experimental data that will not fit. It has just happened again.

The idea that has just been squashed came out of the work of David Munro, a PhD student who is working on the vascularization of the kidney, funded by the Medical Research Council. In his first year, David produced an elegant descriptive study of blood vessel growth in the developing kidney that demonstrated the existence of complex, cyclical choreography of vessel growth and splitting that keeps pace with the splitting of that will become urine-carrying tubes (see the earlier blog, *Bloody cyclists*). One of David's most interesting observations was that, during their intricate dance, developing blood vessels always avoid entering a zone of kidney stem cells called the cap mesenchyme. He wanted to know why, and suspected that the stem cells may be making a 'keep out' signal.

The lab has for some years (2004-2017) curated the GUDMAP database of gene expression in the developing urogenital system. The database contains information on which genes are expressed where, and the genes themselves are tagged with general descriptions of function (eg signalling molecule, signal receptor etc). David therefore interrogated the database to find any examples of signalling molecules produced in the s cap mesenchyme that were capable of binding to signal receptors on developing blood vessels. The approach is similar to the one I took myself in an earlier experiment on the development of urine-carrying tubes (see '*The creative power of self-loathing*'). The results of the search were very interesting: David found that the cap mesenchyme cells express the signalling molecule Semaphorin3f, which is known in other systems to repel blood vessels. What is more, the growing blood vessels express Neuropilin2, which acts as a receptor for Semaphorin3f. The system was therefore set up perfectly for the cap mesenchyme to be using Semaphorin3f to say 'keep out' to growing vessels.

The test of whether Semaphorin3f → Neuropilin2 signalling is critical to keeping vessels out of the caps is simple: block that signalling and see if the caps are now vascularized. Our collaborator, Thomas Coates, had made 'knockout' mice in which the gene for Semaphorin3f was missing, and he sent kidneys from these mice to Edinburgh. At the same time, Prof. Alex Kolodkin and Dr Qiang

Wang generously sent us embryonic kidneys from mice they had made, from which the Neuropilin2 gene had been removed (for studies of their own not connected with the kidney). Neither of these mice would have been able to use Semaphorin3f → Neuropilin2 signalling for any purpose given that each was missing a critical component of the pathway. When David examined the cap mesenchymes of the kidneys, however, they were still completely free of blood vessels. Clearly, something else is keeping them out.

One of the models for progress in research, a model suggested by the philosopher of science, Karl Popper, is that of 'conjecture and refutation': scientists propose ideas, try to prove them wrong, and by discarding the wrong ones they ultimately converge on the surviving, presumable correct explanations. Refutations of hypotheses are therefore important, and we prepared a paper, now accepted for publication in the journal *Developmental Dynamics* (see Links below). Still grieving the death of the original conjecture, we began the introduction to that paper with the T H Huxley quotation; *The great tragedy of Science – the slaying of a beautiful hypothesis by an ugly fact.*

I hope I have succeeded in persuading this bright young PhD student that the whole thing has actually been immensely good experience for a career in research (which, as a colleague of mine once said, can sometimes be a bit like banging your head against the wall, with the twist that you first have to write a grant application to buy the wall).

If we ever do find out what is keeping blood vessels out of the cap mesenchyme, or if anyone else does, I'll devote a future blog article to the story.

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Links

David’s paper: http://onlinelibrary.wiley.com/doi/10.1002/dvdy.24592/full
(This is the accepted manuscript: I’ll replace the link with one to the final published paper when it is available).