IN THE NEWS

Spleen cells could cure diabetes

A report in *Science*, published on World Diabetes Day (14th November 2003), has brought new hope to diabetes sufferers by showing that spleen cells can be used to cure type 1 diabetes in mice — injected spleen cells differentiated into insulin-producing cells in diabetic animals.

Islet cells of the pancreas secrete insulin, which is needed to remove excess glucose from the blood. But in type 1 diabetes, the immune system destroys the islet cells and high sugar levels in the blood cause serious medical complications. Denise Faustman and colleagues, from Massachusetts General Hospital (MGH), injected donor spleen cells from normal mice into diabetic mice and found, surprisingly, that they produce normal insulin-secreting islet cells. According to Faustman "The unanswered question from that study was whether this was an example of rescuing a few remaining islet cells in the diabetic mice or of regeneration of the insulin-secreting islets from another source" (*ScienceDaily*, 17 November 2003).

In an elegant set of experiments using labelled donor spleen cells, Faustman and colleagues now show that, in fact, the insulin-producing cells grow from both the recipient's own cells and the donor cells. "We have found that it is possible to rapidly regrow islets from adult precursor cells, something that many thought could not be done" said Faustman (*ScienceDaily*, 17 November 2003). David Nathan, Director of the MGH Diabetes Center, believes that using this procedure "Patients with fully established diabetes possibly could have their diabetes reversed" (*BBC News Online*, 14 November 2003), and hopes to test the approach in clinical trials.

Emma Croager

ENDOCYTOSIS

A propelling pathway

Budding yeast contain cortical patches, which, in turn, seem to house endocytic adaptors and cytoskeletal proteins. But these patches can vary in composition and in their dynamics, so any relationship between the actin cytoskeleton and endocytosis has, until now, been circumstantial. A recent *Cell* paper by David Drubin's group, though, outlines a role for actin in the internalization of the budding yeast endocytic complex.

The authors began by studying the localization and dynamics of six yeast proteins that are involved in endocytosis using green fluorescent protein (GFP)-tagging experiments. The Arc15 subunit of the Arp2/3 complex (which is required for actin nucleation) and Abp1 (an activator of the Arp2/3 complex) both had lifetimes of ~15 s. On the other hand, two other Arp2/3 activators, Pan1 and Las17, as well as Sla1 (an endocytic adaptor) and Sla2 (which is thought to function at the actin cytoskeleton–endocytic machinery interface), lasted slightly longer (30–40 s). All the patches, except those containing Las17 (which typically remained at their site of formation) had an initial, movement-restricted phase of formation followed by a motile phase. The patches also moved from the cortex towards the cell centre.

Next, the authors observed that different proteins were recruited to patches invariantly and sequentially. Sla1 was an early patch component, and was joined by Abp1 (and Arc15), before both proteins disappeared. Similarly, Las17 and Sla2 were later joined by Abp1 and Arc15. All this regularity indicated to the authors that these changes in patch composition would reflect changes in patch behaviour. For example, Sla1–GFP patches started to move slowly towards the centre when Abp1 was recruited to patches, and, as Sla1–GFP disappeared from the patch, the fast phase of motility began. Because filamentous actin and Arc15 colocalize with Abp1 in patches, Drubin's group proposed that actin polymerization might be responsible for 'propelling' endocytic vesicles into the cell, so they treated cells with latrunculin A to sequester actin monomers. Their results indicated that actin polymerization was required for a presumed endocytic vesicle, plus any associated proteins, to move away from the cortex, and for this complex to later disassemble. Furthermore, *sla2*Δ cells also showed inhibited patch motility, consistent with a role for Sla2 in endocytosis.

As patch motility was inhibited in both *sla2*Δ cells and by latrunculin A, Drubin's group took a look at actin in *sla2*Δ cells. Rather than the expected punctate staining that normally occurs, actin 'comet tails' accumulated at the cortex. At the junction with the cell cortex, the comet tails associated with Sla1, and, because Sla1 has recently been reported to link certain types of cargo to the endocytic machinery, the authors studied the localization of Ste2, a receptor cargo protein, in *sla2*Δ cells. Ste2 accumulated in punctae at the junction between comet tails and the cell cortex, indicating that this site probably represents a blocked endocytic site and that, normally, the function of Sla2 is to join actin with the endocytic site to render it productive for internalization. Finally, the authors used fluorescence recovery after photobleaching (FRAP) experiments to show that actin filaments were nucleated at/near the endocytic complexes at the cortex, and were disassembled inside the cell.

So Drubin and colleagues describe a model for early endocytosis in budding yeast that resolves many previously unaddressed issues. The initial step is the assembly, in a non-motile complex at the plasma membrane, of endocytic adaptors (such as Sla1) and Arp2/3 activators (such as Las17 and Pan1), which interact with each other. After ~20 s, actin, the Arp2/3 complex and Abp1 are recruited to the patch; at this point, Sla1, Sla2 and Pan1 move inwards. The early patch components are then disassembled, and patches containing only late components undergo a transition to a second phase of fast movement, during which the late components are disassembled. The early complex is probably propelled into the cytoplasm by forces that are generated by actin polymerization. These forces might also invaginate the plasma membrane and be involved in the release of the endocytic vesicle. So the pathway is there — finding out how and when each of the steps is regulated is the next challenge.

Katrin Bussell

References and links

ORIGINAL RESEARCH PAPER Kaksonen, M., Sun, Y. & Drubin, D. G. A pathway for association of receptors, adaptors, and actin during endocytic internalization. *Cell* **115**, 475–487 (2003)