Genetics of Epithelial Architecture: Junctions, Polarity and Tumor Suppressors

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Metazoan animals contain a bewildering variety of cell types whose forms are highly specialized for their functions. Yet how cells adopt these diverse shapes and structures remains mysterious. We are exploring this fundamental question of cell biology using a simple cell type -- epithelia-- in a genetically manipulable organism --Drosophila. We have adapted targeted mosaic techniques to screen, in vivo and in an unbiased manner, for genes required for cells to assume the highly regular epithelial organization. Cloning and characterization of these genes will reveal the mechanisms that regulate general cellular elements, such as the cytoskeleton and protein trafficking systems, in order to confer specific cellular architectures.

Initial work has identified the PDZ domain protein Scribble (Scrib) as a fundamental component that the fly uses to generate any cell with an epithelial architecture. Mutations in *scrib* cause a dramatic transformation of embryonic epidermis from a monolayered columnar epithelium to a multilayered pile of isotropically-shaped cells. *scrib* encodes a PDZ domain-containing protein that acts at cell junctions to regulate apicobasal polarity. Genetics and biochemistry have identified two interacting proteins, called Discs-large (Dlg) and Lethal giant larvae (Lgl), that cooperate with Scrib in this process. Homology of Lgl to SNARE-binding proteins from yeast and vertebrates suggests that Scrib, Dlg, and Lgl may regulate polarity by mediating the proper targeting of cargo vesicles. This finding sheds light not only on mechanisms of cell polarization but also on the general role of PDZ proteins in partner localization.

We have recently found that a second set of proteins, which reside in the apical domain of epithelia, act antagonistically to Scrib in polarity determination. These proteins, which include the transmembrane protein Crumbs (Crb) and its PDZ partner Stardust (Sdt), have been previously shown to be required for apical polarity and epithelial cell structure. We find that the epithelial and polarity defects in *crb* and *sdt* mutants can be suppressed by lowering the dose of *scrib* et al., revealing a subordinate system that can regulate apical protein targeting in the absence of Crb and Sdt. This system also appears to be based on PDZ proteins. The emergent picture is of a network of apical and basolateral PDZ proteins that act sequentially, in a finely-tuned balance, to coordinate the maturation of epithelial polarity.

Biochemical and genetic characterization of this network is underway in order to understand in detail the mechanism of PDZ-dependent polarization.

Scrib, Dlg and Lgl are required in not just one specific epithelium but in most epithelial tissues of the fly, as well as in other polarized cell types such as neurons. Scrib plays roles in the assymetric divisions of neuroblasts, and also at the neuromuscular junction (NMJ), where Scrib 'scaffolds' proteins required for synaptic development. In collaboration with Vivian Budnik, we have shown that at the NMJ Scrib and Dlg interact via the 'linker' protein Gukh, and that the three proteins mediate different aspects of synaptic structure. Since vertebrate homologs of Scrib and its partners are also found at both epithelial and synaptic contacts, we believe that Scrib, Dlg, and Lgl represent evolutionarily conserved organizers of cell-cell junctions.

Finally, loss of epithelial structure in *scrib* mutant larval cells is accompanied by severe overproliferation, indicating that *scrib*, like *dlg* and *lgl*, acts as a 'tumor suppressor'. How a mutation in a single fly gene can cause loss of both epithelial organization and growth control similar to that seen in human carcinomas is an intriguing mystery. Because of the correlation between loss of epithelial cytoarchitecture and cancer progression in humans, we are exploring why these mutant fly cells overproliferate, in hopes that these studies may bring a fresh perspective onto studies of human tumorigenesis.

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