Preface

The establishment of planar cell polarity is one of the last frontiers of developmental biology. How individual cells hundreds of cells apart acquire the same polarization within an epithelial field, or how mesenchymal cells establish a uniform polarization prior to their intercalation, is a fascinating biological problem. Much progress has been made in recent years, although the molecular aspects of the process are still far from being understood. Strikingly, the mechanisms appear largely conserved across all metazoa. In this book, we are trying to provide an overview of the current knowledge and features of planar cell polarization (PCP) in different contexts. As this book is being completed several papers have appeared that further extend our understanding of PCP or challenge the views presented here. Thus by no means do we claim this compilation is either complete or fully up-to-date. Nevertheless, we hope it provides a general overview and attempts to cover many different tissues and models of PCP establishment. We apologize to the research areas and view points that have not been included here for whatever reason.

Different aspects of cellular polarization

The establishment and maintenance of cellular polarization is of general importance for the proper development and functioning of most organs and all organisms. Cells in many distinct contexts show different aspects of polarization. Apical-basolateral polarity, for example, is evident in all epithelial cells. Additionally, some epithelial cell types are polarized within the plane of the epithelium, perpendicular to the apical-basal axis. This type of polarity is generally referred to as tissue polarity or planar cell polarity (PCP). PCP has been most studied and is best understood in *Drosophila*, where all adult epithelial cuticular structures show tissue polarity features and are polarized within the plane. Genetic studies in *Drosophila* led to the identification of the first PCP genes some twenty years ago, this first set included also the *frizzled* (*fz*) gene. The study of PCP establishment has recently also been extended to vertebrates, with emphasis on the polarization of the sensory cells in the inner ear epithelium and the morphology and behavior of mesenchymal cells undergoing the morphogenetic process of convergent extension during gastrulation. The combination of the study of PCP establishment in *Drosophila* and vertebrates (in several organisms and tissues) has thus accelerated our insights into and understanding of the process. Nevertheless, much work remains to be accomplished before we can claim of at least partly understanding what is going on.

Whereas apical-basolateral polarity is often established simply through local extracellular clues like cell adhesion properties, PCP establishment requires long range, complex signal propagation in order to ensure that all cells in a tissue are

precisely oriented. It is thought that the interpretation of the polarizing signal is mediated by a core Fz/PCP signaling cassette in all PCP responsive cells. The specific cellular interpretations of this signaling cassette, however, are very diverse, resulting in cytoskeletal rearrangements, changes in cell adhesion properties, reorientation of the mitotic spindle, or other modifications depending on the specific cell type involved. Additionally, PCP signaling effects these changes not only on individual cells, but also on large multicellular units, altering their polarity as a group with respect to the surrounding environment.

Cellular polarization is critical for almost all cell types and is often associated with diseases when disturbed. Epithelial cells and tissues require the apical-basolateral polarity to perform vectorial functions, including the directed transport of fluid and the directed secretion of specialized components. The reported functions of PCP in vertebrates include aspects of body hair orientation and skin development, polarization of the sensory epithelium in the inner ear, and the directed movement of mesenchymal cell populations during gastrulation. Other vertebrate PCP functions can easily be envisioned and/or already have been proposed, including the polarization of cilia in the oviduct and respiratory tract. I will try here to give first a brief account of the historical development of the field, and the follow this with general comments about the state of the PCP field and its many potential impacts on other signaling pathways and/or medical disease states.

The history of the tissue polarity field

The study of PCP originates from work in the fruit fly *Drosophila*, where it was initially called 'tissue polarity'. Elegant work pioneered by the group of Paul Adler has put the problem cleanly on the map some 20 years ago. This was accompanied by the genetic identification of other genes and the emergence of the first models, mostly through work by Paul Adler, David Gubb, and Peter Lawrence and their colleagues. The genetic characterization of *Drosophila* tissue polarity genes was soon followed by the first molecular cloning of a PCP gene in *Drosophila: frizzled*. Again this ground breaking work was performed by Paul Adler and colleagues (see also chapter by Paul Adler in this book). These initial insights were followed up by systematic genetic screens by several research teams in fruit flies and molecular cloning and analysis of several PCP factors. Major advances have thus been made in our understanding of the signaling circuitry and mechanistic aspects of PCP establishment in flies.

The analysis of PCP is now an important feature of developmental studies in many organisms. Starting in Xenopus and zebrafish, with the analysis of the process of convergent extension during gastrulation and neurulation, related processes where discovered and analyzed in vertebrates. Strikingly, the large genome wide forward genetic screens in the zebrafish identified several mutants affecting convergent extension, and following their cloning these turned out to be orthologues of several *Drosophila* PCP genes. Among these are both components of the core primary PCP group, as well as components of a Frizzled PCP specific signaling pathway.

The parallels and conservation of the PCP gene cassette are now extended also to vertebrate epithelia. In particular, the mammalian inner ear is a beautiful example of a neural epithelium with PCP features. Strikingly, the homologues of the fly PCP genes are showing up again when mutants with ear PCP defects are being analyzed (see chapter by Dabdoub et al. in this book). Importantly, some of the vertebrate homologues affect both the inner ear as well as convergent extension during gastrulation and neurulation, supporting the common mechanism of planar polarization not only across species but also between different polarized cell types and organs.

Recent work by Jeremy Nathans and colleagues (Guo, N., Hawkins, C., Nathans, J. 2004. *Frizzled6* controls hair patterning in mice. Proc. Natl. Acad. Sci. USA 101, 9277–9281) closes the circle of PCP features and beautifully demonstrates that the PCP principle can be extended to the mammalian epidermis. The whole field started with the discovery and analysis of *Drosophila frizzled* based on its phenotype in the fly cuticle (generally speaking the cuticle could be considered an equivalent to the mammalian epidermis as it represents the body 'skin') and the work by Guo et al. (2004) now shows that the mammalian epidermis requires also a Frizzled gene for its regular pattern. Even more striking is that the pattern defects in the mouse *Frizzled6* mutant are very much reminiscent of the phenotypic abnormalities of fly *frizzled* mutants. Thus, this is exciting not only for the genetic demonstration of the epidermal PCP features in mammals, but also (1) for the discovery that the same evolutionarily conserved protein family regulates this process from flies to mammals, and (2) that the defects observed in the respective mutants are virtually identical between flies and mammals, suggesting very similar principles at work in all contexts.

Same signaling cassette, but different read-outs

The conserved core PCP gene cassette is conserved. Nevertheless, as it is used in many different contexts in flies or vertebrates, the cellular read-outs are very distinct from tissue to tissue. The effector pathways include cytoskeletal organization, nuclear signaling or orientation of the mitotic spindle. These basic read-outs are again likely conserved from across the animal kingdom.

Cytoskeletal organization is likely the main 'target' of PCP in cuticular cells in Drosophila, the inner ear epithelium in mammals, or the skin in vertebrates (see also chapters by Adler and Dabdoub et al. in this book). A nuclear signaling response is prominent in all multicellular units, e.g., in the *Drosophila* eye (each ommatidium forming a unit, see chapter by Mlodzik in this book) or likely also in feather buds in birds. The PCP regulation of the orientation of mitotic spindles is known in *Drosophila* and *C. elegans*, and likely occurs in other animals as well. In addition, there are PCP controlled processes where the read-out is not yet obvious. These include the cellular behavior and movements during convergent extension in gastrulation and neurulation (see also chapter by Jessen et al. in this book). Although the PCP cassette is clearly required in this context, it is not yet determined what it really regulates in the mesenchymal cells as a cellular read-out. It is likely that other shared PCP read-outs will emerge in the near future.

Establishing signaling 'centers' and long-range PCP patterning

Despite the recent insight into the cellular short range interactions among the core PCP genes, the aspect of long range regulation of Fz-PCP signaling and the establishment of polarizing sources (that could regulate whole fields of cells) remain largely obscure. In particular, the fact that a PCP dedicated Wnt is still elusive in *Drosophila* raises many questions. Although several Wnt family members have been linked to PCP signaling in vertebrates (e.g., see chapter by Jessen and Solnica-Krezel and Dabdoub et al.), no evidence for a PCP specific *Drosophila* Wnt member exists todate. Thus, the long-range regulation of PCP signaling remains largely an unresolved problem.

Nevertheless, some progress has been made in *Drosophila* in this regard. In the fly eye the establishment of a presumed signaling source (the equator) has been quite extensively studied, and suggests that several signaling pathways lead to a restriction of expression of several factors, which could act as potential long range regulators of PCP or at least contribute to the long range regulation of Frizzled-PCP activity (see chapter by Singh et al.). The observation that several factors, implicated in PCP establishment upstream of Frizzled, are expressed in a graded manner within the developing eye disc might provide an insight and explanation into the long range regulation of PCP signaling. Most notably the proto-cadherins Fat and Dachsous, and the transmembrane protein Four-jointed have received attention in this context. Functional analyses have provided support for their general requirement and conserved function as long range regulators of PCP (see chapter by Strutt and Strutt). Interestingly, their graded expression is not restricted to the developing eye but is also observed in other tissues, e.g., during the polarization of the fly wing. Thus, although the jury is still out on this, Fat, Dachsous, and Four-jointed are attractive candidates for being long range PCP regulators, or at least to contribute to the long range regulation in this context.

Multiple Wnt/Frizzled signaling pathways and multiple ways to generate PCP

One striking shared feature of the analysis of Frizzled-PCP signaling is that it utilizes a distinct pathway from canonical Wnt/Frizzled signaling. The Frizzled receptor family has a major function in the reception and transduction of Wnt signals leading to the activation of the β-Catenin signaling pathway, commonly now referred to as canonical Wnt signaling. In contrast, the Wnt/Frizzled-PCP pathway does not affect β-Catenin signaling, and does not share any other components of the canonical pathway downstream of Dishevelled, which appears to be the only cytoplasmic component shared between the PCP and β-Catenin signaling pathways. The analysis and molecular dissection of the signaling specificity is an active area of research and likely to be critical in our understanding of either signaling outcome.

The Wnt/Frizzled signaling complexities do not stop with the PCP and β-Catenin pathways. Recent work has added several other potential effector pathways

downstream of Wnt/Frizzled (see chapter by Pandur in this book). The ongoing analysis suggests an increasingly complex interaction network and potential cross-talk between these distinct effector pathways. The Wnt/Frizzled 'circuitry' is however getting even more complex with the discoveries of new non-Wnt ligands, e.g., Norrin (Xu, Q., Wang, Y., Dabdoub, A., Smallwood, P.M., Williams, J., Woods, C., Kelley, M.W., Jiang, L., Tasman, W., Zhang, K., Nathans, J. 2004. Vascular development in the retina and inner ear: control by Norrin and *Frizzled-4*, a high-affinity ligand-receptor pair. Cell 116, 883–895) and new non-Frizzled Wnt receptors, e.g., Derailed/Ryk (Inoue, T., Oz, H.S., Wiland, D., Gharib, S., Deshpande, R., Hill, R.J., Katz, W.S., Sternberg, P.W., 2004. *C. elegans* LIN-18 is a Ryk ortholog and functions in parallel to LIN-17/*Frizzled* in Wnt signaling. Cell 118, 795–806; and Lu, W., Yamamoto, V., Ortega, B., Baltimore, D. 2004. Mammalian Ryk is a Wnt coreceptor required for stimulation of neurite outgrowth. Cell 119, 97–108). Thus although our understanding of Fz-mediated PCP establishment has increased dramatically warranting several reviews and this book, it appears that we have some far only been scratching the surface of the many complexities to be discovered in this context.

Lastly, although this book focuses on aspects of PCP establishment that are mediated by Frizzled signaling and the associated gene cassette, there is clearly more to planar cell polarization processes and establishment than just this pathway. Recent work in *Drosophila* embryogenesis has nicely shown that a polarization of cells within the embryonic ectodermal cell plane uses molecular mechanisms that do not rely on any of the components of the Frizzled-PCP gene cassette (Zallen, J.A., Wieschaus, E. 2004. Patterned gene expression directs bipolar planar polarity in *Drosophila*. Dev. Cell 6, 343–355; and Bertet, C., Sulak, L., Lecuit, T. 2004. Myosin-dependent junction remodelling controls planar cell intercalation and axis elongation. Nature 429, 667–671). Despite this surprising observation, the cellular aspects of the early cell intercalation during Drosophila germ band extension look very reminiscent of the convergent extension process during vertebrate development, which requires the Fz-PCP signaling pathway.

These observations again show that we know very little about PCP in general and there much more to be discovered in cellular polarization mechanisms. In addition, to trying to understand Frizzled mediated PCP with all its complications, obviously we have to look now also outside the "Frizzled signaling box".

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