Chapter 21. Cell shape, Ap/Ba cellular compartments and morphogenetic field boundaries.

The coupling of the plane of cell division with spindle orientation is dependent on the active transport of Rho GTPase along the mitotic spindle and cortical, membrane-anchored Rho, see above **18**. Similarly, RhoA activity maintains the diffusion barrier between Ap and Ba cellular compartments during interphase via the active transport of RhoGEF2 and the microtubule plus-end tracking protein Eb1 [1] [2]. During embryonic development, the Ap/Ba boundary of individual cells may rotate with respect to the radial embryonic axis, the embryonic surface plane, or any intermediate vector. In general, Ap epithelial surfaces face outwards around the radial (D/V, L/R) and oblate (A/P) embryonic axes, but some Ap surfaces begin to fold inwards during gastrulation. Throughout this process, epithelial axial coupling takes place through an E-Cad/Par-3 (aka Baz) complex at the cortical actin/membrane interface with the extracellular matrix. Constitutive expression of the E-Cad/Par3 complex blocks convergent-extension movements and the mobility of migrating cells [3]. As E-Cad is a major component of AJs, the localisation of Rho1 may maintain the epithelial plane of individual cells during interphase, as well as regulating dynamic cell-shape alterations. In particular, Rho1 activation via RhoGEF2 along the ventral furrow, triggers the localisation of MyoII and E-Cad [4]. Apically localised Rho1 recruits MyoII before the surface contraction of ventral mid-line cells; with contractile filaments tethered to AJs [5] [6]. Notably, the membrane localisation of Rho1 is driven by the (reversible) lipidation of its C-terminal CAAX motif, while RhoGEF2 delivery along microtubules stabilises MyoII accumulation [7].

Thus, during the segmentation cascade epithelial folding and invagination are regulated via asymmetric Par/Cdc42 partitioning. In particular, the cell shape changes at the *runt*-2 and *runt*-3 pair-rule stripes are dependent on Baz and Par-1. In the absence of these functions, pair-rule folds do not form; while increasing the of Baz/Par-1 ratio drives ectopic folding along the *runt*-3 and *runt*-5 stripes [8]. At the epithelial folds, the Ap/Ba cellular compartments, with the associated ring of AJs, are displaced basally, towards the fold, and apically, away from the fold: rotating the centriole anchoring sites prior to the next division. Taken together, these interactions provide a mechanistic link between cell shape changes, matrix anchoring and mitotic spindle alignment. In addition, the infolds may trap accumulated morphogens at a segmental boundary, to form a morphogen source, before endocytotic recycling and lateral translocation. Cytoskeletal remodelling at morphogenetic field boundaries deploys additional interactions between RhoGEF2 and Dsh; linked to NEK2 kinase activity and Wg [9] [10]. The spindle orientation is dependent on the Dsh/Dlg1 interaction; while the transport of Arm (β-catenin) along microtubules is coupled to the Kinesin-II (Klp64D) motor [11] [12], see above, **17**. By implication, field boundary cells require both microtubule and microfilament anchoring through AJs during assembly of the mitotic spindle. Notably, ventral furrow formation and germ band extension require RhoGEF2 activity, without which morphogenetic remodelling is blocked [13]. The linear alignment of the L/R embryonic flanks with the ventral furrow may be transmitted through elongated, oblong cells; with E-Cad concentrated along their D/V boundaries. The rigid, straight interfaces between these cells may be maintained by contractile tension along actomyosin filaments, while their oblong shape may polarise MVBs transmission around the D/V (L/R) axis of the embryo. Similarly, during larval growth, RhoGEF2 is asymmetrically localised to the D/V margin of the wing disc, with rotation around the Ap/Ba epithelial axis regulated by Dpp, the MyoII light chain (Sqh) and Rho1 [14] [15].

Summary:

Dynamic interactions between labile actin and tubulin microfilaments drive cytoplasmic flux and the separation mitotic spindles. A membrane diffusion barrier separates Ap and Ba compartments of epithelial cells during interphase. This Ap/Ba boundary may be displaced by cytoplasmic remodelling, which will rotate the mitotic spindle plane of subsequent divisions. The ventral furrow forms by contraction of the Ap surface of midline cells initiates formation of a furrow, which becomes a L/R AMS. These cell shape changes spread across flanking cells, which elongate along the A/P axis and may transmit a polarised flux of MVBs, and exocytotic vesicles, around the L/R flanks of the embryo. The localisation of E-Cad to the lateral interfaces of these oblong cells allows actomyosin contractions to drive the convergent-extension movements of gastrulation. Meanwhile the A/P embryonic axis is divided by an orthogonal set of parasegmental AMSs, with infolding segmental boundaries. Thus, cytoskeletal remodelling is driven by the asymmetric localisation of RhoGEF2 and the Par-3/Par-1 boundary, which may rotate the Cartesian axial system of cells with respect to the embryonic surface topography.

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