Adhesive Crosstalk in Gastrulation

Recent studies show that signaling through integrin receptors is required for normal cell movements during *Xenopus* gastrulation. Integrins function in this process by modulating the activity of cadherin adhesion molecules within tissues undergoing convergence and extension movements.

During gastrulation, the three germ layers, ectoderm, mesoderm, and endoderm, are formed, and progenitor cells are brought into the positions from which they later form more complex tissues and organs. There is substantial information about the mechanisms that establish cell fates during gastrulation, but much less is known about the molecular and cellular mechanisms underlying gastrulation movements.

The formation and elongation of the embryonic body axis during *Xenopus* gastrulation is triggered by cellular rearrangements commonly termed "convergent extension" (CE) (Wallingford et al., 2002). During CE, cells move from ventral and lateral positions within the gastrula toward the dorsal side (convergence) and, at the same time, redistribute along the anterior-posterior extent of the forming body axis (extension). The elongation and concomitant intercalation of cells along the mediolateral axis of the gastrula is thought to constitute the prime cellular rearrangement underlying CE movements.

Much recent attention has been given to the role of Wnt signals in coordinating gastrulation movements (Wallingford et al., 2002). Downstream of Wnt and its receptor, Frizzled, both canonical and noncanonical Wnt signaling have been implicated in the regulation of mediolateral cell polarization and intercalation in *Xenopus* and zebrafish CE. Although a steadily increasing number of components associated with these pathways are being identified, the mechanisms by which these components control cell morphology and movements are still largely unclear.

Distinct adhesive properties constitute a key feature of cells that undergo gastrulation movements, pointing to a central role of adhesion molecules in the regulation of those movements (Winklbauer et al., 1996). This notion is further supported by studies which demonstrate that interfering with the function of various members of the integrin, (proto)cadherin, and syndecan families of adhesion molecules during gastrulation has profound effects on CE movements (Marsden and DeSimone, 2003, and the references therein). However, the precise contribution of these adhesion molecules and, most importantly, their interaction in regulating CE movements have not yet been determined.

In a recent study published in *Current Biology*, Marsden and DeSimone (2003) start to address this issue. They first show that fibronectin, through its integrin β1 receptor, is needed for mediolateral cell elongation and intercalation underlying CE movements during *Xenopus* gastrulation (see also Figure 1 illustrating the distribution of fibronectin in the early *Xenopus* gastrula). They also demonstrate that C-cadherin functions as a downstream target of integrin β1 signaling in this process. This latter finding is particularly interesting, as it opens up new ways of looking at the interaction of different adhesion molecules in regulating cell movements during gastrulation.

As a first step in describing the nature of the crosstalk between integrin and C-cadherin, Marsden and DeSimone present evidence that integrin-mediated homotypic cell binding in different types of cell sorting and invasion assays is dependent on C-cadherin activity during *Xenopus* gastrulation. These observations indicate that C-cadherin functions downstream of integrin β1 and that integrin β1 is able to modulate the adhesive state of C-cadherin.

Several questions remain about the mechanisms by which integrin signaling modulates C-cadherin activity and about the precise function of C-cadherin in regulating CE movements during *Xenopus* gastrulation. First of all, although it is clear from the study by Marsden and DeSimone that C-cadherin activity can vary in response to integrin signaling, the molecular mechanisms by which this is achieved are still unclear. The authors show that the levels of C-cadherin at the plasma membrane are not affected by integrin signaling, and activation/phosphorylation of focal adhesion kinases (FAK) also does not appear to be involved.

One possible explanation is that integrin regulates the rate of endocytosis and recycling of C-cadherin molecules from the plasma membrane without interfering with the total levels of C-cadherin at the membrane. This model would be in agreement with recent studies showing that clathrin-mediated endocytosis of C-cadherin is needed for proper CE movements during *Xenopus* gastrulation (Jarrett et al., 2002). Alternatively, integrins control the stability of adherens junctions containing C-cadherin by phosphorylating C-cadherin itself or associated components, such as β-catenin (Gumbiner, 2000). For testing these different possibilities, it will be necessary to obtain more information about the phosphorylation levels and subcellular distribution of C-cadherin and other components associated with adherens junctions in the absence or presence of integrin signaling.

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es the molecular mechanisms by which integrin modulates C-cadherin activity, the function of C-cadherin in regulating CE movements during gastrulation also remains to be elucidated. Downregulation of cadherin activity is required for activin-induced elongation of animal cap tissue (Lee and Gumbiner, 1995) but has also been associated with the epithelial to mesenchymal transition (EMT) of mesendodermal progenitor cells during early stages of gastrulation (Hay, 1995). As inactivation of C-cadherin leads to both involution and CE defects during *Xenopus* gastrulation (Lee and Gumbiner, 1995), it is possible that C-cadherin is required for the proper separation of mesodermal from ectodermal cells at the onset of gastrulation and that the defect observed in CE movements is due to an unbalanced adhesion both within and between the different germ layers.

Interestingly, in an earlier paper, Marsden and DeSimone reported that, during *Xenopus* gastrulation, integrin signaling is required for the localization of the intracellular Wnt signal mediator Dishevelled (Dsh) to
the plasma membrane (Marsden and DeSimone, 2001). Dsh localization to the plasma membrane has also been associated with the activation of noncanonical Wnt signaling (Rothbacher et al., 2000), while interfering with noncanonical Wnt signaling through the Wnt receptor Frizzled 7 leads to defects in both cadherin-mediated cell adhesion and separation of the ectodermal and mesodermal germ layers at the onset of Xenopus gastrulation (Winklbauer et al., 2001, and the references therein). It is therefore likely that integrin and noncanonical Wnt signaling cooperate in the regulation of cadherin-mediated cell adhesion, which controls tissue separation and CE movements during gastrulation.

Understanding the crosstalk, not only between different adhesion molecules, but also between secreted signals, such as Wnts and adhesion molecules, will be extremely helpful in elucidating the molecular and cellular mechanisms underlying gastrulation movements. The study by Marsden and DeSimone provides a first step in this direction.

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Selected Reading

Picket and Other Fences in Biological Membranes

Nakada et al. revisit the controversial question of whether membrane lipids are able to diffuse from the axon of a neuron into the soma. Using single molecule imaging of a fluorescent phospholipid, the authors show that a diffusion barrier in the axon initial segment blocks the diffusion of lipids.

Neurons are highly compartmentalized, with distinct domains such as synapses, nodes of Ranvier, axons, and dendrites. The membrane proteins present in these domains are frequently distinct. How is the differential distribution of membrane proteins established and maintained? The initial segment of the axon (AIS) has been reported to act as a diffusion barrier impeding the lateral mobility of several membrane proteins, largely immobilizing them (Winckler et al., 1999). The actin-based membrane skeleton containing special ankyrin G (Kordeli et al., 1995) and spectrin isoforms (Berghs et al., 2000) has been implicated in the integrity of the AIS diffusion barrier based on the effect of actin-disrupting drugs. An ankyrin binding membrane protein and a GPI-linked protein, which does not bind the ankyrin/spectrin network, are both immobilized in the AIS in an actin-dependent manner, suggesting that ankyrin binding proteins enriched in the AIS might act as immobile obstacles that reduce the long-range diffusibility even of proteins not themselves bound to cytoskeletal elements (Winckler et al., 1999).

A recent paper in Nature Cell Biology by Akihiro Kusumi’s group now investigates the diffusion of membrane lipids in the AIS (Nakada et al., 2003). The paper makes two significant contributions. First, stunningly, even the phospholipid phosphatidylethanolamine cannot escape the diffusion barrier and is immobile in the AIS. Second, the progressive immobilization of lipids in the AIS correlates in time and space with the progressive assembly of ankyrin G and the voltage-gated sodium channel (VGNC) in the AIS, and the immobilization of both lipids and VGNCs is actin dependent. Importantly, a diffusion barrier to lipids at the AIS could provide a mechanism for maintaining lipid asymmetry between the axon and the soma.