19 Integrating Cells into Tissues

Review the Concepts

1. The diversity of adhesive molecules has arisen from 1) duplication of a common ancestor gene followed by divergent evolution producing multiple genes encoding related isoforms; and 2) alternative splicing of a single gene to yield many mRNAs, each encoding a distinct isoform.

2. Homophilic interactions are those between like cell types, e.g., epithelial cells with epithelial cells. One approach to demonstrating homophilic cell interactions experimentally is to use L cell lines transfected with E-cadherin and P-cadherin. L cells adhere poorly to each other and express no cadherins. When transfected with E- and P-cadherin, L cells adhere tightly to E-cadherin-positive cells and P-cadherin-positive cells, respectively. Cadherins directly cause homotypic interactions among cells.

3. Actin and myosin filaments form a circumferential belt in a complex with adherins junctions. This belt functions as a tension cable that can internally brace the cell and control its shape.

4. Tight junctions help to hold cells together in tissues and control the flow of solutes between cells in an epithelial sheet. Several things can happen to cells when tight junctions do not function. In hereditary hypomagnesemia, defects in tight junctions prevent the normal flow of magnesium through tight junctions in the kidney. Low blood-magnesium levels result and this can lead to convulsions. Altering tight junctions in hair cells of the cochlea of the inner ear can result in deafness.

5. Collagen is a major component of the extracellular matrix in animal cells. It is a protein that has a trimeric structure with rodlike and globular domains that form a two-dimensional network. Collagen is synthesized in its precursor form by ribosomes attached to the endoplasmic reticulum. These pro-α chains undergo a series of covalent modifications and are folded into a triple helical procollagen molecule. The folded procollagen is transported through the Golgi and the chains are secreted to the outside of the cell. Once outside the cell, peptidases cleave the N- and C-terminal propeptides. The triple helices are then able to form larger structures called collagen fibrils.

6. Structural studies have shown that integrin exists in both a non-active, low-affinity or "bent" form and an active, high affinity or "straight" form. In outside-in signaling, molecules of the ECM can bind to the extracellular portion of inactive integrin and induce conformational changes that lead to the straightening of the intracellular, cytoplasmic tails of integrin. The straightening of the cytoplasmic tails can stimulate intracellular components such as the cytoskeleton and parts of signaling pathways. This structure also facilitates inside-out signaling. For example, when the metabolic state of the cell is altered, adapter proteins inside the cell can interact with the cytoplasmic tails of integrin and cause straightening from the inside. This would result in new ECM interactions on the outside of the cell.

7. *Proteoglycans* are highly viscous glycoproteins that cushion cells and bind to a wide variety of extracellular molecules. *Collagen* is fibrous and provides structural integrity, mechanical strength, and resilience. Soluble extracellular matrix proteins such as *laminin* and *fibronectin* bind and cross-link cell-surface receptors and othr ECM components.

8. Sydecans in the hypothalmic region of the brain participate in the binding of antisatiety peptides to cell surface receptors. In the "fed" state, the extracellular domain of sydecans is released from

the surface by proteolysis. When this happens, the activity of antisatiety peptides is suppressed along with feeding behavior.

9. The RGD sequence on fibronectin mediates binding to integrin proteins. If RGD-containing peptides were added to a layer of fibroblasts grown on a fibronectin substrate in tissue culture, the RGD peptides would compete with fibronectin for binding to the integrins present in the fibroblast extracellular matrix. As a result, the fibroblasts would likely lose adherence to the fibronectin substrate.

10. Fibronectin contains fibrin-binding domains. Since fibrin is a major constituent of blood clots, this domain allows fibronectin to recruit blood clots.

11. The dystrophin gene, which is defective in Duchenne muscular dystrophy, is an adapter protein that binds to cytoskeletal components such as actin and to the cell-adhesion molecule dystroglycan. Normally, dystrophin and dystroglycan function in an important part of the signaling relay linking the extracellular matrix on the outside of the muscle cell to the cytoskeleton and signaling components inside the muscle cell. When any of these components is defective, the muscle cells do not develop or function properly and muscular dystrophy results.

12. Inflammatory signals including chemokines are released in the area of infection. These signals activate the endothelial cells lining blood vessels in the area. P-selectin exposed on the surface of activated endothelial cells mediates weak adhesion of passing leukocytes. Weakly bound leukocytes roll along the surface of the endothelium.

    At the same time, chemokines and other signaling molecules including the platelet activating factor (PAF) also activate β2-containing integrins on the cell surface of the leukocytes. Upon activation, the integrins change conformation in to their high-affinity form. Activated integrins bind to IgCAMs on the surface of endothelial cells. Tightly bound leukocytes stop rolling, spread out on the surface of the endothelium, and eventually crawl between adjacent endothelial cells into the underlying tissue

13. Small molecule hormones, called auxins, induce the weakening of the cell wall. This permits expansion of the intracellular vacuole by uptake of water, leading to cell elongation.

14. Both plasmodesmota and gap junctions are channels that directly connect the cytosol of one cell to that of an adjacent cell. However, in plasmodesmota, the plasma membranes of the adjacent cells are merged to form a continuous channel, the annulus. Membranes of the cells at a gap junction are not continuous. Plasmodesmota may also contain an extension of the endoplasmic reticulum, the desmotube, that passes through the annulus. Animal cells do not contain desmotubes.

Analyze the Data

a. Cells transfected with wild-type E-cadherin aggregate more than untransfected cells because the increase in E-cadherin allows for more cell-cell interactions via the ECM.

b. Mutant A behaves almost identically to the wild-type E-cadherin in the aggregation assay; therefore, this mutation does not change the function of E-cadherin as far as homophilic interactions are concerned. Expression of mutant B, however, does not result in increased aggregation, so this particular mutation does alter the adhesive qualities of E-cadherin.

c. The monoclonal antibody specific for E-cadherin blocks the resulting aggregation because it binds to E-cadherin and blocks homophilic interactions. In contrast, the nonspecific antibody does not specifically interact with E-cadherin, and does not block homophilic interactions.

d. Since cadherins require calcium for function, lowering the calcium in the media during the assay would lower the aggregation ability.