In This Issue

A synaptic scaffold quiets T cells

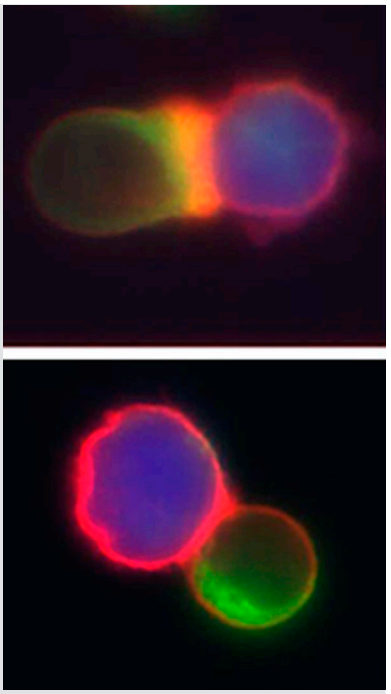
A scaffolding protein at neuronal synapses has found its way to another kind of synapse—the immune synapse, which forms at the interface between antigen presenting cells and T cells. As shown by Xavier et al. on page 173, this multidomain protein, called Dlg1, tempers the activity of both types of synapses.

On the T cell side of the immune synapse, activated T cell antigen receptors (TCRs) congregate with many cytoskeletal and signaling molecules that spread the word of antigen recognition to the rest of the cell. Recognition of TCR engagement must be transmitted somehow to these other proteins to coordinate their synaptic congregation.

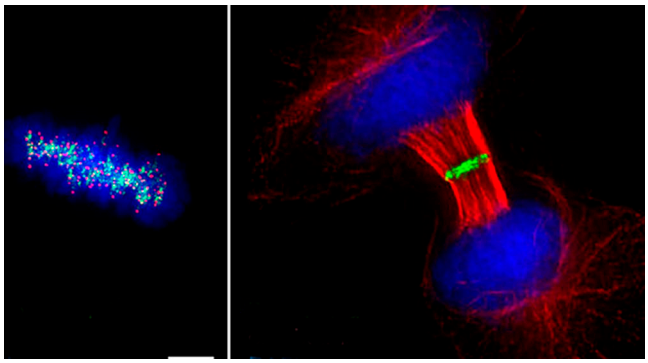
Based on its multidomain structure and role as a scaffold in neuronal synapses and epithelial junctions, Dlg1 seemed a likely candidate for this coordination job in T cells. The authors' localization data support this idea—Dlg1 moves from the cytosol to cortical actin at the immune synapse within five minutes of antigen recognition. There, Dlg is complexed with proteins needed for T cell activation, including a Src kinase, a TCR subunit, and the Cbl ubiquitin ligase, suggesting that it might bring these and perhaps other signaling proteins to the synapse.

Once the complex has assembled, however, Dlg1's job seems to be to prevent overactivity of T cells, which could potentially cause autoimmune problems. Dlg1 overexpression limited T cell activation, whereas loss of Dlg1 caused an overactive response to antigen.

In flies, Dlg1-like proteins both bring AMPA receptors to synapses and promote their degradation, which requires AMPA ubiquitination. Dlg1 might similarly cause immune receptor down-regulation, possibly via its interaction with Cbl. However, as Dlg1 left synapses within 15 min of TCR engagement, it might instead down-regulate T cell signaling by taking some other synaptic components with it. ■



Dlg1 (green) is at the immune synapse within 5 min of T cell activation (top), but leaves by 20 min (bottom).



Borealin (green) moves from metaphase chromosomes (left) to the spindle midzone at telophase (right).

Another passenger on chromosomes

Chromosomes are getting crowded. On page 179, Gassmann et al. present their identification of a fourth chromosomal passenger protein. As part of the Aurora B kinase complex, this new protein, Borealin, helps stabilize the mitotic spindle. In a paper soon to appear in *Cell*, Sampath et al. identify what seems to be the same protein.

Chromosomal passenger proteins, which until now included only Aurora B, INCENP, and Survivin, have several functions during mitosis. As mitosis begins, they are dispersed along chromosomes and phosphorylate histone H3. By metaphase, they gather at centromeres, where they are needed for kinetochore function and to correct spindle attachment errors. Later they move to the spindle midzone and the plasma membrane for cytokinesis.

The identification of Borealin in human cells reveals that not all these jobs are done by the same complex. Borealin was found in a complex containing all three other passengers. Some Aurora B, however, associated with INCENP but not Borealin or Survivin. The smaller complex probably phosphorylates histone H3, as loss of Borealin did not affect this Aurora B function.

As is common for passenger proteins, Borealin localization depended on its partners, INCENP and Survivin. Expression of Borealin fragments, in turn, perturbed the localization of other passengers to the centromeres, but did not affect spindle midzone targeting. Different subcomplexes with or without Borealin may thus bring Aurora B to the appropriate places for its various functions.

Cells that were depleted of Borealin, and that therefore mislocalized the other passengers, were delayed in prometaphase and unable to correct errors in spindle attachments to the kinetochore. Many of these cells nonetheless built normal-looking bipolar spindles but, once anaphase began, formed ectopic asters that caused the chromosomes to segregate in several directions. The extra asters seem to derive from the poles, but the reason for their generation remains unclear. This is the first hint that the chromosomal passengers are involved in spindle assembly and function. ■

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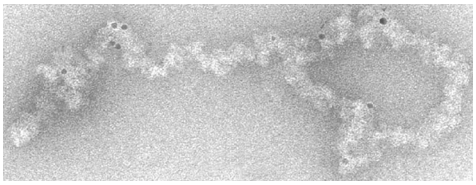
Loopy for telomeres

On page 161, Nikitina and Woodcock present the first isolation of telomeres complete with their chromatin protein complement.

Telomeres protect chromosome ends from degradation and shortening, but they must themselves be protected from repair systems that recognize free DNA ends. Images of telomere DNA, lacking its chromatin components, suggested that its protection might be enhanced by looping and insertion of the telomere's single-stranded overhang into the double-stranded telomeric repeats. Wrapping this loop up in chromatin proteins probably hides the overhang from agents that detect free ends, but because telomere chromatin is difficult to extract from the nuclear matrix, whether chromatin looping occurs was unknown.



TRF1-labeled chromatin reveals that telomeres form loops.

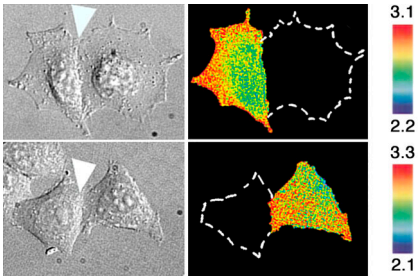
The authors have conquered the chromatin extraction problem by using two types of blood cells that have fewer proteins gluing the chromatin to the matrix. After digesting away non-telomeric DNA, the authors labeled the remainder with biotinylated TRF1, a telomere-binding protein, thus identifying the telomeric chromatin. Like naked telomeric DNA, many of the TRF1-labeled structures formed loops.

As expected, the chromatin loop is smaller than that of the naked DNA due to nucleosomal packing. Nucleosomal repeat length is shorter on telomeres than on other DNA, but the authors did not note any changes in fiber morphology resulting from the difference. Future 3-D reconstructions will address this problem. An up-close look is also needed to determine how the overhang associates with the double-stranded DNA and how the nucleosome arrangement allows the loop to open and close during replication. ■

Integrins talk to N-cadherin

On page 283, Yano et al. show how some cells use signals from cell-matrix contacts to maintain cell–cell contacts as they migrate.

Although most cells travel in isolation, some epithelial cell types migrate as a sheet, as during gastrulation or in carcinoma metastasis. Yano et al. find that HeLa cells (derived from a carcinoma) need the focal adhesion proteins Fak and paxillin to maintain their N-cadherin–based cell contacts.



The down-regulation (top) of Rac1 activity (colored) at cell contact sites is lost if Fak is inhibited (bottom).

When integrins at focal adhesions bind to the matrix, paxillin joins the complex. The new results suggest that paxillin recruits Fak to focal adhesions. Then, activated Fak somehow down-regulates only the Rac1 at sites of cell contact, although the direct Fak substrates are not yet known. When either paxillin or Fak was inhibited, the resulting Rac1 activity caused large overactive membrane protrusions and reduced the number of N-cadherin–based cell–cell adhesions. The mechanism is unclear, but perhaps the strong membrane motion caused by Rac1 makes cadherin bond formation unlikely.

Fak inhibition also increased migration rates and disrupted wound healing because cells broke away from the sheet. The studies were done using cancer cells, and Fak inhibition has the opposite effect on fibroblast migration, so the authors next need to see if their studies extend to normal epithelial cells. ■

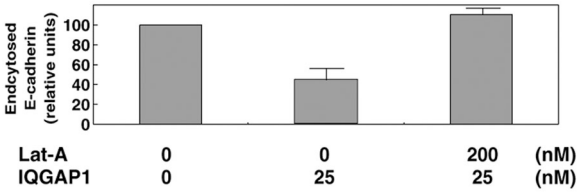
E-cadherin evades endocytosis

E-cadherin interacts with other E-cadherin molecules on neighboring cells to form cell–cell adhesions. E-cadherin that is not involved in these trans interactions is removed from the cell surface and replaced with newly synthesized molecules to maintain dynamic adhesions. Now, Izumi et al. (page 237) show that E-cadherins that are involved in trans interactions are excused from this endocytosis by small GTPases. Disruption of this system may free cells for migration.

By reconstituting endocytosis in membrane bilayers, the group shows that clathrin-dependent endocytosis removes E-cadherin that is not interacting in trans with other E-cadherins. E-cadherins engaged in trans interactions, however, activated Rac and Cdc42, which blocked their internalization. So far it is unclear how the trans interactions activate the G proteins.

The endocytic block is enhanced by IQGAP1, an effector of Rac and Cdc42. IQGAP1 cross-links actin filaments into bundles, and the group shows that F-actin is needed to inhibit endocytosis. The bundles might press up against the bilayer, thus preventing the membrane invagination needed for vesicle budding.

Because the activation of Rac strengthens cell–cell interactions, the authors plan to determine whether down-regulation of Rac at adhesion sites—and thus reactivation of endocytosis—is essential for HGF-induced cell dissociation. ■



E-cadherin endocytosis is blocked by IQGAP1 unless actin filaments are disrupted by Lat-A.