**In This Issue**

**Joined at the Hip**

A direct link between clathrin cages and actin is described on page 1209 by Engqvist-Goldstein et al.

The protein that forms the link is huntingtin interacting protein 1 related (Hip1R), which was characterized previously as an actin-binding protein that associates with clathrin-coated pits and vesicles. The related protein Hip1 has been implicated in Huntington’s disease, where loss of an interaction between normal huntingtin protein and Hip1 may cause problems with membrane–cytoskeletal integrity in the brain.

Engqvist-Goldstein et al. now show that Hip1R tightly associates with clathrin during endocytosis and that purified Hip1R both binds to purified clathrin cages and stimulates cage assembly in vitro. They also show that clathrin cages assembled in vitro with Hip1R copellet with F-actin at low speed.

Other workers have induced the formation of clathrin coated pits using only clathrin, two coat proteins, and membranes. Actin and Hip1R may be needed, however, for the pits to mature into vesicles. The importance of the actin interaction can now be tested in cell culture systems.

**Cooperation in the cerebellum**

By crossing knockout mice deficient for two cell adhesion molecules, Sakurai et al. have obtained the first genetic evidence for the molecules’ functional overlap, and have explained why earlier tissue culture results were not replicated in single knockout mice (page 1259).

Sakurai and colleagues started by engineering mice deficient for NrCAM, which had only mild (~11%) growth defects in two cerebellar lobes. When the authors crossed the NrCAM-deficient mice with existing mice that are deficient for the related cell adhesion molecule L1, the cerebellum of the double knockout was drastically reduced in size, and the mice were small and never survived later than eight days after birth. The mice probably die because their lack of coordination does not allow them to compete successfully for food.

The cerebellar defect may be largely a result of reduced migration or decreased survival of granule cells. In the double knockout, these interneurons initially differentiate and start migrating correctly, but they reach their final destination in vastly reduced numbers. There is no accumulation of these cells in other areas of the cerebellum, suggesting a survival problem.

The survival hypothesis would be consistent with tissue culture experiments, in which Sakurai et al. added L1 antibodies to cells isolated from NrCAM-deficient mice, and observed a dramatic reduction in viability at around day 14 of culture. More experiments along these lines should reveal the specific functions of L1, NrCAM, and the two other related CAMs (neurofascin and CHL1) in processes such as cell migration, axon bundling, and synaptogenesis.

**Sticky utrophin messages**

On page 1173, Gramolini et al. report that utrophin mRNA is immobilized by binding to an actin-dependent structure. Manipulation of this system may be important for the therapy of Duchenne muscular dystrophy (DMD).

DMD is characterized by a weakened linkage from muscle cells to the extracellular matrix because of a compromised dystrophin complex. Urophin may be able to substitute for dystrophin, but utrophin’s expression is normally limited to the areas near where nerves contact muscle cells. This is due, at least in part, to the concentration of utrophin mRNA to these regions.

In multinucleated muscle cells, the utrophin gene is preferentially expressed from nuclei that are close to the sites of nerve contact. Gramolini et al. now find that this high localized concentration of mRNA is then maintained by the binding of utrophin polysomes, via the utrophin 3′ untranslated region, to actin-dependent structures.

The possible relevance of these results to DMD therapy arises because individuals with DMD may mount an immune response to dystrophin introduced by gene therapy, given that the individual’s body has never seen the protein before. Although increased expression of utrophin should not cause such an autoimmune reaction, the localization of utrophin translation may limit utrophin’s usefulness. If, however, the tethering system described by Gramolini et al. can be manipulated with small molecules, the dispersed expression of utrophin may act as one element of a DMD therapy.
Mitosis without wind

Kinesin motor called Kid that sticks to chromosome arms and is thought to mediate chromosome movement out along spindle microtubules. This polar ejection force has been proposed as a mechanism for pushing chromosomes away from spindle poles and thus aligning chromosomes on the metaphase plate.

The importance of Kid and the polar ejection force has been shown in fly germ cells mutant for the Nod kinesin, and in frog extracts, where depletion of Kid resulted in widespread wandering of chromosomes away from in vitro assembled metaphase plates. Levesque and Compton set out to see if Kid was equally important in a mitotic rather than meiotic system.

After injection of Kid antibodies into cells with monopolar spindles, chromosomes clustered tightly around the pole, whereas after control injections chromosomes kept a polite distance from the pole because of polar ejection forces. When the authors inhibited Kid in cells with bipolar spindles, most chromosomes moved to the metaphase plate correctly. The chromosome arms trailed toward the poles, but nevertheless, the chromosomes moved in the right direction, and only 17.5% of cells had chromosome disorganization sufficient to cause a noticeable cell cycle delay. Delayed cells tended to have one or more sister chromatid pairs hung up very close to one pole.

The relatively small numbers of problems suggests that mitotic kinetochores can usually be captured even by microtubules from a very distant pole, and once captured are smart enough to sense the midpoint of the spindle without the help of polar ejection forces. The kinetochore microtubules, like a rope in a tug-of-war, may be subject to forces that are proportional to the length of the microtubule, so that the opposing forces will cancel out when a chromosome reaches the metaphase plate.

From nuclear pore to kinetochore

Nuclear proteins are turning up in some odd places lately. First, the nuclear-transport factor Ran was implicated in spindle formation. Then, the mitotic checkpoint proteins Mad1 and Mad2 turned up at the nuclear pore, in a switch with the mRNA export factor hRae1, which appeared with the mitotic checkpoint protein mBUB1 at the kinetochore. Now, Belgareh et al. report that two structural nuclear pore complex constituents also localize to the kinetochore (page 1147).

Belgareh et al. were interested in characterizing the human version of a budding yeast nuclear pore subcomplex that is involved in mRNA export. They identified hNup133 by homology, and identified three other members of the subcomplex through immunoprecipitation experiments. The surprise came in the localization studies, when both antibody staining and GFP fluorescence indicated that at least two of these nucleoporins are found at kinetochores from prophase through to late anaphase.

This simple observation has many possible interpretations, none of which (as yet) comes with any significant supporting evidence. First, nucleoporins at the kinetochore could act to seed the formation of a specific subset of nuclear pores at the end of mitosis, while the nuclear envelope reforms around chromosomes. Second, the nucleoporins could have some unspecified function at the kinetochore, although a role in spindle formation is not obvious as none of the nucleoporins under discussion has the FG repeats characteristic of proteins that link indirectly to Ran.

Finally, the nucleoporins may visit the kinetochore merely to pick up mitotic checkpoint proteins, so that those checkpoint proteins can be sequestered at the nuclear pores during the following cell cycle. Why that might be necessary is anyone’s guess.