Hyperdynamic proteins keep stem cells on their toes

Chromatin has a staid and static image. But Eran Meshorer, Tom Misteli (National Cancer Institute, Bethesda, MD), and colleagues suggest that ES cells are kept pluripotent thanks to hyperactively mobile chromatin proteins.

Chromatin proteins provide architectural integrity to DNA. Despite their structural role, they do not stay statically bound but release and reform their bonds continuously. Using FRAP to measure protein dynamics, the group showed that chromatin proteins in differentiated cells are exchanged within minutes to hours, whereas ES cells contain a pool of such proteins that turns over at the rate of seconds. The fast-moving protein fraction is present in several types of pluripotent cells but strikingly absent in committed precursor cells. This quick exchange may be what keeps the genome breathing—open and ready to take on any fate.

Frenetic protein activity also seems to be necessary for differentiation. When chromatin proteins were mutated to bind more tightly to DNA, cells failed to differentiate, whereas mutations increasing the pool of loose proteins led to faster than normal differentiation. The team postulates that in ES cells the proteins may be acting as building blocks where regions of silent (nontranscribing) chromatin are formed as the cell differentiates and shuts down unused areas of the genome.

Misteli hints that chromatin protein dynamics is probably just the tip of the iceberg in the search for what distinguishes pluripotent cells from committed ones, and says the group is looking for other properties of chromatin, such as histone modifications, gene activity, and chromatin structure that may differ in ES cells. They are also examining the underlying question of what the mobility might mean for gene expression. “If [ES] chromatin is really more open,” he says, “there might be more transcription.” JCB

Reference: Meshorer, E., et al. 2006. Dev. Cell. 10:105–116.

Making HoxD waves

When developmental biologists first got their hands on DNA, perhaps the most remarkable finding was that the order of genes on the chromosome reflected the genes’ time and location of expression. Genes at the tail end of each cluster were expressed proximally and early; those at the front of the cluster were expressed distally and later. Now, Basile Tarchini and Denis Duboule (University of Geneva, Switzerland) find that simple placement of two gene regulatory elements explains the entire expression sequence for the HoxD cluster, which patterns forelimb development.

Two waves of HoxD expression control limb formation in vertebrates: an early wave, which generates proximal structures such as the forearm, and a later wave, which forms distal structures such as digits. By breeding mouse strains with targeted meiotic recombinations, Duboule and colleagues created deletions and reevaluations of the HoxD genes, and they looked for changes in regulation.

Earlier work by the group showed that the later wave of digit development is controlled by a positive regulatory element located outside the HoxD cluster. Analysis of the 19 strains of mice generated for this study identified a different mechanism for the forelimb, in which gene expression is regulated by a positive and a negative regulator, each located on either side of the cluster. The asymmetry in HoxD expression is thus simply a factor of a gene’s relative distance from the positive or negative influence, and the balance between the two forces.

The group is now applying the same technique to tease out the trunk patterning mechanism, a more complicated endeavor because many more genes are involved. “But we have good evidence that it’s quite close to the one that’s organizing the proximal limbs,” he says.

If the similarity holds, it would support the idea that proximal limb structures are phylogenetically much older ones. Duboule proposes that when proximal limbs evolved, nature co-opted the similar trunk patterning mechanism to also regulate limb development. But digit development, which is thought to have occurred about 300 million years ago, required nature to come up with a new approach. JCB

Reference: Tarchini, B., and D. Duboule. 2005. Dev. Cell. 10:93–103.
Two-speed cell specification

N euron subtypes are specified through diversity: each subtype gets its own transcription factor. But in the lateral glia of Drosophila, specification of all glial cell types is controlled by a single fate-determining gene called Glide/Gcm (gcm).

How can just one gene make many glial cell types? Rossana De Iaco, Angela Giangrande (National Center for Scientific Research in Strasbourg, France), and colleagues provide the solution for one type of glial cell. They show that although low expression of the gcm gene make cells competent to become glia, a boost in expression level regulates the type of glia that cells will become.

Glia fail to form in mutant embryos lacking gcm. In mutants lacking a patterning protein called Huckebein (Hkb), one type of glia—the product of one specific neuroglioblast lineage—fails to form. Following this lineage under different conditions of gcm and hkb expression, the researchers found that Hkb binds to the Gcm protein to up-regulate gcm expression. Hkb thus provides a molecular link between gcm’s general role in specifying all glia and its subdividing role in specifying different glial cell types.

Although Hkb itself can be seen as a lineage-specific factor, the fact that it acts by altering levels of a cell fate determinant is unusual. Giangrande speculates that such quantitative regulation of cell types might operate in animals besides flies.

Reference: De Iaco, R., et al. 2005. EMBO J. doi:10.1038/sj.emboj.7600907.

E. coli squeezed into action

E. coli is always on the look-out for a better environment. As it swims, chemoattractant receptors talk to the flagellar motors, thus orienting the bacterium’s travels. Using fluorescence polarization to image the receptors’ position in living cells, Vaknin and Berg found that increased osmolarity caused receptors, joined in triplets like the legs of a tripod, to move closer together by about 10%. The squeeze stimulates kinase activity and the subsequent signaling pathway, prompting the bacterium to swim away from the potentially damaging environment.

This compression can be explained by simple cell membrane dynamics. As osmotic stress increases, water leaves the cell. Reduced pressure from within causes a slackness in the membrane and an increase in its thickness—much as a rubber balloon acts as some air is let out. “We think that when the membrane thickens in response to osmotic stress, that changes the orientation of the receptors, making them move closer together,” says Berg.

How changes in relative receptor position stimulate kinase activity is still unknown. The group is now investigating whether chemoattractant stimuli cause receptors to move further apart. They are also looking downstream at the effect of such mechanical perturbations on the flagellar motor’s control of direction.

Reference: Vaknin, M., and H.C. Berg. 2005. Proc. Natl. Acad. Sci. USA. doi:10.1073/pnas.0510047103.

Aging on the clock

T he same genes that regulate timed events during development also regulate aging and lifespan in C. elegans, according to Michelle Boehm and Frank Slack (Yale University, New Haven, CT).

The lin-4 microRNA is required for the correct timing of cell fate specification at larval stage L2. It acts by blocking the expression of lin-14, a putative transcription factor. But both genes are also expressed in the adult. The Yale duo engineered temperature-sensitive mutations that allowed expression changes after the proteins had functioned in development. Mutants with decreased lin-4 activity lived half a normal lifespan, whereas lin-14 mutants lived 31% longer than normal. This biological clock seems to regulate adult lifespan in the same way it regulates development: via lin-4 repression of lin-14.

Further experiments suggested that lin-4 and lin-14 exert their effect on lifespan by linking into a well-characterized insulin-like signaling pathway. This pathway, regulated by the protein Daf-2, combats aging by fighting damage from heat-shock and oxidative stress. Both lin-4 and lin-14 are widely conserved genes, so the Yale group is examining whether the murine homologues also control aging.

The function of lin-14—and whether this function is the same in both development and aging—is not known. But Slack notes that the involvement of heterochronic genes in adulthood is not surprising, as many events in adulthood occur in a timed manner, including reproduction, signs of aging, and death.

Reference: Boehm, M., and F. Slack. 2005. Science. 310:1954–1957.