lysosomes to be degraded and recycled. However, cultured cells components are engulfed by autophagosomes and delivered to thereby up-regulates the autophagy pathway, in which cellular A lack of amino acids also inactivates the mTOR kinase and production of proteins that synthesize or import free amino acids. The GAAC pathway therefore prevents cells from degrading nutrient deprivation in a number of different ways. Low amino acid levels up-regulate the general amino acid control (GAAC) pathway, in which the transcription factor ATF4 induces the production of proteins that synthesize or import free amino acids. A lack of amino acids also inactivates the mTOR kinase and thereby up-regulates the autophagy pathway, in which cellular components are engulfed by autophagosomes and delivered to lysosomes to be degraded and recycled. However, cultured cells larvae lacking the only somatic SUN protein, UNC-84. Surprisingly, the absence of UNC-84 had no effect on nuclear envelope spacing in most worm tissues. In the body wall muscle cells of unc-84 mutants, however, the nuclear membranes became widely separated in regions predicted to undergo mechanical strain. Cain et al. therefore think that LINC complexes are only required to hold nuclear membranes together in cells subjected to high mechanical stress. Consistent with this, HeLa cells experience intracellular tension when plated onto tissue culture dishes.

Cain et al. wondered whether the size of LINC complexes nevertheless sets the width of the perinuclear space. An UNC-84 mutant lacking most of its luminal domain formed functional LINC complexes, and, even though these complexes are predicted to be much shorter, the nuclear membranes of worms expressing this mutant were still spaced 50 nm apart. Senior author Daniel Starr thinks that inherent properties of the nuclear membranes (and the contiguous ER) might instead determine nuclear envelope spacing. Cain, N.E., et al. 2014. J. Cell Biol. http://dx.doi.org/10.1083/jcb.201405081.

Deubiquitination helps Rad18 grow more tolerant

Zeman et al. describe how deubiquitination of the ubiquitin ligase Rad18 fine-tunes the cell’s response to specific types of DNA damage. 

Rad18 helps the cell’s DNA replication machinery “tolerate” DNA damage and duplicate the genome with a minimal number of mutations. The ubiquitin ligase induces slightly different tolerance pathways in response to different types of DNA lesions, and it may even promote mutagenesis if activated in the absence of any damage at all. Zeman et al. therefore investigated how Rad18 is regulated.

Around a quarter of a cell’s Rad18 molecules are usually ubiquitinated, probably by the enzyme itself. Rad18 was deubiquitinated, however, in response to the DNA alkylating agent MMS or H2O2 but not in response to other insults such as UV irradiation. MMS and H2O2 promote Rad18’s interaction with another ubiquitin ligase, SHPRH, that is crucial for the cell’s ability to tolerate these sources of DNA damage. Accordingly, ubiquitinated Rad18 didn’t bind SHPRH and instead preferred to dimerize with other, nonubiquitinated Rad18 molecules, potentially sequestering them in an inactive state. Ubiquitinated Rad18 failed to localize to sites of DNA damage and was unable to ubiquitinate its downstream targets. Cells overexpressing a constitutively ubiquitinated form of Rad18 were less able to tolerate MMS and thus showed an increased rate of mutagenesis.

Ubiquitination therefore inhibits Rad18’s activity in undamaged cells, but this posttranslational modification is quickly removed in response to MMS or H2O2, allowing the ubiquitin ligase to bind SHPRH and promote tolerance of the ensuing DNA damage. The authors now want to investigate how Rad18 is deubiquitinated in response to these DNA damage agents. Zeman, M.K., et al. 2014. J. Cell Biol. http://dx.doi.org/10.1083/jcb.201311063.