Mdm1 helps the ER and vacuole stay in touch

Mdm1 (green) localizes to the sites where vacuoles (V) contact the ER (red) surrounding nuclei (N).

Henne et al. reveal that a protein linked to human neurological disease regulates sphingolipid metabolism in yeast by tethering vacuoles to the ER.

Little is known about yeast Mdm1, but it is homologous to several mammalian sorting nexins, including SNX14, which is mutated in an autosomal-recessive form of cerebellar ataxia. Henne et al. identified Mdm1 in a screen for proteins that regulate endocytic trafficking to the yeast lysosome, or vacuole.

Surprisingly, however, Mdm1 didn’t localize to endosomes. Instead, the protein accumulated at the sites where vacuoles contact the ER membranes surrounding the nucleus. Overexpressing Mdm1 enlarged these nuclear ER–vacuole junctions (NVJs), suggesting that the protein helps tether these two organelles to each other.

miR-7 loss is hard to stomach

Compared with control cells (left), miR-7 overexpression (right) reduces the expression of IKKε (red), decreasing the amount of active RELA (green) in the nucleus.

Zhao et al. reveal that the microRNA miR-7 suppresses gastric cancer by inhibiting NF-κB signaling, and that this protective mechanism is compromised by the cancer-causing bacterium Helicobacter pylori.

miR-7 is frequently down-regulated in gastric cancers and can suppress gastric cell metastasis by inhibiting the growth factor receptor IGF1R. Whether miR-7 also suppresses earlier stages of gastric carcinogenesis is unknown, however, so Zhao et al. screened for new targets of the microRNA.

The researchers found that miR-7 directly targets RELA and FOS, which encode transcription factors involved in the pro-oncogenic NF-κB and AP-1 signaling pathways, respectively.

Gauging Gag assembly

The concentration and diffusion coefficient of cytoplasmic Gag (green) can be measured by fluorescence correlation spectroscopy.

Hendrix et al. use fluorescence fluctuation imaging to reveal that HIV-1 particles start assembling in the cytoplasm of infected cells.

The HIV-1 polyprotein Gag contains three different domains that promote viral assembly: the membrane-binding matrix domain, the RNA-binding nucleocapsid domain, and the capsid domain that mediates Gag oligomerization. Gag assembles into new viral particles at specialized regions of the plasma membrane, but whether the protein only oligomerizes after it is recruited to these sites, or whether it begins to assemble while still in the cytoplasm, remains unclear.

Hendrix et al. used a series of fluorescence fluctuation imaging techniques to identify two populations of Gag molecule diffusing in the cytosol: a faster-moving monomeric form and a slowly moving oligomeric species. Even the Gag monomers diffused more slowly than most proteins of a similar size, a property the researchers mainly attributed to the nucleocapsid domain’s transient interactions with cytoplasmic RNAs.

Gag oligomerization depended on the protein’s concentration in the cytosol and was promoted by both the capsid domain’s self-association and the nucleocapsid domain’s interaction with RNA. A small number of Gag molecules therefore assemble on viral RNAs in the cytoplasm. Senior author Don Lamb now wants to test the idea that these small oligomers then associate with the plasma membrane and nucleate the assembly of viral particles by recruiting additional Gag monomers.

Hendrix, J., et al. 2015. J. Cell Biol. http://dx.doi.org/10.1083/jcb.201504006

In human gastric cancer samples, low miR-7 levels correlated with elevated RELA and FOS expression and poor patient survival. Overexpressing miR-7 reduced RELA and FOS levels and inhibited both gastric cell proliferation and tumor growth in vivo.

Zhao et al. found that, as well as directly suppressing RELA expression, miR-7 reduced the transcription factor’s activity by targeting its upstream kinase IKKε. Yet IKKε and RELA were themselves able to repress miR-7 transcription, forming a feedback loop between the NF-κB and miR-7 pathways.

Chronic H. pylori infection is a major risk factor for gastric cancer, in part because the bacterium can hyperactivate the NF-κB pathway. Co-culturing H. pylori with gastric epithelial cells induced the expression of IKKε and RELA, and down-regulated the expression of miR-7, a potentially key step in gastric cell transformation. Senior author Dai-Ming Fan now wants to identify drugs capable of inducing miR-7 and suppressing tumorigenesis.

Zhao, X.-D., et al. 2015. J. Cell Biol. http://dx.doi.org/10.1083/jcb.201501073

Indeed, Henne et al. found that two N-terminal transmembrane domains anchored Mdm1 in the ER, while a C-terminal PX domain bound to phospholipids on the vacuole surface. A truncated version of Mdm1 that, similar to a disease-causing SNX14 allele, lacked the PX domain failed to localize to NVJs.

NVJs have previously been implicated in lipid metabolism, and the cells of patients with SNX14 mutations appear to accumulate lipids in cytoplasmic granules. Henne et al. found that yeast expressing the truncated version of Mdm1 were hypersensitive to an inhibitor of sphingolipid synthesis, suggesting they were somehow defective in sphingolipid metabolism. Because sphingolipids are required for efficient endocytosis, this could explain why endosomal trafficking is delayed in mdm1Δ yeast. Lead author Mike Henne now wants to investigate how defects in vacuole–ER contacts and sphingolipid metabolism contribute to the symptoms of humans with SNX14 mutations.

Henne, W.M., et al. 2015. J. Cell Biol. http://dx.doi.org/10.1083/jcb.201503088