Lysosome ‘talks’ to mitochondria using vitamin B12 in *C. elegans*

Preview on “Lysosomal activity regulates Caenorhabditis elegans mitochondrial dynamics through vitamin B12 metabolism”

By Tao Chen, Ahmet Yavuz, Qian Zhao

Introduction

Mitochondria plays a central role in modulating physiology and metabolism in eukaryotic cells, by carrying out energy production as well as signal transduction. Interestingly, mitochondria do not function as static double-membrane bound cellular compartments. On the contrary, they are constantly in the transition states of fusion and fission that are tightly regulated by several GTPase. Mitochondria fission is mediated by dynamin-related protein 1(DRP1), while mitochondria fusion requires optic atrophy 1(EAT-3 in *C.elegans*) and mitofusin 1/2 (FZO-1 in *C.elegans*) for outer membrane fusion and inner membrane fusion, respectively. Disruption of the balance of mitochondria dynamics has been linked to a number of diseases, however, the intricate regulatory mechanism of mitochondria dynamics largely remains mysterious.

Despite being an organelle of structural confinement, mitochondria do not only perform stand-alone function but also constantly interact with other organelles. In a very recent study, physical contact between lysosome and mitochondria has been observed in healthy untreated Hela cells using electron microscopy, structured illumination microscopy and high spatial and temporal resolution confocal live cell imaging. The contact formation and subsequent untethering was regulated by a small GTPase Rab7 which also regulated lysosome dynamics. When the Rab7 GTPase-activating protein TBC1D15 was recruited to mitochondria by FIS1 to accelerate Rab GTP hydrolysis, the contact was released. The discovery that mitochondria-lysosome contacts marks sites of mitochondrial fission allowing lysosomal Rab7 to regulate mitochondrial dynamics, conversely Rab7 hydrolysis is regulated by mitochondrial TBC1D15, provide a mechanism for bidirectional regulation of mitochondria and lysosome dynamics(Wong, Ysselstein et al. 2018).

In addition to direct physical contact, the crosstalk between lysosome and mitochondria also happens on a more complicated and intertwined level. In this study, Wei et al show that *Caenorhabditis elegans* lysosomal activity regulates mitochondrial fission through the metabolism vitamin B12. This provides an insight on the communication between mitochondrial dynamics and lysosomal degradation.

Results

The authors started from searching for genes that can mitigate mitochondrial fission defects when inactivated. They took advantage of the *dhc-1* mutant, which has similar mitochondrial dynamic phenotype as *drp*-1. From a pool of 49 genes already identified in *dhc-1* RNAi screening, spe-5/vacuolar ATPase turned to increase the viability of *drp-1* for 5 folds through inactivation. To validate the result, the authors deployed several characterizations, including mitochondrial morphology by periodic pattern (peaks), max length, and tubular percentage. Since mitochondrial dynamic abnormality can lead to several cellular responses, including mitochondrial unfolded protein response (mtUPR), it is adopted for characterization as well. These characterizations were used throughout the paper.

Since *spe-5* encodes a subunit of v-ATPase, they then checked other subunits of v-ATPase, and found the inactivation of them can lead to improvement on viability, implying the lysosome activity involved in mitochondrial dynamics. Furthermore, two drugs, bafilomycinA1 (BafA1) and concanamycin A (CMA), were applied to the worms to inhibit v-ATPase activity, showing effect on viability improvement on *drp-1*. These data lead to investigation on lysosome biogenesis and activity to affect mitochondria dynamics. A master lysosome biogenesis regulator, HLH-30, and the two major lysosomal membrane proteins, LMP-1 and LMP-2 were chosen to evaluate this purpose. The inactivation of *hlh-30*, *lmp-1* and *lmp-2* through RNAi all mitigated the mitochondrial fission defects at *drp-1* background. Supporting results were shown in mitochondria morphology, membrane potential and homeostasis (indicated by *hsp-6* and *hsp-60* expression). Meanwhile, the inhibition of lysosome activity, which can affect mitophagy, showed little effect on *drp*-1 fission defects. When inactivating v-ATPase subunits, lysosome biogenesis and membrane protein genes in wild type background, more fragmented mitochondria were observed. But in *eat-3*, the fragmentation effect was slightly stronger than the others, indicating another pathway involved.

Food source was then investigated. Comparison between the two bacteria strains, OP50 and HT115 showed that OP50 fed worms had less fission defects in *drp-1*. Hence, vitamin B12 was found as one of the major molecular differences between these two food sources. A cellular B12 deficiency reporter P*acdh-1*::GFP showed significantly higher signal in OP50 fed worms than HT115 fed worms, confirming the lack of B12 in OP50 as a food source. Then the correlation between lysosome and vitamin B12 was investigated, since lysosome mediated the cellular uptake of B12. The RNAi on v-ATPase subunits or inhibition of lysosome function can both promote P*acdh1*::GFP signal, supporting that insufficient B12 releasing from lysosome may cause cellular B12 deficiency. Further investigation on enzymes that B12 serves as cofactor confirmed the inactivation *metr-1* can lead to B12 deficiency. Finally, mitochondrial biogenesis was found to be enhanced with lysosomal dysfunction.

Perspectives

This paper shows the link between mitochondrial dynamics and micronutrient metabolism through lysosomal dysfunction and vitamin B12 metabolism. The lysosome plays an important role in vitamin B12 intake. The paper shows that reducing lysosomal activity causes vitamin B12

levels to go down and in turn reducing methionine synthase (MTR) activity, one of two enzymes that requires vitamin B12 as cofactor. The reduction in MTR output affects overall methionine levels and low levels of methionine affect mitochondria biogenesis through yet to be identified mechanism. Previously, Revtovich et al. shown that vitamin B12 levels affects mitochondrial homeostasis through the other gene that uses B12 as cofactor methylmalonyl-CoA mutase gene (MUT) suggesting vitamin B12 playing important role in mitochondria health (Revtovich et al.,2019) .

References:

Revtovich AV, Lee R, Kirienko NV (2019) Interplay between mitochondria and diet mediates pathogen and stress resistance in Caenorhabditis elegans. PLoS Genet 15(3): e1008011

Wong, Y. C., D. Ysselstein and D. Krainc (2018). "Mitochondria-lysosome contacts regulate mitochondrial fission via RAB7 GTP hydrolysis." Nature 554(7692): 382-386.