Uncovering the principles coordinating systems-level organelle biogenesis with cellular growth

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***Short Abstract* — Deciphering the principles by which the eukaryotic cell coordinates growth across its multiple scales of organization is a frontier question in quantitative cell biology. Here we aim to map out the correlation structure of systems-level organelle biogenesis with cellular growth. To this end, we generated "rainbow yeast", a strain of *Saccharomyces cerevisiae* that allows simultaneous visualization of 6 major organelles. By carrying out hyperspectral imaging of rainbow yeast, we suggest that cellular growth excites specific systems-level organelle biogenesis modes that characterize the response to changes in nutrient availability. Contrary to expectations derived from scaling relationships relating organelle size to cell size, our findings suggest the following design rule for the eukaryotic cell: systems-level organelle biogenesis is driven by cellular growth rate rather than cell size.**

***Keywords* — systems cell biology, organelles**

I. INTRODUCTION

The principles by which cells coordinate organelle growth with overall cellular growth have been examined primarily through scaling relationships of organelle sizes with the sizes of their host cells[1]. It is widely appreciated, for example, that the volume of the nucleus scales linearly with the size of its host cell. Similar scaling relationships have been documented for the endoplasmic reticulum, vacuoles/lysosomes, and mitochondria. However our knowledge of similar relationships even at the level of pairs of organelles is sparse, let alone multibody interactions from strong organelle interconnectivity that are presumed to be important for regulating cellular scale physiology [2]. We reasoned that a combination of systems-scale organelle imaging, chemical biology tools, and simple data reduction methods would yield deep insights into how organelle biogenesis couples to the interrelated variables of cell growth rate and cell size.

II. METHODS

A. Strategy to dissect systems-level organelle biogenesis program

In order to visualize the 6 major organelles in budding yeast, we transcriptionally fused genes encoding fluorescent proteins to genes encoding organelle-resident proteins known with high confidence to localize to either the mitochondria, endoplasmic reticulum, Golgi apparatus, vacuole/lysosome, peroxisome, or lipid droplet. We then applied a hyperspectral imaging strategy [3] with machine learning based image analysis tools [4,5] to characterize the organelle profile of thousands of single cells.

We proceeded to use this tool to dissect the contributions of cell size and cell growth to the regulation of systems-level organelle biogenesis.

B. Response of systems-level organelle biogenesis to cell size versus cell growth rate

To characterize the response of systems-level organelle biogenesis to natural inputs driving changes in cellular growth, we examined rainbow yeast cultured in defined amounts of the sugar glucose. For each glucose concentration the cells were cultured in, we profiled the cellular organelle composition. We then applied principal components analysis to these organelle profiles to uncover collective organelle modes responsive to input glucose concentrations.

We proceeded to profile the organelle biogenesis patterns in rainbow yeast cells grown with specific perturbations to their growth rates and their cell sizes and thereby uncover the organelle modes excited by these perturbations.

C. Orchestration of systems-level organelle biogenesis by key growth signaling pathways

To gain mechanistic insight into how growth rate drives systems-wide organelle biogenesis, we examined the response to variations in the main growth rate regulating signal transduction pathways in yeast: the PKA and TOR pathways. Using small molecule inhibitors, we tuned the activity levels of these pathways, profiled the systems-level organelle biogenesis response, and compared the organelle modes excited by PKA and TOR signaling to those excited by glucose, cell size, and cell growth rate.

III. CONCLUSION

The geometry of organelle modes excited by changes in input glucose concentrations were most co-linear with those modes excited by variations in cell growth rate and orthogonal to modes excited by variations in cell size. We suggest, therefore, that systems-level organelle biogenesis responds to signals from growth rate rather than cell size.

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