Increased heme synthesis in yeast induces a metabolic switch from fermentation to respiration even under conditions of glucose repression

Tiantian Zhang, Pengli Bu, Joey Zeng, and Ales Vancura

From the Department of Biological Sciences, St. John’s University, Queens, New York 11439

Edited by Joel Gottesfeld

Regulation of mitochondrial biogenesis and respiration is a complex process that involves several signaling pathways and transcription factors as well as communication between the nuclear and mitochondrial genomes. Under aerobic conditions, the budding yeast Saccharomyces cerevisiae metabolizes glucose predominantly by glycolysis and fermentation. We have recently shown that altered chromatin structure in yeast induces respiration by a mechanism that requires transport and metabolism of pyruvate in mitochondria. However, how pyruvate controls the transcriptional responses underlying the metabolic switch from fermentation to respiration is unknown. Here, we report that this pyruvate effect involves heme. We found that heme induces transcription of HAP4, the transcriptional activation subunit of the Hap2/3/4/5p complex, required for growth on nonfermentable carbon sources, in a Hap1p- and Hap2/3/4/5p-dependent manner. Increasing cellular heme levels by inactivating ROX1, which encodes a repressor of many hypoxic genes, or by overexpressing HEM3 or HEM12 induced respiration and elevated ATP levels. Increased heme synthesis, even under conditions of glucose repression, activated Hap1p and the Hap2/3/4/5p complex and induced transcription of HAP4 and genes required for the tricarboxylic acid (TCA) cycle, electron transport chain, and oxidative phosphorylation, leading to a switch from fermentation to respiration. Conversely, inhibiting metabolic flux into the TCA cycle reduced cellular heme levels and HAP4 transcription. Together, our results indicate that the glucose-mediated repression of respiration in budding yeast is at least partly due to the low cellular heme level.

The yeast Saccharomyces cerevisiae, even under aerobic conditions, metabolizes glucose predominantly by glycolysis and fermentation, producing ethanol and carbon dioxide (1–4). This metabolic specialty of the budding yeast is partly due to the high activity of pyruvate decarboxylase Pdc1p (5, 6), which leads to cytoplasmic conversion of pyruvate to acetaldehyde and subsequently to ethanol. Because the majority of the glycolytically produced pyruvate in glucose-grown cells is converted to acetaldehyde, only a small fraction of pyruvate is translocated into mitochondria and used for the tricarboxylic acid (TCA) cycle (7–9) (see Fig. 1). Under these conditions of low metabolic flux into the TCA cycle, the expression of genes encoding enzymes of the TCA cycle, electron transport chain (ETC), and oxidative phosphorylation (OXPHOS) is low. After glucose is exhausted during diauxic shift, yeast cells activate TCA cycle, ETC, and OXPHOS genes and switch metabolism from fermentation to respiration, utilizing the produced ethanol as a carbon source (3, 10–12).

The transition from fermentation to respiration is controlled by PKA, target of rapamycin (TOR), Sch9p, Snf1p, and Mec1p/Rad53p signaling pathways and requires several transcription factors, including Hap1p, Hap2/3/4/5p, and Rtg1/3p (10, 13–19). However, despite the central position of glycolysis, the TCA cycle, ETC, and OXPHOS in cell metabolism and physiology, the signaling mechanisms and transcriptional regulation by which these pathways are coordinated and aligned with nutritional conditions are not well understood.

The expression of TCA cycle, ETC, and OXPHOS genes is regulated by the heterotetrameric transcription complex Hap2/3/4/5p (heme-activated protein (HAP) complex) independently of PKA and SNF1, suggesting that the HAP complex provides an additional, separate mechanism of transcriptional regulation of mitochondrial respiration (4, 20) (Fig. 1). The HAP complex binds to DNA through the Hap2, -3, and -5 subunits, which are constitutively expressed (21). The activation domain of the complex is contained within the Hap4p subunit (22). HAP4 expression increases upon glucose depletion, and overexpression of HAP4 induces respiration even in a glucose-repressed state, resulting in an extension of replicative life span (23–26). The HAP complex is activated by heme and is required for growth on non-fermentable carbon sources (27, 28). The human homolog of the HAP complex is the heterotrimeric NF-Y complex (29). Interestingly, the NF-Y complex is also regulated by heme (30).

Heme stimulates the transcriptional activity of Hap1p, and as such, Hap1p acts as the key sensor of heme (20, 31). Heme synthesis in yeast is regulated by the availability of oxygen, and heme serves as an intermediate in the signaling mechanism for

This work was supported by National Institutes of Health Grant GM120710 (to A.V.). The authors declare that they have no conflicts of interest with the contents of this article. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

1 To whom correspondence should be addressed: Dept. of Biological Sciences, St. John’s University, 8000 Utopia Parkway, Queens, NY 11439. Tel.: 718-990-1679; Fax: 718-990-5958; E-mail: vancuraa@stjohns.edu.

2 The abbreviations used are: TCA, tricarboxylic acid; ETC, electron transport chain; OXPHOS, oxidative phosphorylation; HAP, heme-activated protein; ALA, 5-aminolevulinic acid; NF-Y, nuclear transcription factor Y; YPD, yeast extract-peptone-dextrose.