The transcription factors Swi4 and Hap4 contribute to the regulation of the transcriptional response to cold shock in Saccharomyces cerevisiae.

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Recommended Citation
Hong, Monica V.; Wyllie, Kevin W.; McGee, Kevin P.; and Dahlquist, Kam D., "The transcription factors Swi4 and Hap4 contribute to the regulation of the transcriptional response to cold shock in Saccharomyces cerevisiae." (2016). Grants & Fellowships. Paper 2.  
http://digitalcommons.lmu.edu/honors-grants-and-fellowships/2
Abstract Title: The transcription factors Swi4 and Hap4 contribute to the regulation of the transcriptional response to cold shock in *Saccharomyces cerevisiae*.

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Abstract: The effects of cold shock on *Saccharomyces cerevisiae* have not been extensively studied. Cold shock treatment causes changes in gene expression, but which transcription factors regulate this response is still unknown. The purpose of this study was to determine how the transcription factors Swi4 and Hap4 regulate the transcriptional response to cold shock in yeast. Yeast cells deleted for each transcription factor were subjected to cold shock at 13 degrees Celsius, followed by recovery at 30 degrees Celsius. Samples were collected before cold shock (t0), after 30 and 60 minutes of cold shock (t30, t60), and after 30 and 60 minutes (t90, t120) of recovery. Then, total RNA from these cell samples was isolated and aRNA was synthesized and indirectly labeled with Cy3 and Cy5 dye, followed by hybridization to DNA microarrays. Four replicates of this experiment were performed for each deletion strain, and microarray analysis was performed on all replicates, swapping the Cy3 and Cy5 dyes for two replicates of each strain to minimize error due to discrepancies in degradation rates of the dyes. An ANOVA test performed on the ∆swi4 strain showed that 2233 out of 6189 (36%) genes saw a significance change in expression, using a significance criteria of a Benjamini-Hochberg adjusted P value lower than 0.05. For the ∆swi4 strain, the two most significant expression profiles were up-regulation followed by down-regulation, and down-regulation followed by up-regulation, including gene ontological categories such as ribosome biogenesis and glycogen metabolic processing, respectively. Using the same significance criteria, the ANOVA test performed on ∆hap4 showed significant expression changes for 1749 genes (28%). ∆hap4’s two most significant expression profiles were up-regulation followed by a steeper, over-compensatory down-regulation which then went back to baseline expression, and a brief period of down-regulation which also returned to baseline expression. Examples of gene ontologies in included in these two profiles are carbohydrate transport, and apoptotic processes, respectively. Additionally, we successfully genotyped six strains deleted for the transcription factors Nrg1, Phd1, Rsf2, Rtg3, Yhp1 and Yox1 using both colony PCR and DNA sequencing. Then we investigated the effects of temperature stress on these six strains to determine which of them were impaired for growth at 15°C, 20°C, 30°C and 37°C. We discovered that the ∆phd1 strain was impaired for growth at all temperatures, while the ∆nrg1 strain was impaired at 30°C and 37°C, but grew more quickly than the wild-type at 15°C and 20°C. While the microarray results for ∆swi4 and ∆hap4 suggest an involvement of their respective transcription factors in *S. cerevisiae*’s response to cold shock, the ∆phd1 and ∆nrg1 growth plate results suggest that these strains may be worthy of future investigation. In addition, experimentation with another transcription factor, Yap1, has begun, with three replicates remaining for microarray analysis.
**Registration:** $20

**Conference:** Experimental Biology 2016, San Diego. April 2\textsuperscript{nd}-6\textsuperscript{th}.
   San Diego Convention Center
   111 West Harbor Drive, San Diego, CA 92101

**Meals:** $120 ($15 for lunch per day and $25 for dinner per day for 3 days)

**Hotel:** Sommerset Suites Hotel.
   606 Washington Street, San Diego, CA 92103.
   For two nights (Saturday/Sunday) the total is $322.75.