Post-transcriptional regulation after stress in *Schizosaccharomyces pombe*

By Daniel Nilsson

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This thesis is based on following papers:

I: **Fission yeast mitogen-activated protein kinase sty1 interacts with translation factors**. Eva Asp[1], Daniel Nilsson[1] and Per Sunnerhagen.
Eukaryotic Cell 7:328-338 (2008)
[1]Shared first authorship

II **Cellular stress induces cytoplasmic RNA granules in fission yeast**
Daniel Nilsson and Per Sunnerhagen
RNA 17:120-133 (2011)

III **Impact of oxidative stress and the MAP kinase Sty1 on mRNA stability in *S. pombe*.** Eva Asp, Rebecka Jörnsten, Daniel Nilsson, Alexandra Jauhiainen, Olle Nerman, and Per Sunnerhagen.
Manuscript (2011)

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ABSTRACT

Post transcriptional regulation is part of the gene expression control and is important for many cellular processes. It influences how mRNAs are selected for translation, degradation or storage. In this thesis, I describe some of the known mechanisms for transcriptional regulation in *S. pombe* including MAP kinase (MAPK) signaling, translation and mRNA localization to cytoplasmic RNA granules. The MAPK Sty1 in *S. pombe* is activated in response to a wide range of stresses and regulates transcription as well as translation. In a screen for novel interaction partners to Sty1 we identify translation factors eEF2 and eIF3a. The Sty1-eIF3a interaction weakened upon stress treatment but Sty1-eEF2 remained unchanged. Translation initiation is impaired in response to stress in *sty1* cells and the Atf1 transcription factor, which is a known target for Sty1 contributes to translation recovery after osmotic stress but in no other stress investigated. Under conditions of nitrogen limitation we found that both interactions with eEF2 and eIF3a disappeared and that eIF3a is degraded at a time point correlating with the time of translation re-initiation. Both phosphorylation and protein levels of eIF3a in *sty1* cells were reduced. *S. pombe* forms cytoplasmic granules in response to stress positive for the RNA-binding and translation proteins Csx1, Dcp2, eIF4G, eIF3a, Pabp (polyA-binding protein), and mRNA. Pabp and Dcp2 almost exclusively co-localize after glucose starvation but not after osmotic stress. Ca^{2+} perturbations affect the formation of granules after glucose starvation and the Ca^{2+} chelator EGTA alone induced granules. Pathway regulating granules are under control of eIF2a and Protein kinase A (Pka1). eIF2α is not a requirement for granule formation but appear to be important for the disaggregation of granules after osmotic stress and EGTA but not after glucose starvation. *pka1* cells were unable to form Pabp positive granules after glucose starvation and EGTA. Ribosomes in *pka1* cells failed to fully dissociate in response to glucose starvation. In a whole genome mRNA stability analysis we find that mRNAs that are transcriptionally upregulated also become stabilized in the early response to oxidative stress and that this is largely Sty1 dependent.

Keywords: *S. pombe*, MAPK, stress, translation, RNA granules, Sty1, Pabp, mRNA stability
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