## Supplemental Materials

Molecular Biology of the Cell
Basch et al.

## Supplementary Figure 1

Msp1 expression levels from various plasmids. The strains were grown in glucose-containing medium at $30^{\circ} \mathrm{C}$. Whole cell extracts were analyzed for levels of Msp1. Pgk1 was decorated as loading control.

## Supplementary Figure 2

UPR ${ }^{m t}$ induction in wild type and an alternative mspn-1 mutant. Wild type (+/+) and mspn1(tm3831) animals carrying the $\mathrm{P}_{\text {hsp-6 }}$ GFP transcriptional reporter (strains MD4433 and MD4431, respectively) were analyzed by brightfield and fluorescence microscopy. The $\mathrm{P}_{\text {hsp- }}$ ${ }_{6}$ GFP fluorescence intensities were quantified. Error bars indicate standard deviations. The difference between the two strains is statistically significant by unpaired $t$ test with Welch's correction ( $\mathrm{p}=0.0002$; $\mathrm{n}=6$ ).

## Supplementary Figure 3

A) Msp1-dependent binding of the proteasome to isolated mitochondria. Mitochondria were isolated from a W303 and $\Delta m s p 1$ strain and subsequently subjected to sucrose gradient purification. Gradient purified mitochondria were analyzed by SDS-PAGE, Western blot and immuno decoration with an antibody raised against the whole purified proteasome.
B) Rpn10-dependent chase of arrested precursor from isolated mitochondria. Recombinant Cytb2- $\Delta$ TM-DHFR was pre-folded in the presence of methotrexate and bound to mitochondria of the indicated strains. After indicated time points mitochondria were re-isolated, washed and analyzed by SDS-PAGE, Western blot and immuno decoration and quantified.





