FIGURE S1.—Overproduction of chaperone proteins in yeast cells was confirmed by Western blotting analysis. Strain BY241 [ure-o], bearing either control vector or high copy plasmid with a chaperone gene, was grown in liquid SD supplemented with required amino acids. Yeast cells were collected and lysates were prepared and equalized by the protein amount. Levels of Ssa1p, Ydj1p, Sse1p, Sis1p and Hsp104p were compared by immunoblotting with corresponding antibodies. The level of actin served as a loading control. Antibody to Ssa's detects Ssa1p, Ssa2p, Ssa3p and Ssa4p, with Ssa2p being the major cytoplasmic Hsp70 in unstressed cells. Thus Ssa1p is actually elevated more than one would judge by the small increase in the intensity of the band in the upper left panel.