Quantitation of the ribosomal protein autoregulatory network using mass spectrometry

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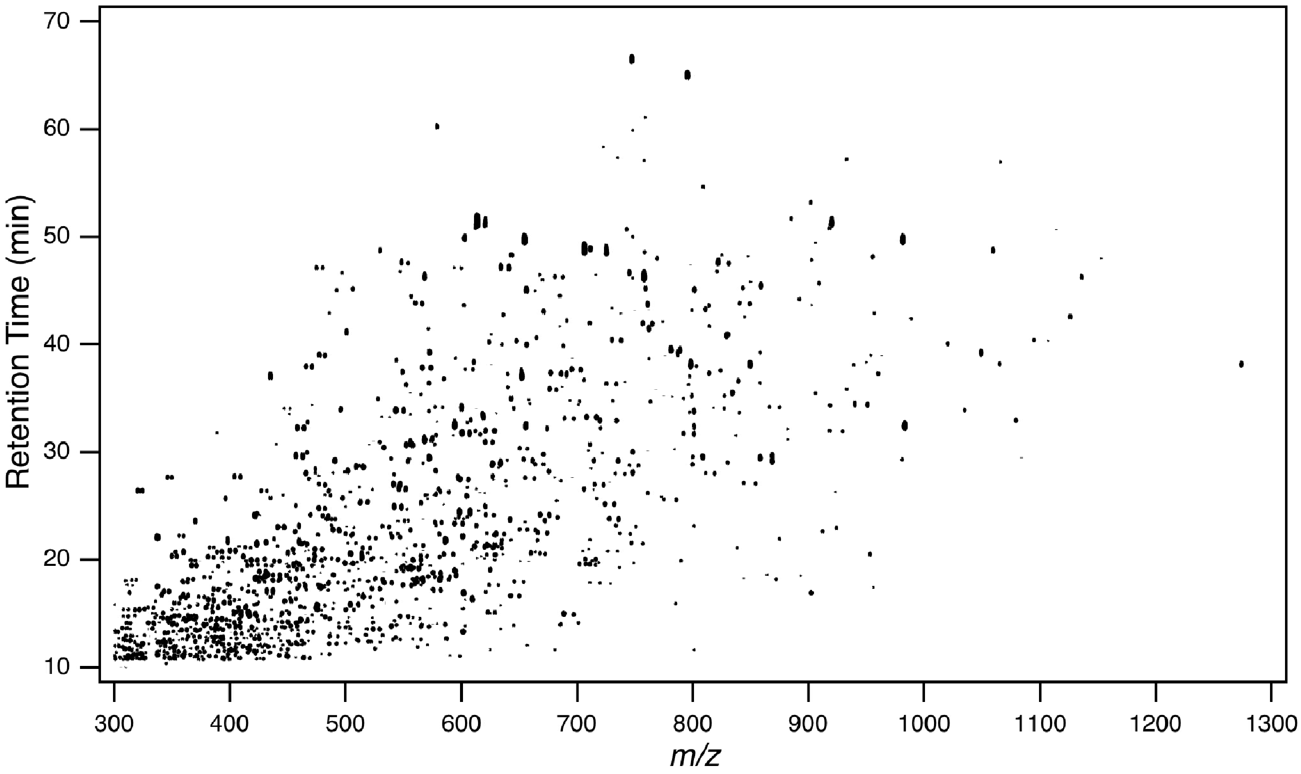
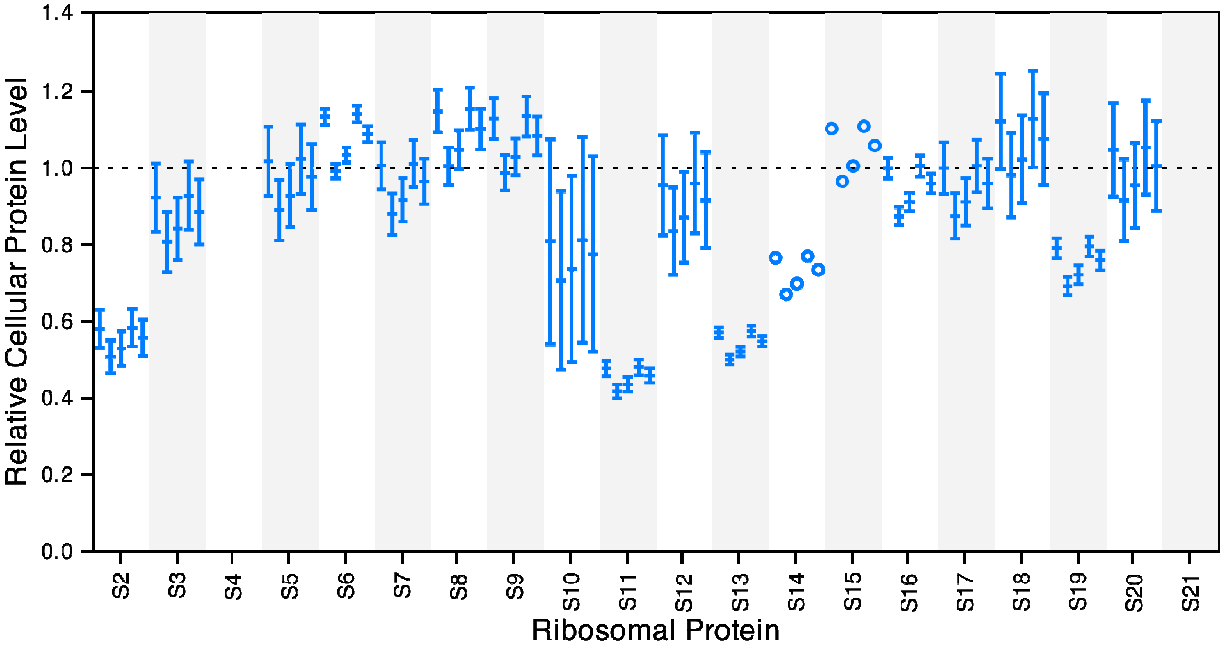
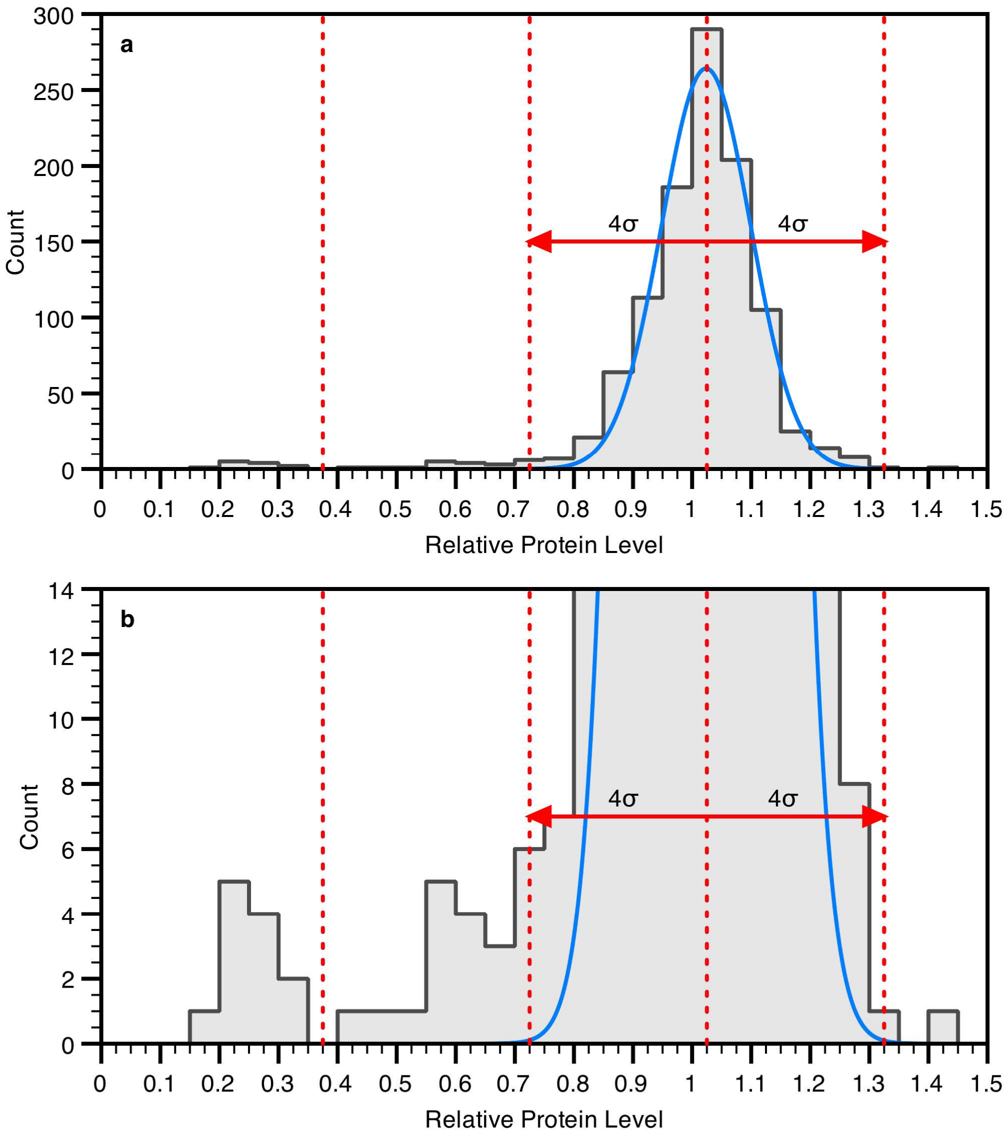
Supporting Information



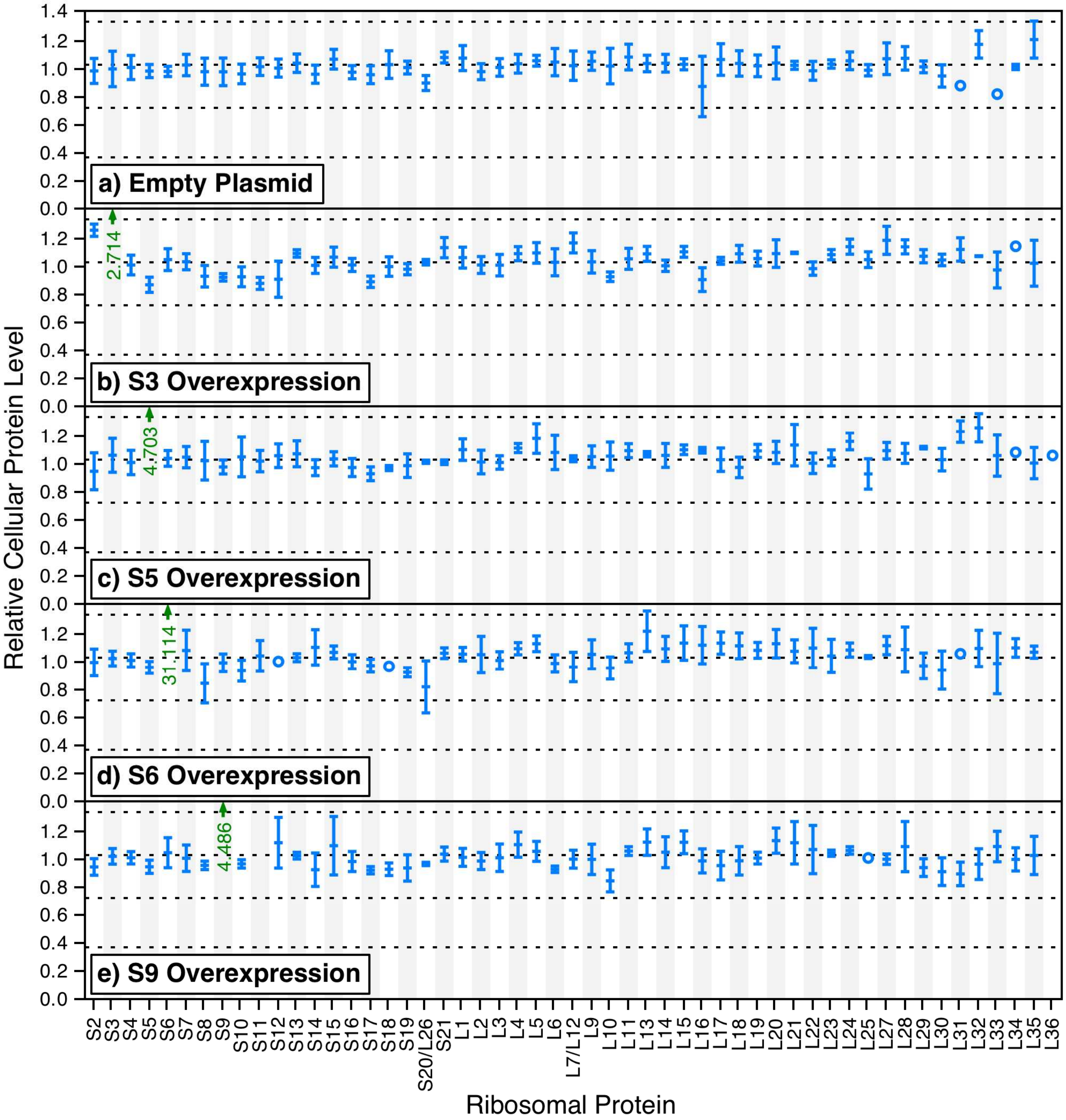
Supplementary Figure S1: A low-resolution contour plot of the mass spectrum of crude *E. coli* cell lysate spiked with ^{15}N-labeled ribosomes. The isotopic envelope consisting of the monoisotopic peak and isotopomers is not resolved in this representation. Pairs of peaks represent unlabeled and ^{15}N-labeled pairs of peptides from ribosomal proteins.



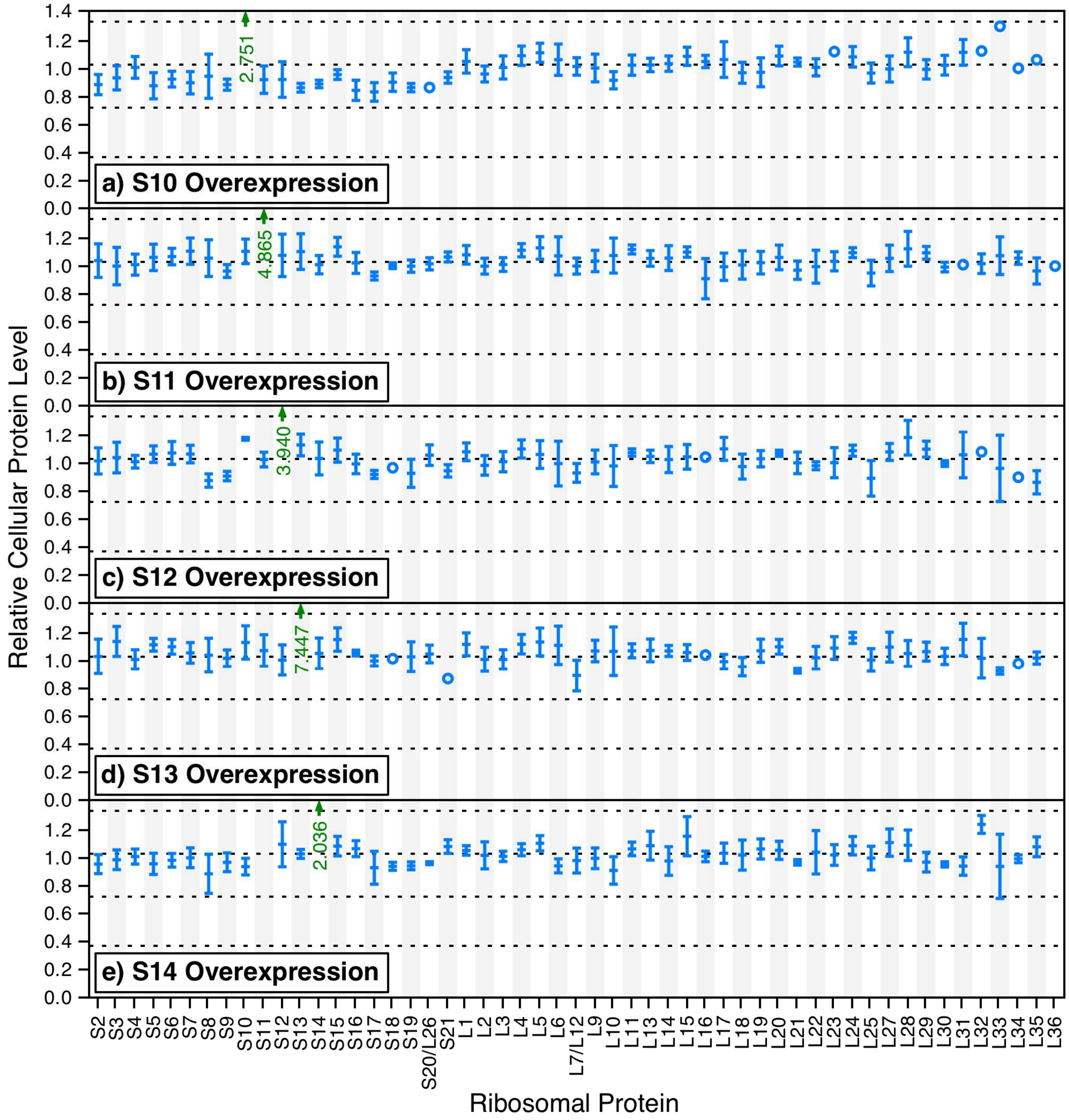
Supplementary Figure S2: Comparison of uniform scaling by different reference proteins. Error bars reflect the standard deviation of measurements obtained from different peptides or from different charge states of the same peptide. Values for the S4 overexpression experiment (small ribosomal subunit proteins only) are shown scaled by, from left to right, S7, S8, S15, S17 and S20. A dashed line indicates a value of 1. Values are generally insensitive to the choice of reference protein.



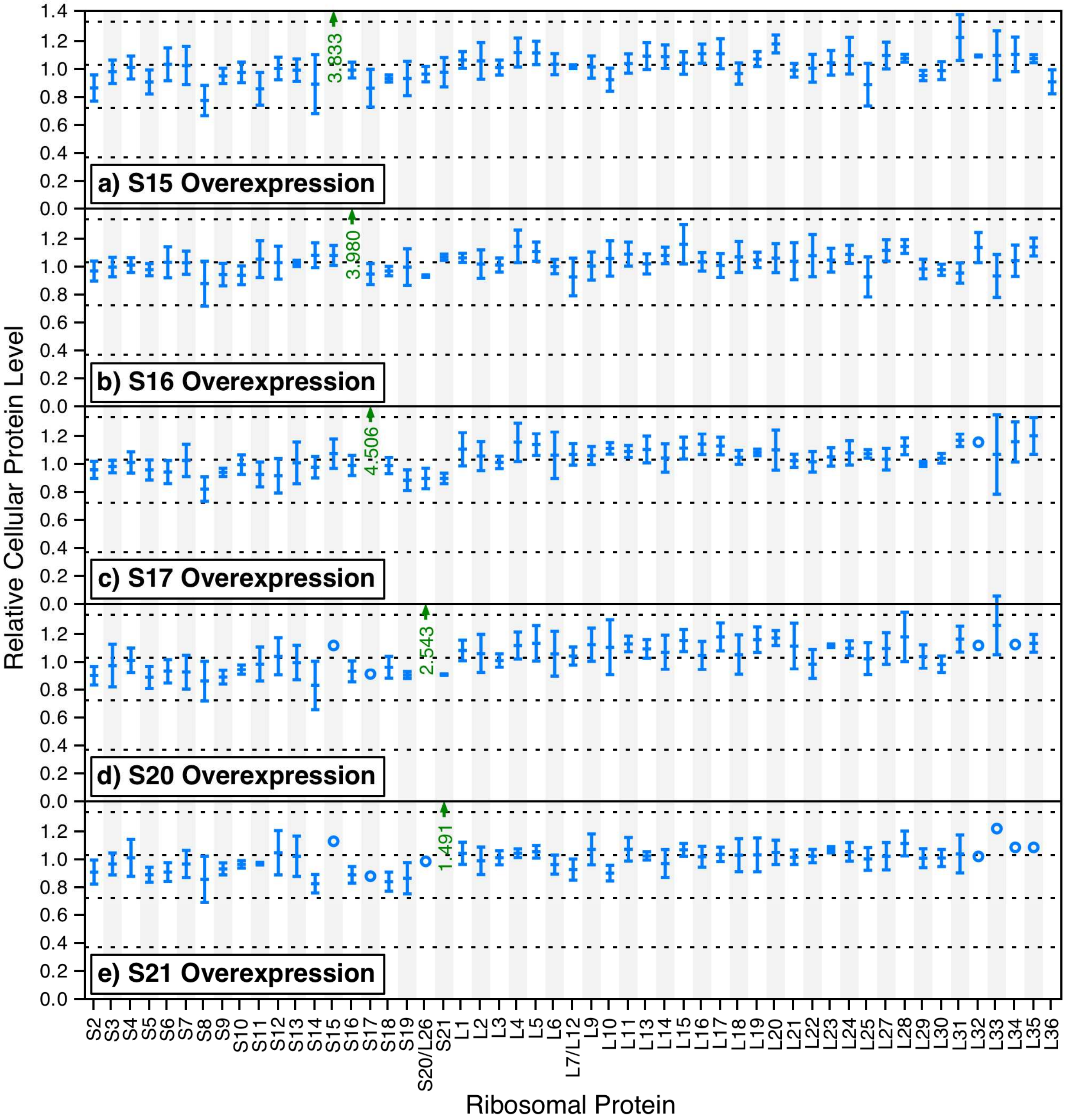
Supplementary Figure S3: A histogram of relative protein level values recorded in the 21 samples, with a bin size of 0.05. Values for proteins specifically overexpressed are omitted. A single Gaussian distribution fit to the histogram is shown in blue (Mean = 1.025, Standard Deviation = 0.075). Four dashed red lines indicate the cutoffs for highly decreased relative protein level (0.375), significantly decreased (0.725) and significantly increased (1.325) as well as the mean of the Gaussian (1.025). The significantly increased and significantly decreased protein levels are defined by taking values four standard deviations (0.075) from the mean. a) The full extent of the histogram. b) Scaled to highlight a shoulder on the left edge of the main distribution and a distinct population below 0.375.



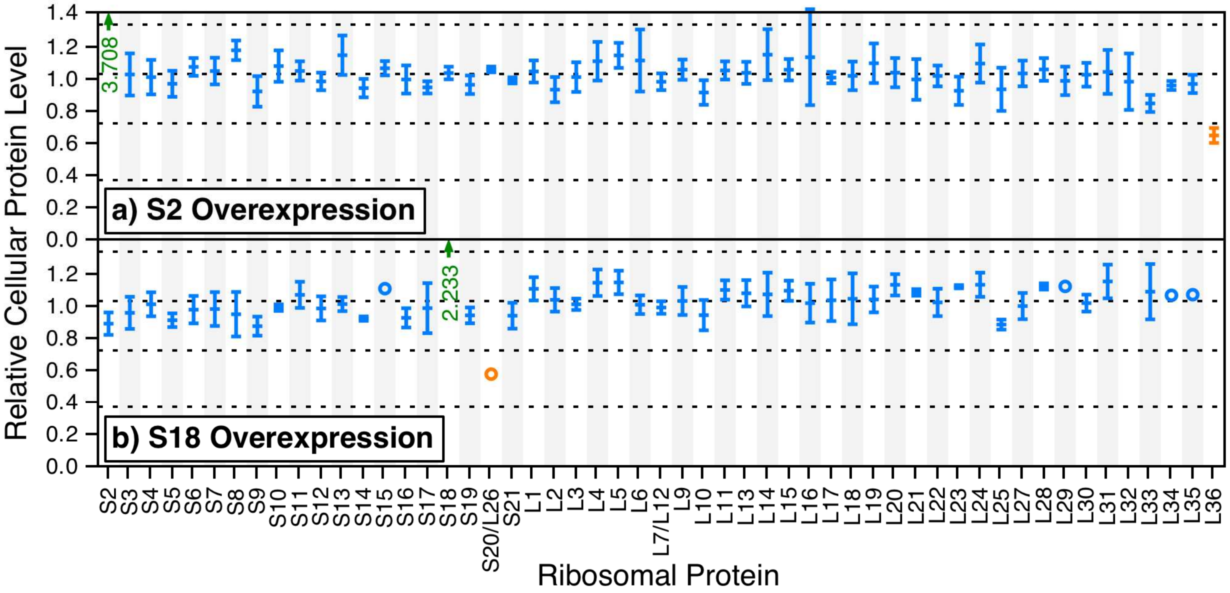
Supplementary Figure S4: Relative cellular protein levels. Dashed lines indicate global mean of 1.025 and cutoffs of 1.325, 0.725 and 0.375 for significantly increased (green), significantly decreased (orange) and highly decreased (red) respectively. Values outside the range of the plots are indicated by arrows. a) *E. coli* with an empty plasmid. b) S3 overexpression. c) S5 overexpression. c) S6 overexpression. d) S9 overexpression.



Supplementary Figure S5: Relative cellular protein levels. Dashed lines indicate global mean of 1.025 and cutoffs of 1.325, 0.725 and 0.375 for significantly increased (green), significantly decreased (orange) and highly decreased (red) respectively. Values outside the range of the plots are indicated by arrows. a) S20 overexpression. b) S11 overexpression. c) S12 overexpression. c) S13 overexpression. d) S14 overexpression.



Supplementary Figure S6: Relative cellular protein levels. Dashed lines indicate global mean of 1.025 and cutoffs of 1.325, 0.725 and 0.375 for significantly increased (green), significantly decreased (orange) and highly decreased (red) respectively. Values outside the range of the plots are indicated by arrows. a) S15 overexpression. b) S16 overexpression. c) S17 overexpression. c) S20 overexpression. d) S21 overexpression.



Supplementary Figure S7: Relative cellular protein levels. Dashed lines indicate global mean of 1.025 and cutoffs of 1.325, 0.725 and 0.375 for significantly increased (green), significantly decreased (orange) and highly decreased (red) respectively. Values outside the range of the plots are indicated by arrows. a) S2 overexpression. b) S18 overexpression.