Cell Reports, Volume *25*

Supplemental Information

Precise Post-translational Tuning Occurs for Most Protein Complex Components during Meiosis

Amy Rose Eisenberg, Andrea Higdon, Abdurrahman Keskin, Stefanie Hodapp, Marko Jovanovic, and Gloria Ann Brar

Supplemental Figures:

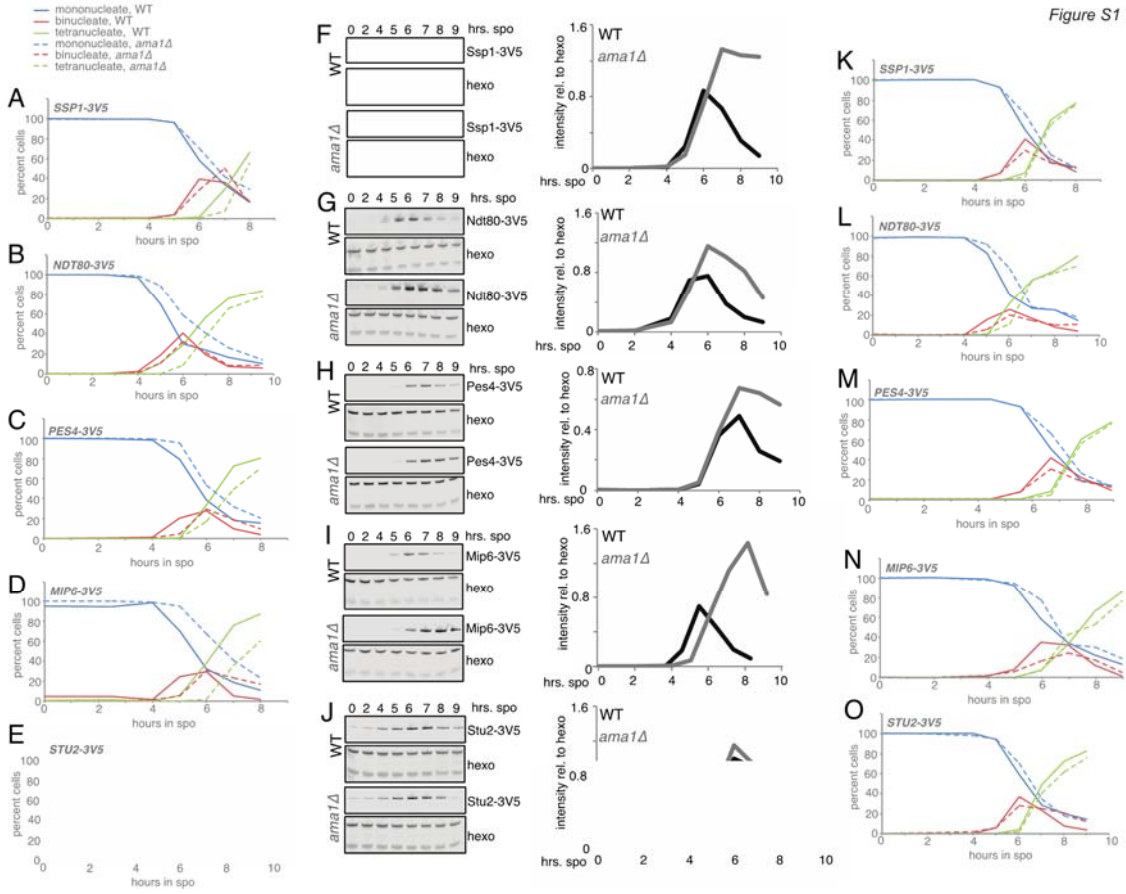

Figure S1: Meiotic staging and replicate for experiment pictured in Figure 2, Related to Figure 2. A-E) Cells were staged by DAPI quantification at hourly timepoints (n=200 per strain per timepoint). Note a slight delay in cells deleted for *AMA1* in all cases that can not account for the larger delay in protein depletion in these strains. A) Progression data for experiment shown in 2D B) Progression data for experiment shown in 2E C) Progression data for experiment shown in 2F D) Progression data for experiment shown in 2G E) Progression data for experiment shown in 2H. F-J) Replicate timecourses were run for experiments shown in Figure 2D-2I and western blotting and quantification were performed. F) Replicate for experiment shown in 2D G) Replicate for experiment shown in 2E H) Replicate for experiment shown in 2F I) Replicate for experiment shown in 2G J) Replicate for experiment shown in 2H K-O) Cells were staged as in S1A-E. Note a slight delay in

cells deleted for *AMA1* in all cases that can not account for the larger delay in protein depletion in these strains. K) Progression data for experiment shown in S1F L) Progression data for experiment shown in S1G M) Progression data for experiment shown in S1H N) Progression data for experiment shown in S1I O) Progression data for experiment shown in S1J

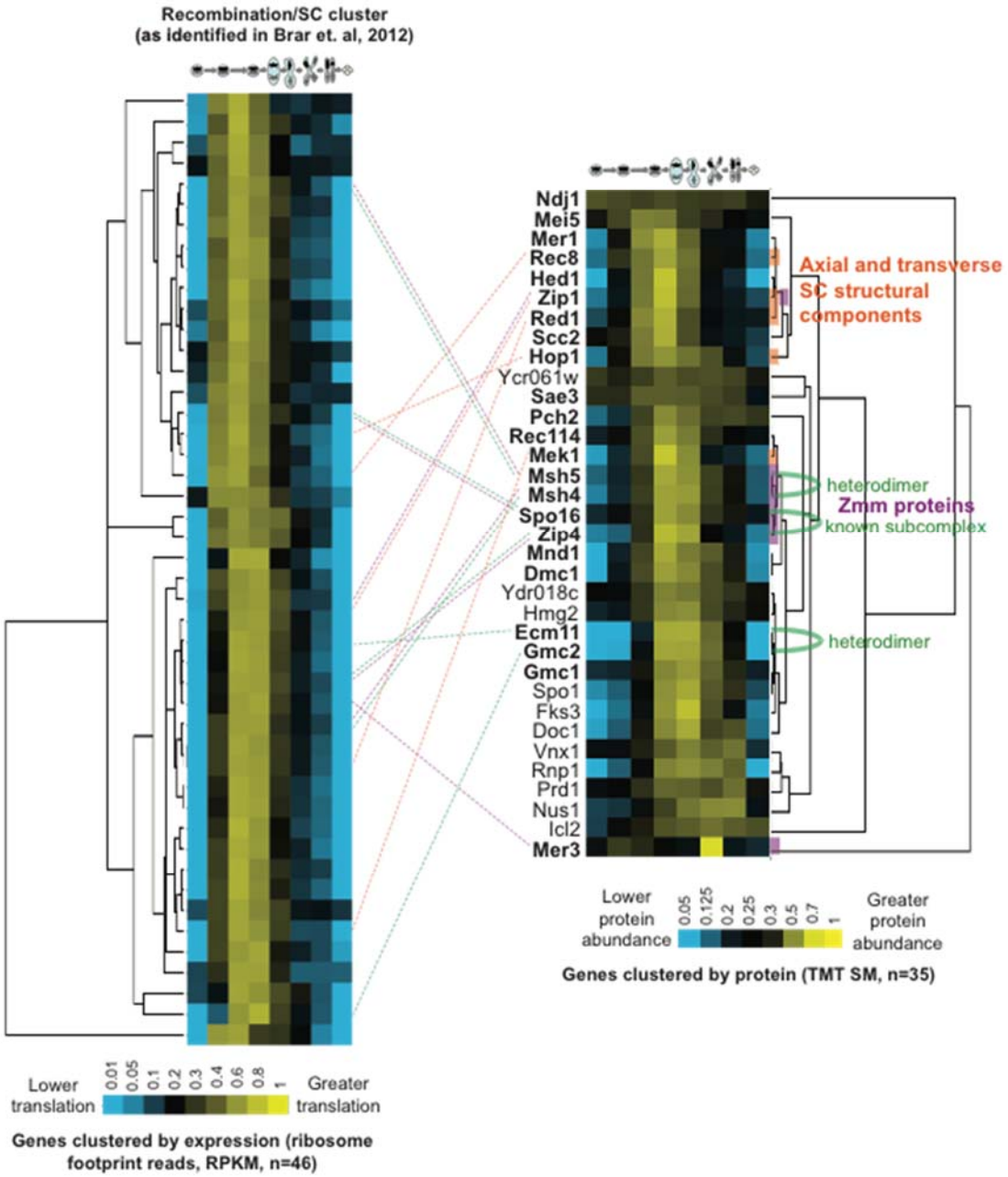

Figure S2: Protein-level clustering of genes translated during recombination and SC formation reveals interaction- and function-related substructure, Related to Figures 3 and 4. At left is the translation pattern for a group of 46 genes

that co-clustered translationally in our original meiotic ribosome profiling study (Brar et al., 2012). At right, clustering of the protein-level data for the 35 of these genes that were quantified by mass spectrometry. Note that bold names represent genes with known relationships to recombination and SC formation. Also note that groups of genes with similar function or similar physical interactions tend to cluster together at the protein level but not the translation level. This effect is especially striking for the four genes that encode so-called “Zmm proteins”, with functions in linking recombination and SC formation (Lynn et al., 2007), which cluster tightly together at the protein level and consist of two heterodimers, which cluster even more tightly than the group as a whole.

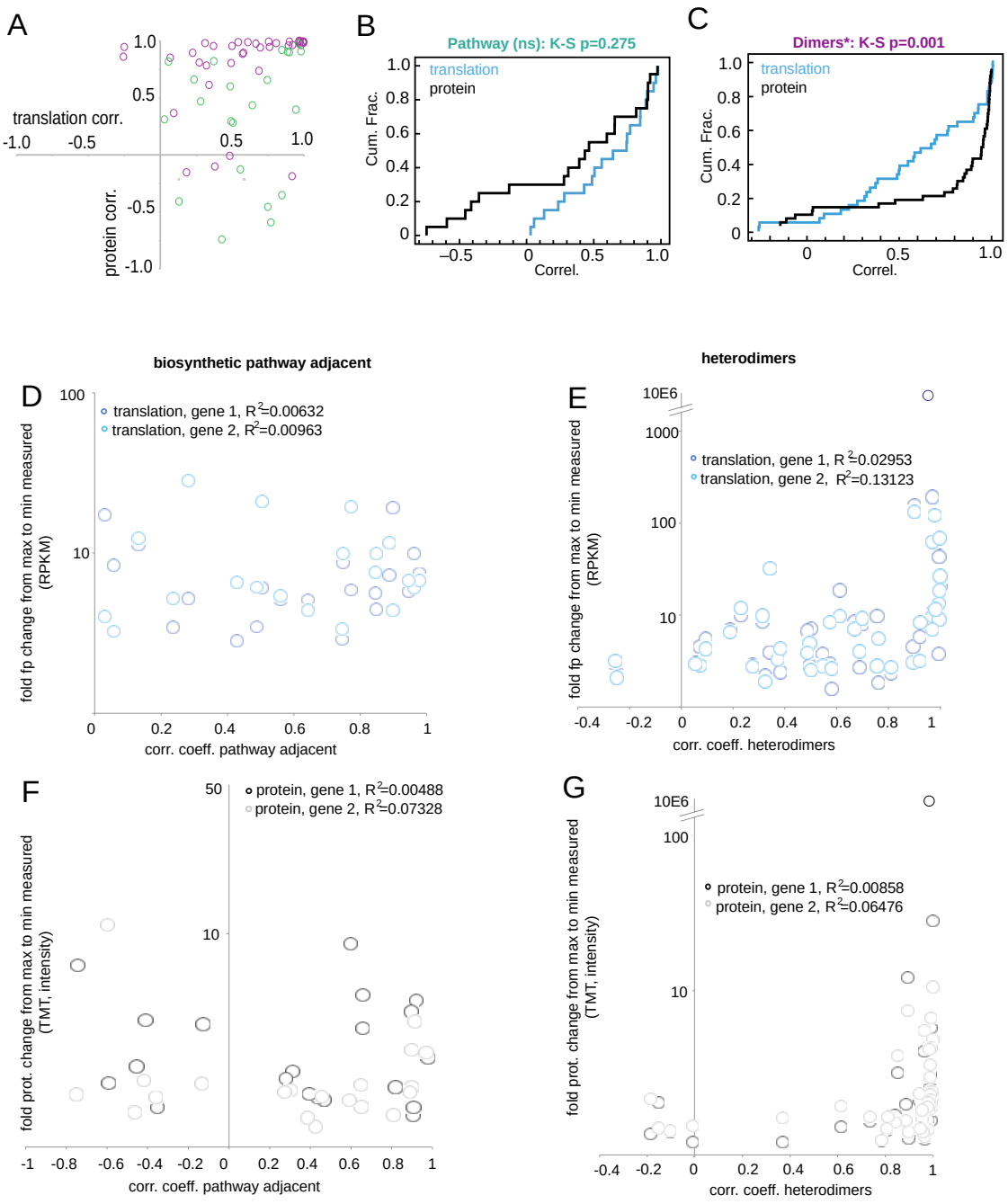

Figure S3: Sequential enzyme pairs and heterodimer partners behave differently, but correlation differences are not associated with differences in fold-changes, Related to Figure 3. A-C) Alternative representations of the data in

Fig. 3G, 3H. A) The correlation between heterodimer pairs (purple) and adjacent enzymes (turquoise) is plotted with the x-axis representing translation and the y-axis representing protein. Note that the former skew heavily towards the upper right (and particularly very high y values), while the latter skew rightward, representing high correlation of synthesis only. B) A cumulative distribution plot of the data in Fig. 3G is shown comparing translation and protein correlations among adjacent enzymes in biosynthetic pathways. The difference is not significant by K-S test. C) A cumulative distribution plot of the data in Fig. 3G is shown comparing translation and protein correlations among heterodimer pairs. The difference is significant by K-S test (p-value= 0.001). D-G) Differences in the degree of regulation are not associated with skewed correlation values. D) Fold changes from highest to lowest values measured for translation for the set of adjacent pathway genes shown in Fig. 3G are plotted against translation correlation coefficients and no significant association is seen. E) The same analysis for the heterodimers in Fig. 3G is shown. Note a propensity for very highly regulated pairs to show a high correlation in translation, likely due to highly similar mechanisms of regulation of synthesis and evolution for cellular efficiency. The outlier here is, for example, Msh4/Msh5, which are expressed in a highly meiosis-specific manner. No similarly highly regulated (or meiosis-specific) biosynthetic pathway genes were found to allow comparison. F) Fold changes from highest to lowest values measured for protein for the set of adjacent pathway genes shown in Fig. 3G are plotted against protein correlation coefficients and no significant association is seen. G) The same analysis for the heterodimers in Fig. 3G is shown. Note a propensity for very highly regulated pairs to show a high correlation in protein abundance, likely due to highly similar mechanisms of regulation of and evolution for cellular efficiency. No similarly highly regulated biosynthetic pathway genes were found for comparison (see note above for E).

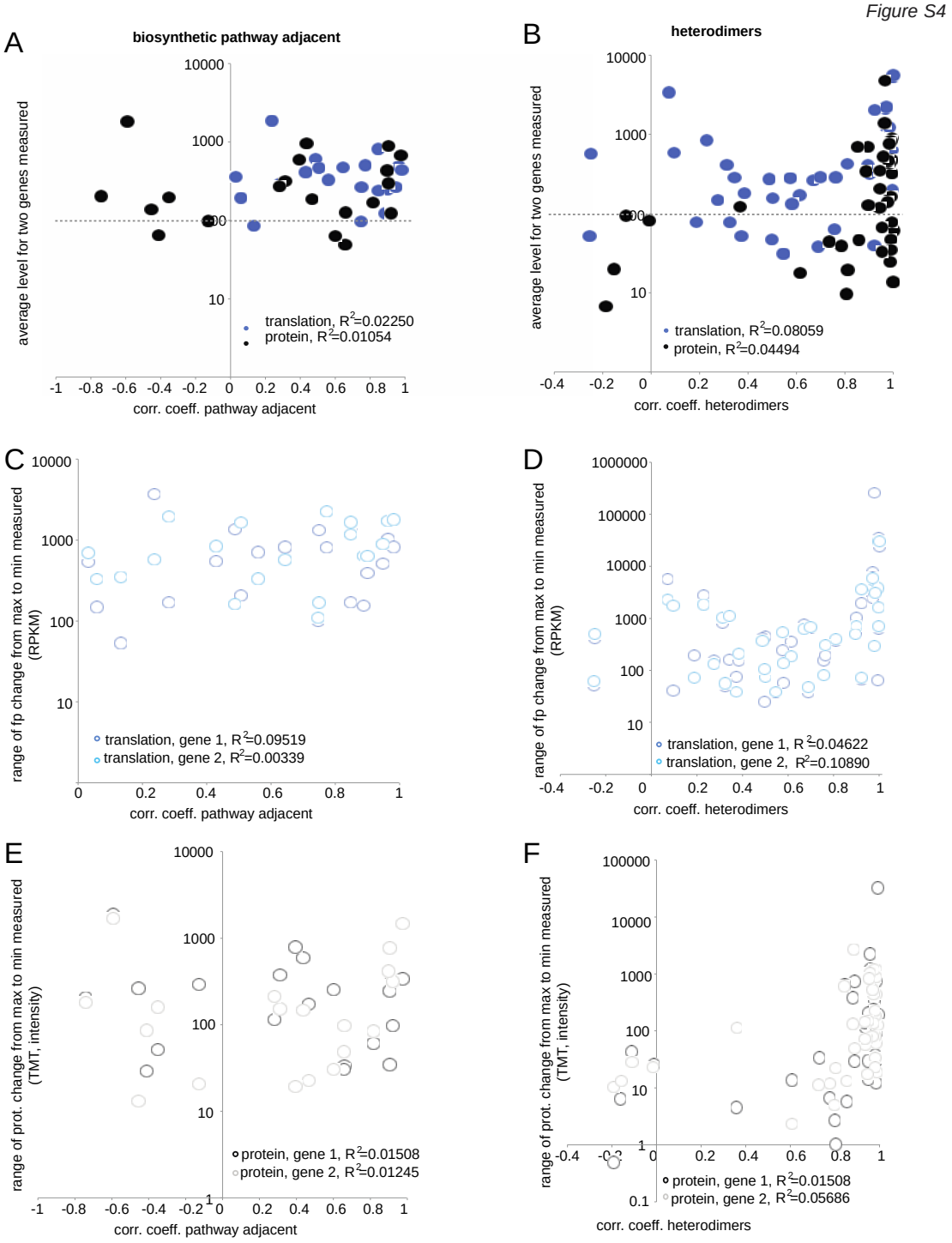

Figure S4: Levels of expression do not skew correlation analyses for the genes analyzed in our study, Related to Figure 3. A) Absolute average levels of translation and protein for the set of adjacent pathway genes shown in Fig. 3G are plotted against the appropriate translation or protein correlation coefficients and no

significant association is seen. B) Absolute average levels of translation and protein for the set of heterodimer pairs shown in Fig. 3G are plotted against the appropriate translation or protein correlation coefficients and no significant association is seen. C) Absolute changes in translation level measured in our dataset for the set of adjacent pathway genes shown in Fig. 3G are plotted against translation correlation coefficient and no significant association is seen. D) The same analysis is shown for the set of heterodimer pairs shown in Fig. 3G. E) Absolute changes in protein level measured in our dataset for the set of adjacent pathway genes shown in Fig. 3G are plotted against protein correlation coefficient and no significant association is seen. F) The same analysis is shown for the set of heterodimer pairs shown in Fig. 3G. Note that few poorly correlated protein pairs are available for analysis and none show evidence for big shifts in absolute levels, but that small absolute changes alone do not preclude or associate with any particular correlation coefficient.

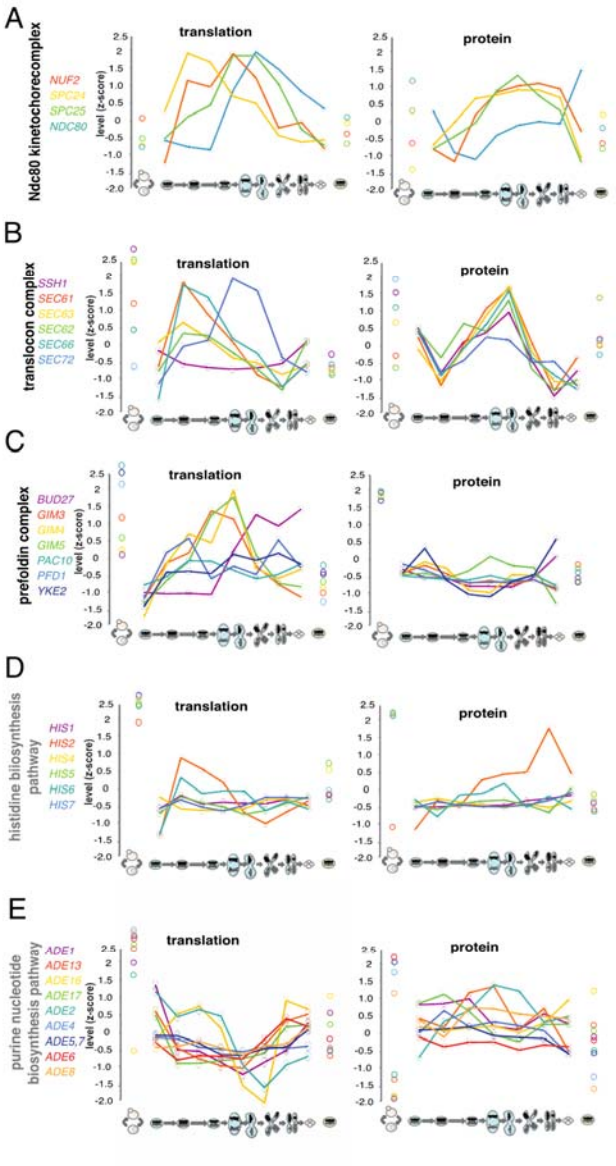

Figure S5: Additional examples of differences in translation- and protein-level trends among complex and biosynthetic pathway members, Related to Figures 4-6. For all panels A-E, translation levels (z-score) are plotted at the left and protein at the right. A) Plotted are data for the Ndc80 complex members. The regulation of this subcomplex is well characterized and it is established that Ndt80 synthesis alone is delayed in meiosis to limit kinetochore activity (Chen et al., 2017; Chia et al.,

2017; Miller et al., 2012). Our data recapitulate this result. Translation data do not match well for any of the four subunits, but protein data trends match well for Nuf2, Spc24, and Spc25, while Ndc80 accumulation is delayed relative to the others. B) Plotted are data for the translocon complex components. Note that Sec72 is the only outlier at the protein level and is the only non-essential component of this complex. C) Plotted are data for the prefoldin complex components. D) Plotted are data for the histidine biosynthesis pathway components. E) Plotted are data for the purine nucleotide biosynthesis pathway.

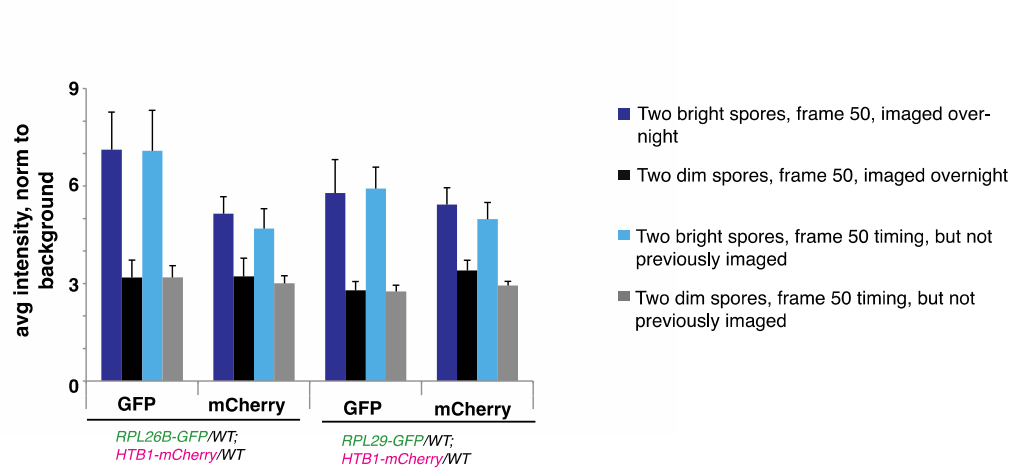

Figure S6: The low signal in two spores without fluorescently-tagged RP genes is not due to photo-bleaching, Related to Figure 6. Average integrated intensity of the two brightest or two dimmest spores from tetrads are compared for strains carrying heterozygously tagged Rpl26b and Htb1 (left) or Rpl29 and Htb1 (right). Plotted values are either for cells imaged after 50 frames or for not previously imaged cells at the same time point. 10 tetrads were counted per strain.