



Fig S4C) Pob3 and Spt16 levels in strains with point mutations in the Spt16 NTD integrated into the genome were tested alone or in combination with *pob3-Q308K*. Columns with error bars indicate comparison of 3-5 independent experiments, those without are the average of 2 independent experiments. 260 indicates the IIQ260DIR mutation, 268 is DLR268KDD, and 371 is EFR371AAA. 399 is a G399V mutation isolated in a screen for Ts- mutants in *SPT16* (Huyen Bui, personal communication), and affects a buried residue in the Spt16 NTD. The point mutations in surface residues do not cause the Ts- phenotype and both Spt16 and Pob3 levels are stable, even though these three mutations all cause temperature sensitivity at 37° when combined with *pob3-Q308K* (Table 2). In contrast, the G399V mutation causes Ts- by itself, and leads to gross instability of the Spt16 protein at elevated temperatures in a *pob3-Q308K* strain. This analysis shows that the point mutants in surface residues cause a defect in a function that overlaps with the middle domain of Pob3, rather than causing instability of yFACT.