



**Figure S3** Confirmation of *duo1-S115F* and *ndc80-K204E* alleles. Cultures of ascospore clones were serially diluted onto YPD medium and imaged after 28-30 hr at the designated temperatures. (A) Haploid spore clones from a diploid strain KT3349 (*ipl1-2/IPL1 duo1Δ::kan/DUO1* pRS303:*duo1-S115F*). All the clones are *ipl1-2* and contain either wild-type *DUO1* or pRS303:*duo1-S115F*. As expected the pRS303-*duo1-S115F* clones are also cold sensitive, arresting as large budded cells at 14°. (B) Haploid spore clones from a diploid strain KT3350 (*ipl1-2/IPL1 ndc80Δ::kan/NDC80* pRS316:*ndc80-K204E*). All the clones are *ipl1-2* and contain either wild-type *NDC80* or pRS316:*ndc80-K204E*.