



Figure S2 Mating scheme for preparing strains that differ only at a single locus. Evolved strains with the *HXT6/7* CNV were sporulated to create a large number of haploid spores. The spores were genotyped for the *HXT6/7* CNV by qPCR and for secondary mutations, if present in the evolved diploid sporulated, by PCR. We chose four spores from the same evolved strain (4.2L) that had all combinations of mating types and presence-absence of *HXT6/7* CNV. We then performed the appropriate matings and created an ancestral genotype (*HXT6/7* / *HXT6/7*), a heterozygote for the *HXT6/7* CNV ($[HXT6/7]_{10}$ / *HXT6/7*) and a homozygote ($[HXT6/7]_{10}$ / $[HXT6/7]_{10}$). Haploids are shown as circles, and diploids as more flattened ovals with pointed ends.