

## WORKING WITH H90 STRAINS

The h90 marker can be used as either h<sup>-</sup> or h<sup>+</sup> in crosses (since half of the population at any given time is h<sup>-</sup>, and half is h<sup>+</sup>). That is, you can cross your h90 strain with an h<sup>+</sup> or h<sup>-</sup> partner. But the h90 colony is likely to provide its own partner rather than use the one you give it, simply because of proximity. Thus, the best way to cross it is to make a diploid first, using complementing markers; e.g., h90 ade6-m210 X h<sup>+</sup> ade6-M216 (see diploid section for more detail; any pair of complementing markers can be used). Upon subsequent sporulation, the h90 allele will segregate in normal Mendellian fashion, giving a 2:2 ratio with the h<sup>+</sup> allele. The same sort of cross can be done with h<sup>-</sup>.

If you are used to working with cerevisiae, you may think h90 is similar to using an HO strain. However, in pombe, the mutations in mating type are not in the switching apparatus (as in an ho mutant, which lacks an endonuclease), but in the mating type locus itself. That is, h<sup>+</sup> and h<sup>-</sup> are still potentially switchable, they just don't have the correct silent information to switch into place. Most h<sup>+</sup> strains have rearranged the mating type loci so that they switch plus to plus; these can revert to h90 at about 1 in 103. In contrast, most h<sup>-</sup> strains have lost all P information and are constitutively minus.

Also, unlike cerevisiae, pombe only mates if it is starved, and the diploid generally is unstable and sporulates immediately. Thus unless you make an effort to keep cells diploid, for example by complementing markers, they will be haploid, regardless of their mating type!

(copied from PombeWeb)