Mating of baker's yeast

Introduction:

The baker's yeast (also called beer or wine yeast) *Saccharomyces cerevisiae* was established as a eukaryotic model organism with pioneer characteristics in fundamental research mainly due to its genetics. For example, in 1996 the yeast genome was the first eukaryotic genome to be completely sequenced. A very practical genetic characteristic of *S. cerevisiae* is its haplo-diploid life-cycle i.e. its ability to reproduce asexually (by budding) in the haploid as well as in the diploid state (see figure below). The stable diplophase is advantageous particularly for the characterisation of mutants (complementation analysis). Yeast only possesses two mating types, a and alpha, and mating (plasmo- and karyogamy) can only take place between individuals of different mating types.



Figure 1. Haplo-diploid life-cycle of the baker's yeast *Saccharomyces cerevisiae* (budding yeast).

Some notes to the nomenclature of the yeast genome

Yeast genes are designated with a 3-letter code and a number. The agreement is as follows:

ABC1 = wildtype gene abc1 = mutant gene

Some examples:

HIS1, HIS3 = genes coding for two different enzymes of histidine biosynthesis

LEU2 = gene coding for a specific enzyme of leucine biosynthesis *URA3* = gene coding for a specific enzyme of uracil biosynthesis *MET15* = gene coding for a specific enzyme of methionine biosynthesis *LYS2* = gene coding for a specific enzyme of lysine biosynthesis

Material:

Per 2 Students:

1 YPD (yeast complete medium)-plate with 4 yeast strains:

- Nr. 14 (DC14): MATa ade2-101 his3Δ200 tyr1 ura3-52 LEU2 LYS2 TRP1
- Nr. 17 (DC17): MATalpha ade2-101 his3Δ200 lys2-801 ura3-52 LEU2 TYR1 TRP1
- Nr. 41 (BY4741): MATa leu2-3,112 trp1-1 can1-100 ura3-1 ade 2-1 his3-11,15 TYR1 LYS2
- Nr. 42 (BY4742): *MAT*alpha leu2-3,112 trp1-1 can1-100 ura3-1 ade 2-1 his3-11,15 TYR1 LYS2

1 empty YPD-plate

2 empty yeast minimal medium (MV)-plates

Procedure:

- 1. You are provided with a plate containing 4 yeast strains
- 2. As in figure 2 (YPD-plate crossings), all 6 possible crossings of the four yeast strains are set up on the empty YPD-plate with an inoculating loop.
- 3. Incubation of the plate at 30°C.
- 4. Streak out the combinations as well as the 4 initial strains in sectors on the two MV-plates as in figure 2. Do not take too much material.
- 5. Incubation of the plates for two days at 30 °C.



Figure 2. Setup of a yeast crossing experiment: streaking of crossings on a YPD plate and on minimum media (MV) plates.

Evaluation:

For which combinations do you observe clear growth on the MV-plates?

Do you observe single colonies with some of the other combinations? Do you have an explanation for the phenomenon?

The outcome of the experiment will be analyzed and discussed in the lecture on October 24th.