Efficient meiosis for timelapse

(adapted from Fission Yeast Handbook by Paul Nurse Lab)

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h90 haploids

- 1. Thaw cells in YES plates and incubate for 1-2 days at 25-32°C.
- 2. Grow a culture in liquid YES at 25-30°C with 200 rpm agitation up to saturation.
- 3. Subculture in liquid MM 1% Glu + Sup to reach O.D. 0.8-1 with 200 rpm agitation.

For the generation time, you can estimate:

Genotype	T (°C)	Gen. time (h)
wt	25°C	6.5
wt	28°C	4.5
bqt 1Δ sad 1.2	25°C	7
$imp1\Delta bqt1\Delta sad1.2$	25°C	9.5

- 4. Centrifuge for 3 min at 3000 rpm.
- 5. Discar supernatant by pouring and wash pellet with equal volume of MM N.
- 6. Centrifuge for 3 min at 3000 rpm.
- 7. Remove supernatant completely by pipetting.
- 8. Resuspend pellet in 100-200 uL MM N (depending on pellet size).
- 9. Plate a drop of 20 uL of suspension in a MEA plate (tempered at room temperature) and incubate for 4-5 h at 28°C.