

# Yeast Colony PCR protocol

derived from [http://sequence-www.stanford.edu/group/yeast\\_deletion\\_project/deletions3.html](http://sequence-www.stanford.edu/group/yeast_deletion_project/deletions3.html)

- Wash a matchhead amount of cells with 500  $\mu$ l dH<sub>2</sub>O.
- Resuspend the cells in 100  $\mu$ l solution containing 60 U/ml of Zymolyase (Make by mixing 60  $\mu$ l 10mg/ml Zymolyase in 1 ml of water).
- Add 25  $\mu$ l of glass beads to sample.
- Repeat these steps for each isolate and control.
- Incubate the samples at 37 ° C for 30 minutes.
- Vortex for 1 minute.
- Incubate the samples at 95 ° C for 10 minutes.
- Chill the samples on ice for 5 minutes.
- Spin for 1 minute to pellet cellular debris.
- Store at -20 ° C, or use immediately in PCR.
- Use 1-10  $\mu$ l per 50  $\mu$ l PCR reaction.

\*The Zymolyase treated cells can be stored at -20 ° C indefinitely and still be used in PCR. The cells should be thawed and kept on ice during use.