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## Glass Bead Transformation of Chlamydomonas

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## Abstract

A short and relatively cheap method for non-homologous nuclear transformation of *Chlamydomonas reinhardtii*. Works best with linearized DNA. Requires 500 micron glass beads for DNA transformation.

## Troubleshooting



## Prep

- 1 Grow cell culture to an OD750 of 0.15 to 0.4
- 2 Centrifuge at 400g for 5 minutes at room temperature  
 00:05:00
- 3 Resuspend in 1/100th the original volume of TAP


- 4 Add the following:

	8000PEG to 5% final conc.
	3 ug of DNA
	0.3g of 500micron glass beads
	0.4mL Chlamy cell suspension

- 5 Mix with a pipette

## Vortex


- 6 Vortex at max speed for 15 seconds

 00:00:15

## Plating & Selection

- 7 Take 25uL of the cell suspension and add to 100uL of TAP with an appropriate antibiotic
- 8 Spread on a TAP or YA plate, with an appropriate antibiotic, using large glass beads
- 9 Allow the liquid to dry while avoiding light
- 10 Seal the plates with parafilm



- 11 Allow the colonies to grow (colonies will appear in 1-3 weeks)
- 12 Transfer the remaining cell/vortex culture to a 125mL flask with 20mL of TAP (w/o antibiotic)
- 13 Incubate for 6 hours on an orbital shaker at 70rpm  
 06:00:00
- 14 Add antibiotic to an appropriate concentration
- 15 Take 50uL of the cell suspension and spread on a TAP or YA plate with an appropriate antibiotic with large glass beads
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