

How to Use an Avestin Emulsiflex C3 Homogenizer to Disrupt Cells

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[Abstract] The EmulsiFlex-C3 homogenizer is powered by an electric motor. The pump does not require a compressor for it to run. This equipment can be used to disrupt cells at a large scale. The EmulsiFlex-C3 has a fixed flow-through capacity of 3L/hr. It has the ability to process samples as small as 10 ml. The homogenizing pressure is adjustable between 500 and 30,000 psi. In this protocol, we describe the use of the Avestin Emulsiflex C3 Homogenizer to disrupt *S. pombe* and *S. cerevisiae* cells.

Materials and Reagents

1. *S. pombe* cells
2. *S. cerevisiae* cells
3. DI water

Equipment

1. Avestin Emulsiflex C3 homogenizer (Avestin®)



Figure 1. Avestin Emulsiflex C3 homogenizer

2. Standard laboratory bench-top light microscope

Procedure

1. Switch on the homogenizer.
2. Turn on nitrogen. Pressure reads 80 psi.
3. Unscrew the funnel cap. Check if the funnel cap is on to make sure ethanol does not evaporate.
4. Turn red stop knob clockwise and push green knob to start.
5. Pump residual ethanol out of the tubing.
6. Pour DI water into the funnel to wash ethanol out. Keep air pressure on occasionally to make sure no cell debris is left from the last user.
7. Before loading samples, take the funnel off and roll it on ice to keep it cool. Install the funnel back to the top.
8. Put the steel coil heat exchanger into ice to cool down the samples.
9. Load your samples into the funnel. Turn on the homogenizer. Let the samples run through the tubing back to the funnel before air pressure is on.
10. Turn on air pressure. Air pressure at 40 psi, gauge pressure \geq 20,000 psi and $<25,000$ psi. The maximum pressure is 30,000 psi. Leave the tubing in a sample collection tube chilled on ice.
11. *S. pombe* samples need to be passed through 5~6 times to reach 80~90% efficiency. *S. cerevisiae* samples need to be passed through 8~9 times to reach 80~90% lysis efficiency.
12. Check samples under a standard light microscope.
13. If the homogenizer is clogged by the samples, cap the funnel and blow with nitrogen tube.
14. After samples are done, take off the funnel and rinse it with DI water.
15. Run more water to flush out cell debris. Keep the air pressure on occasionally.
16. Run ethanol and leave 1/3 of a funnel volume of ethanol in the funnel.

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References

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