Bullet Blender® 50
Homogenization Protocol for E. coli Cultures

The protocol described in this document is for the use of the Bullet Blender® 50 for the homogenization of *Escherichia coli* (or other bacterial) cultures. This protocol does not specify a particular buffer - you may choose which is most appropriate for your downstream application (nucleic acid isolation, protein extraction, etc.).

Materials Required: E. coli, Bullet Blender® 50, homogenization buffer, pipettor, 50mL skirted centrifuge tubes (Axygen® or Corning® brand), 0.5mm zirconium oxide beads (part number ZROB05).

Instructions

1. Pour bacterial culture into 50mL centrifuge tube.
2. Centrifuge culture (2000g for one minute) to yield a cell pellet. Pellet should be 4 mL or less to achieve efficient homogenization.
3. Completely aspirate supernatant liquid. Place tube on ice.
4. Add a volume of beads to the tube approximately equal to the volume of the pellet.
5. Add 0.2 mL to 8mL buffer (2 volumes of buffer for every volume of sample).
6. Screw caps onto centrifuge tubes **TIGHTLY**.
7. Place tubes into the Bullet Blender® 50.
8. Set controls for **SPEED 8** and **TIME 12** minutes.
9. Remove tubes from the instrument.
10. Visually inspect samples, if homogenization is unsatisfactory, run for another six minutes at the **SPEED 9**.
11. Proceed with your downstream application.

SAFETY NOTE!!!

When using a centrifuge to separate your homogenate from the debris and beads, make sure your tubes are balanced.

Date 05/06/2011