Bullet Blender[®] 5 Homogenization Protocol for Saccharomyces

The protocol described in this document is for the use of the Bullet Blender[®] 5 for the homogenization of *Saccharomyces* cultures (*cerevisiae*, *pombe*, etc.). 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: yeast, Bullet Blender® 5, homogenization buffer, pipettor, 5mL

Axygen® brand tubes, and 0.5mm zirconium oxide beads (part

number ZROB05)

Instructions

- **1.** Pour overnight yeast culture into a 5ML tube.
- 2. Centrifuge culture to yield a cell pellet (1000g for two minutes).
- **3.** Completely aspirate the supernatant liquid. Place tube on ice.
- **4.** Inspect the volume of the pellet. It should be 0.75ML or less in order to get efficient homogenization.
- **5.** Add a volume of beads equal to the volume of cells.
- **6.** Add 0.2mL to 1.5mL buffer (2 volumes of buffer for every volume of sample).
- 7. Tightly screw the centrifuge tubes closed and place them into the Bullet Blender[®].
- **8.** Set controls for **SPEED 8** and **TIME 3** minutes. Press start.
- **9.** After the run, remove the tubes from the instrument.
- **10.** Visually inspect samples, if homogenization is unsatisfactory, run for another two minutes at **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.