

Bullet Blender[®] 5

Homogenization Protocol for Lymph Node

The protocol described in this document is for the use of the Bullet Blender[®] 5 for the homogenization of lymph node tissue (from a variety of animals). If you have difficulty with this protocol, cutting your tissue into smaller pieces will help. 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: lymph node tissue, Bullet Blender[®] 5, homogenization buffer, pipettor, 5mL Axygen[®] brand tubes, and [3.2 mm stainless steel beads \(part number SB32\)](#).

Instructions

1. Cut lymph node into appropriately sized pieces for analysis (0.1g – 1g).
2. OPTIONAL: If desired, wash the tissue 3x with 5mL PBS to remove blood and other contaminants from the tissue.
3. Place sample in 5ML tube and add beads to the tube. Use a volume of beads equal to the mass of tissue. NOTE: 100mg \approx 100 μ L.
4. Add 0.2mL to 2.0mL buffer (2 volumes of buffer for every volume of sample).
5. Tightly screw the centrifuge tubes closed and place them into the Bullet Blender[®].
6. Set controls for SPEED 8 and TIME 3 minutes. Press start.
7. After the run, remove the tubes from the instrument.
8. Visually inspect samples, if homogenization is unsatisfactory, run for another two minutes at SPEED 9
9. 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.