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Protocol for Adipose Tissue Homogenization in the Bullet Blender™

The protocol described in this document is for the use of the Bullet Blender™ for the homogenization of fat / adipose tissue (from a variety of animals). Note that the time and speed settings may differ due to the variation in consistency/texture of fatty tissue from species to species. 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: adipose tissue, saline, aspirator, Bullet Blender™, zirconium oxide beads (0.5mm), microcentrifuge tubes, homogenization buffer, and pipetor.

Instructions

1. Cut adipose tissue into appropriately sized pieces for analysis (30mg-300mg) and place into a microcentrifuge tube.
2. **OPTIONAL:** Wash tissue with ~1mL PBS. Aspirate. **NOTE:** This step removes external contaminants (blood, etc.).
3. Add a mass of 0.5mm zirconium oxide beads equal to the mass of sample in each tube (for 100mg sample, add 100mg beads). One full scoop of beads ≈ 180mg.
4. Add 2 - 4 volumes of buffer for every volume of tissue.
5. Close the microcentrifuge tubes.
6. Place tubes into the Bullet Blender™.
7. Set controls for **SPEED 7** and **TIME 2** minutes. Press **Start**.
8. After the run, remove tubes from the instrument.
9. Inspect samples. Fatty tissue homogenate will be difficult to see through due to the light scattering of lipid micelles formed, so it may be necessary to employ a pipette tip to check inside the tube for remaining pieces of intact tissue. If homogenization is unsatisfactory, run for another minute at **SPEED 9**.
10. Proceed with your downstream application.

SAFETY NOTE!!! – Make sure your tubes are balanced before placing them into a centrifuge!

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