Protocol for Meconium Homogenization in the Bullet Blender®

The protocol described in this document is for the use of the Bullet Blender® for the homogenization of human meconium. 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 meconium, Bullet Blender®, homogenization buffer, pipettor, microcentrifuge tubes and Red bead lysis kit/Pink bead lysis kit/0.5mm zirconium oxide beads (part number ZrOB05).

Instructions

1. Weigh a mass of meconium (10mg-300mg) to be homogenized. Try to keep the walls of the tube as clean as possible, and ensure the meconium sits as low in the tube as possible.
2. a. *Samples 100mg or greater*
   Place the sample in Red bead lysis kit tube.
   b. *Samples less than 100mg*
   Place the sample in Pink bead lysis kit tube.
   c. *Alternate protocol step for bulk beads*
   Place sample in microcentrifuge tube and add beads to the tube. Use a volume of beads equal to the mass of tissue. **NOTE:** 100mg ≅ 100µL.
3. Securely close the microcentrifuge tubes.
4. Place tubes into the Bullet Blender®.
5. Set controls for **SPEED 8** and **TIME 5** minutes. Press **Start**.
6. After the run, remove tubes from the instrument.
7. Verify that all samples have a uniform appearance. A smooth semi-liquid should exist. If homogenization is unsatisfactory, run for another five minutes at speed 8.
8. 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.

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