



Protocol for *A. thaliana* Homogenization in the Bullet Blender™

The protocol described in this document is for the use of the Bullet Blender™ for the homogenization of *Arabidopsis thaliana* tissue (leaves, stems, seeds, or roots). 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: plant material, Bullet Blender™, homogenization buffer, pipettor, microcentrifuge tubes, and [0.5mm zirconium oxide beads \(part number ZrOB05\)](#).

Instructions

1. Try to remove as much dirt or other debris from the plant by gently shaking.
2. If desired, wash the with saline/distilled water to remove any soil or other particulate matter that is not part of the plant.
3. Blot excess liquid from the soft tissue using a Kimwipe® or other lint free cloth.
4. Cut the tissue into appropriately sized pieces (0.05 to 0.3g).
5. Place the plant tissue into a microcentrifuge tube.
6. Add 0.1g to 0.6g of 0.5mm zirconium oxide beads to the tube (twice as much beads as sample). One scoop of beads ≈ 170mg.
7. Add 0.1m to 0.6mL buffer, i.e. twice as much buffer as sample.
8. Close the microcentrifuge tubes.
9. Place tubes into the Bullet Blender™.
10. Set controls for **SPEED 8** and **TIME 2 to 3** minutes. Press **Start**.
11. After the run, remove tubes from the instrument.
12. Visually inspect samples. If homogenization is unsatisfactory, run for another five minutes at the **SPEED 10**.
13. 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.

The text above describes a generic protocol. A specific measure of materials from two different protocols are listed here:

Protocol A

Leaf tissue: ~0.05g
(snap centrifuge cap closed on leaf)
Buffer: 500µL
ZrO beads
SPEED 8, TIME 3 minutes

Protocol B

Leaf tissue: ~0.05g
(snap centrifuge cap closed on leaf)
Buffer: 450-500µL
Stainless Steel beads (1.6mm)
SPEED 8, TIME 2 to 3 minutes