Bladder

- Extract molecules (DNA, RNA, protein, chemicals)
- Wet final product
- Sample sizes: up to 300 mg.

Notes on the protocol: 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.).

User note: This protocol was developed using mouse tissue. Homogenization times, speeds, and beads may need to be adjusted if you are working with material from other species, especially larger animals.

Materials Required

One of these Bullet Blenders

- **Bullet Blender** (BBX24)*
- **Bullet Blender Blue** (BBX24B)*
- **Bullet Blender Storm 24** (BBY24M)
- **Bullet Blender 24 Gold** (BB24-AU)

*To use these models, set the speed to 10 and use no more than two tubes in the machine.

Reagents

Homogenization buffer
    2 x volume of sample
PBS (optional)
    3 x volume of sample

Bead choices

- **GREEN bead lysis kit** (GREEN) (for samples up to 50 mg.)
- **NAVY bead lysis kit** (NAVY) (for samples between 50 and 300 mg.)
- **Bead combination:**
  0.9 - 2.0 mm stainless steel blend (SSB14B) Use a volume of beads equivalent to 1 x the volume of the sample **plus**
  3.2 mm stainless steel beads (SSB32)

Procedure
1. Cut the sample into appropriately sized pieces. For larger samples, we recommend cutting the material into long, thin strips for faster homogenization.
2. (Optional) Wash the sample 3x with 1/2 tube volume of PBS to remove surface contaminants.
3. Place the sample in the tube with the beads.

4. Add a volume of buffer that is twice the volume of the sample. Sample volume may be approximated by sample weight. E.g., for a 100 mg. sample, add 0.2 ml. buffer.
5. Close the tubes tightly and place them in the Bullet Blender.
6. Set the controls for Speed 12 and Time 3. Press Start.
7. After the run, remove the tubes from the instrument and visually inspect the samples. If homogenization is incomplete, repeat the homogenization step at a higher speed.
8. Proceed with your downstream application.