Xenopus Homogenization Using the Bullet Blender

Materials

RS18-0238B.AXEN

- <u>Bullet Blender</u>[®] for 1.5/2 mL tubes using 5 mL adapters
- Homogenization Buffer
- <u>FoamBlocker</u> (Optional)
- Lysis Kit or Lysis Beads
 - GREEN or NAVY Lysis Kit (from <u>PrecisionPak™</u> or purchased separately)
 - 1.4 mm blend + 3.2 mm Stainless Steel Beads in Eppendorf, GATOR, or RINO tubes
- Sample up to 300 mg

Table 1. Proper sample, bead and buffer volume ratios for 1.5/2 mL tubes using 5 mL adapters.

| Lysis Kit and Bead Choices | Sample Volume | Bead Volume | Buffer Volume |
|---|---------------|--------------------|---------------|
| GREEN | Up to 100 mg | Pre-filled | 200 - 300 μL |
| NAVY | 100 - 300 mg | Pre-filled | 300 - 600 μL |
| 1.4 mm blend + 3.2 mm Stainless Steel Beads | Up to 300 mg | 100 - 200 μL | 200 - 600 μL |

Procedure

- 1. Use the pre-filled bead lysis kit tubes OR prepare a tube with the recommended volume of bead choices from the table above.
- 2. Add the appropriate volume of buffer according to the table above
- 3. Prepare the sample by cutting it into small thin pieces and then transfer it into the buffer-filled tubes.
- 4. (Optional) To avoid excess foaming, add FoamBlocker up to 1-2% of the total volume of the homogenization buffer.
- 5. Close the tubes tightly and place into the Bullet Blender sample chamber. If using the Gold or Gold⁺ models, pre-cool the chamber before adding sample tubes.
- 6. Set the controls to speed 16, time 5 minutes then press Start. *Note: Using single-size beads instead of pre-filled lysis kits may require additional time.*
- 7. After the run, remove the tubes from the instrument and visually inspect the samples. If homogenization is incomplete, homogenize for an additional 30 seconds, or repeat the homogenization step with a higher speed.
- 8. Using a pipette, transfer the homogenized samples into new tubes.
- 9. Proceed with downstream application.

Notes

This protocol does not specify a particular buffer – choose a buffer that is most appropriate for the downstream application or use the lysis buffer provided in a <u>PrecisionPak™</u>, a simplified workflow solution which also includes a bead lysis kit, supplemental reagents for high quality nucleic acids isolations, and an optimized protocol for specific samples.

The provided homogenization conditions serve as a general guideline. Homogenization times, speeds, or beads may need to be optimized based on sample characteristics and desired outcomes.



Page 1 of 1

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