

BULLET BLENDER® USER GUIDE: TABLE OF CONTENTS

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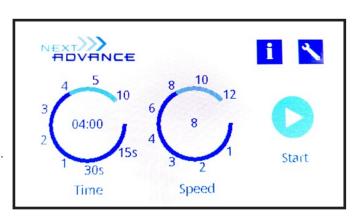
GENERAL OPERATION

BULLET BLENDER® USER GUIDE ADJUSTING THE SPEED & DURATION

➤ NOTES

Set the desired speed and time by adjusting the values on the front of the instrument.

Homogenizing tougher tissue requires longer durations at full speed, while mixing or cell dissociation requires lower speeds. Suggested speeds and times for different tissues are available on the Next Advance website.







SAMPLE PREPARATION & HANDLING

BULLET BLENDER® USER GUIDE PROPER FILL PROPORTION

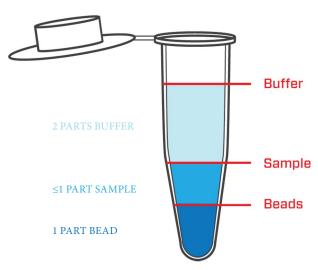
> NOTES

Proportions are volumetric.

Always keep the bead-to-buffer ratio the same. The amount of sample must be less than or equal to half of the buffer volume. Low sample to buffer ratios are fine.

Different ratios may be required for certain sample types.
See specific protocols on our website for details.





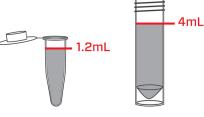
BULLET BLENDER® USER GUIDE MAXIMUM TOTAL VOLUME

> NOTES

Overloading the tube will result in poor homogenization and can cause tube leakage.

Some sample types homogenize more efficiently with smaller tube loads.

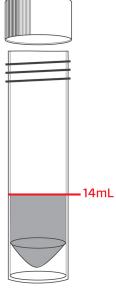
*Volumes shown are for total bead, buffer, and sample volumes







5 mL Tubes



50 mL Tubes

BULLET BLENDER® USER GUIDE USE OF DETERGENT

> NOTES

Homogenizing samples in buffer containing detergent may result in excessive sample foaming.

For DNA and proetin applications, we recommend adding an antifoam agent, such as <u>Next Advance</u> <u>FoamBlocker</u>.







BULLET BLENDER® USER GUIDE CUTTING A SAMPLE PROPERLY

> NOTES

Thin, narrow cuts of sample will homogenize faster than thick, wide cuts.



EXAMPLE: CITRUS LEAVES CUT INTO THIN NARROW PIECES

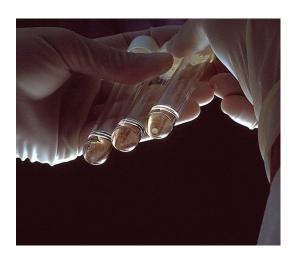


BULLET BLENDER® USER GUIDE CELL CULTURE PREPARATION

> NOTES

Cells should be pelleted and then resuspended in the recommended volume of buffer before homogenization.

Total packed volume of cells should be 300 uL or less for microcentrifuge tube models, 1 mL or less for 5 mL tube models, and 3.5 mL or less for 50 mL tube models.



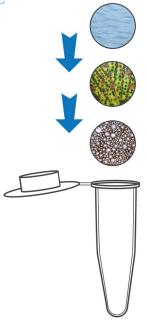


BULLET BLENDER® USER GUIDE ALTERING TUBES

> NOTES

Altering homogenization tubes will likely reduce the efficiency of homogenization.

Parafilm should not be used on tubes going into your instrument. Using parafilm will reduce the ability of the tube to oscillate in the Bullet Blender, thereby hindering homogenization.



BUFFER

SAMPLE

BEADS

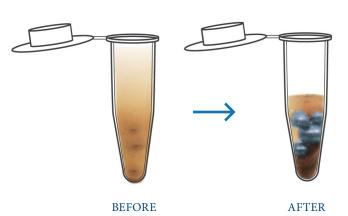


BULLET BLENDER® USER GUIDE RETRIEVING SAMPLES FROM TUBES

> NOTES

To remove the homogenate, either carefully remove as much of the homogenate as possible from around the beads with a pipette or centrifuge at a low speed not exceeding 6,000 x g for 2 minutes.

CENTRIFUGATION







BEAD SELECTION

BULLET BLENDER® USER GUIDE BEAD LYSIS KITS

> NOTES

Bead Lysis Kits are a convenient all-in-one solution: beads are pre-loaded into homogenization tubes. Just add sample and buffer.

Choose <u>bead lysis kits</u> based on your sample type and size.



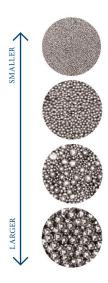


BULLET BLENDER® USER GUIDE BEAD SELECTION: SIZE

> NOTES

Using beads that are too large for the tube (e.g. 4.8 mm beads in a microcentrifuge tube) can result in inefficient homogenization and tube breakage and sample loss.

Refer to your protocol or contact <u>customer</u> support for bead recommendations.





BULLET BLENDER® USER GUIDE BEAD SELECTION: MATERIAL

> NOTES

Use denser beads for tougher samples.

Lighter beads, such as glass, can be used for soft samples. Denser beads such as zirconium oxide or stainless steel provide more homogenizing power.





BULLET BLENDER® USER GUIDE BEAD SELECTION: SPECIALTY BEADS

> NOTES

Use special bead types to homogenize difficult samples.

For resilient samples that contain a lot of connective tissue or fibers, consider using "UFO" beads. The sharper edges of these beads are excellent for cutting up tough samples.

To crush dry grains into powder, use large stainless steel beads (3.5 or 5.6 mm) in the 5 mL tube model Bullet Blenders.



STAINLESS STEEL UFO BEADS





CONSIDERATIONS BY ANALYTE

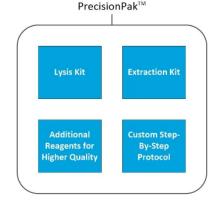
BULLET BLENDER® USER GUIDE CONSIDER ATIONS: PRECISIONPAK™

> NOTES

We offer a streamlined, customized solution for biomolecule extractions, called the PrecisionPak™.

The pack bundles together the bead lysis kits with the extraction kits and an optimized protocol for your specific sample type.

Extraction kits use magnetic bead based technology for DNA and RNA, and come with additional reagents and enzymes for removing unwanted proteins, RNA, and/or DNA from the homogenate.



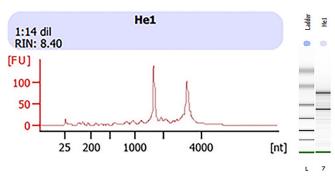


BULLET BLENDER® USER GUIDE CONSIDERATIONS: RNA

> NOTES

Bullet Blenders can be used in cold rooms. For maintaining sample temperature at 4 °C, consider Gold+ models with dry ice cooling or liquid nitrogen cooling.

Use RNase-free beads and tubes to limit sample degradation. Consider RNAse and DNase-free Bead Lysis Kits to reduce handling.



MOUSE HEART RNA ISOLATED USING PRECISIONPAK™



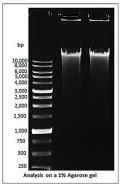
BULLET BLENDER® USER GUIDE CONSIDER ATIONS: DNA

> NOTES

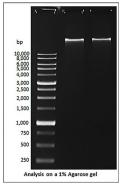
Using the Bullet Blender at high speeds and long durations may cause DNA shearing during processing.

Consider adding a short dry homogenization step before homogenizing sample with buffer.

Full length chromatin can be extracted using the Bullet Blender at lower speeds.



MOUSE SKIN DNA FROM PRECISIONPAK®







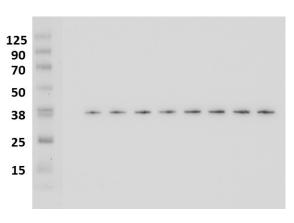
BULLET BLENDER® USER GUIDE CONSIDER ATIONS: PROTEIN

> NOTES

For protein application, it is especially important to avoid foaming.

Adding an antifoam agent, such as <u>FoamBlocker</u>, to the lysis tube can significantly reduce foaming.

All of our Gold and Gold+ models have a <u>4°C cooling</u> option to keep your samples cool during homogenization and prevent protein degradation.



BLOT SHOWING GAPDH FROM BULLET BLENDER HOMOGENATE

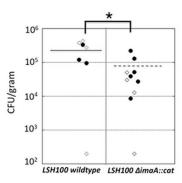


BULLET BLENDER® USER GUIDE BACTERIAL & VIRAL EXTRACTION

> NOTES

Bullet Blenders can isolate bacteria and viruses from infected tissue and plant material.

Specific protocols vary by sample and analyte. Contact <u>customer support</u> for protocols.





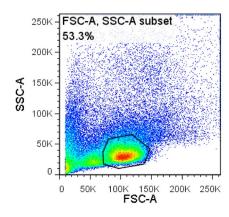
BULLET BLENDER® USER GUIDE CELL DISSOCIATION

> NOTES

Use large dense beads and homogenize samples at low speeds.

There will be some loss of viability. Tougher samples have greater loss.

Pre-incubating with a digestion buffer (e.g. containing collagenase) will aid in dissociation.



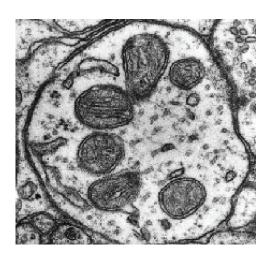


BULLET BLENDER® USER GUIDE ORGANELLES (E.G. NUCLEUS, MITOCHONDRIA, CHLOROPLASTS)

➤ NOTES

Some organelles, such as mitochondria, can be isolated from cells using a Bullet Blender.

To minimize damage to the organelles, use low speed settings that will still result in cell lysis.



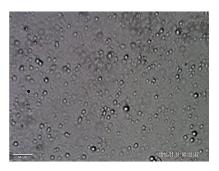


BULLET BLENDER® USER GUIDE NANO PARTICLES/LIPOSOMES

> NOTES

The Bullet Blender can generate consistent nano particles or liposomes.

Particle size of nanoparticles can be decreased by increasing homogenization time.



Scale: 50 µm



BULLET BLENDER® USER GUIDE REDUCE PARTICLE SIZE

> NOTES

The Bullet Blender can break down coarse material into a fine powder and break up clumps.

To finely grind material, use large stainless steel beads. To break up clumps, large stainless steel or zirconium oxide beads may be used.





CONSIDERATIONS BY SAMPLE TYPE

BULLET BLENDER® USER GUIDE PROTOCOL LIBRARY

> NOTES

Our experienced staff of molecular biologists have worked to provide you with a set of optimized protocols for the homogenization of various tissue, cell types, and organisms so you can spend less time troubleshooting and more time getting results.

Click on this <u>link</u> to get to the protocol page on our website.



BULLET BLENDER® USER GUIDE ORGAN TISSUE

> NOTES

Generally larger animals have "tougher" organs, so you may need to increase homogenization cycle times beyond protocol recommendations.

Connective tissue may remain unhomogenized in your samples but this does not reduce quality or yield.



BULLET BLENDER® USER GUIDE PLANT MATTER

> NOTES

For tough, fibrous plant tissue, consider using our UFO beads which are excellent for chopping fibers.

Some plant material homogenizes more efficiently if buffer volume is reduced.

Consider adding a dry homogenization step before homogenizing sample with buffer.





BULLET BLENDER® USER GUIDE DRIED GRAINS, NUTS AND BEANS

> NOTES

Hard samples may need precrushing. See specific <u>protocol</u> for details.

A 5 mL tube with specialized UFO stainless steel beads is the best choice for homogenizing most samples of this type.

Use <u>5 mL Bullet Blender models</u> for these conditions.







BULLET BLENDER® USER GUIDE TISSUE FROZEN IN LIQUID NITROGEN

> NOTES

Immerse the frozen tissue in cold buffer and allow it to thaw, then treat it as you would any other sample.

If the tissue was dehydrated before it was frozen, you can pulverize the tissue by homogenizing with beads only (no buffer), then adding the cold buffer and running again to complete the homogenization.

We do not recommend immersing the lysis tube in liquid nitrogen.

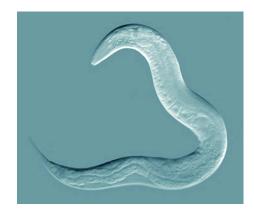


BULLET BLENDER® USER GUIDE SMALL ORGANISMS

> NOTES

Small soft organisms, like fruit flies and nematodes, can be homogenized in the same way tissue samples are.

- Place the samples into the tube dry, or centrifuge them at low speed and remove the supernatant if they were cultured in growth media.
- Add beads and buffer, flick the tubes lightly to resuspend if the samples are pelleted, and homogenize.





BULLET BLENDER® USER GUIDE INSECTS

> NOTES

Heavy-bodied insects, such as ticks, can be homogenized in the same way as animal tissue.

Some insects, such as small ticks, can float on the buffer surface.

To homogenize these samples, perform one run with just beads and sample, and then add buffer and rehomogenize to finish.





BULLET BLENDER® USER GUIDE HAIR

> NOTES

Pre-incubate hair with a demineralization buffer.

Do not densely pack hair into the sample tube. Overloading the tube will cushion the bead impacts and prevent good homogenization.

Hair is best homogenized dry. Buffer can be added after homogenization. Use 2.0 mm zirconium oxide beads and run at top speed.







BULLET BLENDER® USER GUIDE RESILIENT SAMPLES

➤ NOTES

Tough resilient samples include skin, umbilical cord, tendon, etc...

Shave skin samples to remove the hair.

Slice the sample into thin strips. Homogenize using stainless steel UFO beads.







BULLET BLENDER® USER GUIDE DEHYDRATED SAMPLES

> NOTES

Dehydrated samples can be homogenized dry, to form a powder, or wet. For best results with wet homogenization, rehydrate the sample fully before processing.

Efficiency will be improved if the sample is cut into thin strips or precrushed before homogenization.





BULLET BLENDER® USER GUIDE REFERENCES & PUBLICATIONS

> NOTES

The Bullet Blender has been used in a wide variety of applications for many years. Check our publication reference section to see if previous studies similar to yours are available. The Bullet Blender has been cited in over thousands of publications!

Click on this <u>link</u> to see the publications citing the Bullet Blender.

