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Manufacturer and Distributor of Scientific Equipment

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Mini-BeadBeater 96 Instructions

The Mini-BeadBeater 96 is high-energy, high-throughput cell disrupter. The Mini-BeadBeater 96 disrupts cells and tissue by violently shaking a sample inside a classic 2 ml microvial or standard deep well microplate partially filled with tiny ceramic or steel beads. Complete cell disruption is achieved in 2 to 3 minutes of beadbeating.

Our variable RPM controller allows greater operational flexibility of your Mini-BeadBeater 96.

This controller offers timing cycles from 0 to 5 minutes, and includes digital RPM control from 1400 to 2400 rpm. The controller also provides presets that allow for up to three time/speed combinations to be stored for easy retrieval.

Operating the Mini-BeadBeater 96

When using the microvial rack, insert a ***minimum of four*** microvials (vials may be empty if processing less than four samples) into the four corners of the rack. Failure to do so may result in damage to the instrument. Distribute additional tubes symmetrically. Place the rack of microvials or the microplate (if using 1 ml deep well microplates you can stack two at a time) into the chamber holder, top side toward you. Screw down the two black plastic knobs until the top stainless steel retainer plate is in contact with the microvial caps or the top of the microplate(s). Tighten knobs firmly, but do not over tighten. Finally, screw down the white nylon wing nuts.

Turn on the Mini-BeadBeater 96 with the rocker switch located at the appliance inlet at the rear of the machine and in front above the timer.

When power is applied to the controller it reads the last time and RPM values used and displays the time. Pressing the **[Start/Stop]** button will run another timing cycle for the preset speed to

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the motor controller. During the timing cycle the LED above the display flashes and the display changes back and forth from the countdown time and the programmed RPM. Pressing **[Start/Stop]** during the timing cycle stops the machine, shows the time, and blinks the LED display to show that it is paused. Pressing **[Start/Stop]** again continues the timing cycle. When paused, pressing and holding the **[Start/Stop]** button for 2 seconds will cause the controller to reset to the beginning of the cycle and it emits a double chirp.

Editing Time & Speed

The controller always operates from the current time and RPM value in memory. To edit the time and RPM values, simultaneously press the **[▲]** and **[▼]** buttons and the display shows Edit. Release the buttons and the controller displays the current time with the last digit highlighted

- Pressing the **[▲]** or the **[▼]** button will increase or decrease that digits value.
- Pressing the **◄[A]** button moves the highlight to the next digit to the left, if you are on the leftmost digit it will wrap around to the rightmost digit.
- Pressing the **[C]►** button moves the highlight to the next digit to the right, if you are on the rightmost digit it will wrap around to the leftmost digit.
- Once the time value is set press the **[Start/Stop]SET** button to edit the RPM value.

The RPM value is displayed with only the 100's value highlighted; the left and right arrows are not used when adjusting the RPM value. Use the **[▲]** or the **[▼]** buttons to change the RPM value, increasing the RPM value past the range limits will wrap around to the lowest value and vice versa. Once the RPM value is set, press the **[Start/Stop]SET** button to saves both RPM and time values.

With the time and RPM values set, press the **[Start/Stop]** to start the machine.

Using the Presets

The controller always operates from the latest time and RPM value in memory. The **[A]**, **[B]**, and **[C]** preset buttons can be used to save and recall three sets of values for future use.

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Recalling the saved time and RPM values is easy. Simply touch one of the preset buttons and the controller reads the saved values for that button, displays the time for one second, then the RPM for one second, and then returns to displaying the time. The controller is now ready to run a timing cycle by pressing the **[Start/Stop]** button.

Saving a time and RPM value is also a simple process. Edit the current time and speed per the previous section, then press and hold the preset button for two seconds until the controller chirps twice. The current time and RPM values are now saved to that preset.

Preparation of Sample

- Use 0.1 mm beads for bacteria, 0.5 mm beads for yeast, fungi, and tissue culture cells, and 1.0 mm beads for plant and animal tissue. Up to 400 mg wet weight of biomass can be disrupted per ml of liquid extraction media.* If you have solid tissue, pre-chop it into approximate 1 mm cubes using a scalpel, single-edge razor blade, or fragment the tissue at liquid nitrogen temperatures with a BioPulverizer (see www.biospec.com).

- Fill either 2 ml screw-cap microvials or 1 ml or 2 ml deep-well microplates one-half to two-thirds full with beads (BioSpec has a selection of bead loaders for vials and 96-well microplates which makes most loading easy and accurate, see https://biospec.com/category/bead-loaders for details). Bead size and composition should be matched to the biomaterial you are processing (see our Bead page at www.biospec.com for guidance).

- Then add your extraction media and cells, being sure to fill the microvials or wells almost to the top. Exclude as much air from the microvials or wells as is practical. We recommend using screw-cap microvials with o-ring seals – classic snap-top microvials release aerosol during shaking. Be sure there are no beads on the threads of the microvials or on the lip area of the microplate when sealing the microvials or microplate. If you are sealing your microplate with a flexible mat or with adhesive film, firmly press the mat or film over the entire top surface of the microplate.

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** Seeds and fibrous plant or animal material can be dry ground without added solvent. It is not necessarily a preferred method over wet grinding. Before beadbeating, samples are often pretreated by crushing, air- or freeze-drying, or freezing in liquid nitrogen. Two or three steel beads (6.4 or 3.2 mm diameter, respectively) are added to the vial containing the sample material. Shaking time is generally less than 20 seconds. We recommend our XXTuff or stainless steel microvials or Porvair microplates for all dry grinding applications. See https://biospec.com/category/vials-and-plates for details.*

Comments on Microvials, Beads, and Techniques

Microvials

All 1.5 ml and 2.0 ml microvials (conical, skirted, Eppendorf®, screw-cap, snap-top, etc.), but not some cryovials, can be used in the Mini-BeadBeater 96. The user should be aware that sealed snap-top type microvials still ***may release aerosol*** during the very high-energy shaking in a Mini-BeadBeater 96. We recommend using screw-cap microvials with a rubber o-ring in the cap. Even then, if the user is working with biohazardous material, the Mini-BeadBeater 96 should be operated in a bio-hood for safety.

Even smaller capacity microvials can be used. However, cell disruption may not be as efficient when using smaller microvials due to wall effects.

Beads

Do not acid wash the beads - It is a waste of time. If you must clean them (it is usually not necessary) do so with a detergent used to clean lab glassware. Rinse thoroughly, of course, and dry in an oven at about 50 C. Beads can be washed and reused about ten times. Beads can be sterilized in an autoclave or oven.

There is a quick, inexpensive way to rid beads of ***all*** nucleic acid and nuclease contamination. Soak the beads in a 1:10 dilution of ordinary household bleach (Clorox®) for one minute. See Biotechniques, Vol 12, No 3, p.358-360 (1992). Rinse thoroughly afterward.

Plates & Mats

The high-energy Mini-BeadBeater 96 is designed for microplates made of polypropylene or polycarbonate. Do not use plates made of polystyrene. Our testing has shown that Porvair

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Science brand plates and mats offer superior performance with regards to sealing ability and strength of any plates we have tested. See https://biospec.com/product/deep-well-micro_plates-and-mats.

Testing has also shown the Axygen Scientific plates and mats perform well. Microplates from other manufactures may also be acceptable. We welcome feed-back on this. We do know that mats made of polymer material other than silicone rubber do not seal well. Finally, microplates can be sealed with plastic or aluminum membranes. The integrity of some, but not all, of these membrane seals held up to shaking in the Mini-BeadBeater 96.

Temperature

During cell disruption, there will be a 10 degree increase in temperature per minute of operation. If you are isolating heat-sensitive material such as native proteins, consider pre-cooling your beads and sample, homogenizing for 1 minute only, and then removing the vials or plate and re-cooling in ice-water for 1 minute. Cycle in this manner for a total runtime of 2-3 minutes. Cooling is not needed when extracting nucleic acids in nucleic acid extraction media.

Another effective way to keep vials cold is to use a pre-chilled, solid aluminum vial holder. BioSpec offers two such holders for use with the Mini-BeadBeater 96. One for use with 2 ml microvials (see https://biospec.com/product/aluminum-vial-rack-2ml) and for use with 50 ml tubes (see https://biospec.com/product/aluminum-vial-holder-50ml).

The Mini-BeadBeater 96 is not designed to operate in a cold room. Cold air does little to cool the sample during homogenization.

Safety Concerns

The Mini-BeadBeater 96 will only operate with the black plastic hood lowered over the samples holder. A magnetic safety switch cuts all power to the Mini-BeadBeater 96 should the hood open during operation. The hood prevents the user from coming in contact with moving components and retains any component should it break free during operation.

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Trouble Shooting

If cell disruption does not meet expectations, there are several factors to consider.

1) The microvial must be filled *at least* half full of beads.
2) The rest of the volume should be filled with the extraction solution and sample - fill the microvial to the top.
3) Run for 3-5 minutes total "ON" time. If you are working with native proteins, see the Temperature section above.

When working with plant or animal tissue, pre-chop the tissue before attempting cell disruption. Strive for pieces smaller than 1 mm in cross-section. A popular option is to "powder" the tissue at liquid nitrogen temperatures (see our BioPulverizers, https://biospec.com/product/biopulverizer). Denser and/or larger beads can sometimes improve ***tissue*** cell disruption (see the Beads section https://biospec.com/category/lysis-beads-guidelines).

BioSpec Products is the inventor of the bead mill method of cell disruption. Please contact us if you need additional advice.