

### Bead Mill Tube Handbook for OHAUS Bead Mill Homogenizers



# The Right Tube for the Right Job

Having the correct type of grinding media is critical for successful lysing.This handbook includes the complete line of OHAUS Bead Mill tubes and describes their appropriate use for specific applications.

Look inside for NEW Lysing Tubes for OHAUS Homogenizers!



# **Ingeniously Practical**

### Contents

<i>Overview</i> 1
Bead Beating Tips1
Lysis Buffers
Yeast Lysing Bead Tubes2
Fungi Lysing Bead Tubes (cells and mycelia)3
Soft Sample Lysing Bead Tubes
Plant Lysing Bead Tubes4
Animal Tissue Lysing Bead Tubes4
Environmental Lysing Bead Tubes5
Lysing Bead Tube Sample Kit5
Safety and Handling Instructions5



### Overview

OHAUS' convenient, ready-to-use Lysing Bead Tubes are a widely-used tool for sample homogenization. Each 2 mL self-standing tube contains pre-measured molecular biology grade grinding media\* to effectively disrupt samples using high throughput homogenizers (e.g., reciprocating OHAUS HT Lysing Homogenizer), popular oscillating homogenizers, and smaller reciprocal bead beaters.

Pre-treated grinding beads and balls are processed to remove potential contaminants, including nucleic acids, nucleases, and proteases, which could negatively impact downstream applications\*. Quality control testing is performed to ensure that the lysing bead tubes meet OHAUS' strict quality requirements.

# **Bead Beating Tips**

#### **Lysis Buffers**

Bead beating is often performed using lysis buffers (i.e., "wet grinding").

Buffer composition changes significantly depending on the application. The components of those buffers can dramatically impact bead beating effectiveness. Buffers used to isolate DNA and RNA will typically contain detergents which can cause excessive foaming during bead beating that may impair the motion (and grinding mechanism) of the grinding beads. Increasing homogenization time can be used to compensate for foaming and improve homogenization. Detergents can often be added after the processing.

Small molecule or protein extractions may require the use of organic solvents (e.g., methanol, acetonitrile). Use caution when handling organic solvents. It is best to use organic solvent mixtures containing small amounts of water when bead beating. Heat buildup from the collision of beads can pressurize the tubes, particularly when organic solvents are used without dilution.

Stainless steel grinding balls may oxidize when exposed to organic solvents (e.g., phenol), in which case a suitable alternative, such as 3 mm zirconium beads in the Plant Lysing Bead Tubes, can be used.

#### If Making a Homogenization Buffer

- For **DNA** isolations, use of Tris-EDTA ("TE") is recommended to chelate magnesium ions, rendering DNases inactive.
- For RNA isolations, use of a reducing agent (e.g., β-mercaptoethanol, dithiothreitol) and a chaotrope (e.g., guanidine hydrochloride, guanidine thiocyanate) is recommended to denature RNases present in the sample.
- For Protein isolations, use of protease inhibitors is recommended.

#### **General Guidelines**

Do not overfill Lysis Bead Tubes. Overfilling the tubes will impede homogenization by limiting the motion of the grinding media. Total volume of sample, grinding media, and buffer should never exceed 50% of tube volume.

Homogenize in short bursts with pauses when homogenizing temperature-sensitive samples, such as proteins, to dissipate heat generated during grinding.

Do not use Lysing Bead Tubes for cryogenic grinding. Tubes will become brittle if dipped in liquid nitrogen.

\*Disclaimer: Not for clinical use. For research purposes only.



# **Bacteria Lysing Bead Tubes**

#### Product Number: 30391402 (100 each)

Contains: 100 µm Zirconium Beads Button Setting #1: 5 min. @ 1500 rpm

#### Protocol:

- 1. Spin down 2 mL of sample or cultured bacteria.
- 2. Decant the supernatant and resuspend the pellet in 500  $\mu l$  of a suitable buffer.
- 3. Transfer the solution to a Bacteria Lysis Bead Tube.
- 4. Homogenize for 5 minutes at 1500 rpm (press preset button).
- 5. Centrifuge for 5 minutes at  $12,000 \times g$ .
- 6. Transfer the supernatant to a clean tube and store/process.

#### May be used for: bacteria/spores

#### **Helpful Hints:**

- Increase the homogenization time if obtaining low DNA concentrations from gram positive bacteria.
- Assays can be performed to assess optimal homogenization conditions.

### Yeast Lysing Bead Tubes

#### Product Number: 30391404 (100 each)

Yellow

Contains: 400 µm Zirconium Beads Button Setting #1: 5 min. @ 1500 rpm

#### Protocol:

- 1. Spin down 1 mL of sample or cultured yeast.
- 2. Decant supernatant and resuspend the pellet in 500 µl of a suitable buffer.
- 3. Transfer the yeast solution to a Yeast Lysing Bead Tube.
- 4. Homogenize for 5 minutes at 1500 rpm (press preset button).
- 5. To remove debris, centrifuge for 5 minutes at  $12,000 \times g$ .
- 6. Transfer the supernatant to a clean tube for processing or storage.

#### May be used for: yeast, algae, spores

#### **Helpful Hints:**

- Longer processing times may be required to homogenize smaller yeasts (i.e. Pichia).
- Homogenization can be evaluated by comparing homogenized yeast to a non-homogenized sample under a microscope. Yeast cells that are cracked open will appear dark gray.



### Fungi Lysing Bead Tubes (cells and mycelia)

#### Product Number: 30391405 (100 each)

Contains: 800 μm Zirconium Beads Button Setting #1: 5 min. @ 1500 rpm

#### Protocol:

- Cultured fungal cells, pseudomycelia, and small pellicle formations can be processed with Fungi Lysis Bead Tubes. Samples can be concentrated by centrifugation and resuspended in lysing buffer, or, if solid, added directly to Fungi Lysing Bead Tubes. Sample volume should be approximately 50 µl. Add 500 µl of a suitable buffer.
- 2. Homogenize for 5 minutes at 1500 rpm (press preset button).
- 3. To remove debris, centrifuge for 5 minutes at  $12,000 \times g$ .
- 4. Transfer the supernatant to a clean tube and process or store.

May be used for: fungal mycelium, cells, spores, eukaryotic algae



Blue

### Soft Sample Lysing Bead Tubes

#### Product Number: 30391406 (100 each)

Contains: 1.4 mm Zirconium Beads Button Setting#1: 2 min. @ 1500 rpm

#### Protocol:

- For cultured cells or blood, centrifuge sample and decant supernatant. Resuspend the pellet in 500 µl of a suitable buffer and transfer to a Lysing Bead Tube. For solid samples, place approximately 50 mg in a Lysing Bead Tube and 500 µl of a suitable buffer. Homogenize for 2 minutes at 1500 rpm (press present button).
- 2. Remove debris by centrifuging for 5 minutes at  $12,000 \times g$ .
- 3. Transfer the supernatant to a clean tube for processing or storage.

May be used for: soft tissues (e.g., brain, adipose, liver, spleen); cultured cells and blood, as well as thin sections of fungal thallus and soft plant material







# Plant Lysing Bead Tubes

#### Product Number: 30391408 (100 each)

Contains: 3.0 mm Zirconium Beads Button Setting #3: 2 min. @ 1500 rpm

#### Protocol:

- 1. Add 50 to 70 mg sample to a Lysing Bead Tube along with 500  $\mu$ l of a suitable buffer.
- 2. Start with a 2 minute homogenization step at 1500 rpm (press preset button). Check the sample for lysis. If the sample isn't fully homogenized, repeat the homogenization.
- 3. Pellet cellular debris by centrifuging for 5 minutes at  $12,000 \times g$ .
- 4. Transfer the supernatant to a clean tube for processing or storage.

#### May be used for: plant, stems, roots, and leaves

#### **Helpful Hints:**

- Plant sample mass should not exceed 70 mg.
- If using a CTAB buffer, increase the homogenization time, as foaming may impair movement of the beads.
- If plant stem does not fully homogenize, use the stainless-steel Animal Tissue Lysing Bead Tubes, with 600 μl homogenization buffer.
- To homogenize plant pollen, use the Fungi Lysing Bead Tubes with 600  $\mu l$  homogenization buffer.
- Lysing Bead Tubes are not intended to homogenize seeds. Larger vials and grinding balls are better suited for this purpose.



### Animal Tissue Lysing Bead Tubes

#### Product Number: 30391409 (100 each)

Contains: 3 mm Stainless Steel Balls Button Setting #2: 2 min. @ 1500 rpm

#### Protocol:

- 1. Animal tissues can be extremely difficult to homogenize. Tissues high in collagen are very difficult. Hair is very difficult. For most tissues, place up to 50 mg sample in a Lysing Bead Tube. Add 500  $\mu$ l of a suitable buffer.
- 2. Homogenize for 2 minutes at 1500 rpm (press preset button). If the tissue isn't fully homogenized, repeat the homogenization step.
- 3. Remove insoluble debris by centrifuging for 5 minutes at  $12,000 \times g$ .
- 4. Transfer the supernatant to a clean tube for processing or storage.

May be used for: animal tissue, muscle, tumors, lungs, insects

#### **Helpful Hints:**

- Stainless Steel may oxidize when exposed to organic solvents in some buffers.
- Plant Lysing Bead Tubes may be substituted for Animal Tissue Lysing Bead Tubes, though slightly longer homogenization times may be necessary.





# **Environmental Lysing Bead Tubes**

#### Product Number: 30391410 (100 each)

Contains: Mixture of 100  $\mu m$  / 800  $\mu m$  / 3.0 mm Zirconium Beads Button Setting #4: 2 min. @ 1500 rpm

#### Protocol:

- 1. Add approximately 50 mg sample and 500  $\mu l$  of a suitable buffer to the Lysing Bead Tube.
- 2. Homogenize for 4 minutes at 1500 rpm (press preset button).
- 3. Centrifuge for 6 minutes at  $12,000 \times g$  to pellet debris.
- 4. Transfer the supernatant to a clean tube and process or store.

May be used for: environmental samples, including feces, soil, sediments, biofilms

#### Helpful Hints:

 Environmental sample types (e.g., soil, sediment, biofilms) can vary widely. Sample volume needs to be adjusted based upon the target biomolecule's concentration in the sample. The protocol used in the subsequent extraction procedure will also significantly influence sample size.

# Lysing Bead Tube Sample Kit

#### Product Number: 30391433 (14 each)

**Contains:** Two (2) Bacteria Lysing Bead Tubes, two (2) Yeast Lysing Bead Tubes, two (2) Fungi Lysing Bead Tubes, two (2) Soft Sample Lysing Bead Tubes, two (2) Plant Lysing Bead Tubes, two (2) Animal Tissue Lysing Bead Tubes, and two (2) Environmental Lysing Bead Tubes



### Safety and Handling Instructions

- Tubes should be stored tightly-capped in a cool, dry, well-ventilated area, protected from moisture and away from incompatible substances.
- Wash thoroughly after handling. Use with adequate ventilation and avoid ingestion and inhalation of tube contents.
- Do not leave tubes uncapped when not in use in order to avoid contamination.
- Do not submerge the tubes in liquid nitrogen.
- Please refer to your homogenizer manual and institution's safety guidelines/best practices to ensure that the tubes are safely loaded into the homogenizer.





#### **OHAUS Corporation**

Headquartered in Parsippany, NJ, OHAUS Corporation manufactures an extensive line of weighing scales, lab equipment and lab instruments that meet the weighing, sample processing and measurement needs of multiple industries. We are a global leader in the laboratory, industrial and education markets, as well as a host of specialty markets, including the food preparation, pharmacy and jewelry industries. An ISO 9001:2008 manufacturer, OHAUS lab balances, industrial scales, lab equipment and lab instruments are precise, reliable and affordable, and backed by industry-leading customer support.

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