



**ZYMO RESEARCH**

*The Beauty of Science is to Make Things Simple*

# INSTRUCTION MANUAL

## **Quick-DNA™ Fungal/Bacterial Midiprep Kit** Catalog No. **D6105**

### **Highlights**

- Simple, efficient isolation of DNA from all types of tough-to-lyse fungi (e.g., yeast) and bacteria in as little as 20 minutes.
- State-of-the-art, ultra-high density **BashingBeads™** are fracture resistant and chemically inert.
- Omits the use of organic denaturants as well as proteinases.

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Satisfaction of all Zymo Research products is guaranteed. If you should be dissatisfied with this product please call 1-888-882-9682.

## Product Contents

<b>Quick-DNA™ Fungal/Bacterial Midiprep Kit (Kit Size)</b>	<b>D6105 (25 preps.)</b>	<b>Storage Temperature</b>
<b>ZR BashingBead™ Lysis/Filtration Tubes</b>	25	Room Temp.
<b>Genomic Lysis Buffer<sup>1</sup></b>	2 x 100 ml	Room Temp.
<b>DNA Pre-Wash Buffer<sup>2</sup></b>	15 ml	Room Temp.
<b>g-DNA Wash Buffer</b>	50 ml	Room Temp.
<b>DNA Elution Buffer</b>	16 ml	Room Temp.
<b>Zymo-Spin™ V-E Columns w/ Zymo-Midi Filters™</b>	25	Room Temp.
<b>Collection Tubes</b>	50	Room Temp.
<b>Instruction Manual</b>	1	Room Temp.

Note - Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide maximal performance and reliability.

<sup>1</sup> For optimal performance, add beta-mercaptoethanol to 0.5%(v/v) i.e., 500 µl per 100 ml.

<sup>2</sup> A precipitate may have formed in the DNA Pre-Wash Buffer during shipping. To completely resuspend the buffer, incubate the bottle at 30 – 37 °C for 30 minutes and mix by inversion. DO NOT MICROWAVE.

## Specifications

- **Format** – Bead Beating, Spin/Vacuum Filtration, and Spin Column Purification
- **Sample Sources** – 250-500 mg (wet weight) fungi or bacteria or up to 1 g tissue. This equates to approximately  $5 \times 10^9$  bacterial cells,  $5 \times 10^8$  yeast cells and  $5 \times 10^7$  mammalian cells. Spores, pollen, nematodes, as well as other microorganisms can also be sampled.
- **DNA Purity** – High quality DNA is eluted with **DNA Elution Buffer** making it perfect for PCR.  $A_{260}/A_{280} \geq 1.8$
- **DNA Size Limits** – Capable of recovering genomic DNA sized fragments from 100 bp to  $\geq 40$  kb. Typical fragment sizes range from 25 kb-35 kb. If present, parasitic and viral DNA will also be recovered.
- **DNA Recovery** – Typically, up to ~125 µg total DNA is eluted into  $\geq 150$  µl **DNA Elution Buffer** per sample.
- **Equipment** – Centrifuge, Vacuum Source and Manifold, Microcentrifuge, Cell Disrupter/Pulverizer w/ 50 ml Tube Adapter

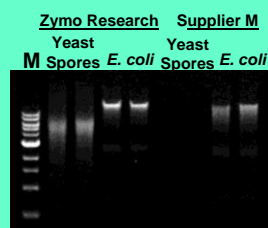
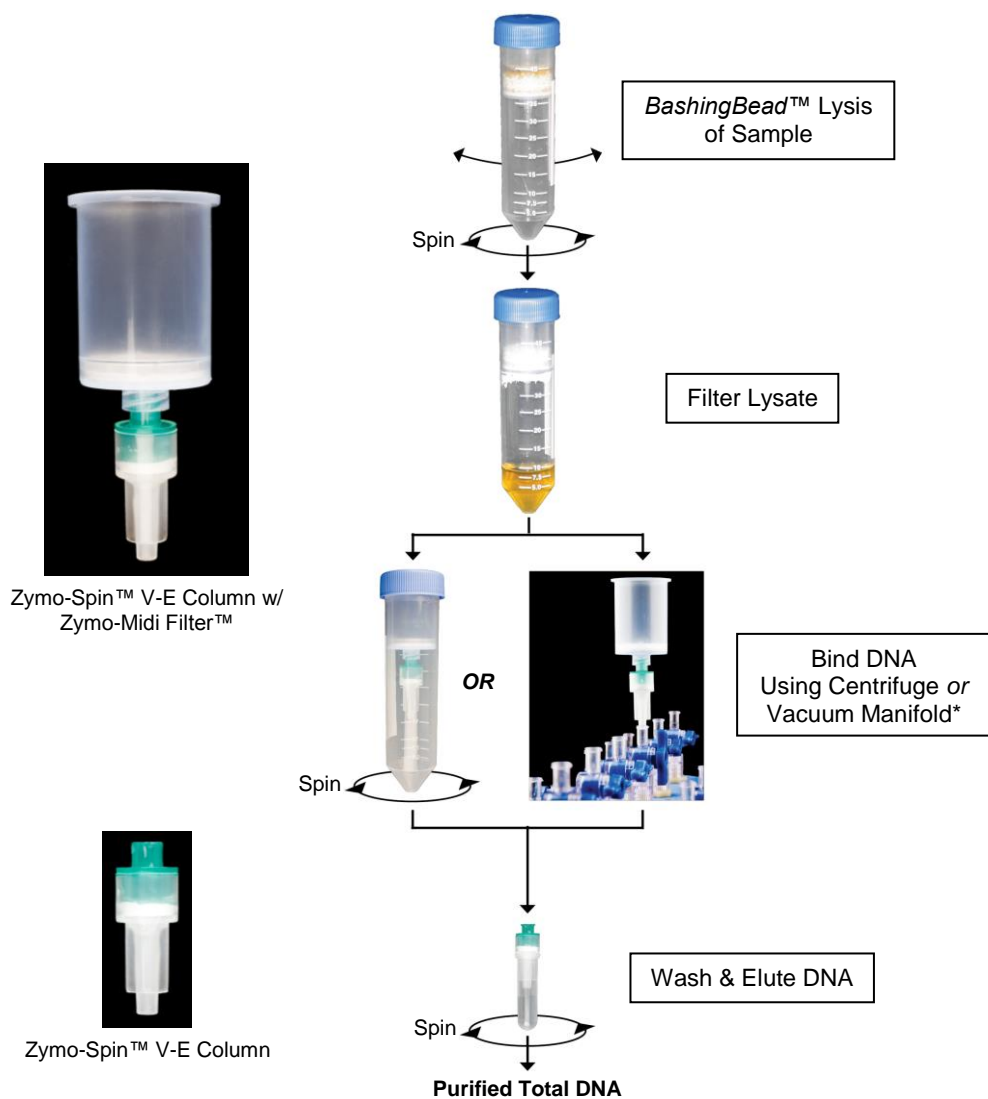
For purification of up to 25 µg DNA/prep use the **Quick-DNA™ Fungal/Bacterial Miniprep Kit (D6005)**. For high throughput purification (96-well) use the **Quick-DNA™ Fungal/Bacterial 96 Kit (D6006)**.

Note - ™ Trademarks of Zymo Research Corporation. This product is for research use only and should only be used by trained professionals. It is not intended for use in diagnostic procedures. Some reagents included with this kit are irritants. Wear protective gloves and eye protection. Follow the safety guidelines and rules enacted by your research institution or facility.

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## Product Description

The **Quick-DNA™ Fungal/Bacterial Midiprep Kit** is designed for the simple, rapid isolation of DNA from tough-to-lyse fungi, including *A. fumigatus*, *C. albicans*, *N. crassa*, *S. cerevisiae*, *S. pombe*, as well as from mycelium and Gram (+) and (-) bacteria. The procedure is easy and can be completed in as little as 20 minutes: fungal and/or bacterial samples are added directly to a **ZR BashingBead™ Lysis/Filtration Tube** and rapidly and efficiently lysed by bead beating without using organic denaturants or proteinases. The DNA is isolated and purified using our *Fast-Spin* column technology and is ideal for downstream molecular-based applications including PCR, array, etc. A schematic of the **Quick-DNA™ Fungal/Bacterial Midiprep Kit** procedure is shown below.



High yield DNA is successfully isolated from *Saccharomyces cerevisiae* (spores) and *E. coli* cells using the **Quick-DNA™ Fungal/Bacterial Kit**. Equivalent amounts of yeast or bacteria were processed using the **Quick-DNA™ Fungal/ Bacterial Kit** or the kit from supplier M. Equal volumes of eluted DNA were then analyzed in a 0.8% (w/v) agarose/ethidium bromide gel. The size marker "M" is a 1 kb ladder (Zymo Research).

For **Technical Assistance**, please contact those at **Zymo Research's Technical Department** at 1-888-882-9682 or E-mail to [tech@zymoresearch.com](mailto:tech@zymoresearch.com).

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## Protocol

For optimal performance, add beta-mercaptoethanol (user supplied) to the **Genomic Lysis Buffer** to a final dilution of 0.5%(v/v) i.e., 500 µl per 100 ml.

### Notes:

<sup>1</sup> This equates to approximately  $5 \times 10^9$  bacterial cells,  $5 \times 10^8$  yeast cells or  $5 \times 10^7$  mammalian cells.

<sup>2</sup> Cap tube tightly to prevent leakage.

<sup>3</sup> Be careful to avoid the pelleted material at the bottom of the tube when transferring the supernatant.

<sup>4</sup> Alternatively, the **Zymo-Spin™ V-E Column/Zymo-Midi Filter™** assembly can be placed in a 50 ml tube and centrifuged at 2,000 x g max. for 5 minutes. Filtration of the entire mixture will require several spins. Empty the flow through from the tube after each spin. **CAUTION: Make sure the connection between the column and filter is secure (finger tight) prior to centrifugation!**

<sup>5</sup> Leave the rotor cover off the microcentrifuge if clearance with the column tops is a problem.

<sup>6</sup> DNA yields can be increased by performing a second elution and pooling the eluates.

1. Add 250-500 mg (wet weight) fungal or bacterial cells that have been resuspended in up to 1 ml of water or isotonic buffer (e.g., PBS) or up to 1 g of tissue to a **ZR BashingBead™ Lysis/Filtration Tube**<sup>1</sup>. Add 6 ml **Genomic Lysis Buffer** to the sample, cap tube<sup>2</sup>, and process.

*(To prevent the **Lysis Solution** from leaking into the bottom of the 50 ml tube, place the **ZR BashingBead™ Lysis/Filtration Tube** on its side prior to processing).*

2. Secure in a bead beater fitted with a 50 ml tube holder assembly (see page 5) to process samples. Optimization of processing time/speed will be necessary for complete sample lysis.

Processing times may be as little as 40 seconds when using high-speed cell disrupters (e.g., FastPrep®-24, Geno/Grinder®, page 5). See manufacturer's literature for operating information.

3. Centrifuge the **ZR BashingBead™ Lysis/Filtration Tube** in a centrifuge at  $\geq 3,000 \times g$  ( $5,000 \times g$  max.) for 5 minutes.
4. Filter the entire mixture from Step 3 using a **Zymo-Spin™ V-E Column/Zymo-Midi Filter™** assembly mounted on a vacuum manifold<sup>3,4</sup> (see diagram on page 2) with a vacuum source set at  $\geq 600$  mm Hg.
5. Disconnect the **Zymo-Spin™ V-E Column/Zymo-Midi Filter™** assembly and transfer the **Zymo-Spin™ V-E Column** to a **Collection Tube**. Spin the column at  $10,000 \times g$  for 1 minute in a microcentrifuge<sup>5</sup>, then add 300 µl **DNA Pre-Wash Buffer** to the column and spin at  $10,000 \times g$  for 1 minute. Discard the flow through.
6. Add 400 µl **g-DNA Wash Buffer** to the column and centrifuge at  $10,000 \times g$  for 1 minute. Discard flow through and repeat wash step.
7. Transfer the **Zymo-Spin™ V-E Column** to a 1.5 ml microcentrifuge tube and add 150 µl **DNA Elution Buffer** directly to the column matrix<sup>6</sup> and allow column to stand for 1 minute at room temperature. Centrifuge at  $10,000 \times g$  for 1 minute to elute the DNA.

Ultra-pure DNA is now ready for use in your experiments.

**Ordering Information**

Product Description	Catalog No.	Kit Size
<b>Quick-DNA™ Fungal/Bacterial Miniprep Kit</b>	D6005	50 preps.
<b>Quick-DNA™ Fungal/Bacterial 96 Kit</b>	D6006	2x96 preps.
<b>Quick-DNA™ Fungal/Bacterial Midiprep Kit</b>	D6105	25 preps.

For Individual Sale	Catalog No.	Amount
<b>ZR BashingBead™ Lysis/Filtration Tubes (50 ml) w/ 0.5 mm Beads</b>	S6010	25 tubes
<b>Genomic Lysis Buffer</b>	D3004-1-100	100 ml
<b>DNA Pre-Wash Buffer</b>	D3004-5-15	15 ml
<b>g-DNA Wash Buffer</b>	D3004-2-50	50 ml
<b>DNA Elution Buffer</b>	D3004-4-16	16 ml
<b>Zymo-Spin™ V-E Columns w/ Zymo-Midi Filters™</b>	C1021-25	25 columns/filters
<b>Collection Tubes</b>	C1001-50	50 tubes
	C1001-500	500 tubes
	C1001-1000	1,000 tubes

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## Compatible Lysis Instruments...

### Bullet Blender™



**NEXT >>> ADVANCE**

Homogenize tissue or disrupt/lyse cells in minutes. **The Bullet Blender™**, is a vortexer (at a low setting), a cell disruptor and a tissue homogenizer (at a high setting) all in one unit. No parts contact the samples, eliminating any possibility of cross contamination.

Description	Cat No.
<b>DX50B Bullet Blender™ Blue 50</b> Accommodates 9 x 50 ml tubes. Features fan cooling.	S6007-1

### FastPrep®



The **FastPrep®-24 Instrument** is a unique, high-speed benchtop homogenizer that employs a powerful, proprietary technology for the rapid lysis of almost any sample in 40 seconds or less. The FastPrep® Instrument makes it possible to isolate DNA, RNA and protein from sources that are virtually impossible to lyse without the use of its rapid reciprocating motion.

Description	Cat No.
<b>FastPrep® Instrument</b>	S6005
<b>BigPrep™ Attachment</b> Accommodates 2 x 50 ml tubes.	S6005-4



### 2010 Geno/Grinder®



The **2010 Geno/Grinder Instrument** is a unique instrument that provides vigorous up-and-down grinding/pulverizing action. The Geno/Grinder instrument makes it possible to prepare plant materials such as seeds, stems, roots, leaves, and certain animal tissue. Can accommodate (2) 96-well plates/blocks for high-throughput sample processing.

Description	Cat No.
<b>2010 Geno/Grinder Instrument</b>	S6006
<b>50 ml Tube Holder/Cryo Block Assembly</b> Accommodates 12 x 50 ml tubes/block.	S6006-3



GenoGrinder and accessories for sale in USA only. Visit [www.spexcsp.com](http://www.spexcsp.com) for a distributor near you.

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