

INSTRUCTION MANUAL

ZR Soil Microbe DNA KitTM Catalog No. **D6001**

Highlights

- Simple, efficient isolation of humic-free DNA from soil microbes in as little as 15 minutes including tough-to-lyse bacteria, fungi, algae, and protozoa.
- State-of-the-art, ultra-high density **BashingBeads**[™] are fracture resistant and chemically inert.
- Can be used with <u>any</u> bead mill, disrupter, or vortex that can accommodate standard 2.0 ml tubes.
- Omits the use of organic denaturants as well as proteinases.

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For Research Use Only

Ver. 1.0.4

Product Contents

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

ZR Soil Microbe DNA Kit™ (Kit Size)	D6001 (50 preps.)	Storage Temperature
ZR BashingBead™ Lysis Tubes	50 tubes	Room Temp.
Soil DNA Binding Buffer	100 ml	Room Temp.
DNA Pre-Wash Buffer*	15 ml	Room Temp.
Soil DNA Wash Buffer	50 ml	Room Temp.
DNA Elution Buffer	10 ml	Room Temp.
Zymo-Spin™ IV Spin Filters (Orange Tops)	50 filters	Room Temp.
Zymo-Spin™ IV-HRC Spin Filters (Green Tops)	50 filters	Room Temp.
Zymo-Spin™ IIC Columns	50 columns	Room Temp.
Collection Tubes	200 tubes	Room Temp.
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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.

* 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 Column.
- **Sample Sources** DNA is isolated from bacteria, fungi, protozoa, and algae in up to 0.25 g of soil or from fungal/bacterial cells directly (see protocol).
- DNA Purity High quality, humic-free DNA is eluted with DNA Elution Buffer making it perfect for PCR. Absorbance_{260 nm/280 nm} > 1.8
- DNA Size Limits > 1 kb
- DNA Recovery Typically, up to 25 μg total DNA is eluted into 100 μl (25 μl minimum) DNA Elution Buffer per sample. For DNA 75 bp to 10 kb, the recovery is 70-90%. For DNA 11 kb to 23 kb the recovery is 50-70%.
- **Equipment** Microcentrifuge, vortex, cell disrupter/pulverizer (optional)

Note - TM Trademarks of Zymo Research Corporation. This product is for research use only and should only be used by trained professionals. 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.

Product Description

The **ZR Soil Microbe DNA Kit**TM is designed for the simple, rapid isolation of humic-free, PCR-quality DNA from microbes in soil. The kit can be used to successfully isolate DNA from tough-to-lyse bacteria, fungi, protozoa, and algae that inhabit a range of samples including clay, sandy, silty, peaty, chalky, and loamy soils. Soil samples are added to a **ZR BashingBead**TM Lysis Tube where microbes are rapidly and efficiently lysed by bead beating in a uniquely designed lysis buffer. Our *Fast-Spin* column technology is then used to isolate the DNA which is subsequently filtered to remove humic acids/polyphenols that inhibit PCR. The entire procedure can be performed in as little as 15 minutes, and there is no need for organic denaturants or proteinases. The **ZR Soil Microbe DNA Kit**TM can also be used to successfully isolate DNA from cultured bacteria, fungi, and yeast. A schematic of the procedure is shown below.





amounts of yeast were processed using the **ZR Soil Microbe DNA KitTM** or the kits from suppliers M and E. Equal volumes of eluted DNA were then analyzed in a 0.8% (w/v) agarose/ethidium bromide gel. The size marker is a 1 kb ladder (NEB).

References:

please contact those at

Technical Department at 1-888-882-9682 or E-mail to

tech@zymoresearch.com.

Zymo Research's

1. Soil and Plant Laboratory, Inc. P.O. Box 11744, Santa Ana, California 92711

Protocol

<u>Before Starting</u>: (soil samples only) **Zymo-Spin**TM **IV-HRC Spin Filters** (green tops) need to be prepared prior to use by: 1) snapping off the base, 2), inserting into a **Collection Tube**, and 3), spinning in a microcentrifuge at exactly $8,000 \times g$ for 3 minutes.

1. Add up to 0.25 grams of soil sample to a **ZR BashingBead™ Lysis Tube**.

Alternatively, add 50-100 mg (wet weight) fungal cells that have been resuspended in up to 200 µl of water or isotonic buffer (e.g., PBS) or up to 200 mg of tissue to a **ZR BashingBead™ Lysis Tube**.

2. Secure in a bead beater fitted with a 2.0 ml tube holder assembly (e.g., Scientific Industries' Disruptor Genie[™], <u>Cat. No. S6001-2 from Zymo Research Corp.</u>) and process at maximum speed for 5 minutes.

Processing times may be as little as 1 minute when using high-speed cell disrupters (e.g., FastPrep[®] and Precellys[®]). See manufacturers' literature for operating information.

- 3. Centrifuge the **ZR BashingBead™ Lysis Tube** in a microcentrifuge at ≥10,000 x *g* for 1 minute.
- 4. Transfer up to 400 µl supernatant to a **Zymo-Spin™ IV Spin Filter** (orange top) in a **Collection Tube** and centrifuge at 7,000 rpm (~7,000 x g) for 1 minute.
- Add 1,200 µl of Soil DNA Binding Buffer to the filtrate in the Collection Tube from Step 4.
- 6. Transfer 800 µl of the mixture from Step 5 to a **Zymo-Spin™ IIC Column** in a **Collection Tube** and centrifuge at 10,000 x g for 1 minute.
- 7. Discard the flow through from the **Collection Tube** and repeat Step 6.
- 8. Add 200 µl **DNA Pre-Wash Buffer** to the **Zymo-Spin™ IIC Column** in a new **Collection Tube** and centrifuge at 10,000 x g for 1 minute.
- 9. Add 500 µl **Soil DNA Wash Buffer** to the **Zymo-Spin™ IIC Column** and centrifuge at 10,000 x *g* for 1 minute.
- 10. Transfer the **Zymo-Spin™ IIC Column** to a clean 1.5 ml microcentrifuge tube and add 100 µl **DNA Elution Buffer** directly to the column matrix. Centrifuge at 10,000 x g for 30 seconds to elute the DNA.

If fungi or bacterial cultures were sampled, the DNA is now suitable for PCR as well as other downstream applications.

11. Transfer the eluted DNA from Step 10 to a prepared **Zymo-Spin™ IV-HRC Spin Filter** (green top) (see above) in a clean 1.5 ml microcentrifuge tube and centrifuge at exactly 8,000 x g for 1 minute. The filtered DNA is now suitable for PCR and other downstream applications.

Disruptor Genie[™] is a trademark of Scientific Industries, Inc. FastPrep[®] is a registered trademark of Qbiogene, Inc. Precellys[®] is a registered trademark of Bertin Technologies. Cap tube tightly to prevent leakage.

Alternatively, a standard bench top vortex can be used although the overall yield of DNA may be lower.

Snap off the base of the Zymo-Spin™ IV Spin Filter prior to use.

The **Zymo-Spin™ IIC Column** has a maximum capacity of 800 µI.

In some cases a browncolored pellet may form at the bottom of the tube after centrifugation. Avoid this pellet when collecting the eluted DNA.

Product Description	Catalog No.	Kit Size
ZR Soil Microbe DNA Kit™	D6001	50 preps.
For Individual Sale	Catalog No.	Amount
BashingBead™ Lysis Tubes (dry beads only)	S6002-50	50 tubes
Soil DNA Binding Buffer	D6001-1-100	100 ml
DNA Pre-Wash Buffer	D3004-5-15	15 ml
Soil DNA Wash Buffer	D6001-2-50	50 ml
DNA Elution Buffer	D3004-4-10	10 ml
Zymo-Spin™ IV Spin Filters (Orange Tops)	C1007-50	50 filters
Zymo-Spin™ IV-HRC Spin Filters (Green Tops)	C1010-50	50 filters
Zymo-Spin™ IIC Columns	C1011-50	50 columns
Collection Tubes	C1001-50 C1001-500 C1001-1000	50 tubes 500 tubes 1,000 tubes
Disruptor Genie™, 120V w/ 2.0 ml Tube Holder Assembly	S6001-2	1 unit
TurboMix Attachment, 2.0 ml (Permanently mounts to most existing Vortex-Genie 2 and Vortex-Genie 2T mixers converting them to a Disruptor Genie [™] .	S6004-2	1 unit



The **Disruptor Genie™** with 2.0 ml Tube Holder Assembly from Scientific Industries, Inc. (Cat. No. S6001-2 from Zymo Research Corp.)

Popular DNA Purification & Analysis Products from Zymo Research

Product	Description	Kit Size (Preps)	Catalog No. (column format)
DNA Clean & Concentrator-5™	Clean & concentrate DNA from any reaction or "crude" preparation in 2 minutes. A 6 μ l minimum elution volume allows for highly concentrated DNA. Designed for samples containing up to 5 μ g of DNA.	50 200 50 200	D4003 (uncapped) D4004 (uncapped) D4013 (capped) D4014 (capped)
DNA Clean & Concentrator-25™	Clean & concentrate DNA in minutes. 25 μl minimum elution volume allows for highly concentrated DNA. Designed for purifying up to 25 μg of DNA.	50 200 50 200	D4005 (uncapped) D4006 (uncapped) D4033 (capped) D4034 (capped)
DNA Clean & Concentrator-100™	Clean & concentrate DNA in minutes. 100 μI minimum elution volume allows for highly concentrated DNA. Designed for purifying up to 100 μg of DNA.	25 50	D4029 D4030
DNA Clean & Concentrator-500™	Clean & concentrate DNA in minutes. 1 ml minimum elution volume allows for highly concentrated DNA. Designed for samples containing up to 500 μg of DNA.	10 20	D4031 D4032
ZR-96 DNA Clean & Concentrator-5™	Quick (15 minute), high-output recovery of pure DNA from PCR, endonuclease digestions, plasmid preparations, etc. 10-15 μ I minimum elution volume allows for highly concentrated DNA. Designed for samples containing up to 5 μ g of DNA.	2x96 4x96	D4023 D4024
Zymoclean™ Gel DNA Recovery Kit	Purify DNA from high and low-melting agarose gels in minutes	50 200	D4001 D4002
ZR-96 Zymoclean™ Gel DNA Recovery Kit	High-throughput DNA purification from high and low-melting agarose gels.	2x96 4x96	D4021 D4022
Pinpoint Slide DNA Isolation System™	Recover genomic DNA from paraffin-embedded or fresh tissue sections for PCR. Ideal for isolating DNA from clinical tissue samples.	50	D3001
Zyppy™ Plasmid Miniprep Kit	Pellet-Free™ plasmid DNA purification in minutes: (alkaline lysis/spin column format for low 30 µl elution volume).	50 100 400	D4036 D4019 D4020
Zyppy™ Plasmid Midiprep Kit	Pellet-Free [™] plasmid DNA purification in minutes: (alkaline lysis/spin column format and 150 µl minimum elution volume).	25 50	D4025 D4026
Zyppy™ Plasmid Maxiprep Kit	High-purity plasmid DNA purification in minutes: (alkaline lysis/spin column format and 2 ml minimum elution volume).	10 20	D4027 D4028
ZR Genomic DNA I Kit™	Genomic DNA isolation from whole blood, tissue culture cells, solid tissue and liquid samples. (Silica bead format is scalable to fit your requirements).	100 400	D3004 D3005
ZR Genomic DNA II Kit™	Genomic DNA purification from whole blood, tissue culture cells, solid tissue and liquid samples. No requirement for beads or phenol chloroform.	50 200 50 200	D3006 (uncapped) D3007 (uncapped) D3024 (capped) D3025 (capped)
ZR-96 Genomic DNA Kit™	High-output genomic DNA purification from whole blood, tissue culture cells, solid tissue and liquid samples. No requirement for beads or phenol chloroform.	2x96 4x96	D3010 D3011
ZR Soil microbe DNA Kit™	Simple, rapid isolation of humic-free, PCR-quality genomic DNA from soil microbes.	50	D6001
ZR Fungal/Bacterial DNA Kit™	Simple, rapid isolation of PCR-quality genomic DNA from fungi.	50	D6005
ZR Fecal DNA Kit™	Simple, rapid isolation of PCR-quality genomic DNA from feces.	50	D6010
ZR Viral DNA Kit™	Isolation of viral DNA from cell-free body fluids or sample mixtures containing cells at concentrations less than 10^5 cells per ml.	50 200	D3015 D3016
ZR-96 Viral DNA Kit™	High-output (96-well) isolation of viral DNA from cell-free body fluids or sample mixtures containing cells at concentrations less than 10^5 cells per ml.	2x96 4x96	D3017 D3018
EZ DNA Methylation™ Kit	Streamlined kit for the conversion of unmethylated cytosines in DNA to uracil via the <u>chemical-denaturation</u> of DNA using our specially designed CT Conversion Reagent [™] . DNA is then desulphonated and subsequently cleaned using <i>Fast-Spin</i> column technology. Ultra-pure recovered DNA can be used for PCR and bisulfite sequencing applications.	50 200 2x96	D5001 D5002 D5003
EZ DNA Methylation- Gold™ Kit	Streamlined kit for the conversion of unmethylated cytosines in DNA to uracil via <u>heat-denaturation</u> of DNA using our specially designed CT Conversion Reagent [™] . DNA is then desulphonated and subsequently cleaned using <i>Fast-Spin</i> column technology. Ultra-pure recovered DNA can be used for PCR and bisulfite sequencing applications. <i>3 hour processing time!</i>	50 200 2x96	D5005 D5006 D5007

^BUlk quantities are available upon request. Please contact: <u>busdev@zymoresearch.com</u> or call 1-888-882-9682 for assistance.