



ZYMO RESEARCH

The Beauty of Science is to Make Things Simple

INSTRUCTION MANUAL

ZR Fungal/Bacterial DNA MiniPrep™

Catalog No. **D6005**

Highlights

- Simple, efficient isolation of DNA (up to 25 µg/prep) from all types of tough-to-lyse fungi (e.g., yeast) and bacteria in as little as 15 minutes.
- State-of-the-art, ultra-high density **BashingBeads™** are fracture resistant and chemically inert.
- Can be used with any bead mill, disrupter, or vortex that can accommodate standard 2 ml tubes.
- 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.

¹The ZR BashingBead™ Lysis Tubes (0.5 mm) Cat # S6002-50, is still available for individual purchase.

Product Contents

ZR Fungal/Bacterial DNA MiniPrep™ (Kit Size)	D6005 (50 preps.)	Storage Temperature
ZR BashingBead™ Lysis Tubes (0.1 & 0.5 mm)¹	50	Room Temp.
Lysis Solution	40 ml	Room Temp.
Fungal/Bacterial DNA Binding Buffer²	100 ml	Room Temp.
DNA Pre-Wash Buffer³	15 ml	Room Temp.
Fungal/Bacterial DNA Wash Buffer	50 ml	Room Temp.
DNA Elution Buffer	10 ml	Room Temp.
Zymo-Spin™ IV Spin Filters (Orange Tops)	50	Room Temp.
Zymo-Spin™ IIC Columns	50	Room Temp.
Collection Tubes	150	Room Temp.
Instruction Manual	1	-

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.

² For optimal performance, add beta-mercaptoethanol to 0.5%(v/v) i.e., 500 µl per 100 ml.

³ 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 Purification
- **Sample Sources** – 50 - 100 mg (wet weight) fungi or bacteria or up to 200 mg tissue. This equates to approximately 10⁹ bacterial cells, 10⁸ yeast cells and 10⁷ 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 ($A_{260}/A_{280} > 1.8$).
- **DNA Size Limits** – Capable of recovering genomic DNA up to and above 40 kb. In most instances, mitochondrial DNA and viral DNA (if present) will also be recovered.
- **DNA Recovery** – Typically, up to 25 µg total DNA is eluted into 100 µl (35 µl minimum) **DNA Elution Buffer** per sample.
- **Equipment** – Microcentrifuge, Vortex, Cell Disrupter/Pulverizer (recommended)

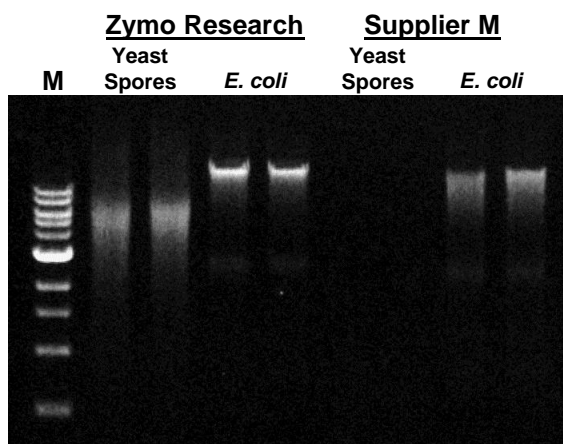
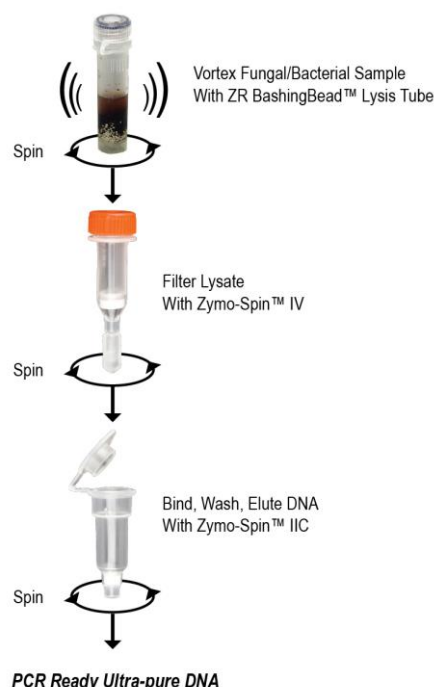
Note - ™ Trademarks of Zymo Research Corporation. This product is for research use only and should only be used by trained professionals. It is not intended for use in diagnostic procedures. Some reagents included with this kit are irritants. Wear protective gloves and eye protection. Follow the safety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

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Product Description

The **ZR Fungal/Bacterial DNA MiniPrep™** 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 15 minutes: fungal and/or bacterial samples are added directly to a **ZR BashingBead™ Lysis Tube (0.1 & 0.5 mm)** and rapidly and efficiently lysed by bead beating (e.g., *Xpedition™* Sample Processor, Page 5) without using organic denaturants or proteinases. The DNA is isolated and purified using our Zymo-Spin™ Technology and is ideal for downstream molecular-based applications including PCR, array, etc. A schematic of the **ZR Fungal/Bacterial DNA MiniPrep™** procedure is shown below.



High yield DNA is successfully isolated from *Saccharomyces cerevisiae* (spores) and *E. coli* cells using the **ZR Fungal/Bacterial DNA MiniPrep™**. Equivalent amounts of yeast or bacteria were processed using the **ZR Fungal/Bacterial DNA MiniPrep™** 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).

Zymo Research offers the following for nucleic acid stabilization, DNA isolation, and RNA isolation.

1) **DNA/RNA Shield™** (R1100-50, R1100-250) can be used to stabilize nucleic acids and inactivate infectious agents in a variety of samples, without the need for reagent removal.

2) **Xpedition™ Fungal/Bacterial DNA MiniPrep** (D6206) incorporates DNA stabilization technology to stabilize fungal/bacterial samples collected in the field, as a part of the DNA isolation workflow.

3) **ZR Fungal/Bacterial RNA MiniPrep™** (R2014) utilizes *BeadBashing* technology, RT-PCR inhibitor removal, and Zymo-Spin Technology for isolation of ultra-pure total RNA from fungal/bacterial samples.

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

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Protocol

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

¹ This equates to approximately 10⁹ bacterial cells, 10⁸ yeast cells or 10⁷ mammalian cells.

²Cap tube tightly to prevent leakage.

1. Add 50 – 100 mg (wet weight) fungal or bacterial cells¹ 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 (0.1 mm & 0.5 mm)**. Add 750 µl **Lysis Solution** to the tube².
2. Secure in a bead beater fitted with a 2 ml tube holder assembly and process at maximum speed for ≥ 5 minutes.

***Note:** Required processing time will vary depending on the device and application and therefore should be evaluated on a case by case basis.*

For example, processing times may be as little as 3 minutes when using high-speed cell disrupters (e.g., the portable TerraLyzer™ Sample Processor, FastPrep® -24, or similar) or as long as 20 minutes when using lower speeds (e.g., Disruptor Genie™, or standard benchtop vortexes). See manufacturer's 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 x g for 1 minute.

***Note:** Snap off the base of the Zymo-Spin™ IV Spin Filter (Orange Top) prior to use.*

5. Add 1,200 µl of **Fungal/Bacterial 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 Column³** 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 **Fungal/Bacterial 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 (35 µl minimum) **DNA Elution Buffer** directly to the column matrix. Centrifuge at 10,000 x g for 30 seconds to elute the DNA.

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

³ The Zymo-Spin™ IIC Column has a maximum capacity of 800 µl.

Ordering Information

Product Description	Catalog No.	Kit Size
ZR Fungal/Bacterial DNA MicroPrep™	D6007	50 preps.
ZR Fungal/Bacterial DNA MiniPrep™	D6005	50 preps.
ZR Fungal/Bacterial DNA MidiPrep™	D6105	25 preps.
ZR-96 Fungal/Bacterial DNA Kit™	D6006	2x96 preps.

For Individual Sale	Catalog No.	Amount
Fungal/Bacterial DNA Binding Buffer	D6005-1-100	100 ml
Lysis Solution	D6001-3-40	40 ml
DNA Pre-Wash Buffer	D3004-5-15	15 ml
Fungal/Bacterial DNA Wash Buffer	D6005-2-50	50 ml
DNA Elution Buffer	D3004-4-10	10 ml
ZR BashingBead™ Lysis Tubes (0.1 & 0.5 mm) ¹	S6012-50	50
Zymo-Spin™ IV Spin Filters (Orange Tops)	C1007-50	50
Zymo-Spin™ IIC Columns	C1011-50	50
Collection Tubes	C1001-50	50
	C1001-500	500
	C1001-1000	1,000

¹ The ZR BashingBead™ Lysis Tubes (0.5 mm) Cat # S6002-50, is still available for individual purchase.

Lysis Instruments



Description	Cat. No.	Amount
Disruptor Genie™, 120V w/ 2 ml tube holder assembly.	S6001-2-120	1 unit
Disruptor Genie™, 230V w/ 2 ml tube holder assembly.	S6001-2-230	1 unit
TurboMix Attachment, 2 ml Permanently mounts to most existing Vortex Genie™ mixers converting them to a Disruptor Genie™.	S6004	1 unit

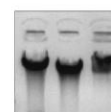
The **Disruptor Genie™** with 2 ml tube holder from Scientific Industries, Inc. (Cat. No. S6001-2-120 from Zymo Research Corp.)

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Xpedition™ Sample Prep & Preservation Technologies: The Complete Solution to “TAKE THE LAB TO THE FIELD”

Workflow (e.g., DNA Purification)



Lyse/stabilize samples during your expedition
using Xpedition™ Sample Processor & Lysis/Stabilization Solution

Store/transport lysates
without refrigeration

Process *later* using
Xpedition™ DNA Prep Kits

Intact, inhibitor-free DNA
ready for analysis!

For Portable Sample Lysis...



S6020

Xpedition™ Sample Processor

1 unit

D6202

Xpedition™ Soil/Fecal DNA MiniPrep

50 preps.

D6206

Xpedition™ Fungal/Bacterial DNA MiniPrep

50 preps.

D6216

Xpedition™ Tissue & Insect DNA MiniPrep

50 preps.

D6221

Xpedition™ Plant/Seed DNA MiniPrep

50 preps.

D6202-1-40

Xpedition™ Lysis/Stabilization Solution

40 ml

All Xpedition™ DNA MiniPrep Kits feature a unique lysis/stabilization solution. This allows high quality DNA to be isolated immediately or when convenient for the researcher following storage/transport of sample lysates.

Other Popular Environmental DNA/RNA Purification Products

ENVIRONMENTAL DNA

	D6003	ZR Soil Microbe DNA MicroPrep™	50 preps.
	D6001	ZR Soil Microbe DNA MiniPrep™	50 preps.
	D6101	ZR Soil Microbe DNA MidiPrep™	25 preps.
	D6002	ZR-96 Soil Microbe DNA Kit™	2 x 96 preps.
	D6007	ZR Fungal/Bacterial DNA MicroPrep™	50 preps.
	D6005	ZR Fungal/Bacterial DNA MiniPrep™	50 preps.
	D6105	ZR Fungal/Bacterial DNA MidiPrep™	25 preps.
	D6006	ZR-96 Fungal/Bacterial DNA Kit™	2 x 96 preps.
	D6012	ZR Fecal DNA MicroPrep™	50 preps.
	D6010	ZR Fecal DNA MiniPrep™	50 preps.
	D6110	ZR Fecal DNA MidiPrep™	25 preps.
	D6011	ZR-96 Fecal DNA Kit™	2 x 96 preps.
	D6015	ZR Tissue & Insect DNA MicroPrep™	50 preps.
	D6016	ZR Tissue & Insect DNA MiniPrep™	50 preps.
	D6115	ZR Tissue & Insect DNA MidiPrep™	25 preps.
	D6017	ZR-96 Tissue & Insect DNA Kit™	2 x 96 preps.
	D6022	ZR Plant/Seed DNA MicroPrep™	50 preps.
	D6020	ZR Plant/Seed DNA MiniPrep™	50 preps.
	D6120	ZR Plant/Seed DNA MidiPrep™	25 preps.
	D6021	ZR-96 Plant/Seed DNA Kit™	2 x 96 preps.

ENVIRONMENTAL RNA

	R2040	ZR Soil/Fecal RNA MicroPrep™	50 preps.
	R2010	ZR Fungal/Bacterial RNA MicroPrep™	50 preps.
	R2014	ZR Fungal/Bacterial RNA MiniPrep™	50 preps.
	R2030	ZR Tissue & Insect RNA MicroPrep™	50 preps.
	R2024	ZR Plant RNA MiniPrep™	50 preps.

ALSO AVAILABLE

D6030	OneStep™ PCR Inhibitor Removal Kit	50 preps.
D6035	OneStep-96™ PCR Inhibitor Removal Kit	2 x 96 preps.
S6002-50	ZR BashingBead™ Lysis Tubes (0.5 mm)	50 tubes
S6003-50	ZR BashingBead™ Lysis Tubes (2.0 mm)	50 tubes
S6002-96-1	ZR-96 BashingBead™ Lysis Rack (0.5 mm)	1 rack
S6002-96-2	ZR-96 BashingBead™ Lysis Rack (2.0 mm)	1 rack



www.zymoresearch.com/xpedition

See the Xpedition™ Sample Processor in action!


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DNA PURIFICATION

What is Clean-Spin™ Technology?

The spin columns from Zymo Research have been designed to ensure complete elution with no binding/wash buffer carryover. The result is ultra-pure inhibitor-free DNA and RNA.

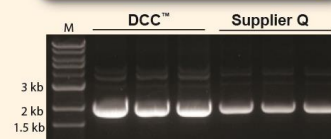


Purify DNA from PCR & other sources

DNA Clean & Concentrator™ (DCC™)

- ✓ Recovery of ultra-pure DNA that is free of salts and contaminants.
- ✓ Small ($\geq 6 \mu\text{l}$) elution volume.
- ✓ DNA is ideal for ligation, PCR, Next-Gen sequencing, etc.

Product	Size (Cat. No.)
DNA Clean & Concentrator™-5	50 Preps. (D4013) 200 Preps. (D4014)
ZR-96 DNA Clean & Concentrator™-5	2 x 96 Preps. (D4023) 4 x 96 Preps. (D4024)
Genomic DNA Clean & Concentrator™	25 Preps. (D4010) 100 Preps. (D4011)



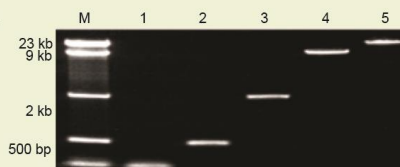
High efficiency DNA recovery with the DCC™-5 compared to Supplier Q.

Boost DNA recoveries from agarose gels to >80%

Zymoclean™ Gel DNA Recovery

- ✓ Rapid (15 min.) recovery of ultra-pure DNA from agarose gels in $\geq 6 \mu\text{l}$.
- ✓ Ultra-pure DNA ideal for DNA ligation, sequencing, etc.
- ✓ Format also available for large DNA >20 kb.

Product	Size (Cat. No.)
Zymoclean™ Gel DNA Recovery Kit	50 Preps. (D4001) 200 Preps. (D4002)
Zymoclean™ Large Fragment DNA Recovery Kit	25 Preps. (D4045) 100 Preps. (D4046)



DNA fragments recovered from an agarose gel using the Zymoclean™ Gel DNA Recovery Kit. Lanes: M: DNA Ladder; 1-5: individual ladder DNA fragments.

Recover transfection-quality plasmid DNA directly from culture

Zyppy™ Plasmid Prep Kits

- ✓ The fastest, simplest method available for purifying high quality plasmid DNA from *E. coli*.
- ✓ Pellet-Free™ procedure omits conventional cell-pelleting and resuspension steps.
- ✓ Transfection quality plasmid DNA directly from culture in under 15 minutes.

Easy, Pellet-free Procedure: Add Lysis Buffer **Directly** to Bacterial Culture



Product	Size (Cat. No.)
Zyppy™ Plasmid Miniprep Kit	50 Preps. (D4036) 100 Preps. (D4019) 400 Preps. (D4020) 800 Preps. (D4037)

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RNA PURIFICATION

Get RNA *directly* from TRIzol® without phase separation



What is Clean-Spin™ Technology?

The spin columns from Zymo Research have been designed to ensure complete elution with no binding/wash buffer carryover. The result is ultra-pure inhibitor-free DNA and RNA.

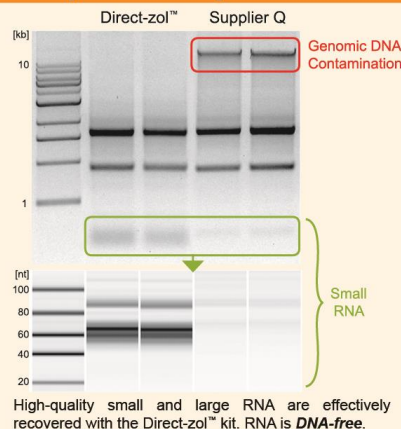
Direct-zol™ RNA

- ✓ For purification of high-quality small and large RNA *directly* from TRIzol®, TRI Reagent®, or similar.
- ✓ Bypasses phase separation and precipitation procedures allowing for unbiased recovery of miRNA

Product	Size (Cat. No.)
Direct-zol™ RNA MiniPrep	50 Preps. (R2050)
	50 Preps. (R2051)*
	200 Preps. (R2052)
	200 Preps. (R2053)*

96-well and MagBead formats also available!

DNase I included in all kits.
* Supplied with TRI-Reagent®

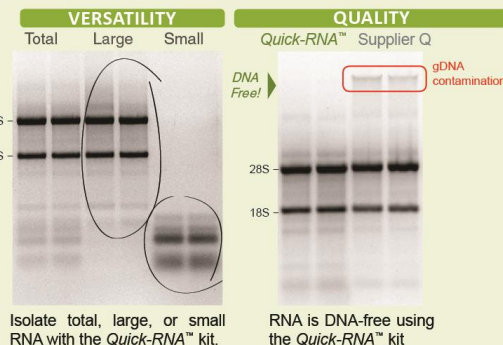


Isolate DNA-free RNA from 1 to 10⁷ cells in minutes

Quick-RNA™

- ✓ Isolation of total, large, or small RNA – *You decide!*
- ✓ Ultra clean, high-quality RNA from a single cell to 10⁷ cells.
- ✓ DNA-free RNA ideal for any downstream application – *DNase I included.*

Product	Size (Cat. No.)
Quick-RNA™ MicroPrep	50 Preps. (R1050)
	200 Preps. (R1051)
Quick-RNA™ MiniPrep	50 Preps. (R1054)
	200 Preps. (R1055)
ZR-96 Quick-RNA™	2 x 96 Preps. (R1052)
	4 x 96 Preps. (R1053)



Purify RNA from enzymatic and labeling reactions in 5 minutes

RNA Clean & Concentrator™

- ✓ Recover ultra-pure RNA in small (≥6 µl) elution volumes.
- ✓ Compatible with TRIzol®, phenol, chloroform, and RNase inhibitors (RNAlater®).
- ✓ RNA is ideal for RT-PCR, q-PCR, hybridization, arrays, RNA interference, etc.

Product	Size (Cat. No.)
RNA Clean & Concentrator™-5	50 Preps. (R1015)
	200 Preps. (R1016)
RNA Clean & Concentrator™-25	50 Preps. (R1017)
	100 Preps. (R1018)
ZR-96 RNA Clean & Concentrator™	2x96 well plates (R1080)
DNA-Free RNA Kit™	50 Preps. (R1013)
	200 Preps. (R1014)



The following are trademarks of other companies: pGEM®, Promega Corp.; TRIzol® and TRI Reagent®, Molecular Research Center, Inc.; DH5® and DH10B™, Life Technologies, Inc.

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OTHER INNOVATIVE PRODUCTS FROM ZYMO RESEARCH...

Competent cells for transformations without heat shock!

Mix & Go! Pre-made Competent *E. Coli*

- ✓ High efficiency: 10^8 - 10^9 transformants/ μ g plasmid DNA
- ✓ Just Mix & Go! Simply add DNA then spread. Transformation in as little as 20 seconds!

Product	Size (Cat. No.)
Zymo 5 α (Same as DH5 α)	10 x 100 μ l aliquots (T3007) 96 x 50 μ l aliquots (T3009) 96 x 50 μ l aliquots PCR-plate (T3010)
Zymo 10B (Same as DH10B)	10 x 100 μ l aliquots (T3019) 96 x 50 μ l aliquots (T3020)
JM109	10 x 100 μ l aliquots (T3003) 96 x 50 μ l aliquots (T3005)
HB101	10 x 100 μ l aliquots (T3011) 96 x 50 μ l aliquots (T3013)
C600	10 x 100 μ l aliquots (T3015)
TG1	10 x 100 μ l aliquots (T3017)

- ✓ No heat shock
- ✓ No incubations
- ✓ No outgrowth
- ✓ No wait!!!

Mix & Go
Competent cells
E. coli + DNA

Mix & Go!
for 20 second*
transformations!

Others
E. coli + DNA

Ice 45 min.

42°C 2 min. (Heatshock)

Place on Ice

Add SOC

Spin to Concentrate Cells

Remove Supernatant

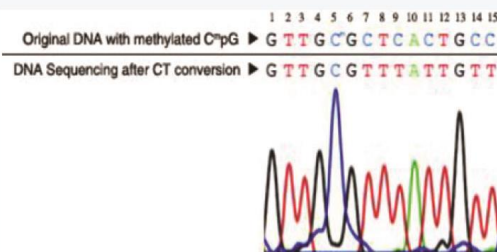


* For Ampicillin selection only.

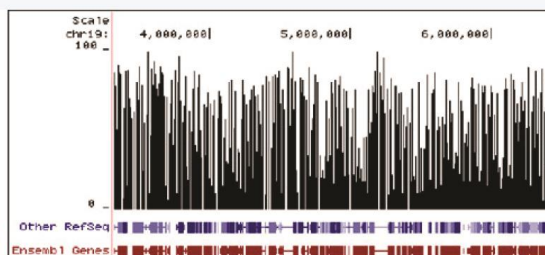
The fastest method for complete bisulfite conversion of DNA

EZ DNA Methylation-Lightning™ Kits

- ✓ The next generation of bisulfite conversion technology by the most cited provider in the industry
- ✓ Guarantees high conversion efficiencies of cytosine (>99.5%)
- ✓ Maintains the highest template integrity following bisulfite conversion
- ✓ Recovered DNA is ideal for PCR, MSP, array, bisulfite, and next-generation sequencing.



DNA Sequencing Results Following Bisulfite Treatment



Methylation Plot From Reduced Representation Bisulfite Sequencing (RRBS)

Product	Size (Cat. No.)
EZ DNA Methylation-Lightning™ Kit	50 rxns. (D5030) 200 rxns. (D5031)
EZ-96 DNA Methylation-Lightning™ Kit	Shallow-Well 2 x 96 rxns. (D5032) Deep-Well 2 x 96 rxns. (D5033)
EZ-96 DNA Methylation-Lightning™ MagPrep	4 x 96 rxns. (D5046) 8 x 96 rxns. (D5047)

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