



**ZYMO RESEARCH**

*The Beauty of Science is to Make Things Simple*

# INSTRUCTION MANUAL

## ZymoPURE™ II Plasmid Maxiprep Kit

Catalog Nos. **D4202 & D4203** (Patent Pending)

### Highlights

- **Fastest:** Simple 20-minute Maxipreps.
- **Highest Yield:** Purify up to 1.2 mg of plasmid DNA directly from a spin-column.
- **Ultra-Pure:** EndoZero, vaccine grade, and transfection ready

### Contents

Product Contents .....	1
Product Specifications.....	1
Product Description.....	2
Procedure Overview.....	3
Buffer Preparation.....	4
Protocol.....	4-6
Appendix.....	7-8
A. Low-Copy Number Protocol.....	7
B. Gram-Positive Bacteria Protocol.....	8
Troubleshooting .....	9
Ordering Information .....	10

For Research Use Only

Version 1.2.9

**ZYMO RESEARCH CORP.**

Phone: (949) 679-1190 ▪ Toll Free: (888) 882-9682 ▪ Fax: (949) 266-9452 ▪ [info@zymoresearch.com](mailto:info@zymoresearch.com) ▪ [www.zymoresearch.com](http://www.zymoresearch.com)

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

#### Notes:

This product is for research use only and should only be used by trained professionals. It is not 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.

<sup>TM</sup> Trademarks of Zymo Research Corporation.

Several ZymoPURE<sup>TM</sup> product technologies are subject to U.S. and foreign patents or are patent pending.

pGL3<sup>TM</sup> is a registered trademark of Promega Corporation.

## Product Contents:

ZymoPURE <sup>TM</sup> II Plasmid Maxiprep Kit (Kit Size)	D4202 (10 preps.)	D4203 (20 preps.)	Storage Temperature
ZymoPURE <sup>TM</sup> P1 <sup>1</sup> (Red)	150 ml	2x 150 ml	4°C
ZymoPURE <sup>TM</sup> P2 <sup>2</sup> (Green)	150 ml	2x 150 ml	Room Temp.
ZymoPURE <sup>TM</sup> P3 (Yellow)	150 ml	2x 150 ml	Room Temp.
ZymoPURE <sup>TM</sup> Binding Buffer	150 ml	2x 150 ml	Room Temp.
ZymoPURE <sup>TM</sup> Wash 1	55 ml	2x 55 ml	Room Temp.
ZymoPURE <sup>TM</sup> Wash 2 (Concentrate)	23 ml	2x 23 ml	Room Temp.
ZymoPURE <sup>TM</sup> Elution Buffer	6 ml	12 ml	Room Temp.
Zymo-Spin <sup>TM</sup> V-P Column Assemblies <sup>3</sup>	10	20	Room Temp.
ZymoPURE <sup>TM</sup> Syringe Filters	10	20	Room Temp.
ZymoPURE <sup>TM</sup> Syringe Plungers	10	20	Room Temp.
EndoZero <sup>TM</sup> Spin-Columns	10	20	Room Temp.
Collection Tubes	10	20	Room Temp.
Instruction Manual	1	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.

<sup>1</sup> ZymoPURE<sup>TM</sup> P1 contains RNase A (100 µg/ml) and is stable at room temperature without loss in RNase activity, however, for long-term storage the product should be stored at 4-8° C.

<sup>2</sup> Caution: ZymoPURE<sup>TM</sup> P2 Buffer contains NaOH. Please use proper safety precautions.

<sup>3</sup> The Zymo-Spin<sup>TM</sup> V-P, 15 ml Conical Reservoir and 50 ml Reservoir are pre-assembled as a single unit.

## Specifications:

- **DNA Purity:** Eluted DNA is ultrapure, endotoxin-free, and well suited for transfection, transformation, sequencing, restriction endonuclease digestion, *in vitro* transcription, *in vivo* studies, and other sensitive applications.
  - Typical Abs<sub>260/280</sub> ≥ 1.8 and Abs<sub>260/230</sub> ≥ 2.0
  - Endotoxin levels: ≤ 1 EU/µg of plasmid DNA using the Standard Protocol  
≤ 0.025 EU/µg of plasmid DNA with optional EndoZero<sup>TM</sup> Spin-Column
- **Plasmid DNA Yield:** Up to 1.2 mg per preparation (*Actual yield is dependent on the plasmid copy number, culture growth conditions, and strain of E. coli utilized*)
- **Plasmid DNA Size:** Up to ~200 kb
- **Recovery Volume:** ≥ 200 µl of ZymoPURE<sup>TM</sup> Elution Buffer or DNase-free water
- **Required Equipment:** Microcentrifuge and vacuum/vacuum manifold (recommended) or swinging bucket centrifuge.
- **Processing Time:** 20 min

### ZYMO RESEARCH CORP.

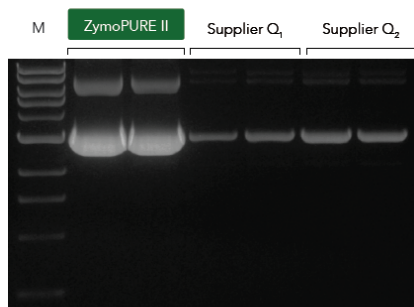
## Product Description

The **ZymoPURE™ II Plasmid Maxiprep Kit** features a simple spin-column based method for the purification of up to 1.2 mg of transfection grade plasmid DNA in less than 20 minutes. The eluted plasmid DNA is EndoZero and ready for immediate use in the most sensitive applications. The unique ZymoPURE methodology removes the need for slow gravity flow anion-exchange columns, alcohol precipitations, lengthy endotoxin removal incubations, and time-consuming centrifugation steps.

ZymoPURE™ technology uses a modified alkaline lysis method and features novel binding chemistry, which enables the highest yields and concentration of plasmid DNA (up to 3 µg/µl) directly from a spin-column. Coupling ZymoPURE with the innovative **EndoZero™ Spin-Columns**, to eliminate endotoxins, achieves EndoZero plasmid DNA ( $\leq 0.025$  EU/µg of plasmid DNA), making it suitable for transfection, restriction endonuclease digestion, *in vivo* studies, bacterial transformation, PCR amplification, DNA sequencing, and other sensitive downstream applications.

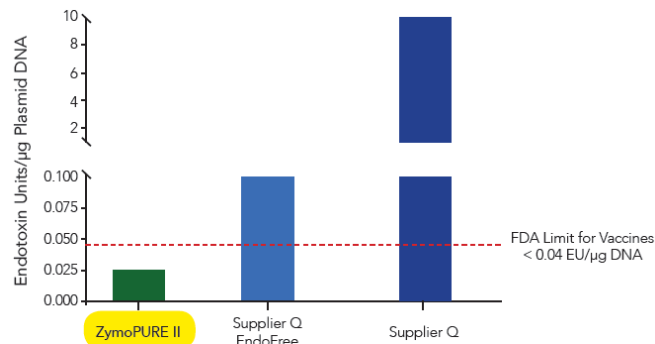
As an added convenience, the **ZymoPURE™ II Plasmid Maxiprep Kit** contains colored buffers that permit error-free visualization and identification of complete bacterial cell lysis and neutralization. Syringe filters are included for rapid clearing of the lysate and the unique spin-column design allows the binding step to be performed using a vacuum or table top centrifuge.

### Highest Recovery



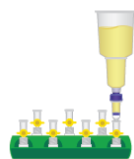
Plasmid DNA concentration and yield from the ZymoPURE II Maxiprep kit compared to two separate kits from Supplier Q. Plasmid DNA (pGL3<sup>®</sup>) was isolated from 150 ml of JM109 *E. coli* culture grown overnight following the manufacturer's suggested protocol (in duplicate). One (1) µl of eluted plasmid DNA was visualized post agarose gel electrophoresis. M, ZR 1 kb DNA Marker (Zymo Research).

### Lowest Endotoxin Levels



Manufacturers' stated endotoxin for two separate Anion-Exchange kits from Supplier Q compared to ZymoPURE II.

### Simplest Workflow



**EZ-Load™**  
No Gravity Flow!



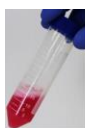
**EZ-Elute™**  
No Alcohol  
Precipitation!



**EndoZero™**  
Endotoxins < FDA  
limit for Vaccines

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

### Procedure Overview:



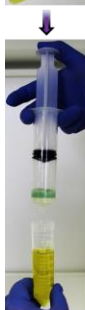
Bacterial cells are resuspended in **ZymoPURE™ P1** (red).



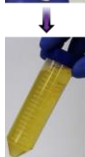
The solution will turn dark purple and viscous following the addition of **ZymoPURE™ P2** (green) indicating bacterial lysis is complete.



The solution will turn yellow and a precipitate will form after adding **ZymoPURE™ P3** (yellow) indicating neutralization is complete.



The neutralized lysate is loaded into the **ZymoPURE™ Syringe Filter** and clarified into a new 50 ml conical tube.



**ZymoPURE™ Binding Buffer** is added to the cleared lysate and mixed thoroughly.



The mixture is loaded into the **Zymo-Spin™ V-P Column** using a vacuum manifold.



The **50 ml Reservoir** is removed and the **Zymo-Spin™ V-P Column** is washed using a vacuum manifold.



Ultra-pure plasmid DNA is eluted from the **Zymo-Spin™ V-P Column** using a microcentrifuge.



The eluted plasmid DNA is passed through the **EndoZero™ Column** using a microcentrifuge.

**Buffer Preparation:**

- ✓ Add 88 ml of 95% ethanol to the 23 ml **ZymoPURE™ Wash 2 (Concentrate)** before use.
- ✓ The **ZymoPURE™ P2** and **ZymoPURE™ Binding Buffer** may have precipitated. If this occurs, dissolve the precipitate by incubating the bottles at 30-37 °C for 10-20 minutes and mix by inversion. Do not microwave!

**Before Starting:**

- ✓ Centrifuge up to 150 ml of bacterial culture at  $\geq 3,400 \times g$  for 10 minutes to pellet the cells<sup>1</sup>. Discard supernatant.

**Protocol:**

The following procedure should be performed at room temperature (15-30°C).

1. Add 14 ml of **ZymoPURE™ P1 (Red)** to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
2. Add 14 ml of **ZymoPURE™ P2 (Green)** and immediately mix by gently inverting the tube 6 times. Do not vortex! Let sit at room temperature for 2-3 minutes<sup>2</sup>.

*Cells are completely lysed when the solution appears clear, purple, and viscous.*

3. Add 14 ml of **ZymoPURE™ P3 (Yellow)** and mix gently but thoroughly by inversion. Do not vortex!

*The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.*

4. Ensure the plug is attached to the Luer Lock at the bottom of the **ZymoPURE™ Syringe Filter**. Place the syringe filter upright in a tube rack and load the lysate into the ZymoPURE™ Syringe Filter<sup>3</sup> and wait 5-8 minutes for the precipitate to float to the top.
5. Remove the Luer Lock plug from the bottom of the syringe and place it into a clean 50 ml conical tube. Place the plunger in the syringe and push the solution through the ZymoPURE™ Syringe Filter in one continuous motion until approximately 33-35 ml of cleared lysate is recovered. Save the cleared lysate!
6. Add 14 ml **ZymoPURE™ Binding Buffer** to the cleared lysate from step 5 and mix thoroughly by inverting the capped tube 10 times.

*To continue processing the lysate using the recommended vacuum protocol, proceed to the next page. If a vacuum is not available, proceed to page 6 for an alternative centrifugation method.*

**Notes:**

<sup>1</sup> A vessel with a minimum volume of 50 ml is required to prepare the bacterial lysate.

<sup>2</sup> Do not allow the lysis reaction to proceed for more than 3 minutes. Excessive lysis can result in denatured plasmid DNA.

<sup>3</sup> If the precipitate has formed a homogenous layer at the surface of the neutralized lysate then invert the tube 3-4 times prior to loading the lysate into the **Zymo PURE™ Syringe Filter**.

**Notes:**

<sup>1</sup> To achieve optimal performance, the vacuum pump should be able to apply at least 400 mm Hg pressure. If less pressure is applied, centrifuge the column prior to washing to remove any residual lysate remaining in the matrix.

<sup>2</sup> The **ZymoPURE™ Elution Buffer** contains 10 mM Tris-HCl, pH 8.5 & 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

<sup>3</sup> The DNA yield can be increased by pre-warming the **ZymoPURE™ Elution Buffer** to 50 °C and/or increasing the incubation period up to 10 minutes prior to centrifugation.

<sup>4</sup> For low copy number plasmids or if higher concentration is desired, the plasmid DNA can be eluted in as little as 200 µl.

<sup>5</sup> This optional step will reduce endotoxin levels from ≤ 1 EU/µg of plasmid DNA to ≤ 0.025 EU/µg of plasmid DNA.

**Vacuum Protocol:** (Recommended)

This product is compatible with any conventional vacuum-based manifold. The vacuum pump should be a single or double-staged unit capable of producing up to 400 mm Hg pressure at the vacuum manifold<sup>1</sup>.

7. Ensure the connections of the **Zymo-Spin™ V-P Column Assembly** are finger-tight and place onto a vacuum manifold. (If vacuum is not available, see page 6 for the centrifugation protocol.)
8. With the vacuum off, add the entire mixture from step 6 into the Zymo-Spin™ V-P Column Assembly, and then turn on the vacuum<sup>1</sup> until all of the liquid has passed completely through the column.
9. Remove and discard the **50 ml Reservoir** from the top of the Zymo-Spin™ V-P Column Assembly.
10. With the vacuum off, add 5 ml of **ZymoPURE™ Wash 1** to the **15 ml Conical Reservoir**. Turn on the vacuum until all of the liquid has passed completely through the column.
11. With the vacuum off, add 5 ml of **ZymoPURE™ Wash 2** to the 15 ml Conical Reservoir. Turn on the vacuum until all of the liquid has passed completely through the column. Repeat this wash step.
12. Remove and discard the 15 ml Conical Reservoir and place the **Zymo-Spin™ V-P Column** in a **Collection Tube**. Centrifuge at ≥ 10,000 x g for 1 minute, in a microcentrifuge, to remove any residual wash buffer.
13. Transfer the column into a clean 1.5 ml microcentrifuge tube and add 400 µl of **ZymoPURE™ Elution Buffer**<sup>2,3,4</sup> directly to the column matrix. Wait 2 minutes, and then centrifuge at ≥ 10,000 x g for 1 minute in a microcentrifuge.
14. *Optional:* For EndoZero Plasmid DNA<sup>5</sup>, remove the Luer Lock cap from the **EndoZero™ Spin-Column** and place the column in a clean 1.5 ml microcentrifuge tube. Add the entire eluate from step 13 into the EndoZero™ Spin-Column, wait 2 minutes, and then centrifuge at 10,000 x g for 1 minute in a microcentrifuge. Store the eluted plasmid DNA at ≤ -20°C.

### **Centrifugation Protocol:** *(Alternative)*

Perform steps 1-6 as indicated in the general protocol, see page 4.

7. Remove the **50 ml Reservoir** from the top of the **Zymo-Spin™ V-P Column Assembly**. Ensure the connection between the **15 ml Conical Reservoir** and **Zymo-Spin™ V-P column** is finger-tight and place the assembly into a 50 ml conical tube.
8. Add 14 ml of the mixture from step 6 into the **15 ml Conical Reservoir/Zymo-Spin™ V-P column assembly**, and then centrifuge the column at 500 x *g* for 2 minutes. Empty the 50 ml conical tube and repeat this step until the entire sample has passed through the column.
9. Add 5 ml of **ZymoPURE™ Wash 1** to the Zymo-Spin™ V-P column assembly and centrifuge the column at 500 x *g* for 2 minutes.
10. Add 5 ml of **ZymoPURE™ Wash 2** to the Zymo-Spin™ V-P column assembly and centrifuge the column for 2 minutes at 500 x *g*. Repeat the wash step.
11. Remove and discard the 15 ml Conical Reservoir and place the Zymo-Spin™ V-P Column into a **Collection Tube**. Centrifuge the column at  $\geq 10,000$  x *g* for 1 minute, in a microcentrifuge, to remove any residual wash buffer.
12. Transfer the column into a clean 1.5 ml microcentrifuge tube and add 400  $\mu$ l of **ZymoPURE™ Elution Buffer**<sup>1,2,3</sup> directly to the column matrix. Wait 2 minutes, and then centrifuge at  $\geq 10,000$  x *g* for 1 minute in a microcentrifuge.
13. *Optional:* For EndoZero Plasmid DNA<sup>4</sup>, remove the Luer Lock cap from the **EndoZero™ Spin-Column** and place the column in a clean 1.5 ml tube. Add the entire eluate from step 12 into the EndoZero™ Spin-Column, wait 2 minutes, and then centrifuge at 10,000 x *g* for 1 minute in a microcentrifuge. Store the eluted plasmid DNA at  $\leq -20^{\circ}\text{C}$ .

#### **Notes:**

<sup>1</sup> The **Zymo PURE™ Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

<sup>2</sup> The DNA yield can be increased by pre-warming the **Zymo PURE™ Elution Buffer** to 50 °C and/or increasing the incubation period up to 10 minutes prior to centrifugation.

<sup>3</sup> For low copy number plasmids or if higher concentration is desired, the plasmid DNA can be eluted in as little as 200  $\mu$ l.

<sup>4</sup> This optional step will reduce endotoxin levels from  $\leq 1$  EU/ $\mu$ g of plasmid DNA to  $\leq 0.025$  EU/ $\mu$ g of plasmid DNA.

Notes:

## **Appendix A: Low-Copy Number Protocol**

When working with low-copy number plasmid DNA, it is possible to increase plasmid DNA yield by processing up to 300 ml of overnight culture grown in LB using the protocol below.

### **Before Starting:**

- ✓ Centrifuge up to 300 ml of bacterial culture at  $\geq 3,400 \times g$  for 10 minutes to pellet the cells. Discard supernatant.

### **Protocol:**

1. Add 14 ml of **ZymoPURE™ P1 (Red)** to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
2. Add 14 ml of **ZymoPURE™ P2 (Green)** and immediately mix by gently inverting the tube 10 times. Do not vortex! Let sit at room temperature for 5 minutes.

*Cells are completely lysed when the solution appears clear, purple, and viscous.*

3. Add 14 ml of **ZymoPURE™ P3 (Yellow)** and mix gently but thoroughly by inversion. Do not vortex! Invert the tube an additional 8 times after the sample turns completely yellow.

*The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.*

4. Centrifuge the neutralized lysate for 10 minutes at  $\geq 3,400 \times g$  at room temperature.
5. Ensure the plug is attached to the Luer Lock at the bottom of the **ZymoPURE™ Syringe Filter**. Place the syringe filter upright in a tube rack and load the supernatant from step 4 into the ZymoPURE™ Syringe Filter.
6. Remove the Luer Lock plug from the bottom of the syringe and place it into a clean 50 ml conical tube. Place the plunger in the syringe and push the solution through the ZymoPURE™ Syringe Filter in one continuous motion until approximately 35 ml of cleared lysate is recovered. Save the cleared lysate!
7. Add 14 ml of **ZymoPURE™ Binding Buffer** to the cleared lysate from step 5 and mix thoroughly by inverting the capped tube 8 times.

*To continue processing the lysate using the recommended vacuum protocol, proceed to page 5. If a vacuum is not available, proceed to page 6 for an alternative centrifugation method.*



## **Appendix B: Gram-Positive Bacteria Protocol**

It is possible to isolate plasmid DNA from Gram-Positive species with the ZymoPURE II Plasmid Midiprep Kit. However, the cell walls of the bacteria must be digested with a lytic enzyme prior to alkaline lysis. The protocol below is for Gram-Positive strains that are sensitive to the lytic enzyme Lysozyme.

1. Add 14 ml of **ZymoPURE™ P1 (Red)** containing lysozyme<sup>1</sup> at a final concentration of 1 mg/ml to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
2. Incubate the resuspended cell pellet at 37°C for 15-60 minutes<sup>2</sup>.
3. Add 14 ml of **ZymoPURE™ P2 (Green)** and immediately mix by gently inverting the tube 6 times. Do not vortex! Let sit at room temperature for 2-3 minutes<sup>3</sup>.

*Cells are completely lysed when the solution appears clear, purple, and viscous. The cell wall digestion is most likely incomplete if the solution remains pink and opaque.*

4. Add 14 ml of **ZymoPURE™ P3 (Yellow)** and mix gently but thoroughly by inversion. Do not vortex!

*The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.*

5. Ensure the plug is attached to the Luer Lock at the bottom of the **ZymoPURE™ Syringe Filter**. Place the syringe filter upright in a tube rack and load the lysate into the ZymoPURE™ Syringe Filter<sup>4</sup> and wait 5-8 minutes for the precipitate to float to the top.
6. Remove the Luer Lock plug from the bottom of the syringe and place it into a clean 50 ml conical tube. Place the plunger in the syringe and push the solution through the ZymoPURE™ Syringe Filter in one continuous motion until approximately 33-35 ml of cleared lysate is recovered. Save the cleared lysate!
7. Add 14 ml of **ZymoPURE™ Binding Buffer** to the cleared lysate from step 6 and mix thoroughly by inverting the capped tube 8 times.

*To continue processing the lysate using the recommended vacuum protocol, proceed to page 5. If a vacuum is not available, proceed to page 6 for an alternative centrifugation method.*

### Notes:

<sup>1</sup> Lytic enzymes other than lysozyme will require optimization and validation in the ZymoPURE P1 buffer prior to use.

<sup>2</sup> Incubation times will vary depending on the cell density and age of cells. Harvesting the cells at early log phase is recommended for optimal cell wall digestion.

<sup>3</sup> Do not allow the lysis reaction to proceed for more than 3 minutes. Excessive lysis can result in denatured plasmid DNA.

<sup>4</sup> If the precipitate has formed a homogenous layer at the surface of the neutralized lysate then invert the tube 3-4 times prior to loading the lysate into the **Zymo PURE™ Syringe Filter**.

## Troubleshooting Guide:

Problem	Possible Causes and Suggested Solutions
<b>Low DNA Yield</b>	
<i>Culture growth conditions</i>	<ul style="list-style-type: none"> <li>• <b>Poor aeration of culture.</b> The optimal culture volume to air volume ratio is 1:5 or less. For best aeration, use baffled culture flasks, or a vented or gas-permeable seal on the culture vessel.</li> <li>• <b>The culture was overgrown, undergrown, contaminated, or antibiotics were omitted from the growth medium.</b> Use a fresh culture for optimal performance. An OD<sub>600</sub> of 0.2-0.35 is the optimal optical density of a tenfold dilution of the culture.</li> </ul>
<i>Cell density is too high</i>	<ul style="list-style-type: none"> <li>• <b>Too much culture used.</b> Lysis and neutralization will be incomplete and the Zymo PURE™ Syringe Filter may clog during filtration. <u>More culture does not always equal more plasmid.</u> Incomplete lysis and neutralization are two of the most common causes of failed plasmid preps and both are caused by too much culture being used.</li> <li>• <b>Incomplete lysis:</b> After addition of ZymoPURE™ P2, the solution should change from opaque pink to a clear viscous purple, indicating complete lysis. Different <i>E. coli</i> strains often require different growth conditions and may vary in their susceptibility to alkaline lysis.</li> <li>• <b>Incomplete neutralization:</b> The solution should not be viscous following neutralization and the yellowish precipitate should appear fluffy and readily float to the surface. Make sure the neutralization is complete prior to filtration. Invert the tube an additional 2-3 times after the sample turns yellow following the addition of ZymoPURE™ P3.</li> </ul>
<i>Lysate Clarification</i>	<ul style="list-style-type: none"> <li>• <b>Less than 33-35 ml of cleared lysate was recovered from the ZymoPURE™ Syringe Filter.</b> For optimal performance, add 14 ml of ZymoPURE™ Binding Buffer to approximately 33-35 ml of clarified lysate.</li> </ul>
<i>ZymoPURE P2 and ZymoPURE Binding Buffer precipitated</i>	<ul style="list-style-type: none"> <li>• <b>Both buffers may have precipitated during shipping.</b> To completely resuspend the buffers, incubate the bottles at 30-37 °C for 10 minutes and mix by inversion. DO NOT MICROWAVE.</li> </ul>
<i>Wash buffer</i>	<ul style="list-style-type: none"> <li>• <b>Ensure that ethanol has been added</b> to the ZymoPURE™ Wash 2.</li> <li>• <b>Ensure that the bottle cap is screwed on tightly</b> after each use to prevent evaporation of the ethanol.</li> </ul>
<i>DNA elution</i>	<ul style="list-style-type: none"> <li>• <b>Incomplete elution:</b> For large size plasmids (&gt; 10 kb), add ZymoPURE™ Elution Buffer and incubate the column for 5-10 minutes before centrifugation. Also, pre-warm the ZymoPURE™ Elution Buffer to 50 °C prior to elution.</li> </ul>
<b>Low DNA Quality</b>	
<i>DNA does not perform well</i>	<ul style="list-style-type: none"> <li>• <b>Incomplete neutralization:</b> Incomplete neutralization generates poor quality supernatant. Ensure that neutralization is complete by inverting the sample an additional 2-3 times after the addition of ZymoPURE™ P3 and extending the incubation.</li> <li>• <b>Ethanol contamination in eluate.</b> Centrifuge the Zymo-Spin™ V-P column as indicated in the protocol prior to adding the ZymoPURE™ Elution Buffer.</li> </ul>
<i>RNA in eluate</i>	<ul style="list-style-type: none"> <li>• <b>Ensure that ZymoPURE™ P1 has been stored at 4°C.</b> RNase A can be purchased separately if necessary.</li> </ul>
<i>Genomic DNA in eluate</i>	<ul style="list-style-type: none"> <li>• <b>Improper handling</b> (Sample was vortexed or handled too roughly). Genomic DNA contamination is usually caused by excessive mechanical shearing during the lysis and neutralization steps. Also, prolonged lysis or incomplete mixing of lysis or neutralization buffers may contribute to genomic DNA contamination in your sample.</li> <li>• <b>Overgrown culture.</b> Overgrown or old cultures may contain more genomic DNA contamination than fresh cultures.</li> </ul>

**Ordering Information**


Product Description	Kit Size	Catalog No.
ZymoPURE™ II Plasmid Maxiprep Kit	10 preps.	D4202
ZymoPURE™ II Plasmid Maxiprep Kit	20 preps.	D4203

For Individual Sale	Amount	Catalog No.
ZymoPURE™ P1 (Red)	150 ml	D4200-1-150
	210 ml	D4200-1-210
	410 ml	D4200-1-410
ZymoPURE™ P2 (Green)	150 ml	D4200-2-150
	210 ml	D4200-2-210
	410 ml	D4200-2-410
ZymoPURE™ P3 (Yellow)	150 ml	D4200-3-150
	210 ml	D4200-3-210
	410 ml	D4200-3-410
ZymoPURE™ Binding Buffer	150 ml	D4200-4-150
	210 ml	D4200-4-210
	410 ml	D4200-4-410
ZymoPURE™ Wash 1	55 ml	D4200-5-55
	410 ml	D4200-5-410
ZymoPURE™ Wash 2 (Concentrate)	23 ml	D4200-6-23
ZymoPURE™ Elution Buffer	6 ml	D4200-7-6
	12 ml	D4200-7-12
	30 ml	D4200-7-30
Zymo-Spin™ V-P Column Assembly w/ 15 ml Conical and 50 ml Reservoir	5	C1042-5
15 ml Conical Reservoir	25	C1031-25
50 ml Reservoir	25	C1032-25
ZymoPURE™ Syringe Filter	5	C1036-5
ZymoPURE™ Syringe Plunger	5	C1037-5
EndoZero™ Spin-Columns	10	C1051-10
Collection Tubes	50	C1001-50
	500	C1001-500
	1000	C1001-1000

**ZYMO RESEARCH CORP.**

# DNA PURIFICATION

**What is Clean-Spin™ Technology?**



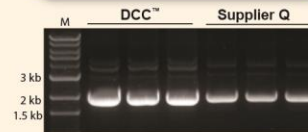
The spin columns from Zymo Research have been designed to ensure complete elution with no binding/wash buffer carryover. The result is ultra-pure inhibitor-free DNA and RNA.

## 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)



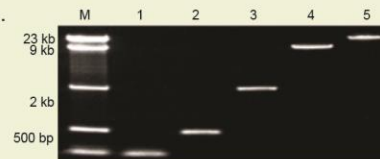
High efficiency DNA recovery with the DCC™-5 compared to Supplier Q.

## 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)

# OTHER INNOVATIVE PRODUCTS FROM ZYMO RESEARCH...

## Competent cells for transformations *without* heat shock!

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

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

Product	Size (Cat. No.)
Zymo 5α (Same as DH5α)	10 x 100 μl aliquots (T3007)
	96 x 50 μl aliquots (T3009)
	96 x 50 μl aliquots PCR-plate (T3010)
Zymo 10B (Same as DH10B)	10 x 100 μl aliquots (T3019)
	96 x 50 μl aliquots (T3020)
JM109	10 x 100 μl aliquots (T3003)
	96 x 50 μl aliquots (T3005)
HB101	10 x 100 μl aliquots (T3011)
	96 x 50 μl aliquots (T3013)
C600	10 x 100 μl aliquots (T3015)
TG1	10 x 100 μl aliquots (T3017)

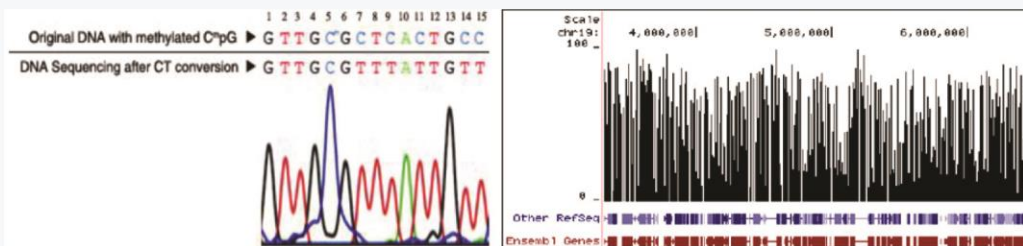
- ✓ No heat shock
- ✓ No incubations
- ✓ No outgrowth
- ✓ No wait!!!



## 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)

**ZYMO RESEARCH CORP.**



**ZYMO RESEARCH**

*The Beauty of Science is to Make Things Simple*

---

**ZYMO RESEARCH CORP.**

Phone: (949) 679-1190 ▪ Toll Free: (888) 882-9682 ▪ Fax: (949) 266-9452 ▪ [info@zymoresearch.com](mailto:info@zymoresearch.com) ▪ [www.zymoresearch.com](http://www.zymoresearch.com)