

## Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

Weitere Information auf den folgenden Seiten! See the following pages for more information!



## Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

## Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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## ZymoPURE™ Plasmid Miniprep Kit

Rapid purification of transfection-grade plasmid DNA from up to 5 ml of overnight E. coli culture.

#### **Highlights**

- Purify up to 100 μg of plasmid DNA in as little as 25 μl directly from a spin-column.
- Purified plasmid DNA contains 50,000 times fewer endotoxins than industry leading minipreps.
- Purify constructs up to ~200 kb in size.

Catalog Numbers: D4208T, D4209, D4210, D4211, D4212



Scan with your smart-phone camera to view the online protocol/video.







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Revised on: 11/15/2023

### **Product Contents**

ZymoPURE <sup>™</sup> Plasmid Miniprep Kit	<b>D4208T</b> (10 prep)	<b>D4209</b> (50 prep)	<b>D4210</b> (100 prep)	<b>D4211</b> (400 prep)	<b>D4212</b> (800 prep)	Storage Temperature
ZymoPURE <sup>™</sup> P1¹ (Red)	3 ml	13 ml	13 ml (2x)	100 ml	210 ml	4°C
ZymoPURE <sup>™</sup> P2 <sup>2,3</sup> (Blue)	3 ml	13 ml	13 ml (2x)	100 ml	210 ml	Room Temp.
ZymoPURE <sup>™</sup> P3 (Yellow)	3 ml	13 ml	13 ml (2x)	100 ml	210 ml	Room Temp.
ZymoPURE <sup>™</sup> Binding Buffer <sup>3</sup>	3 ml	14 ml	14 ml (2x)	110 ml	110 ml (2x)	Room Temp.
ZymoPURE <sup>™</sup> Wash 1	12 ml	20 m (2x)	20 ml (4x)	320 ml	320 ml (2x)	Room Temp.
ZymoPURE <sup>™</sup> Wash 2 <sup>4</sup>	11 ml	12 ml	23 ml	28 ml (3x)	28 ml (6x)	Room Temp.
ZymoPURE <sup>™</sup> Elution Buffer	1 ml	1 ml (2x)	6 ml	12 ml	30 ml	Room Temp.
Zymo-Spin <sup>™</sup> II-PX Columns	10 pcs	50 pcs	100 pcs	400 pcs	800 pcs	Room Temp.
Collection Tubes	10 pcs	50 pcs	100 pcs	400 pcs	800 pcs	Room Temp.
Instruction Manual	1 pc	1 pc	1 pc	1 pc	1 pc	-

<sup>&</sup>lt;sup>1</sup> ZymoPURE<sup>™</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>&</sup>lt;sup>2</sup> Caution: ZymoPURE<sup>™</sup> P2 Buffer contains NaOH. Please use proper safety precautions.

<sup>3</sup> The ZymoPURE<sup>™</sup> P2 and ZymoPURE <sup>™</sup> 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! <sup>4</sup>ZymoPURE<sup>™</sup> Wash 2 included with D4208S and D4208T is supplied ready-to-use and does not require the addition of ethanol prior to use. ZymoPURE™ Wash 2 included with D4209, D4210, D4211, and D4212 are supplied as a concentrate and require the addition of ethanol prior to use. See Buffer Preparation (page 4) for instructions.

## **Specifications**

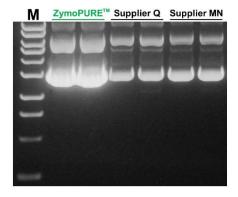
- DNA Purity Eluted DNA is ultra-pure and well suited for transfection, transformation, lentivirus production, adenovirus production, AAV production, CRISPR, genome editing, sequencing, restriction endonuclease digestion, in vitro transcription/translation, PCR, and other sensitive applications.
  - o Typical Abs260/280 ≥ 1.8 and Abs260/230 ≥ 2.0
  - Endotoxin levels: ≤ 1 EU/µg of plasmid DNA using the Standard Protocol. Suitable for transfecting stable, primary, and sensitive cell lines.
- Plasmid DNA Yield Up to 100 μg per preparation. Actual yield is dependent on the plasmid copy number, culture growth conditions, and strain of E. coli utilized. Typical yields from 1 ml of overnight culture grown in LB are 4 – 8 μg for high copy number plasmids and 0.5 – 1 μg for low copy number plasmids.
- Plasmid DNA Size Up to 200 kb
- Recovery Volume ≥ 25 µl of ZymoPURE<sup>™</sup> Elution Buffer or DNase free water
- **Processing Time** 15 min
- Required Equipment Microcentrifuge and/or vacuum manifold (recommended).

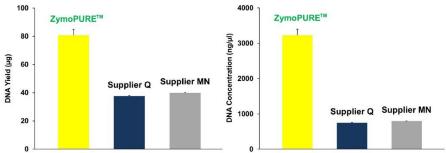
## **Product Description**

The **ZymoPURE**<sup>™</sup> **Plasmid Miniprep Kit** features a spin column-based method for the purification of up to 100 µg of ultra-pure transfection-grade plasmid DNA in less than 15 minutes. The unique spin-column design also provides zero buffer retention and a low elution volume.

ZymoPURE<sup>™</sup> technology uses a modified alkaline lysis method and features novel binding chemistry that yields highly concentrated plasmid DNA (up to 3 μg/μl). In addition, the wash regimen has been optimized to ensure the plasmid DNA is free of endotoxins, salt, protein, and RNA. The result is plasmid DNA suitable for transfection, transformation, lentivirus production, adenovirus production, AAV production, CRISPR, genome editing, sequencing, restriction endonuclease digestion, *in vitro* transcription/translation, PCR and other sensitive downstream applications.

As an added convenience, the **ZymoPURE™ Plasmid Miniprep Kit** contains colored buffers that permit error-free visualization and identification of complete bacterial cell lysis and neutralization.





Plasmid DNA yield and concentration from the ZymoPure™ Miniprep Kit compared to other major suppliers. Plasmid DNA (pGL3®) was isolated from 5 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).

### **Procedure Overview**



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



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



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



**ZymoPURE**<sup>™</sup> **Binding Buffer** is added to the cleared lysate and mixed thoroughly.



The mixture is loaded into the **Zymo-Spin™ II-PX Column** using a vacuum manifold or microcentrifuge.



The **Zymo-Spin**™ **II-PX Column** is washed using a vacuum manifold or microcentrifuge.



Transfection-grade plasmid DNA is eluted from the **Zymo-Spin**™ **II-PX Column** using a microcentrifuge.

### **Protocol**

#### **Buffer Preparation:**

- ✓ Add 46 ml of 95 100% ethanol to the 12 ml ZymoPURE™ Wash 2 (Concentrate) (D4209), 88 ml of 95 100% ethanol to the 23 ml ZymoPURE™ Wash 2 (Concentrate) (D4210), or 107 ml of 95 100% ethanol to the 28 ml ZymoPURE™ Wash 2 (Concentrate) (D4211 & D4212) 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!

#### Plasmid DNA Purification

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

- Centrifuge 0.5-5 ml<sup>1</sup> of bacterial culture in a clear 1.5 ml tube at full speed for 15-20 seconds in a microcentrifuge. Discard supernatant.
- Add 250 µl of cold ZymoPURE™ P1 (Red) to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
- 3. Add 250 µl of **ZymoPURE™ P2 (Blue)** and <u>immediately mix</u> by gently inverting the tube 8-10 times. <u>Do not vortex!</u> Let sit at room temperature for 3 minutes<sup>2</sup>. Cells are completely lysed when the solution appears clear, purple, and viscous
- 4. Add 250 μl of **ZymoPURE™ P3 (Yellow)** and mix thoroughly by inversion. <u>Do not vortex!</u> Invert the tube an additional 5 times after the sample turns completely yellow. The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.
- 5. Centrifuge the neutralized lysate for 5 minutes at 16,000 x g.
- 6. Transfer exactly 600 µl of supernatant from step 5 into a clean 1.5 ml microcentrifuge tube.
- Add 260 µl of ZymoPURE<sup>™</sup> Binding Buffer to the cleared lysate from step 6 and mix thoroughly by vortexing for 15 seconds<sup>3</sup>.

To continue processing the lysate using the recommended vacuum protocol, proceed to the next page. If a vacuum is not available, proceed to page 7 for an alternative centrifugation method.

<sup>&</sup>lt;sup>1</sup> Depending on the volume of bacterial culture it may be necessary to repeat Step 1 several times.

<sup>&</sup>lt;sup>2</sup> Do not allow the lysis reaction to proceed for more than 3 minutes. Excessive lysis can result in denatured plasmid DNA. When processing a large number of samples, work with groups of ≤ 10 at a time.

<sup>&</sup>lt;sup>3</sup> The sample can become hazy/slightly cloudy after this step if a lot of plasmid DNA is present in the lysate.

#### Vacuum Protocol:

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>.

- 8. Place the **Zymo-Spin™ II-PX Column** onto a vacuum manifold. (If vacuum is not available, see page 7 for the centrifugation protocol.)
- Add the entire mixture from step 7 into the Zymo-Spin<sup>™</sup> II-PX Column. Turn on the vacuum until all of the liquid has passed completely through the column.
- 10. Add 800 μl of **ZymoPURE™ Wash 1** to the Zymo-Spin™ II-PX Column. Turn on the vacuum until all of the liquid has passed completely through the column².
- 11. Add 800 μl of **ZymoPURE™ Wash 2** to the Zymo-Spin™ II-PX Column. Turn on the vacuum until all of the liquid has passed completely through the column.
- 12. Add 200 µl of **ZymoPURE™ Wash 2** to the Zymo-Spin™ II-PX Column. Turn on the vacuum until all of the liquid has passed completely through the column.
- 13. Place the **Zymo-Spin™ II-PX Column** in a **Collection Tube** and transfer to a microcentrifuge. Centrifuge at ≥ 10,000 x *g* for 1 minute in order to remove any residual wash buffer.
- 14. Transfer the **Zymo-Spin™ II-PX Column** into a clean 1.5 ml tube and add 25 µl of **ZymoPURE™ Elution Buffer**<sup>2,3</sup> directly to the column matrix. Incubate at room temperature for 2 minutes, and then centrifuge at ≥ 10,000 x g for 1 minute in a microcentrifuge. Store the eluted plasmid DNA at ≤ -20°C.

<sup>&</sup>lt;sup>1</sup>The lysate and wash buffers might take longer to pass through the column when less than 400 mm Hg is used.

<sup>&</sup>lt;sup>2</sup> The matrix bed inside the column will potentially become a Pinkish/Purple color after this step depending on the amount of plasmid DNA that was loaded onto the column. This is normal and not an issue as long as the binding capacity of the spin-column is not exceeded.

<sup>&</sup>lt;sup>3</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>&</sup>lt;sup>4</sup>The DNA yield can be increased by pre-warming the **Zymo PURE**<sup>™</sup> **Elution Buffer** to 50 °C and/or increasing the incubation period up to 10 minutes prior to centrifugation. For high plasmid DNA loads, more plasmid DNA can generally be recovered from the column by performing a second elution.

#### **Centrifugation Protocol:**

Perform steps 1-7 as indicated in the general protocol, see page 5.

- 8. Place a **Zymo-Spin**<sup>™</sup> **II-PX Column** in a Collection Tube.
- 9. Transfer the entire mixture from step 7 into the Zymo-Spin<sup> $\infty$ </sup> II-PX Column. Incubate the **Zymo-Spin<sup>\infty</sup>** II-PX/Collection **Tube** assembly at room temperature for 1 minute and then centrifuge at  $\ge 10,000 \ x \ g$  for 1 min. Discard the flow through<sup>1</sup>.
- 10. Add 800  $\mu$ I of **ZymoPURE™ Wash 1** to the Zymo-Spin™ II-PX Column and centrifuge at ≥ 10,000 x g for 1 min². Discard the flow through.
- 11. Add 800  $\mu$ I of **ZymoPURE™ Wash 2** to the Zymo-Spin™ II-PX Column and centrifuge at ≥ 10,000 x *g* for 1 min. Discard the flow through.
- 12. Add 200 µl of **ZymoPURE™ Wash 2** to the Zymo-Spin™ II-PX Column and centrifuge at ≥ 10,000 x *g* for 1 min. Discard the flow through.
- 13. Centrifuge the Zymo-Spin™ II-PX Column at ≥ 10,000 x g for 1 minute in order to remove any residual wash buffer.
- 14. Transfer the Zymo-Spin™ II-PX Column into a clean 1.5 ml tube and add 25 µl of **ZymoPURE™ Elution Buffer**<sup>3,4</sup> directly to the column matrix. Incubate at room temperature for 2 minutes, and then centrifuge at ≥ 10,000 x g for 1 minute in a microcentrifuge. Store the eluted plasmid DNA at ≤ -20°C.

<sup>&</sup>lt;sup>1</sup>The capacity of the collection tube with the column inserted is 900 μl. Empty the collection tube whenever necessary to prevent contamination on the spin-column with the flow-through.

<sup>&</sup>lt;sup>2</sup>The matrix bed inside the column will potentially become a Pinkish/Purple color after this step depending on the amount of plasmid DNA that was loaded onto the column. This is normal and not an issue as long as the binding capacity of the spin-column is not exceeded.

<sup>&</sup>lt;sup>3</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>&</sup>lt;sup>4</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. For high plasmid DNA loads, more plasmid DNA can generally be recovered from the column by performing a second elution.

## **Appendices**

### **Low-Copy Number Protocol**

When working with low-copy number plasmid DNA, it is possible to increase plasmid DNA yield by processing up to 10 ml of overnight culture grown in LB using the protocol below. Please be advised that using this protocol will reduce the number of preps that can be performed with this kit because it requires using larger volumes of ZymoPURE P1, P2, P3, and binding buffer.

#### Plasmid DNA Purification

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

- Centrifuge up to 10 ml<sup>1</sup> of bacterial culture in a clear 2 ml tube at full speed for 15-20 seconds in a microcentrifuge. Discard supernatant.
- Add 500 µl of cold ZymoPURE™ P1 (Red) to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
- 3. Add 500 µl of **ZymoPURE™ P2 (Blue)** and <u>immediately mix</u> by gently inverting the tube 8-10 times. <u>Do not vortex!</u> Let sit at room temperature for 3 minutes<sup>2</sup>. Cells are completely lysed when the solution appears clear, purple, and viscous.
- 4. Add 500 µl of ZymoPURE™ P3 (Yellow) and mix thoroughly by inversion. Do not vortex! Invert the tube an additional 5 times after the sample turns completely yellow. The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.
- 5. Centrifuge the neutralized lysate for 5 minutes at 16,000 x g.
- Transfer exactly 1,200 µl of supernatant from step 5 into a clean 1.5 ml microcentrifuge tube.
- Add 520 µl of ZymoPURE™ Binding Buffer to the cleared lysate from step 6 and mix thoroughly by vortexing for 15 seconds.

To continue processing the lysate using the recommended vacuum protocol, proceed to page 6. If a vacuum is not available, proceed to page 7 for an alternative centrifugation method. Step 9 in the vacuum and centrifugation protocol must be performed twice because the volume of the lysate and binding buffer mixture is greater than 900  $\mu$ l.

<sup>&</sup>lt;sup>1</sup> Depending on the volume of bacterial culture it may be necessary to repeat Step 1 several times.

<sup>&</sup>lt;sup>2</sup> Do not allow the lysis reaction to proceed for more than 3 minutes. Excessive lysis can result in denatured plasmid DNA. When processing a large number of samples, work with groups of ≤ 10 at a time.

#### **Gram-Positive Bacteria Protocol**

It is possible to isolate plasmid DNA from Gram-Positive species with the ZymoPURE Miniprep 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.

#### **Plasmid DNA Purification**

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

- 1. Centrifuge up to 0.5-5 ml<sup>1</sup> of bacterial culture in a clear 1.5 ml tube at full speed for 15-20 seconds in a microcentrifuge. Discard supernatant.
- 2. Add 250 µl of cold **ZymoPURE™ P1 (Red)** containing lysozyme<sup>2</sup> at a final concentration of 1 mg/ml to the bacterial cell pellet and resuspend completely by vortexing or pipetting.
- 3. Incubate the resuspended cell pellet at 37°C for 15-60 minutes<sup>3</sup>.
- Add 250 µl of ZymoPURE™ P2 (Blue) and immediately mix by gently inverting the tube 8-10 times. Do not vortex! Let sit at room temperature for 3 minutes⁴. Cells are completely lysed when the solution appears clear, purple, and viscous.
- Add 250 µl of ZymoPURE™ P3 (Yellow) and mix thoroughly by inversion. Do not vortex! Invert the tube an additional 5 times after the sample turns completely yellow. The sample will turn yellow when the neutralization is complete, and a yellowish precipitate will form.
- 6. Centrifuge the neutralized lysate for 5 minutes at 16,000 x g.
- 7. Transfer exactly 600 µl of supernatant from step 5 into a clean 1.5 ml microcentrifuge tube.
- Add 260 µl of ZymoPURE<sup>™</sup> Binding Buffer to the cleared lysate from step 6 and mix thoroughly by vortexing for 15 seconds.

To continue processing the lysate using the recommended vacuum protocol, proceed to page 6. If a vacuum is not available, proceed to page 7 for an alternative centrifugation method.

<sup>&</sup>lt;sup>1</sup> Depending on the volume of bacterial culture it may be necessary to repeat Step 1 several times.

<sup>&</sup>lt;sup>2</sup> Lytic enzymes other than lysozyme will require optimization and validation in the ZymoPURE P1 buffer prior to use.

<sup>&</sup>lt;sup>3</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>&</sup>lt;sup>4</sup> Do not allow the lysis reaction to proceed for more than 3 minutes. Excessive lysis can result in denatured plasmid DNA. When processing a large number of samples, work with groups of ≤ 10 at a time.

#### **Growing Overnight Culture**

The plasmid purification protocol has been optimized for Luria-Bertani (LB) media. Enriched culture medias such as Terrific Broth or Super Broth can result in reduced performance and column clogging depending on the cell density and plasmid DNA copy number. Therefore, it might be necessary to reduce the volume of culture being processed when working with enriched culture media

For overnight culture volumes greater than 10 ml, we recommend using a starter culture for optimal growth. This is accomplished by inoculating 10 ml or less of LB with the appropriate antibiotic using a colony on a plate or glycerol stock and shaking at 37°C for 8 hours. After 8 hours, prepare the larger overnight culture by diluting the starter culture 1:500 to 1:1000 with LB containing the appropriate antibiotic.

The size of the culture vessel is also critical for proper aeration of the overnight culture. The optimal culture volume to air volume ratio is 1:5 or less (Example: Use a 250 ml flask for 50 ml of culture). For best aeration, use baffled culture flasks and a vented or gas-permeable seal on the culture vessel and shake at 200 - 300 rpm.

## **Troubleshooting**

**Problem** 

#### Possible Causes and Suggested Solutions

Poor aeration of culture. 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 and shake at 200 - 300 rpm.

The culture was overgrown, undergrown, contaminated, or antibiotics were omitted from the growth medium. 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. Please refer to the Growing Overnight Culture section in the appendix for optimal culture conditions.

Too much culture used. Lysis and neutralization will be incomplete. More culture does not always equal more plasmid. 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.

#### Low DNA Yield

Incomplete neutralization: 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 centrifugation. Invert the tube an additional 3-4 times after the sample turns yellow following the addition of ZymoPURE™ P3.

ZymoPURE P2 and/or ZymoPURE Binding Buffer may have precipitated during shipping. To completely resuspend the buffers, incubate the bottles at 30-37 °C for 10 minutes and mix by inversion. DO NOT MICROWAVE.

ZymoPURE™ Wash 2: Ensure that the correct volume of ethanol was added to the ZymoPURE™ Wash 2 prior to use. Also, ensure that the bottle cap is screwed on tightly after each use to prevent evaporation of the ethanol.

Low copy-number plasmid: Increase the overnight culture processing volume up to 10 ml and use the Low-Copy Number Plasmid Protocol on page 8.

Problem	Possible Causes and Suggested Solutions
Low DNA Yield	Less than 600 µl of supernatant was recovered after pelleting the lysate debris. The ratio of binding buffer to lysate is critical for optimal performance and plasmid DNA yield will be significantly reduced if less than 600 µl of clarified lysate is used.  Incorrect column washing: Using less wash volume than stated to in the protocol, skipping a wash step, or performing
	the wash steps out of order can result in reduced plasmid DNA yields.
	Incomplete elution: For large size plasmids (> 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.
Low DNA Quality	Incomplete neutralization: Incomplete neutralization generates poor quality supernatant. Ensure that neutralization is complete by inverting the sample an additional 3-4 times after the sample turns yellow following the addition of ZymoPURE™ P3.
	<b>Insufficient centrifugation:</b> Make sure that all centrifugation steps are performed at the indicated speed and time. If a lower centrifuge speed is used, then extend the centrifugation time to compensate.
Genomic DNA in eluate	Improper handling: Sample was vortexed or handled too roughly. Genomic DNA contamination is usually caused by excessive mechanical shearing during the lysis and neutralization steps. Also, incomplete lysis or neutralization may contribute to genomic DNA contamination in your eluate.
	Overgrown culture. Overgrown or old cultures may contain

more genomic DNA contamination than fresh cultures.

Problem	Possible Causes and Suggested Solutions
RNA in Eluate	ZymoPURE P1: Ensure that ZymoPURE™ P1 has been stored at 4°C. RNase A can be purchased separately if necessary.  Too much culture used. Using more than the recommended culture volume or using enriched culture media can cause incomplete lysis and the RNase A being overwhelmed by too many cells. Reduce the volume of culture being processed or switch to LB media.  Incorrect Ratio of Lysate to Binding Buffer: Mixing too little ZymoPURE Binding Buffer with the lysate will result in degraded RNA also binding to the spin-column. Ensure the correct volume of ZymoPURE Binding Buffer is used.
Pinkish/Purple Eluate	Incorrect Column Washing: Using less wash volume than stated to in the protocol, skipping a wash step, or performing the wash steps out of order can result in recovering an eluate with a pinkish/purple color.  Exceeded Binding Capacity of Spin-Column: Using more than the recommended culture volume or using enriched culture media when preparing high-copy number plasmids might result in incomplete washing of the column and recovering an eluate with a pinkish/purple color. Reduce the volume of culture being processed or switch to LB media.
Column Clogs	Exceeded Binding Capacity of Spin-Column: Using more than the recommended culture volume or using enriched culture media when preparing high-copy number plasmids can reduce column flow and potentially completely clog the spin-column. Reduce the volume of culture being processed or switch to LB media.  Lysate Debris is loaded onto the Spin-Column: The lysate recovered after centrifugation should be free of debris. Prior to adding the ZymoPURE™ Binding Buffer to the lysate, centrifuge the lysate an additional 5 minutes at 16,000 x g and collect the supernatant if a lot of visible debris is present in the lysate recovered in step 6.

## **Ordering Information**

Product Description	Catalog No.	Size
ZymoPURE™ Plasmid Miniprep Kit	D4208T D4209 D4210 D4211 D4212	10 Preps. 50 Preps. 100 Preps. 400 Preps. 800 Preps.

Individual Kit Components	Catalog No.	Amount
ZymoPURE™ P1 (Red)	3 ml 13 ml 100 ml 210 ml	D4200-1-3 D4200-1-13 D4200-1-100 D4200-1-210
ZymoPURE™ P2 (Blue)	3 ml 13 ml 100 m 210 ml	D4200-2-3 D4200-2-13 D4200-2-100 D4200-2-210
ZymoPURE™ P3 (Yellow)	3 ml 13 ml 100 ml 210 ml	D4200-3-3 D4200-3-13 D4200-3-100 D4200-3-210
ZymoPURE™ Binding Buffer	3 ml 13 ml 100 ml 210 ml	D4200-4-3 D4200-4-13 D4200-4-100 D4200-4-210
ZymoPURE™ Wash 1	20 ml 55 ml 320 ml 420 ml	D4200-5-20 D4200-5-55 D4200-5-320 D4200-5-420
ZymoPURE™ Wash 2 (Concentrate)	10 ml 12 ml 23 ml 28 ml	D4200-6-10 D4200-6-12 D4200-6-23 D4200-6-28
ZymoPURE™ Elution Buffer	6 ml 12 ml 30 ml	D4200-7-6 D4200-7-12 D4200-7-30
Zymo-Spin™ II-PX	50	C1086-50
Collection Tubes	50 500 1000	C1001-50 C1001-500 C1001-1000

## **Complete Your Cloning Workflow**

#### √ 18 Minute Endotoxin-Free Midi & Maxipreps

ZymoPURE™ Plasmid Prep Kits	Size	Catalog No.
ZymoPURE™ II Plasmid Midiprep Kit	25 Preps. 50 Preps.	D4200 D4201
ZymoPURE™ II Plasmid Maxiprep Kit	10 Preps. 20 Preps.	D4202 D4203
ZymoPURE™ II Plasmid Gigaprep Kit	5 Preps.	D4204

#### ✓ High-Throughput purification of Transfection-Grade Plasmid DNA

ZymoPURE™ Plasmid Miniprep Kits	Size	Catalog No.
ZymoPURE™ 96 Plasmid Miniprep Kit	2 x 96 Preps. 4 x 96 Preps.	D4214 D4215

#### ✓ Simple 20 second High Efficiency Transformations

Mix & Go! Competent Cells	Size	Catalog No.
DH5α	10 x 100 μl aliquots 96 x 50 μl aliquots 96 x 50 μl aliquots PCR Plate	T3007 T3009 T3010
JM109	10 x 100 μl aliquots 96 x 50 μl aliquots	T3019 T3020
Zymo10B	10 x 100 μl aliquots 96 x 50 μl aliquots	T3003 T3005
HB101	10 x 100 μl aliquots 96 x 50 μl aliquots	T3011 T3013
TG1	10 x 100 μl aliquots	T3017

#### ✓ Recover ultra-pure highly concentrated DNA from PCR & other sources

DNA Clean & Concentrator™	Size	Catalog No.
DNA Clean & Concentrator™-5	50 Preps. 200 Preps.	D4003 D4004
ZR-96 DNA Clean-Up Kit™	2 x 96 Preps. 4 x 96 Preps.	D4017 D4018
Genomic DNA Clean & Concentrator™-10	25 Preps. 100 Preps.	D4010 D4011

#### ✓ Rapid extraction of ultra-pure DNA from agarose gels

Zymoclean Gel DNA Recovery™	Size	Catalog No.
Zymoclean™ Gel DNA Recovery Kit	50 Preps. 200 Preps.	D4001 D4002
Zymoclean™ Large Fragment DNA Recovery Kit	2 x 96 Preps. 4 x 96 Preps.	D4045 D4046

Notes			



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Reagents are routinely tested on a lot-to-lot basis to ensure they provide the highest performance and reliability.

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.

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