

# PureLink™ Pro 96 PCR Purification Kit

**For rapid, high-throughput purification of PCR  
products**

Catalog no. K3100-96A

**Rev. Date: 7 May 2009**  
Part no. 100007166

MAN0001671



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# Experienced Users Procedure

## Introduction

This quick reference sheet is included for experienced users of PureLink™ Pro 96 PCR Purification Kit. If you are a first time user, follow the detailed protocol in this manual.

Step	Action
Purification Protocol Using Centrifugation	<p>Perform all centrifugation steps at room temperature using a centrifuge with a swinging bucket rotor with plate carriers that have a plate height clearance of 7.0 cm.</p> <ol style="list-style-type: none"><li data-bbox="291 483 851 537">1. Place a PureLink™ 96 Well PCR Filter Plate on a PureLink™ 96 Receiver Plate.</li><li data-bbox="291 553 910 634">2. Add 4 volumes of Binding Buffer with isopropanol (page 4) to 1 volume of PCR product (50–100 µl). Mix well.</li><li data-bbox="291 651 833 678">3. Transfer above sample to the PCR Filter Plate.</li><li data-bbox="291 695 870 743">4. Centrifuge the stacked plates at <math>\geq 2,100 \times g</math> for 1–2 minutes. Discard the flow through.</li><li data-bbox="291 760 923 813">5. Add 600 µl Wash Buffer with ethanol to the PCR Filter Plate.</li><li data-bbox="291 829 870 878">6. Centrifuge the stacked plates at <math>\geq 2,100 \times g</math> for 1–2 minutes. Discard the flow through.</li><li data-bbox="291 894 926 948">7. <b>Repeat</b> the Wash Step one more time. Discard the flow through.</li><li data-bbox="291 964 934 1045">8. Place the PCR Filter Plate on top of a dry Receiver Plate and centrifuge the stacked plates at <math>\geq 2,100 \times g</math> for 10 minutes.</li><li data-bbox="291 1062 950 1115">9. Place the PCR Filter Plate on top of a new PureLink™ Pro 96 Elution Plate.</li><li data-bbox="291 1131 865 1159">10. Add 100 µl Elution Buffer to the PCR Filter Plate.</li><li data-bbox="291 1175 804 1203">11. Incubate at room temperature for 1 minute.</li><li data-bbox="291 1219 963 1268">12. Centrifuge the stacked plates at <math>\geq 2,100 \times g</math> for 1–2 minutes to elute DNA into the Elution Plate.</li><li data-bbox="291 1284 852 1338">13. Store DNA at <math>-20^{\circ}\text{C}</math> or use DNA for the desired downstream application.</li></ol>

## Experienced Users Procedure, Continued

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Step	Action
Purification Protocol Using Vacuum	<ol style="list-style-type: none"><li data-bbox="288 261 937 318">1. Set up the vacuum manifold. If using the EveryPrep™ Universal Vacuum Manifold, see page 16 for details.</li><li data-bbox="288 326 937 383">2. Place a PureLink™ 96 Well PCR Filter Plate on top of the manifold.</li><li data-bbox="288 391 937 480">3. Add 4 volumes of Binding Buffer with isopropanol (page 4) to 1 volume of PCR product (50–100 µl). Mix well.</li><li data-bbox="288 488 830 521">4. Transfer above sample to the PCR Filter Plate.</li><li data-bbox="288 529 905 561">5. Apply vacuum for 1–2 minutes. Release the vacuum.</li><li data-bbox="288 570 937 626">6. Add 600 µl Wash Buffer with ethanol to the PCR Filter Plate. Apply vacuum for 1 minute. Release the vacuum.</li><li data-bbox="288 634 703 667">7. <b>Repeat</b> Wash Step one more time.</li><li data-bbox="288 675 937 732">8. Apply vacuum for 10 minutes. Release the vacuum. Tap the plate on a stack of paper towels to blot the plate dry.</li><li data-bbox="288 740 937 829">9. Place an PureLink™ Pro 96 Elution Plate in the vacuum manifold (in place of the waste collection tray) and place the PCR Filter Plate on top of the manifold.</li><li data-bbox="288 837 862 870">10. Add 100 µl Elution Buffer to the PCR Filter Plate.</li><li data-bbox="288 878 799 911">11. Incubate at room temperature for 1 minute.</li><li data-bbox="288 919 905 976">12. Apply vacuum for 1–2 minutes to elute DNA into the Elution Plate. Release the vacuum.</li><li data-bbox="288 984 852 1040">13. Store DNA at –20°C or use DNA for the desired downstream application.</li></ol>

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# Kit Contents and Storage

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## Shipping and Storage

All components of the PureLink™ Pro 96 PCR Purification Kit are shipped at room temperature. Upon receipt, store all components at room temperature

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## Contents

The components included in the PureLink™ Pro 96 PCR Purification Kit are listed below.

Sufficient reagents are in the kit to perform 384 (4 × 96) reactions.

Component	Amount
PureLink™ Pro 96 Binding Buffer (B2)	120 ml
PureLink™ Pro 96 Wash Buffer (W1)	120 ml
PureLink™ Pro 96 Elution Buffer (E1); 10 mM Tris-HCl, pH 8.5	70 ml
PureLink™ 96 Well PCR Filter Plate	4
PureLink™ Pro 96 Elution Plate	4

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# Accessory Products

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## Additional Products

The following products are also available from Invitrogen. For more details on these products, visit [www.invitrogen.com](http://www.invitrogen.com) or contact Technical Support (page 16).

Product	Quantity	Catalog no.
PureLink™ 96 Receiver Plates (deep-well)	50 pack	12193-025
EveryPrep™ Universal Vacuum Manifold	1 manifold	K211101
PureLink™ PCR Purification Kit	50 reactions	K3100-01
	250 reactions	K3100-02
SequenceRx Enhancer System	25 reactions	12237-012
E-Gel® 96 1% Gels	8 gels	G7008-01
E-Gel® 96 2% Gels	8 gels	G7008-02
UltraPure™ DNase/RNase-free Distilled Water	500 ml	10977-015
Quant-iT™ DNA Assay Kit, High Sensitivity	1000 assays	Q33140
Quant-iT™ DNA Assay Kit, Broad-Range	1000 assays	Q33130
Platinum® <i>Taq</i> DNA Polymerase High Fidelity	100 reactions	11304-011
Platinum® <i>Taq</i> DNA Polymerase	100 reactions	10966-018

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# Introduction

## About the Kit

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### Introduction

The PureLink™ Pro 96 PCR Purification Kit is designed for rapid, efficient, and high-throughput purification of PCR products. The kit is designed to efficiently remove primers, dNTPs, enzymes, and salts from PCR products in less than 20 minutes.

The purified PCR product is suitable for automated fluorescent DNA sequencing, restriction enzyme digestion, cloning.

The PureLink™ Pro 96 PCR Purification Kit can be used with a vacuum manifold or a centrifuge, and is compatible with automated liquid handling workstations (page 5).

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### System Overview

The PureLink™ Pro 96 PCR Purification Kit is based on the selective binding of dsDNA to silica-based membrane in the presence of chaotropic salts.

The PCR product is mixed with the Binding Buffer to adjust conditions for subsequent dsDNA binding to the PureLink™ 96 Well PCR Filter Plate. The dsDNA binds to the silica-based membrane in the plate and impurities are removed by thorough washing with Wash Buffer. The dsDNA is then eluted in low salt Elution Buffer or water into the Receiver Plate.

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### Advantages

The PureLink™ Pro 96 PCR Purification Kit offers the following advantages:

- Ability to isolate up to 40 µg dsDNA per well
  - Designed for high-throughput purification of PCR products within 20 minutes
  - Compatible with automated liquid handling workstation (page 5)
  - Reliable performance of the purified PCR product in downstream applications
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# Product Specifications

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## PureLink™ 96 Well PCR Filter Plate

Dimensions:	Standard SBS (Society for Biomolecular Screening) footprint
Description:	Polypropylene 96-well plate
Volume:	1.0 ml
Binding Capacity:	40 µg dsDNA/well

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## System Specifications

Starting Material:	50–100 µl PCR product (50 ng–10 µg dsDNA)
Elution Volume:	100 µl
Separation Range:	0.1–12 kb from 10–40 mer primers
DNA Recovery:	>70%
Instrument Compatibility	Vacuum manifold or centrifuge (page 5)

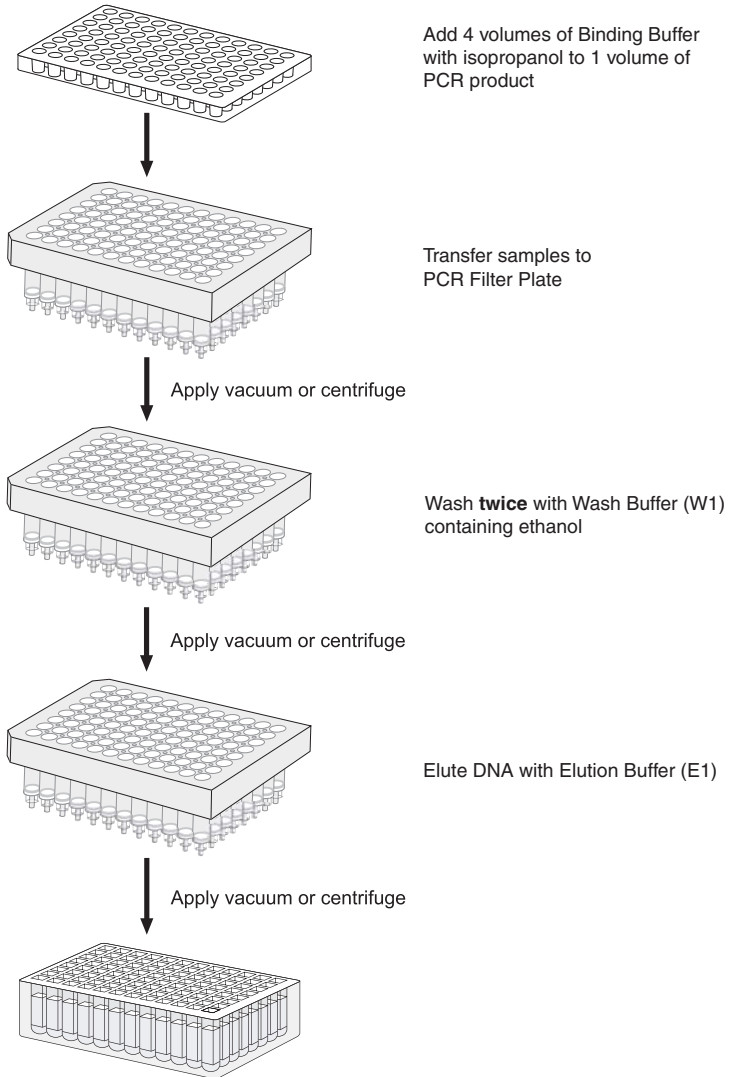
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# Experimental Overview

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## Workflow

The flow chart for purifying PCR products using the PureLink™ Pro 96 PCR Purification Kit is shown below.



# Methods

## Before Starting

### Materials Needed

- 96–100% ethanol
- 100% isopropanol



The PureLink™ Pro 96 PCR Purification Kit buffers contain guanidine hydrochloride and isopropanol. Always wear a laboratory coat, disposable gloves, and eye protection when handling buffers.

Do not add bleach or acidic solutions directly to solutions containing guanidine hydrochloride or sample preparation waste as it forms reactive compounds and toxic gases when mixed with bleach or acids.

### Preparing Buffers

- Add 80 ml isopropanol to 120 ml PureLink™ Pro 96 Binding Buffer (B2). Mark the box on the label to indicate isopropanol has been added. Mix well and store the Binding Buffer with isopropanol at room temperature.
- Add 480 ml 96–100% ethanol to the PureLink™ Pro 96 Wash Buffer (W1) bottle. Mix well and store the Wash Buffer with ethanol at room temperature.

### Recovery of Elution Volume

Based on the volume of elution buffer used for elution, the recovery of the elution volume will vary and is listed below:

Elution Buffer Volume Used	Recovered Elution Volume	
	Centrifuge	Vacuum
120 µl	119 µl	104 µl
100 µl	97 µl	82 µl
50 µl	45 µl	29 µl

*Continued on next page*

## Before Starting, Continued

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### Processing Fewer than 96 Samples

You can use a portion of the PureLink™ 96 Well PCR Filter Plate, if you wish to purify the PCR product from less than 96 samples. **Each well can only be used once.**

1. Cover the entire surface of the PCR Filter Plate with adhesive foil.
2. Just prior to use, use a sharp blade to score the foil around the wells to be used and peel away the foil to expose the clean wells.

**Important:** Keep all unused wells sealed with adhesive foil during purification to obtain uniform vacuum.

You will need a new Elution Plate for each experiment.

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### Instrument Compatibility

The PureLink™ 96 Well PCR Filter Plates are compatible with the following instruments:

- **Vacuum Manifold:** The manifold must accommodate the PureLink™ 96 Well Plates (half-skirted filter plate) and be capable of collecting the filtrate (e.g. EveryPrep™ Universal Vacuum Manifold from Invitrogen).
  - **Centrifuge:** The centrifuge must be capable of centrifuging 96-well plates at  $\geq 2,100 \times g$ , and accommodate a 7.0 cm microtiter plate stack.
  - **Automated Liquid Handling Workstation:** The workstation must be equipped with a vacuum manifold and a vacuum source, and accommodate the PureLink™ 96 Well Plate (half-skirted filter plate).
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# Purification Using Centrifugation

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## Introduction

Instructions for purification using a centrifuge are described below. All steps are performed at room temperature.

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## Materials Needed

- Centrifuge with a swinging bucket rotor with plate carriers that have a plate height clearance of 7.0 cm
  - PureLink™ 96 Receiver Plate (see page viii) or equivalent
  - PureLink™ 96 Well PCR Filter Plate (supplied with kit)
  - PureLink™ Pro 96 Elution Plate (supplied with kit)
  - Binding Buffer (B2) with isopropanol (page 4)
  - Wash Buffer (W1) with ethanol (page 4)
  - Elution Buffer (supplied with kit)
  - Sterile, distilled water (pH>7.0), **optional**
- 



Follow these recommendations to obtain the best results:

- Use 50–100  $\mu$ l of PCR product per well
  - Save an aliquot of the PCR products before purification for gel verification of the amplicon
  - Remove any residual wash buffer prior to elution as traces of wash buffer may inhibit downstream enzymatic reactions
  - Dispense Elution Buffer or water in the center of the well and incubate for 1 minute for proper elution
  - Use sterile water (pH 7–8.5), when using water for elution
  - Increase the elution buffer volume to 120  $\mu$ l to increase DNA yield (note that the DNA will be diluted)
  - **Do not** use Binding Buffer HC (supplied with the PureLink™ PCR Kit) with the PureLink™ Pro 96 PCR Kit as Binding Buffer HC is not compatible with the PureLink™ Pro 96 Plate
  - Perform all centrifugation steps at room temperature.
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# Purification Using Centrifugation, Continued

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## Binding DNA

1. Add 4 volumes of PureLink™ Pro 96 Binding Buffer (B2) with isopropanol (page 4) to 1 volume of PCR product (50–100 µl). Mix well.  
**Example:** Add 200 µl of Binding Buffer to 50 µl PCR product.
  2. Place a PureLink™ 96 Well PCR Filter Plate on a PureLink™ 96 Receiver Plate.  
**Note:** To process less than 96 samples, cover unused wells of the PCR Filter Plate with adhesive foil.
  3. Transfer sample from Step 1 to the PCR Filter Plate using a multichannel pipettor.
  4. Centrifuge the stacked plates at  $\geq 2,100 \times g$  for 1–2 minutes. Discard the flow through.
  5. Proceed to **Washing DNA**, below.
- 

## Washing DNA

1. Add 600 µl PureLink™ Pro 96 Wash Buffer (W1) with ethanol (page 4) to the PCR Filter Plate.
  2. Centrifuge the stacked plates at  $\geq 2,100 \times g$  for 1–2 minutes. Discard the flow through.
  3. **Repeat** Step 1 and Step 2.
  4. Place the PCR Filter Plate on top of a dry PureLink™ 96 Receiver Plate and centrifuge the stacked plates at  $\geq 2,100 \times g$  for 10 minutes to remove any residual wash buffer.
  5. Proceed to **Eluting DNA**, next page.
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# Purification Using Centrifugation, Continued

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## Eluting DNA

1. Place the PCR Filter Plate on top of a new PureLink™ Pro 96 Elution Plate (supplied with the kit).
2. Add 100 µl Elution Buffer (10 mM Tris-HCl, pH 8.5) or sterile, distilled water (pH >7.0) to the center of the well of the PCR Filter Plate.  
**Optional:** Use 50–120 µl of Elution Buffer (see page 4).
3. Incubate the plate at room temperature for 1 minute.
4. Centrifuge the stacked plates at  $\geq 2,100 \times g$  for 1–2 minutes to elute DNA into the Elution Plate.
5. Store purified dsDNA at  $-20^{\circ}\text{C}$  in the Elution Plate or proceed to the downstream application of choice.

See page 13 for an example of efficient primer removal.

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# Purification Using a Vacuum Manifold

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## Introduction

Instructions for purification using a vacuum manifold are described below.

All steps are performed at room temperature. For a protocol using the EveryPrep™ Universal Vacuum Manifold, see page 16.

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## Materials Needed

- Vacuum manifold and vacuum pump (producing pressure of –12 to –15 in. Hg (see next page to calibrate vacuum) or automated liquid handling workstation)
  - PureLink™ 96 Well PCR Filter Plate (supplied with kit)
  - PureLink™ Pro 96 Elution Plate (supplied with kit)
  - Binding Buffer (B2) with isopropanol (page 4)
  - Wash Buffer (W1) with ethanol (page 4)
  - Elution Buffer (supplied with kit)
  - Sterile, distilled water (pH>7.0), **optional**
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Follow these recommendations to obtain the best results:

- Use 50–100 µl of PCR product per well
  - Save an aliquot of the PCR products before purification for gel verification of the amplicon
  - Remove any residual wash buffer prior to elution as traces of wash buffer may inhibit downstream enzymatic reactions
  - Dispense Elution Buffer or water in the center of the well for proper elution
  - Perform a 1 minute incubation with Elution Buffer
  - Use sterile water (pH 7–8.5), when using water for elution
  - Increase the elution buffer volume to 120 µl to increase DNA yield (note that the DNA will be diluted)
  - **Do not** use Binding Buffer HC (supplied with the PureLink™ PCR Kit) with the PureLink™ Pro 96 PCR Kit as Binding Buffer HC is not compatible with the PureLink™ Pro 96 Plate
  - Use the recommended vacuum pressure
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# Purification Using a Vacuum Manifold,

## Continued

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### Calibrating Vacuum

Use a vacuum pressure of -12 to -15 in. Hg to obtain the best results.

Using higher vacuum pressure than the recommended pressure may cause sample splattering or inefficient DNA binding, while using lower vacuum pressure will affect the elution resulting in lower recovery.

To check the vacuum pressure:

1. Place an unused PureLink™ 96 Well PCR Filter Plate on top of the vacuum manifold.
2. Apply vacuum and check the vacuum pressure on the vacuum regulator (usually attached to the manifold or a vacuum pump).
3. Adjust the vacuum pressure on the regulator to obtain the recommended pressure of -12 to -15 in. Hg.

**Note:** During purification the vacuum pressure may exceed the recommended value.

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### Binding DNA

1. Set up the vacuum manifold using manufacturer's recommendations.  
If you are using an automated liquid handling workstation, prepare the workstation deck as recommended by the manufacturer.
2. Add 4 volumes of PureLink™ Pro 96 Binding Buffer (B2) with isopropanol (page 4) to 1 volume of PCR product (50–100 µl). Mix well.

**Example:** Add 200 µl of Binding Buffer to 50 µl PCR product.

3. Place a waste tray at the base of the manifold to collect the flow through, if necessary. Place a PureLink™ 96 Well PCR Filter Plate on top of the manifold.  
**Note:** If you wish to process less than 96 samples, seal unused wells of the PCR Filter Plate with adhesive foil.
  4. Transfer sample from Step 2 to the PCR Filter Plate using a multichannel pipettor or robotic loading device.
  5. Apply vacuum for 1–2 minutes or until all samples pass through the filter. Release the vacuum.
  6. Proceed to **Washing DNA**, next page.
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# Purification Using a Vacuum Manifold,

## Continued

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### Washing DNA

1. Add 600  $\mu$ l PureLink™ Pro 96 Wash Buffer (W1) containing ethanol (see page 4) to the PCR Filter Plate.
  2. Apply vacuum for 1 minute. Release the vacuum.
  3. **Repeat** Step 1 and Step 2.
  4. Apply vacuum for an additional 10 minutes to remove any residual Wash Buffer. Release the vacuum.  
Place the PCR Filter Plate on a stack of paper towels, and pat firmly to blot residual liquid that may be trapped in the nozzles or the bottom of the plate.
  5. Proceed to **Eluting DNA**, below.
- 

### Eluting DNA

2. Place the PureLink™ Pro 96 Elution Plate in the vacuum manifold (in place of the waste collection tray) and place the PCR Filter Plate on the manifold.  
**Note:** To avoid any cross contamination and ensure contact between the PCR Filter Plate and Elution Plate, raise the Elution Plate in the vacuum manifold.
  3. Add 100  $\mu$ l PureLink™ Pro 96 Elution Buffer (10 mM Tris-HCl, pH 8.5) or sterile, distilled water (pH >7.0) to the center of the well of the PCR Filter Plate.  
**Optional:** Use 50–120  $\mu$ l of Elution Buffer (see page 4).
  4. Incubate the plate at room temperature for 1 minute.
  5. Apply vacuum for 1–2 minutes to elute DNA into the Elution Plate. Release the vacuum.
  6. Store purified dsDNA at  $-20^{\circ}\text{C}$  in the Elution Plate or proceed to the downstream application of choice.
- See page 13 for an example of efficient primer removal.
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# Analyzing DNA Yield and Primer Removal

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## DNA Yield

After purification with PureLink™ Pro 96 PCR Purification Kit, the yield of purified dsDNA can be estimated by agarose gel electrophoresis or Quant-iT™ DNA Assay Kits.

### Agarose Gel Electrophoresis

To estimate the yield, perform agarose gel electrophoresis of the purified PCR product and known quantities of a DNA fragment of the same size (standard). Compare the band intensity of the purified PCR product with the standard DNA fragment.

### Quant-iT™ DNA Assay Kits

The Quant-iT DNA Assay Kits (see page viii for ordering information) provide a rapid, sensitive, and specific method for dsDNA quantitation with minimal interference from RNA, protein, ssDNA (primers), or other common contaminants that affect UV absorbance.

The kit contains a state-of-the-art quantitation reagent, pre-diluted standards for standard curve, and a ready-to-use buffer. The assay is performed in a microtiter plate format and is designed for reading in standard fluorescent microplate readers. Follow manufacturer's recommendations to perform the assay.

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## Primer Removal

The efficiency of primer removal can be estimated by agarose gel electrophoresis as described in examples shown on the next page.

If greater sensitivity is required, the WAVE® System is an ideal method to estimate the efficiency of primer removal. The WAVE® System is an automated DHPLC (denatured high-performance liquid chromatography) system.

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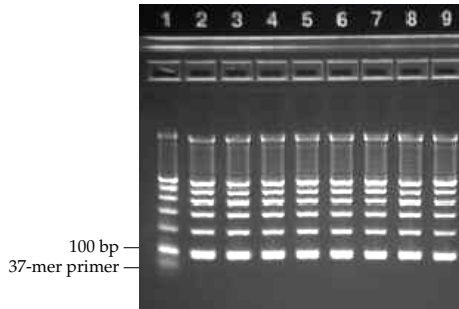
# Analyzing DNA Yield and Primer Removal, Continued

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## Example with Binding Buffer

An example of efficient primer removal is shown in the figure below.

100 bp DNA Ladder (cat. no. 15628-019) was mixed with an excess of a 37-mer primer. The mixture was purified using the PureLink™ Pro 96 PCR Purification Kit as described in the manual. The mixture was analyzed before (lane 1) and after (lanes 2–9) purification using agarose gel electrophoresis.



# Troubleshooting

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## Introduction

Review the information below to troubleshoot experiments with the PureLink™ Pro 96 PCR Purification Kit.

To troubleshoot problems with the vacuum manifold or automated liquid handling workstation, contact the manufacturer.

Problem	Cause	Solution
Low DNA yield	PCR conditions not optimized	Check amplicon on gel to verify the PCR product prior to purification.
	Incorrect binding conditions	For efficient DNA binding always <b>mix</b> 1 volume of PCR (50–100 µl) with 4 volumes of Binding Buffer (B2).  Be sure to add 100% isopropanol to the Binding Buffer as described on page 4.
	Ethanol not added to Wash Buffer	Be sure to add 96–100% ethanol to Wash Buffer (W1) included in the kit before use (page 4).
	Incorrect elution conditions	Add Elution Buffer (E1) to the center of the column and perform incubation for 1 minute before elution. Use the recommended vacuum pressure (page 9).
Low elution volume or sample cross-contamination	Incorrect vacuum pressure	Make sure the vacuum manifold is sealed tightly and there is no leakage. A vacuum pressure of –12 to –15 in. Hg is required to obtain the best results.  To avoid any cross contamination and ensure contact between the PCR Filter Plate and Elution Plate, raise the Elution Plate in the vacuum manifold.

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# Troubleshooting, Continued

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<b>Problem</b>	<b>Cause</b>	<b>Solution</b>
Inhibition of downstream enzymatic reactions	Presence of ethanol in purified DNA	<p>Traces of ethanol from the Wash Buffer can inhibit downstream enzymatic reactions.</p> <p>To remove Wash Buffer by vacuum, apply vacuum for 10 minutes to remove any residual Wash Buffer. Blot the plate dry on a stack of paper to remove residual liquid trapped in the nozzles or on the bottom of the plate.</p> <p>To remove Wash Buffer by centrifugation, discard Wash Buffer flow through and centrifuge the plate at maximum speed for 10 minutes to completely dry the column.</p>
	High salt in sample	Be sure to perform 2 wash steps with Wash Buffer containing ethanol.

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# Appendix

## Purification Using the EveryPrep™ Universal Vacuum Manifold

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### Introduction

Instructions are provided below to purify DNA using the EveryPrep™ Universal Vacuum Manifold (see page viii). Refer to the manual for the EveryPrep™ Universal Vacuum Manifold for detailed instructions on operation with the 96 Well Top Plate. All steps are performed at room temperature.

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### Materials Needed

- Vacuum manifold and vacuum pump (producing pressure of 12–15 in. Hg)
  - PureLink™ 96 Well PCR Filter Plate (supplied with kit)
  - PureLink™ Pro 96 Elution Plate (supplied with kit)
  - Binding Buffer (B2) with isopropanol (page 4)
  - Wash Buffer (W1) with ethanol (page 4)
  - Elution Buffer (supplied with kit)
  - Sterile, distilled water (pH>7.0), **optional**
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# Purification Using the EveryPrep™ Universal Vacuum Manifold, Continued

## EveryPrep™ Universal Vacuum Manifold Assembly

1. Assemble the EveryPrep™ Universal Vacuum Manifold: Place the Waste Tray in the Binding Chamber, cover the top with the 96 Well Top Plate, and place the PureLink™ PCR Filter Plate over the Top Plate.



2. Proceed to **Binding DNA**, below.

## Binding DNA

1. Add 4 volumes of PureLink™ Pro 96 Binding Buffer (B2) with isopropanol (page 4) to 1 volume of PCR product (50–100  $\mu$ l). Mix well.  
**Example:** Add 200  $\mu$ l of Binding Buffer to 50  $\mu$ l PCR product.
2. Transfer samples to the PCR Filter Plate using a multichannel pipettor.
3. Apply vacuum for 1–2 minutes or until all samples pass through the PCR Filter Plate. Release the vacuum.
4. Proceed to **Washing DNA**, next page.

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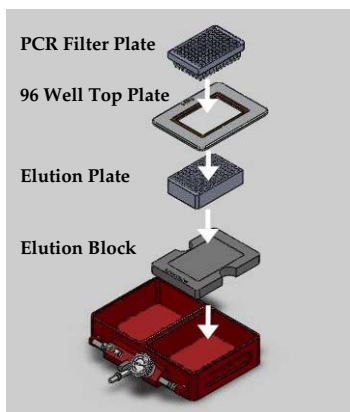
# Purification Using the EveryPrep™ Universal Vacuum Manifold, Continued

## Washing DNA

1. Add 600 µl PureLink™ Pro 96 Wash Buffer (W1) containing ethanol (see page 4) to the PCR Filter Plate.
2. Apply vacuum for 1 minute. Release the vacuum.
3. **Repeat** Step 1 and Step 2.
4. Apply vacuum for an additional 10 minutes to remove any residual Wash Buffer. Release the vacuum. Place the PCR Filter Plate on a stack of paper towels, and pat firmly to blot any residual liquid.
5. Proceed to **Eluting DNA**, below.

## Eluting DNA

1. Prepare the EveryPrep™ Universal Vacuum Manifold for elution: Place the Elution Block and PureLink™ Pro 96 Elution Plate in the Elution Chamber, cover the top with the 96 Well Top Plate, and place the PureLink™ PCR Filter Plate over the Top Plate.



2. Add 100 µl PureLink™ Pro 96 Elution Buffer (10 mM Tris-HCl, pH 8.5) or sterile, distilled water (pH >7.0) to the center of the well of the PCR Filter Plate.  
**Optional:** Use 50–120 µl of Elution Buffer (see page 4).
3. Incubate the plate at room temperature for 1 minute.
4. Apply vacuum for 1–2 minutes to elute DNA into the Elution Plate. Release the vacuum.
5. Store purified dsDNA at –20°C in the Elution Plate or proceed to the downstream application of choice.

See page 13 for an example of efficient primer removal.

# Technical Support

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## World Wide Web



Visit the Invitrogen website at [www.invitrogen.com](http://www.invitrogen.com) for:

- Technical resources, including manuals, vector maps and sequences, application notes, MSDSs, FAQs, formulations, citations, handbooks, etc.
- Complete technical support contact information
- Access to the Invitrogen Online Catalog

Additional product information and special offers

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## Contact Us

For more information or technical assistance, call, write, fax, or email. Additional international offices are listed on our website ([www.invitrogen.com](http://www.invitrogen.com)).

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## MSDS Requests

Material Safety Data Sheets (MSDSs) are available on our website at [www.invitrogen.com/msds](http://www.invitrogen.com/msds).

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## Certificate of Analysis

The Certificate of Analysis provides detailed quality control and product qualification information for each product. Certificates of Analysis are available on our website. Go to [www.invitrogen.com/support](http://www.invitrogen.com/support) and search for the Certificate of Analysis by product lot number, which is printed on the box.

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# Purchaser Notification

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**Notes:**

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