August 2004

MinElute® 96 UF PCR Purification Handbook

For efficient, high-throughput purification of PCR products

QIAGEN

WWW.QIAGEN.COM

Contents

Kit Contents

MinElute 96 UF PCR Purification Kit Catalog no.	(4) 28051	(24) 28053
MinElute 96 UF PCR Purification Plates	4	24
Handbook	1	1

Storage Conditions

MinElute 96 UF PCR Purification Plates should be stored dry and at room temperature. They can be stored for up to 12 months without showing any reduction in performance, capacity, or quality of separation.

Quality Control

As part of the stringent QIAGEN quality assurance program, the performance of MinElute UF PCR Purification Kits is monitored routinely and on a lot-to-lot basis. MinElute UF PCR Purification Kits are tested for primer removal and by isolation of DNA fragments from aqueous solution.

Product Use Limitations

MinElute UF PCR Purification Kits are developed, designed, and sold for research purposes only. They are not to be used for human diagnostic or drug purposes or to be administered to humans unless expressly cleared for that purpose by the Food and Drug Administration in the USA or the appropriate regulatory authorities in the country of use. All due care and attention should be exercised in the handling of many of the materials described in this text.

Safety Information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs). These are available online in convenient and compact PDF format at www.qiagen.com/ts/msds.asp where you can find, view, and print the MSDS for each QIAGEN kit and kit component.

24-hour emergency information

Emergency medical information in English, French, and German can be obtained 24 hours a day from:

Poison Information Center Mainz, Germany

Tel: +49-6131-19240

Product Warranty and Satisfaction Guarantee

QIAGEN guarantees the performance of all products in the manner described in our product literature. The purchaser must determine the suitability of the product for its particular use. Should any product fail to perform satisfactorily due to any reason other than misuse, QIAGEN will replace it free of charge or refund the purchase price. We reserve the right to change, alter, or modify any product to enhance its performance and design. If a QIAGEN product does not meet your expectations, simply call your local Technical Service Department or distributor. We will credit your account or exchange the product — as you wish.

A copy of QIAGEN terms and conditions can be obtained on request, and is also provided on the back of our invoices. If you have questions about product specifications or performance, please call QIAGEN Technical Services or your local distributor (see inside back cover).

Technical Assistance

At QIAGEN we pride ourselves on the quality and availability of our technical support. Our Technical Service Departments are staffed by experienced scientists with extensive practical and theoretical expertise in molecular biology and the use of QIAGEN products. If you have any questions or experience any difficulties regarding MinElute UF PCR Purification Kits or QIAGEN products in general, please do not hesitate to contact us.

QIAGEN customers are a major source of information regarding advanced or specialized uses of our products. This information is helpful to other scientists as well as to the researchers at QIAGEN. We therefore encourage you to contact us if you have any suggestions about product performance or new applications and techniques.

For technical assistance and more information please call one of the QIAGEN Technical Service Departments or local distributors (see inside back cover).

Typical Results*

DNA recovery (≥100 bp):	80–95%
Reproducibility of recovery:	CV≤5%
Elution volume:	20 µl
Volume of eluate:	20 µl
Primers (<20mer):	Removed
DNA binding capacity per well:	15 µg

* Table refers to the manual procedure and results may vary if using an automated procedure.

Introduction

The MinElute 96 UF PCR Purification Kit is designed to provide manual or fully automated high-throughput PCR purification. MinElute 96 UF PCR Purification Kits provide pure nucleic acids, which are highly suited for direct use in many applications, such as:

- Sequencing
- Microarray analysis

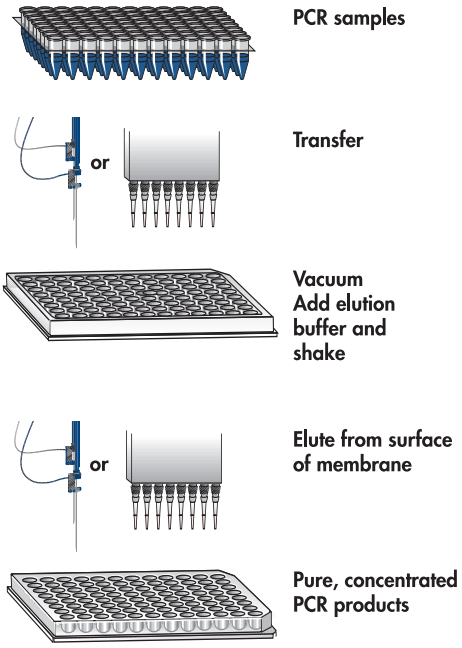
This handbook contains technical information about the MinElute 96 UF PCR Purification system to help users gain maximum benefit from the kit. The handbook explains the technology underlying MinElute 96 UF PCR purification and outlines the major steps of the procedures. Detailed protocols are included, as well as a Troubleshooting Guide to help analyze any difficulties. If you need any further information please contact our Technical Service Department.

Please spend some time reading this handbook and familiarizing yourself with the MinElute 96 UF PCR Purification system.

The MinElute 96 UF PCR Purification Principle

The QIAGEN MinElute 96 UF PCR Purification Kit combines the convenience of multiwell technology and the separation properties of an ultrafiltration membrane with a unique design. The plates enable the fast and efficient recovery of double-stranded DNA fragments ≥100 bp and the removal of primers (<20mers), unincorporated nucleotides, and salts. PCR products are applied to the wells of the ultrafiltration plate and a vacuum is applied. While small molecules such as primers, salts, and unincorporated nucleotides run through the membrane, PCR products are retained. Purified PCR products are eluted from the surface of the membranes in small volumes (down to 20 µl), giving eluates with high concentrations of DNA. It is possible to process PCR sample volumes up to 150 µl using either a manual or fully automated procedure. PCR products are typically purified with a recovery of 80–95%.

MinElute 96 UF PCR Purification Procedure



96 Samples 13 min (manual procedure)

Protocol: MinElute 96 UF PCR Purification (Manual Procedure)

Important notes before starting

This protocol is for purification of up to 96 PCR samples in parallel using a manual procedure.

- The use of MinElute 96 UF PCR Purification Plates requires a suitable vacuum manifold, e.g., the QIAvac Multiwell Unit, cat. no. 9014579.
- A multichannel pipet facilitates handling of PCR samples.
- For elution of DNA from MinElute 96 UF PCR Purification Plates, use of a microplate shaker is recommended. Alternatively, purified DNA can be dissolved by pipetting samples up and down 20 times.
- Deionized water (used for the optional wash step) and elution buffer must be supplied by the user.

Calibrating a microplate shaker for DNA elution

Calibrate a microplate shaker by following the steps below.

1. **Use a 96-well polystyrene microplate with 300 µl round-bottom wells, e.g., 96-Well Microplates RB (24), QIAGEN cat. no. 19581.**
2. **Add 200 µl of a colored aqueous solution (e.g., bromophenol blue)*, to two wells.**
3. **Place the 96-well plate on a microplate shaker.**
4. **Set the speed to the lowest level and slowly increase the speed of the shaker. Ensure that the plate is fixed securely on top of the shaker.**
5. **The recommended shaking speed for elution is the maximum speed at which no liquid is splashed out of the wells.**

Purification procedure

1. **Prepare the vacuum manifold according to the supplier's instructions.**

   Place a waste tray inside the base of the manifold.
2. **Place the MinElute 96 UF PCR Purification Plate on top of the vacuum manifold.**
3. **Pipet the PCR samples onto the MinElute 96 UF PCR Purification Plate.**

   Note: Processing PCR sample volumes larger than 150 µl may lead to increased processing time and incomplete primer removal.

* When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs), available from the product supplier.

4. **Apply a vacuum and maintain at –800 mbar for 5 min or until the wells are completely dry. Switch off vacuum source.**

   Note: If the PCR volume exceeds 50 µl, a longer vacuum time is needed. Apply the vacuum until all wells are dry. Approximately 5 min of vacuum application are needed for each 50 µl PCR volume.

5. **Optional: Add 50 µl deionized water to each well, apply a vacuum, and maintain at –800 mbar for 5 minutes or until the wells are completely dry. Switch off vacuum source.**

   Note: The purity of the DNA obtained after elution is sufficient for most applications without this wash step being required. If a higher purity is needed for a specific application, this step should be carried out.

6. **Carefully remove the MinElute 96 UF PCR Purification Plate from the vacuum manifold.**

7. **Carefully tap the MinElute 96 UF PCR Purification Plate on a stack of clean absorbent paper to remove any liquid that might remain on the bottom of the plate.**

8. **Add 20 µl deionized water to each well.**

   DMSO (50% v/v)*, 3 x SSC*, and EB (10 mM Tris·Cl, pH 8.5)* or similar buffers can be used instead of water for elution.

9. **Elute DNA according to step 9a or 9b.**

9a. **Shake the MinElute 96 UF PCR Purification Plate on a microplate shaker for 2 min at the recommended speed (see "Calibrating a microplate shaker for DNA elution").**

   Note: Ensure that the MinElute 96 UF PCR Purification Plate is fixed securely on top of the shaker.

9b. **Alternatively, purified DNA may be dissolved by pipetting samples up and down 20 times.**

10. **Recover the purified PCR product by pipetting the eluate out of each well.**

    For easier recovery of the eluates, the plate can be held at a slight angle.

* When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs), available from the product supplier.

Protocol: MinElute 96 UF PCR Purification Using the BioRobot® 3000

The following protocol is for MinElute 96 UF PCR Purification using the BioRobot 3000. The recommended minimum elution volume for MinElute 96 UF PCR Purification using the BioRobot 3000 is 30 µl. Before starting with the purification, ensure that the BioRobot is calibrated properly.

Your BioRobot 3000 workstation requires the following features for MinElute 96 UF PCR purification:

- High-speed pipetting system
- Robotic handling system, T-grip
- Automated Vacuum Unit 3000
- Shaker system, 4-plate
- Shaker adapter, microplate, LHS
- Pipetting probe (0.9 mm)
- Precision syringe (0.5 ml)

Procedure

1. **Make sure that the BioRobot 3000 is switched on.**
2. **Switch on the computer and the monitor.**
3. **Launch QIAsoft, if necessary.**
4. **Select "MinElute 96 UF PCR Purification" from the protocol field and click "RUN" on the toolbar.**
5. **The "Layout Configuration" dialog box appears. Click "OK".**
6. **The "Select Samples" dialog box appears. Choose the number of samples and click "OK".**
7. **Follow the instructions provided by the QIAsoft Wizard.**

   The Wizard gives a step-by-step guide to entering parameters and placing items on the worktable.

Protocol: MinElute 96 UF PCR Purification Using the BioRobot 8000

The following protocol is for MinElute 96 UF PCR Purification using the BioRobot 8000. The recommended minimum elution volume for MinElute 96 UF PCR Purification using the BioRobot 8000 is 30 µl. Before starting with the purification, ensure that the BioRobot is calibrated properly.

Your BioRobot 8000 workstation requires the following features for MinElute 96 UF PCR purification:

- Robotic Handling System, (LabHand 8000)
- Automated Vacuum System
- High-Speed Shaker System
- Probe Set 8000, 09/500

Procedure

1. **Make sure that the BioRobot 8000 is switched on.**
2. **Switch on the computer and the monitor.**
3. **Launch QIAsoft, if necessary.**
4. **Select "MinElute 96 UF PCR Purification" from the protocol field and click "RUN" on the toolbar.**
5. **The "Layout Configuration" dialog box appears. Click "OK".**
6. **The "Select Samples" dialog box appears. Choose the number of samples and click "OK".**
7. **Follow the instructions provided by the QIAsoft Wizard.**

   The Wizard gives a step-by-step guide to entering parameters and placing items on the worktable.

Recommendations for working with an automated system

- The use of MinElute 96 UF PCR Purification Plates requires a suitable vacuum manifold capable of maintaining a vacuum of –800 mbar.
- Set times and parameters as recommended for the manual procedure.
- For elution of DNA from MinElute 96 UF PCR Purification Plates, use of a microplate shaker is recommended. Alternatively, purified DNA can be dissolved by pipetting samples up and down 20 times.
- For automated systems, it is recommended to start protcol development using an elution volume of 30 µl.

Troubleshooting Guide

	Comments and suggestions
Low DNA recovery	Modify the elution conditions (e.g., by increasing the elution volume or checking optimal shaking speed).
Insufficient primer removal	Ensure the vacuum is maintained at –800 mbar. A weaker vacuum may result in insufficient removal of primers. The optional wash step helps remove primers more effectively.
PCR contains detergents	Some detergents, such as Triton® X-100, may not be completely removed by ultrafiltration. Where possible, omit detergents from PCR setup.

Ordering Information

Product	Contents	Cat. no.
MinElute 96 UF PCR Purification Kit (4)	4 MinElute 96 UF PCR Purification Plates	28051
MinElute 96 UF PCR Purification Kit (24)	24 MinElute 96 UF PCR Purification Plates	28053
Vacuum manifold		
QIAvac Multiwell	Vacuum manifold for manual processing of MinElute 96 UF PCR Purification Plates: includes QIAvac Multiwell Top and Base	9014579

Notes

QIAGEN Companies

Please see the back cover for contact information for your local QIAGEN office.

QIAGEN Distributors

Argentina
Tecnolab S.A.
Tel: (011) 4555 0010
Fax: (011) 4553 3331
E-mail: info@tecnolab.com.ar
Web site: www.tecnolab.com.ar

Austria
VWR International GmbH
Tel: (01) 576 00 0
Fax: (01) 576 00 600
E-mail: info@at.vwr.com
Web site: www.vwr.com

Brazil
Uniscience do Brasil
Tel: 011 3622 2320
Fax: 011 3622 2323
E-mail: info@uniscience.com
Web site: www.uniscience.com

China
Gene Company Limited
Tel: (852)2896-6283
Fax: (852)2515-9371
E-mail:
Hong Kong:
info@genehk.com
Beijing:
info_bj@genecompany.com
Shanghai:
info_sh@genecompany.com
Chengdu:
gene@public.cd.sc.cn
Guangzhou:
info_gz@genecompany.com

Croatia
INEL Medicinska Tehnika d.o.o.
Tel: (01) 2984-898
Fax: (01) 6520-966
E-mail:
inel-medicinska-tehnika@zg.htnet.hr

Cyprus
Scientronics Ltd
Tel: 02-357 22 765416
Fax: 02-357 22 764614
E-mail: a.sarpetsas@biotronics.com.cy

Czech Republic
BIO-CONSULT spol. s.r.o.
Tel/Fax: (420) 2 417 29 792
E-mail: info@bioconsult.cz
Web site: www.bioconsult.cz

Denmark
VWR International A/S
Tel: 43 86 87 88
Fax: 43 86 87 90
E-mail: info@dk.vwr.com
Web site: www.vwr.com

Egypt
Clinilab
Tel: 52 57 212
Fax: 52 57 210
E-mail: Clinilab@link.net

Finland
VWR International Oy
Tel: (09) 804 551
Fax: (09) 8045 5200
E-mail: info@fi.vwr.com
Web site: www.vwr.com

Greece
BioAnalytica S.A.
Tel: (210)-640 03 18
Fax: (210)-646 27 48
E-mail: bioanalyt@hol.gr
Web site: www.bioanalytica.gr

Hungary
Kasztel-Med Co. Ltd.
Tel: (01) 385 3887
Fax: (01) 381 0695
E-mail: info@kasztel.hu
Web site: www.kasztel.hu

India
Genetix
Tel: (011)-2542 1714
or (011)-2515 9346
Fax: (011)-2546 7637
E-mail: genetix@nda.vsnl.net.in

Israel
Westburg (Israel) Ltd.
Tel: 08-6900655
or 1-800 20 22 20 (toll free)
Fax: 08-6900650
E-mail: info@westburg.co.il
Web site: www.westburg.co.il

Korea
LRS Laboratories, Inc.
Tel: (02) 924-86 97
Fax: (02) 924-86 96
E-mail: webmaster@lrslab.co.kr
Web site: www.lrslab.co.kr

Malaysia
RESEARCH BIOLABS SDN. BHD.
Tel: (603)-8070 3101
Fax: (603)-8070 5101
E-mail: biolabs@tm.net.my
Web site: www.researchbiolabs.com

Mexico
Quimica Valaner S.A. de C.V.
Tel: (55) 55 25 57 25
Fax: (55) 55 25 56 25
E-mail: ventas@valaner.com
Web site: www.valaner.com

New Zealand
Biolab Ltd
Tel: (09) 980 6700
or 0800 933 966
Fax: (09) 980 6788
E-mail:
biosciences@nzl.biolabgroup.com
Web site: www.biolabgroup.com/nzl

Norway
VWR International AS
Tel: 02 29 0
Fax: 22 90 00 40
E-mail: info@no.vwr.com
Web site: www.vwr.com

Poland
Syngen Biotech Sp.z.o.o.
Tel: (071) 351 41 06
or 0601 70 60 07
Fax: (071) 351 04 88
E-mail: info@syngen.pl
Web site: www.syngen.pl

Portugal
IZASA PORTUGAL, LDA
Tel: (21) 424 7312
Fax: (21) 417 2674
E-mail: consultasbiotec@izasa.es

Singapore
Research Biolabs Pte Ltd
Tel: 67775366
Fax: 67785177
E-mail: sales@researchbiolabs.com
Web site: www.researchbiolabs.com

Slovak Republic
BIO-CONSULT Slovakia spol. s.r.o.
Tel/Fax: (02) 5022 1336
E-mail: bio-cons@cdicon.sk
Web site: www.bioconsult.cz

Slovenia
MEDILINE d.o.o.
Tel: (01) 830-80-40
Fax: (01) 830-80-70
or (01) 830-80-63
E-mail: mediline@siol.net

South Africa
Southern Cross Biotechnology
(Pty) Ltd
Tel: (021) 671 5166
Fax: (021) 671 7734
E-mail: info@scb.co.za

Spain
IZASA, S.A.
Tel: (93) 902.20.30.90
Fax: (93) 902.22.33.66
E-mail: consultasbiotec@izasa.es

Sweden
VWR International AB
Tel: (08) 621 34 00
Fax: (08) 760 45 20
E-mail: info@se.vwr.com
Web site: www.vwr.com

Taiwan
TAIGEN Bioscience Corporation
Tel: (02) 2880 2913
Fax: (02) 2880 2916
E-mail: taigen@ms10.hinet.net

Thailand
Theera Trading Co. Ltd.
Tel: (02) 412-5672
Fax: (02) 412-3244
E-mail: theetrad@samart.co.th

Turkey
Medek Medikal Ürünler
ve Saglik Hizmetleri A. S.
Tel: (216) 302 15 80
Fax: (216) 302 15 88
E-mail: makialp@med-ek.com

All other countries
QIAGEN GmbH, Germany

Australia ▪ **QIAGEN Pty Ltd** ▪ PO Box 641 ▪ DONCASTER VIC 3108
Orders 03-9840-9800 ▪ Fax 03-9840-9888 ▪ Technical 1-800-243-066

Belgium ▪ **QIAGEN Benelux B.V.** ▪ Spoorstraat 50 ▪ 5911 KJ Venlo ▪ The Netherlands
Orders 0800-79612 ▪ Fax 0800-79611 ▪ Technical 0800-79556

Canada ▪ **QIAGEN Inc.** ▪ 2800 Argentia Road ▪ Unit 7 ▪ Mississauga ▪ Ontario ▪ L5N 8L2
Orders 800-572-9613 ▪ Fax 800-713-5951 ▪ Technical 800-DNA-PREP (800-362-7737)

France ▪ **QIAGEN S.A.** ▪ 3 avenue du Canada ▪ LP 809 ▪ 91974 COURTABOEUF CEDEX
Orders 01-60-920-920 ▪ Fax 01-60-920-925 ▪ Technical 01-60-920-930

Germany ▪ **QIAGEN GmbH** ▪ QIAGEN Strasse 1 ▪ 40724 Hilden
Orders 02103-29-12000 ▪ Fax 02103-29-22000 ▪ Technical 02103-29-12400

Italy ▪ **QIAGEN S.p.A.** ▪ Via Grosio, 10/10 ▪ 20151 Milano
Orders 02-33430411 ▪ Fax 02-33430426 ▪ Technical 02-33430414

Japan ▪ **QIAGEN K.K.** ▪ Forefront Tower II ▪ 13-1, Kachidoki 3 Chome ▪ Chuo-ku, Tokyo 104-0054
Telephone 03-5547-0811 ▪ Fax 03-5547-0818 ▪ Technical 03-5547-0811

Luxembourg ▪ **QIAGEN Benelux B.V.** ▪ Spoorstraat 50 ▪ 5911 KJ Venlo ▪ The Netherlands
Orders 8002-2076 ▪ Fax 8002-2073 ▪ Technical 8002-2067

The Netherlands ▪ **QIAGEN Benelux B.V.** ▪ Spoorstraat 50 ▪ 5911 KJ Venlo
Orders 0800-0229592 ▪ Fax 0800-0229593 ▪ Technical 0800-0229602

Switzerland ▪ **QIAGEN AG** ▪ Auf dem Wolf 39 ▪ 4052 Basel
Orders 061-319-30-30 ▪ Fax 061-319-30-33 ▪ Technical 061-319-30-31

UK and Ireland ▪ **QIAGEN Ltd.** ▪ QIAGEN House ▪ Fleming Way ▪ Crawley ▪ West Sussex, RH10 9NQ
Orders 01293-422-911 ▪ Fax 01293-422-922 ▪ Technical 01293-422-999

USA ▪ **QIAGEN Inc.** ▪ 27220 Turnberry Lane ▪ Suite 200 ▪ Valencia ▪ CA 91355
Orders 800-426-8157 ▪ Fax 800-718-2056 ▪ Technical 800-DNA-PREP (800-362-7737)