QIAsymphony® DNA Investigator® Handbook

QIAsymphony DNA Investigator Kit

For purification of DNA from surface and buccal swabs
FTA® and Guthrie cards
body fluid stains
chewing gum
cigarette butts
nail clippings and hair
paper and similar materials
small volumes of blood or saliva
bones and teeth
sexual assault specimens
using the QIAsymphony SP



QIAGEN Sample and Assay Technologies

QIAGEN is the leading provider of innovative sample and assay technologies, enabling the isolation and detection of contents of any biological sample. Our advanced, high-quality products and services ensure success from sample to result.

QIAGEN sets standards in:

- Purification of DNA, RNA, and proteins
- Nucleic acid and protein assays
- microRNA research and RNAi
- Automation of sample and assay technologies

Our mission is to enable you to achieve outstanding success and breakthroughs. For more information, visit www.qiagen.com.

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Kit Contents

QIAsymphony DNA Investigator Kit	(192)*
Catalog no.	931436
Number of preps	192
Reagent Cartridge ^{†‡}	2
Enzyme Rack	2
Piercing Lid	2
Buffer ATE [‡]	20 ml
Buffer AVE [‡]	20 ml
Buffer ATL	2 x 50 ml
Carrier RNA	310 μg
Proteinase K	3 x 1.4 ml
Reuse Seal Set§	2
Trough Cover (for magnetic particles)	1
Handbook	1

^{*} For 192 x 200 μ l preps, 144 x 500 μ l preps, or 96 x 1000 μ l preps.

Storage

QIAsymphony DNA Investigator Kits should be stored at room temperature (15–25°C). Do not store reagent cartridges at temperatures below 15°C.

QlAsymphony DNA Investigator Kits contain ready-to-use proteinase K solution that can be stored at room temperature. To store for extended periods of time, we recommend storing the proteinase K at 2–8°C.

When stored properly, the kit is stable until the expiration date on the kit box.

Partially used reagent cartridges can be stored for a maximum of two weeks, enabling cost-efficient reuse of reagents and more flexible sample processing. If a reagent cartridge is partially used, replace the cover of the trough containing the magnetic particles, seal the buffer troughs with the provided Reuse Seal

[†] Contains guanidine salts. Not compatible with disinfectants containing bleach. See page 6 for safety information.

[‡] Contains sodium azide as a preservative.

[§] A Reuse Seal Set contains 8 Reuse Seal Strips.

Strips, and close the carrier RNA tubes with screw caps immediately after the end of the protocol run to avoid evaporation.

To avoid evaporation, the reagent cartridge should be open for a maximum of 15 hours (including run times) at a maximum environmental temperature of 30°C.

Running batches with low sample numbers (<24) will increase both the time that the reagent cartridge is open and the required buffer volumes, potentially reducing the total number of sample preparations possible per cartridge.

Avoid exposure of the reagent cartridges (RC) to UV light (e.g., used for decontamination) as exposure may cause accelerated aging of the reagent cartridges (RC) and buffers.

Intended Use

The QIAsymphony DNA Investigator Kit is intended for molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of a disease.

All due care and attention should be exercised in the handling of the products. We recommend all users of QIAGEN® products to adhere to the NIH guidelines that have been developed for recombinant DNA experiments, or to other applicable guidelines.

The QIAsymphony DNA Investigator Kit is not intended to be used for isolation and purification of RNA.

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. Separate conditions apply to QIAGEN scientific instruments, service products, and to products shipped on dry ice. Please inquire for more information.

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 back cover or visit www.qiagen.com).

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 sample and assay technologies and the use of QIAGEN products. If you have any questions or experience any difficulties regarding the QIAsymphony DNA Investigator Kit 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 see our Technical Support Center at www.qiagen.com/Support or call one of the QIAGEN Technical Service Departments or local distributors (see back cover or visit www.qiagen.com).

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of QIAsymphony DNA Investigator Kit is tested against predetermined specifications to ensure consistent product quality.

Safety Information

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



DO NOT add bleach or acidic solutions directly to the sample-preparation waste.

Buffers in the reagent cartridge contain guanidine salts, which can form highly reactive compounds when combined with bleach. If liquid containing these buffers is spilt, clean with suitable laboratory detergent and water. If the spilt liquid contains potentially infectious agents, clean the affected area first with laboratory detergent and water, and then with 1% (v/v) sodium hypochlorite.

24-hour emergency information

Chemical emergency or accident assistance is available 24 hours a day from:

CHEMTREC

USA & Canada = Tel: 1-800-424-9300

Outside USA & Canada ■ Tel: +1-703-527-3887 (collect calls accepted)

Introduction

The QIAsymphony DNA Investigator Kit is designed for automated purification of total DNA from samples encountered in forensic, human identity, and biosecurity applications. Proven, performance-leading magnetic-particle technology provides high-quality DNA, which is suitable for direct use in downstream applications, such as quantitative PCR or STR analyses, or for storage for later use. Purified DNA is free of proteins, nucleases, and inhibitors. The QIAsymphony SP performs all steps of the sample extraction procedure after lysis according to the pretreatment protocols. Up to 96 samples are processed in a single run.

Principle and procedure

QIAsymphony technology combines the speed and efficiency of silica-based DNA purification with the convenient handling of magnetic particles (Figure 1). The purification procedure is designed to ensure safe and reproducible handling of precious or potentially infectious samples, and comprises 4 steps: lyse, bind, wash, and elute (see flowchart, next page). The user can choose different elution volumes between 30 μ l to 400 μ l of water or modified TE Buffer (Buffer ATE), depending on the protocol. DNA yields depend on sample type, age, and storage.

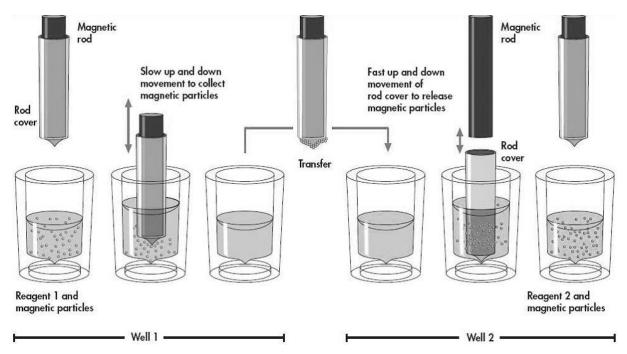
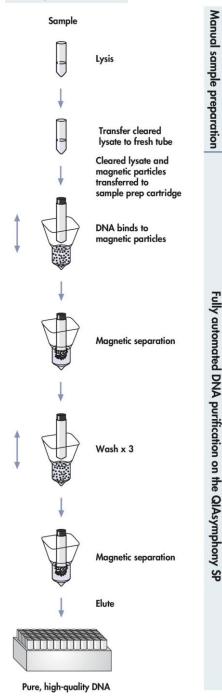


Figure 1. Schematic of the QIAsymphony SP principle. The QIAsymphony SP processes a sample containing magnetic particles as follows. A magnetic rod protected by a rod cover enters a well containing the sample and attracts the magnetic particles. The magnetic rod cover is positioned above another well and the magnetic particles are released. The QIAsymphony SP uses a magnetic head containing an array of 24 magnetic rods, and can therefore process up to 24 samples simultaneously. Steps 1 and 2 are repeated several times during sample processing.

QIAsymphony DNA Investigator Procedure



Fully automated DNA purification on the QIAsymphony SP

Description of protocols

There are two different kinds of DNA purification protocols, which can be used in conjunction with the pretreatment protocols (see protocol sheets at www.qiagen.com/goto/qsdnainvestigator).

- Casework protocols purify genomic and mitochondrial DNA from 200 μ l, 500 μ l, or 1000 μ l of lysate obtained from the sample lysis procedures detailed in the corresponding pretreatment protocols. DNA can be eluted in 30–200 μ l of either water or modified TE Buffer (Buffer ATE).
- Casework HE (High Efficiency) protocols are optimized for elution in small volumes (30–80 μ l). The protocols use TopElute Fluid (TOPE) to overlay eluates during the elution process. TopElute Fluid is not transferred to the elution labware.
- Casework ADV (Advanced) protocols are optimized for maximum recovery of DNA. ADV protocols use an extended and heated binding step.
- Reference protocols purify DNA from database samples, such as buccal swabs or dried blood. DNA can be eluted in $100-400 \mu l$ of Buffer ATE.

The purified DNA is ready to use in downstream applications.

Equipment and Reagents to Be Supplied by User

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

All protocols

- Sample Prep Cartridges, 8-well cartridges (cat. no. 997002)
- 8-Rod Covers (cat. no. 997004)
- Filter-Tips, 200 μ l and 1500 μ l (cat. nos. 990332 and 997024)
- Sample tubes or plates (e.g., 2 ml sample tubes with screw caps, Sarstedt cat. no. 72.693, or without caps, Sarstedt cat. no. 72.608, or S-Blocks, QIAGEN cat. no. 19585). Compatible primary and secondary tube formats are listed at www.giagen.com/QIAsymphony/Resources
- Elution tubes. Compatible elution tube formats are listed at www.qiagen.com/QIAsymphony/Resources
- Pipets and pipet tips (to prevent cross-contamination, we strongly recommend the use of pipet tips with aerosol barriers)
- Microcentrifuge tubes, 2 ml
- Vortexer
- Thermomixer or shaker–incubator
- For additional materials required for specific sample preparations, please refer to protocol sheets at www.qiagen.com/goto/qsdnainvestigator.

All casework HE (High Efficiency) protocols

TopElute Fluid (60 ml) (cat. no. 1055628)

Important Notes

Automated purification with the QIAsymphony SP

The QIAsymphony SP makes automated sample preparation easy and convenient. Samples, reagents and consumables, and eluates are separated in different drawers. Simply load samples, reagents provided in special cartridges, and preracked consumables in the appropriate drawer before a run. Start the protocol and remove purified DNA from the "Eluate" drawer after processing. Refer to the user manual provided with the instrument for operating instructions.

Loading reagent cartridges (RC) into the "Reagents and Consumables" drawer

Reagents for purification of DNA are contained in an innovative reagent cartridge (see Figure 2). Each trough of the reagent cartridge contains a particular reagent, such as magnetic particles, lysis buffer, wash buffer, or elution buffer. Partially used reagent cartridges can be reclosed with Reuse Seal Strips for later reuse, which avoids generation of waste due to leftover reagents at the end of the purification procedure.

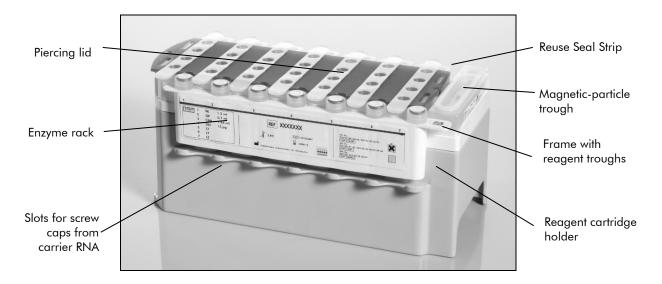


Figure 2. QIAsymphony reagent cartridge (RC). The reagent cartridge contains all reagents required for the protocol run.

Before starting the procedure, ensure that the magnetic particles are fully resuspended. Remove the magnetic-particle trough from the reagent cartridge frame, vortex it vigorously for at least 3 minutes, and replace it in the reagent cartridge frame before the first use. Place the reagent cartridge into the reagent cartridge holder. Place the enzyme rack with the diluted carrier RNA into the reagent cartridge holder. Before using a reagent cartridge for the first time, place the piercing lid on top of the reagent cartridge (Figure 3, next page).

Important: The piercing lid is sharp. Take care when placing it onto the reagent cartridge. Make sure to place the piercing lid onto the reagent cartridge in the correct orientation.

After the magnetic-particle trough cover is removed and the carrier RNA tubes are opened (screw caps can be stored in dedicated slots, see Figure 2), the reagent cartridge is subsequently loaded into the "Reagents and Consumables" drawer.

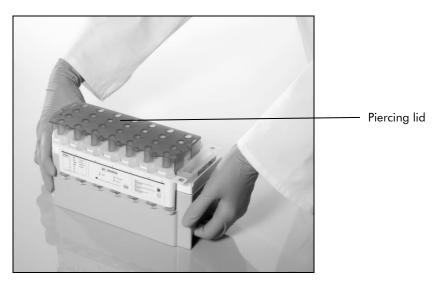


Figure 3. Easy worktable setup with reagent cartridges.

Partially used reagent cartridges can be stored until needed again, see "Storage", page 4.

TopElute Fluid (TOPE)

The casework HE (High Efficiency) protocols require TopElute Fluid (TOPE). An opened 60 ml bottle containing TopElute Fluid is placed into the "Reagents and Consumables" drawer.

Loading plasticware into the "Reagents and Consumables" drawer

Sample prep cartridges, 8-Rod Covers (both preracked in unit boxes), and disposable filter-tips (200 μ l tips provided in blue racks, 1500 μ l tips provided in gray racks) are loaded into the "Reagents and Consumables" drawer.

See protocol sheets at www.qiagen.com/goto/qsdnainvestigator for the consumables required for QIAsymphony DNA Investigator protocols.

For plasticware ordering information, see page 27.

Note: Both types of tips have filters to help prevent cross-contamination.

Tip rack slots on the QIAsymphony worktable can be filled with either type of tip rack. The QIAsymphony SP will identify the type of tips loaded during the inventory scan.

Note: Do not refill tip racks, unit boxes for sample prep cartridges, or 8-Rod Covers manually before starting another protocol run. The QIAsymphony SP can use partially used tip racks.

Loading the "Waste" drawer

Sample prep cartridges and 8-Rod Covers used during a run are re-racked in empty unit boxes in the "Waste" drawer. Make sure that the "Waste" drawer contains sufficient empty unit boxes for plastic waste generated during the protocol run.

Note: Ensure that the covers of the unit boxes are removed before loading the unit boxes into the "Waste" drawer. If you are using 8-Rod Cover boxes for collecting used sample-prep cartridges and 8-Rod Covers, ensure that the box spacer has been removed.

A bag for used filter-tips must be attached to the front side of the "Waste" drawer.

Note: The presence of a tip disposal bag is not checked by the system. Make sure that the tip disposal bag is properly attached before starting a protocol run. For more information, see the user manual provided with the instrument.

A waste container collects all liquid waste generated during the purification procedure. The "Waste" drawer can only be closed if the waste container is in place.

Loading the "Eluate" drawer

Load the required elution rack into the "Eluate" drawer. Do not load a 96-well plate onto "Elution slot 4". If eluates should be cooled, use "Elution slot 1" with the corresponding cooling adapter. As long-term storage of eluates in the "Eluate" drawer may lead to evaporation of eluates, we strongly recommend using the cooling position.

Inventory scan

Before starting a run, the instrument checks that sufficient consumables for the queued batch(es) have been loaded into the corresponding drawers.

Determining the amount of starting material

The amount of starting material for use in QIAsymphony DNA Investigator procedures can vary greatly, depending on the amount of DNA in the sample. Specific guidance for starting amounts is given in the individual protocols (see protocol sheets at www.qiagen.com/goto/qsdnainvestigator). The QIAsymphony SP can process $200 \, \mu$ l, $500 \, \mu$ l, or $1000 \, \mu$ l pretreated sample lysates using the casework protocols for purification of DNA or $200 \, \mu$ l or $500 \, \mu$ l pretreated samples lysates using the reference protocols.

Table 1 and Table 2 provide additional information about protocol options. Note that for all casework protocols listed in Table 1 there are two protocol versions that can be selected, depending on the elution volume to be used (see Table 2).

Table 1. Starting materials, elution buffers, and DNA purification protocols used in QIAsymphony DNA Investigator procedures

Sample type*	Elution buffer	QIAsymphony SP protocol
Surface swabs	Buffer ATE	Casework 500 µl
	or water	Casework 500 μl HE
Buccal swabs	Buffer ATE	Reference 500 μ l
FTA and Guthrie cards	Buffer ATE	Reference 200 μ l
Body fluid stains	Buffer ATE	Casework 500 μ l
	or water	Casework 500 μ l HE
Chewing gum	Buffer ATE	Casework 200 μ l
	or water	Casework 200 μ l HE
Cigarette butts	Buffer ATE	Casework 200 μ l
	or water	Casework 200 μ l HE
Nail clippings and hair	Buffer ATE	Casework 200 μ l
	or water	Casework 200 μ l HE
Paper and similar materials	Buffer ATE	Casework 500 μ l
	or water	Casework 500 μ l HE
Blood and saliva, $< 10 \mu$ l	Buffer ATE	Casework 200 μ l
	or water	Casework 200 μ l HE
Blood and saliva, $10-50 \mu l$	Buffer ATE	Reference 200 μl
Bones and teeth	Buffer ATE or water	Casework 500 μl
		Casework 500 μ l HE
Sexual assault specimen	Buffer ATE or water	Casework 200 μ l
		Casework 200 μ l HE
Large-volume samples	Buffer ATE	Casework 1000 μ l
	or water	Casework 1000 μ l HE

^{*} Specific guidance for processing each sample type is provided in the individual protocols (see protocol sheets at www.qiagen.com/goto/qsdnainvestigator).

Table 2. Protocol options for elution and number of samples per kit

Protocol	Elution buffer	Elution volume, μ l	Number of samples*
Casework 200 μ l HE	Buffer ATE or water	30, 40, 50, 60, 70, 80	192
Casework 200 μ l	Buffer ATE or water	100, 150, 200	192
Casework 500 μ l HE	Buffer ATE or water	30, 40, 50, 60, 70, 80	144
Casework 500 μ l	Buffer ATE or water	100, 150, 200	144
Casework 1000 μ l HE	Buffer ATE or water	30, 40, 50, 60, 70, 80	96
Casework 1000 μl	Buffer ATE or water	100, 150, 200	96
Reference 200 μ l	Buffer ATE	100, 150, 200, 400	192
Reference 500 μ l	Buffer ATE	100, 150, 200, 400	144

^{*} Calculations are based on batch sizes of 24 samples. Note that running batches smaller than 24 samples may reduce the number of preparations.

Carrier RNA

The kit is supplied with lyophilized carrier RNA to be used in QIAsymphony DNA Investigator Casework protocols. Carrier RNA enhances binding of DNA to the magnetic particles, especially if there are very few target molecules in the sample. The concentration of carrier RNA allows the procedure to be used as a generic purification system that is compatible with many different amplification systems. Note that eluates contain both carrier RNA and DNA from the sample, with the amount of carrier RNA greatly exceeding the amount of DNA. For details about using carrier RNA, refer to the section "Things to do before starting" in the DNA purification protocols.

Lysis with proteinase K

The QIAsymphony DNA Investigator Kit contains proteinase K, which possesses a high specific activity that remains stable over a wide range of temperatures and pH values, with substantially increased activity at higher temperatures. Proteinase K is a recombinant protein expressed in *Pichia pastoris* and is particularly suitable for short digestion times.

Use of TopElute Fluid

The casework HE (High Efficiency) protocols are optimized for maximum recovery of low elution volumes. Casework protocols eluting in small volumes of water or buffer ATE (30–80 μ l) use TopElute Fluid during the elution process. Casework protocols using 100 μ l or more for elution do not use TopElute Fluid.

Storage and quality of purified DNA

DNA eluted in Buffer ATE or water is immediately ready for use in amplification reactions or can be stored at 2–8°C, –20°C, or at –80°C.

QIAsymphony DNA Investigator procedures yield DNA free of proteins, nucleases, and inhibitors.

Quantification of DNA

Depending on the sample type, the yields of DNA obtained in the purification procedure might be below 1 μ g and therefore difficult to quantify using a spectrophotometer. In addition, eluates prepared with carrier RNA might contain much more carrier RNA than target nucleic acids. We recommend using quantitative amplification methods to determine yields.

Carryover of magnetic particles may affect the absorbance reading at 260 nm (A_{260}) of the purified DNA. The measured absorbance at 320 nm (A_{320}) should be subtracted from all absorbance readings. To remove magnetic-particle carryover, see Appendix B, page 25.

Note: For accurate quantification of DNA eluted in Buffer ATE by absorbance at 260 nm, we recommend diluting the sample in elution buffer (Buffer ATE). Dilution of the sample in water may lead to inaccurate values. The elution buffer has a high absorbance at 220 nm, which can lead to high background absorbance levels if the spectrophotometer is not properly zeroed. We therefore strongly recommend using elution buffer as a blank. Extra elution buffer to blank the spectrophotometer is provided in a separate bottle with QIAsymphony DNA Investigator Kits.

Protocol: Purification of DNA from Casework and Reference Samples

This protocol is designed for isolation of total (genomic and mitochondrial) DNA from various forensic casework and reference samples that have been prepared as described in the pretreatment protocols (see protocol sheets at www.qiagen.com/goto/qsdnainvestigator). The protocol describes the procedure for setting up the QIAsymphony SP and starting a run. See Tables 1 and 2, pages 16 and 17, for a summary of protocol options.

Casework protocols are designed to ensure efficient extraction of DNA from a wide range of demanding samples. The fully automated procedure processes sample lysate volumes of $200 \,\mu$ l, $500 \,\mu$ l, or $1000 \,\mu$ l. DNA is eluted in 30– $200 \,\mu$ l of either Buffer ATE or water. Carrier RNA is added to the samples during the automated procedure to maximize yields from very small samples.

Reference protocols allow robust and time-optimized extraction of database samples. DNA is eluted in $100-400 \mu l$ Buffer ATE.

Important points before starting

- Ensure that you are familiar with operating the QIAsymphony SP. Refer to the user manual provided with the instrument for operating instructions.
- Before beginning the procedure, read "Important Notes" starting on page 12.
- Try to avoid vigorous shaking of the reagent cartridge otherwise foam may be generated, which can lead to liquid-level detection problems.
- Ensure you are familiar with the protocol sheet corresponding to the procedure you want to use (<u>www.qiagen.com/goto/qsdnainvestigator</u>).
- Before using a reagent cartridge for the first time, check that Buffer QSL3 does not contain a precipitate. If necessary, remove the trough containing Buffer QSL3 from the reagent cartridge and incubate for 30 minutes at 37°C with occasional shaking to dissolve precipitate. Make sure to replace the trough in the correct position. If the reagent cartridge is already pierced, make sure that the troughs are sealed with Reuse Seal Strips and incubate the complete reagent cartridge for 30 minutes at 37°C with occasional shaking in a water bath.

Things to do before starting

- Remove any solid material from the sample tube. Note that there is no need to replace lysate volumes absorbed into the material. Optional: QIAshredder spin columns can be used to harvest lysate remaining in absorbent materials. See page 27 for ordering information.
- Dissolve the lyophilized carrier RNA in 1.6 ml Buffer ATE (provided in the QIAsymphony DNA Investigator Kit) before using the kit for the first time. Transfer 400 μ l to each of the tubes in positions 1 and 2 of the enzyme rack on the reagent cartridge. Add additional 1.2 ml Buffer ATE to each tube and mix by pipetting up and down several times.

Note: It is important that the final volume of carrier RNA in the tubes of the enzyme rack is exactly 1.6 ml. Dissolved carrier RNA is stable for 4 weeks when stored at $2-8^{\circ}$ C. For longer periods, store carrier RNA at -20° C.

Note: For the inventory scan to be completed successfully, tubes containing carrier RNA must be opened and placed in the enzyme rack which is placed in the reagent cartridge. The carrier RNA, however, will not be used for reference protocols.

- For information about sample tubes compatible with a certain protocol, see the corresponding labware list (available at www.qiagen.com/goto/qsdnainvestigator).
- For information about minimum sample volumes for samples in primary and secondary tubes for a certain protocol, see the corresponding labware list (available at www.qiagen.com/goto/qsdnainvestigator). This information also indicates which tubes can be used for different protocols.
- Casework HE (High Efficiency) protocols require TopElute Fluid (TOPE). Place an opened 60 ml bottle containing TopElute Fluid (TOPE) into the "Reagents and Consumables" drawer.
- Before starting the procedure, ensure that the magnetic particles are fully resuspended. Vortex the trough containing the magnetic particles vigorously for at least 3 minutes before first use.
- Before loading the reagent cartridge remove the cover from the trough containing the magnetic particles and open the carrier RNA tubes. Make sure that the piercing lid is placed on the reagent cartridge or, if using a partially used reagent cartridge, make sure the Reuse Seal Strips have been removed.
- If samples are bar coded, orient samples in the tube carrier so that the bar codes face the bar code reader at the left side of the QIAsymphony SP.

Procedure

1. Ensure that the QIAsymphony SP is switched on.

The power switch is located at the bottom, left corner of the QIAsymphony SP.

- 2. Ensure the "Waste" drawer is prepared properly, and perform an inventory scan of the "Waste" drawer, including the tip chute and liquid waste. Replace the tip disposal bag if necessary.
- 3. Load the required reagent cartridge(s) and consumables into the "Reagents and Consumables" drawer.
- 4. For casework HE protocols only: Press the "R+C" button in the touchscreen to open the screen that shows the consumables status ("Consumables/8-Rod Covers/Tubes/Filter-Tips/Reagent Cartridges"). Press the "Scan Bottle" button to scan the bar code of the bottle of TopElute Fluid (TOPE) with the handheld bar code scanner. Press "OK".

Ensure that the bottle of TopElute Fluid (TOPE) is scanned, opened, and placed into the "Reagents and Consumables" drawer before starting the inventory scan. Otherwise the inventory scan must be repeated after scanning, opening, and placing the bottle of TopElute Fluid (TOPE) into the "Reagents and Consumables" drawer.

- 5. Perform an inventory scan of the "Reagents and Consumables" drawer.
- 6. Load the required elution rack into the "Eluate" drawer.

Do not load a 96-well plate onto "Elution slot 4".

If eluates should be cooled, use "Elution slot 1" with the corresponding cooling adapter.

- 7. Place the samples into the appropriate sample carrier, and load them into the "Sample" drawer.
- 8. Using the touchscreen, enter the required information for each batch of samples to be processed.

Enter the following information:

- Sample information (depending on sample racks used)
- Protocol ("Assay Control Set") to be run
- Elution volume and output position

After information about the batch has been entered, the status changes from "LOADED" to "QUEUED". As soon as one batch is queued the "Run" button appears.

9. Press the "Run" button to start processing.

All processing steps are fully automated. At the end of the protocol run, the status of the batch changes from "RUNNING" to "COMPLETED".

10. Retrieve the elution rack containing the purified DNA from the "Eluate" drawer.

The DNA is ready to use, or can be stored at 2–8°C, –20°C, or –80°C.

In general, magnetic particles are not carried over into eluates. If carryover does occur, magnetic particles in eluates will not affect most downstream applications. If magnetic particles need to be removed before performing downstream applications, tubes or plates containing eluates should first be placed in a suitable magnet and the eluates transferred to a clean tube (see Appendix B, page 25).

If the eluate drawer is closed when a batch is running (e.g., if elution racks which contain the eluates are removed), the run will be paused and an inventory scan of the "Eluate" drawer will be performed. A message appears during the scan and must be closed (by pressing the "Close" button) before the run can be restarted.

Result files are generated for each elution plate.

11. If the reagent cartridge(s) is only partially used, seal it with the provided Reuse Seal Strips and close the carrier RNA tubes with screw caps immediately after the end of the protocol run to avoid evaporation.

Note: For more information about storage of partially used reagent cartridges, see "Storage" on page 4.

12. Discard used sample tubes, plates, and waste according to your local safety regulations.

See page 6 for safety information.

13. Clean the QIAsymphony SP.

Follow the maintenance instructions in the user manual provided with the instrument.

14. Close the workstation drawers, and switch off the QIAsymphony SP.

Troubleshooting Guide

This troubleshooting guide may be helpful in solving any problems that may arise. For more information, see also the Frequently Asked Questions page at our Technical Support Center: www.qiagen.com/FAQ/FAQList.aspx. The scientists in QIAGEN Technical Services are always happy to answer any questions you may have about either the information and protocols in this handbook or sample and assay technologies (for contact information, see back cover or visit www.qiagen.com).

Comments and suggestions

General handling

Error message displayed in the touch screen

If an error message is displayed in the touchscreen during a protocol run, refer to "Troubleshooting" in the user manual provided with the instrument.

Precipitate in reagent trough of opened cartridge

a) Buffer evaporation Excessive evaporation can lead to increased salt

concentration in buffers. Discard reagent

cartridge.

Make sure to seal buffer troughs of partially used reagent cartridge when not being used for DNA

purification.

water bath.

b) Storage of reagent cartridge

Storage of reagent cartridge under 15°C may lead to the formation of precipitates. If necessary, remove the trough containing Buffer QSL3 from the reagent cartridge and incubate for 30 min at 37°C with occasional shaking to dissolve the precipitate. If the reagent cartridge is already pierced, make sure that the reagent cartridge is reclosed with the Reuse Seal Set and incubate the complete reagent cartridge for 30 min at 37°C with occasional shaking in a

Comments and suggestions

Low DNA yield

 a) Magnetic particles were not completely resuspended Before starting the procedure, ensure that the magnetic particles are fully resuspended. Vortex for at least 3 min before use.

b) Incomplete lysis of samples

Proteinase K was stored at high temperatures for a prolonged time. Repeat the procedure using new samples and fresh proteinase K.

c) Clogging of pipet tip due to insoluble material Solid sample material was not removed from the digested sample prior to starting the QIAsymphony DNA purification procedure.

DNA does not perform well in downstream applications

 a) Insufficient DNA used in downstream application See "Low DNA yield" (page 24) for possible reasons. If possible, increase the amount of eluate used in the reaction.

b) Reduced sensitivity

Determine the maximum volume of eluate suitable for your amplification reaction. Reduce or increase the volume of eluate added to the reaction accordingly.

Appendix A: Working with DNA

General handling

Proper microbiological aseptic technique should always be used when working with small sample sizes. Hands and dust particles may carry bacteria and molds, and are the most common sources of contamination. Always wear latex or vinyl gloves while handling reagents and samples to prevent contamination from the surface of the skin or from dusty laboratory equipment. Change gloves frequently and keep tubes closed.

Disposable plasticware

The use of sterile, disposable polypropylene tubes is recommended throughout the purification procedure. These tubes are generally DNase-free.

Appendix B: Removing Magnetic-Particle Carryover

In general, magnetic particles are not carried over into eluates. If carryover does occur, magnetic particles in eluates will not affect most downstream applications.

Carryover of magnetic particles in the eluate may affect the absorbance at 260 nm (A_{260}) in a spectrophotometer. To remove particles, the tube containing the eluate should first be applied to a suitable magnetic separator and the eluate transferred to a clean tube:

- Apply the tube containing the nucleic acids to a suitable magnetic separator (e.g., QIAGEN 12-Tube Magnet, cat. no. 36912) until the magnetic particles are separated. If eluate is in microplates, apply the microplate to a suitable magnetic separator (e.g., QIAGEN 96-Well Magnet Type A, cat. no. 36915) until the magnetic particles are separated.
- If a suitable magnetic separator is not available, centrifuge the tube containing the DNA for 1 minute at full speed in a microcentrifuge to pellet any remaining magnetic particles.
- Once separation is complete, carefully withdraw an aliquot for quantification and dilute as necessary.
- Measure the absorbance at 320, 280, and 260 nm. Subtract the absorbance reading obtained at 320 nm from the readings obtained at 260 and 280 nm to correct for the presence of magnetic particles.

References

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Ordering Information

Product	Contents	Cat. no.
QIAsymphony DNA Investigator Kit (192)	For $192 \times 200 \mu$ l preps, $144 \times 500 \mu$ l preps, or $96 \times 1000 \mu$ l preps.: Includes 2 reagent cartridges enzyme racks and accessories	931436
Related products		
Accessory Trough (10)	Accessory troughs for use with the QIAsymphony SP	997012
Reagent Cartridge Holder (2)	Reagent cartridge holder for use with the QIAsymphony SP	997008
Sample Carrier, plate, Qsym	Plate carrier for sample input. For use with the QIAsymphony SP	9017660
Tube Insert, 11 mm, sample carrier, Qsym	Primary tube adapter (11 mm) for use with the QIAsymphony tube carrier	9241033
Tube Insert, 13 mm, sample carrier, Qsym	Primary tube adapter (13 mm) for use with the QIAsymphony tube carrier	9241034
Tube Insert, 2 ml, sample carrier, Qsym	Secondary tube adapter (for 2 ml screw-cap tubes) for use with the QIAsymphony tube carrier	9241032
Cooling Adapter, tubes, 2 ml, Qsym	Cooling adapter for 2 ml screw-cap tubes. For use in the QlAsymphony "Eluate" drawer	9018088
Cooling Adapter, EMT, Qsym	Cooling adapter for EMT racks. For use in the QIAsymphony "Eluate" drawer	9018086
Cooling Adapter, MTP, RB, Qsym	Cooling adapter for round bottom microtiter plates (MTP). For use in the QIAsymphony "Eluate" drawer	9018085
Cooling Adapter, PCR, Qsym	Cooling adapter for PCR plates. For use in the QIAsymphony "Eluate" drawer	9018087
Adapter, tubes, 2 ml, Qsym	Adapter for 2 ml screw-cap tubes. For use in the QIAsymphony "Eluate" drawer	9018577

Product	Contents	Cat. no.
Sample Prep Cartridges, 8-well (336)	8-well sample prep cartridges for use with the QIAsymphony SP	997002
8-Rod Covers (144)	8-Rod Covers for use with the QIAsymphony SP	997004
Filter-Tips, 200 μ l (1024)	Disposable Filter-Tips, racked; (8 x 128). For use with the QIAcube and the QIAsymphony SP	990332
Filter-Tips, 1500 μ l (1024)	Disposable Filter-Tips, racked; (8 x 128). For use with the QIAsymphony SP	997024
Tip Disposable Bags (15)	Tip disposal bags for use with the QIAsymphony SP	9013395
TopElute Fluid (60 ml)	60 ml TopElute Fluid for elution process in casework HE protocols.	1055628
Buffer ATL (200 ml)	200 ml Tissue Lysis Buffer for 1000 preps	19076
QIAGEN Proteinase K (2 ml)	For protease digestion during DNA and RNA preparation. Contents: 2 ml (>600 mAU/ml, solution)	19131
QIAshredder (50)	50 disposable cell-lysate homogenizers for use in nucleic acid minipreps, caps	79654
TissueLyser II	Universal laboratory mixer-mill disruptor, 100–120/220–240 V 50/60 Hz	85300
TissueLyser Adapter Set 2 x 24	2 sets of Adapter Plates and 2 racks for use with 2 ml microcentrifuge tubes on the TissueLyser II	69982
Stainless Steel Beads, 5 mm (200)	Stainless Steel Beads, suitable for use with the TissueLyser system	69989
QlAcard FTA One Spot (100)	For 100 samples: 100 QIAcard FTA One Spots	159201
QIAcard FTA Two Spots (100)	For 100 x 2 samples: 100 QIAcard FTA Two Spots	159203

Product	Contents	Cat. no.
QIAcard FTA Four Spots (100)	For 100 x 4 samples: 100 QIAcard FTA Four Spots	159205
QIAcard FTA Indicator Four Spots (25)	For 25 x 4 samples: 25 QIAcard FTA Indicator Four Spots	159214
12-Tube Magnet	Magnet for separating magnetic particles in 12 x 1.5 ml or 2 ml tubes	36912
96-Well Magnet Type A	Magnet for separating magnetic particles in wells of 96-well plates, 2 x 96-Well Microplates FB	36915
S-Blocks (24)	96-well blocks with 2.2 ml wells, 24 per case	19585

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