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February 2015

# QIAamp<sup>®</sup> 96 Virus QIAcube<sup>®</sup> HT Handbook

For automated purification of viral RNA and  
DNA from human samples using QIAcube HT  
or QIAxtractor<sup>®</sup>

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# Kit Contents

<b>QIAamp 96 Virus QIAcube HT Kit</b>	<b>(5)</b>
<b>Catalog no.</b>	<b>57731</b>
<b>Number of preps</b>	<b>480</b>
QIAamp 96 plates	5
Buffer ACL*	1 x 220 ml
Buffer ACB*† (concentrate)	3 x 60 ml
QIAGEN Proteinase K	3 x 6 ml
Carrier RNA (poly A)	2 x 310 µg
Buffer ATL	2 x 50 ml
Buffer AW1 *‡ (concentrate)	1 x 190 ml
Buffer AW2‡ (concentrate)	1 x 127 ml
Buffer AVE§	1 x 125 ml
TopElute Fluid	1 x 60 ml
Quick-Start Protocol	2

\* **CAUTION:** Contains a chaotropic salt. Take appropriate laboratory safety measures and wear gloves when handling. Not compatible with disinfectants containing bleach. See page 6 for safety information.

† Before using for the first time, add isopropanol as indicated on the bottle to obtain a working solution.

‡ Before using for the first time, add ethanol (96–100%) as indicated on the bottle to obtain a working solution.

§ **CAUTION:** Contains sodium azide as a preservative.

<b>QIAcube HT Plasticware</b>	<b>(480)</b>
<b>Catalog no.</b>	<b>950067</b>
<b>Number of preps</b>	<b>480</b>
S-Blocks	5
Filter-Tips OnCor C	9 x 96
Tape Pad	1
Elution Microtubes RS (EMTR)	5
8-Well Strip Caps for EMTR	120

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## Storage

QIAamp 96 plates, buffers and lyophilized carrier RNA are stable until the expiration date on the kit box at room temperature (15–25°C) and dry conditions without affecting performance.

Freeze Carrier RNA dissolved in Buffer AVE immediately in aliquots at –30 to –15°C. Do not subject aliquots of carrier RNA to more than 3 freeze–thaw cycles. Carrier RNA dissolved in Buffer AVE is stable at room temperature (15–25°C) for up to 48 hours (see “Preparing reagents and instrument” on page 17 for further information).

QIAGEN Proteinase K can be stored at room temperature (15–25°C). To store for extended periods of time, or if the ambient temperature often exceeds 25°C, we recommend storing at 2–8°C.

## Intended Use

The QIAamp 96 Virus QIAcube HT Kit is intended for the automated extraction of viral RNA and DNA from human whole blood, serum, plasma, body fluids, swabs, washes, tissue and stool using the QIAcube HT instrument. The QIAamp 96 Virus QIAcube HT 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.

# 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](http://www.qiagen.com/safety) where you can find, view and print the SDS for each QIAGEN kit and kit component.



**CAUTION: DO NOT add bleach or acidic solutions directly to the sample preparation waste.**

Buffer ACL and Buffer ACB contain guanidine thiocyanate and Buffer AW1 contains guanidine hydrochloride, which can form highly reactive compounds if combined with bleach.

If liquid containing these buffers is spilled, clean with suitable laboratory detergent and water. If the spilled liquid contains potentially infectious agents, clean the affected area first with laboratory detergent and water, and then with 1% (v/v) sodium hypochlorite.

## Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of QIAamp 96 Virus QIAcube HT Kit is tested against predetermined specifications to ensure consistent product quality.

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# Introduction

The QIAamp 96 Virus QIAcube HT Kit uses well-established technology to enable efficient purification of viral RNA and DNA from a broad range of sample types (see Table 1 on page 8).

The kit combines the selective binding properties of a silica-based membrane with a high-throughput 96-well format, and is designed for fully-automated, simultaneous processing of 96 samples on the QIAcube HT. Purified nucleic acids are eluted in Buffer AVE, and are free of proteins, nucleases and other impurities, ready for use in downstream applications. The kit is not intended for host RNA or host DNA preparation.

## Principle and procedure

Samples are lysed under highly denaturing conditions at room temperature in the presence of QIAGEN proteinase K and Buffer ACL, which together ensure the inactivation of nucleases. Adding Buffer ACB adjusts the binding conditions for the co-purification of DNA and RNA. The lysate is then transferred to a QIAamp 96 plate. During vacuum, nucleic acids are adsorbed onto the silica membranes while contaminants pass through. Three efficient wash steps remove the remaining contaminants and enzyme inhibitors, and nucleic acids are eluted in Buffer AVE.

Performance is not guaranteed for every combination of starting material and virus species and must be validated by the user. Some samples may require a pretreatment (see Table 1 on page 8).

## Description of protocols

Samples will either directly undergo nucleic acid purification, or undergo pretreatment followed by nucleic acid purification.

Many sample types can be directly processed without pretreatment. However, depending on the starting material, one of the pretreatments may be needed. Table 1 provides an overview of pretreatment protocols suited to different starting materials.

- Protocol: Purification of Viral Nucleic Acids from Diverse Samples (page 23)
- Sample Pretreatment Protocols (pages 30–35)

**Table 1. Pretreatment protocols for various sample types**

Sample	Name	Page
Fluids (e.g., plasma and serum*, cell-free body fluids†, whole blood†‡)	No pretreatment necessary; proceed directly to “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”	23
Respiratory samples and sputum§	Pretreatment B1 for Respiratory Samples	30
Liquid transport media	Pretreatment B2 for Liquid Transport Media	31
Urine	Pretreatment B3 for Urine	31
Dried blood spots	Pretreatment B4 for Dried Blood Spots	32
Swabs and buccal cells	Pretreatment B5 for Swabs and Buccal Cells	32
Stool suspensions	Pretreatment F1 for Stool Suspensions	33
Tissues (e.g., liver, spleen, kidney, lymph node)	Pretreatment T1 Mechanical Disruption of Tissue Pretreatment T2 for Enzymatic Digestion of Tissue¶	33 35

\*Samples can be fresh or frozen, provided they have not been frozen and thawed more than once.

† Includes amniotic fluid, cerebrospinal fluid, ocular fluid, synovial fluid and pleural fluid.

‡ Includes samples containing EDTA or citrate.

§ Includes nasopharyngeal aspirates (NPA), bronchoalveolar lavage (BAL) fluid, sputum and nasal and throat swabs.

¶ Not suitable for viral RNA as the lysis conditions do not sufficiently conserve RNA integrity.

For further pretreatment recommendations, contact QIAGEN Technical Services.

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## Nucleic acid purification protocol

The protocol “Purification of Viral Nucleic Acids from Diverse Samples” (page 23) is optimized for purification of viral RNA and DNA from up to 200 µl of fluid material. Suitable starting materials for direct processing using this method include:

- Cell-free body fluids such as plasma, serum and CSF
- Whole blood

## Pretreatments

The various pretreatments included in this handbook are optimized for specific combinations of starting material. The choice of pretreatment depends on the workflow focus, and is to be followed by nucleic acid purification.

Table 1 on page 8 summarizes the pretreatments and their applications.

Some of the pretreatments may require additional components (see pages 10–11).

# 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.

For all protocols:

- Pipets and disposable pipet tips with aerosol barriers (20–1000 µl)
- Isopropanol
- Ethanol (96–100%)\*
- Phosphate-buffered saline (PBS) may be required for sample dilution
- QIAcube HT Instrument†
- QIAcube HT Software version 4.17.1 or higher
- QIAcube HT Reagent troughs
- Vortexer

Pretreatment B1 – for respiratory samples

- Sputasol (Oxoid Limited) and 37°C water bath, or
- NAC buffer (10 g N-acetylcysteine per liter of 0.9% NaCl solution), or
- PBS or Buffer AE (cat. no. 19077), DTT and 37°C water bath

\* Do not use denatured alcohol, which contains other substances such as methanol or methylethylketone.

† To process dedicated QIAcube HT kits on QIAextractor instruments, QIAcube HT Software version 4.17.1 or higher is required, along with the Accessories Pack, QXT. See “Ordering Information” on page 43).

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Pretreatment B2, B3, B4 and B5 – for liquid transport media, urine and dried blood spots

- Buffer ATL (cat. no. 19076)
- QIAGEN Proteinase K (cat. nos. 19131 or 19133)

Pretreatment F1 – stool samples

- 0.9% NaCl solution

Pretreatment T1 – mechanical disruption of tissue

- TissueLyser II (QIAGEN, cat. no. 85300) with a TissueLyser II Adapter Set 2 x 24 (QIAGEN, cat. no. 69982), or TissueLyser LT (QIAGEN, cat. no. 85600) with the TissueLyser LT Adapter for 12 tubes (QIAGEN, cat. no. 69980), or other bead-mill homogenizer\*
- 5 mm stainless steel beads (cat. no. 69989)
- Buffer ATL (cat. no. 19076)
- QIAGEN Proteinase K (cat. nos. 19131 or 19133)

Pretreatment T2 – enzymatic digestion of tissue

- Thermoshaker suitable for 2 ml collection tubes
- Buffer ATL (cat. no. 19076)
- QIAGEN Proteinase K (cat. nos. 19131 or 19133)

\* This is not a complete list of suppliers and does not include many important vendors of biological supplies.

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# Important Notes

## Starting material

Do not overload the QIAamp membrane, as this can lead to impaired nucleic acid extraction and/or performance in downstream assays. For samples with very high host nucleic acid contents (e.g., for certain tissues, such as spleen or blood samples with highly increased cell counts), use less than the maximum amount of sample recommended in the protocol or pretreatments. In some downstream applications such as PCR and RT-PCR, very high background concentrations of nucleic acids may impair the reaction. Use appropriate controls (e.g., an internal control) to verify successful PCR amplification.

Avoid transferring solid material to the S-Block that could reduce flow through the membrane (e.g., blood clots, solid tissue, swab fibers, etc.). When working with difficult samples, use a user confirmation step to check if all liquid has passed the membrane. See “Troubleshooting Guide” on page 37 and the *QIAcube HT User Manual* for guidance.

Highly viscous fluids may require a treatment to reduce their viscosity to allow for efficient extraction of viral nucleic acids. Please contact QIAGEN Technical Services for recommendations.

Avoid repeated thawing and freezing of samples since this may reduce nucleic acid yield and quality.

## **Serum, plasma, other body fluids and swab media**

Up to 200 µl serum, plasma, other body fluid or swab media supernatant can be processed.

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Carrier RNA should be used in the nucleic acid purification protocol to prevent the loss of nucleic acids during the procedure (see page 16 for information about the use of carrier RNA).

The processing of samples with very high inhibitor contents, such as urine or stool, may require a reduction in sample input volume and/or an extra pretreatment to remove inhibitors. To reduce the input volume, use 25–50  $\mu\text{l}$  of the sample and adjust the volume to 200  $\mu\text{l}$  with PBS or 0.9% NaCl.

### **Plasma and serum samples**

After collection, centrifugation and phase separation, plasma or serum can be stored at 2–8°C for up to 6 hours. For long-term storage, freezing at –30 to –15°C or –70°C in aliquots is recommended. Frozen plasma or serum samples must not be thawed more than once. Repeated freeze–thawing leads to reduced viral titers and therefore reduced yields of viral nucleic acids.

Freezing of samples can lead to denaturation and precipitation of proteins that may aggregate on the QIAamp membrane. If cryo-precipitates are visible, they can be pelleted by centrifugation at 6800  $\times g$  for 3 minutes. The cleared supernatant should be transferred to a new sample tube without disturbing the pellet and processed immediately. This step will not reduce viral titers.

### **Swabs**

Swabs may be processed on the same day as collection or stored for future processing. While storage at –30 to –15°C is recommended, DNA of suitable quality for single-copy gene amplification has been documented from swabs stored at room temperature for 24 months.

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QIAGEN provides sterile foam-tipped swabs for collection of saliva and buccal cells. The nonabrasive foam head is the same size as the sample area on QIAcard FTA Indicator Four Spots to facilitate sample application. See “Ordering Information” on page 43.

Alternatively, samples can be collected using plastic swabs with cotton or Dacron® tips. Puritan® applicators with plastic shafts and cotton or Dacron tips are available from Hardwood Products Company ([www.hwppuritan.com](http://www.hwppuritan.com), cat. nos. 25-806 1PC and 25-806 1PD) and from Daigger ([www.daigger.com](http://www.daigger.com), cat. nos. EF22008D and EF22008DA). \* Nylon cytology brushes and other swab types may also be used.

**Note:** Solid pieces remaining in the sample fluid may aggregate on the QIAamp membrane, which may decrease nucleic acid yield.

## Whole blood

Blood samples treated with EDTA or citrate as anticoagulant can be used for nucleic acid purification. Samples can be either fresh or frozen, provided that they have not been frozen and thawed more than once. After collection, whole blood samples can be stored at 2–8°C for up to 6 hours. For longer storage, we recommend freezing aliquots at –30 to –15°C or –70°C.

We recommend using 50–200 µl blood per sample. Typically, 200 µl of blood can be used with most blood samples. However, highly elevated cell counts due to inflammatory or neoplastic diseases may strongly increase the host nucleic acid content of a sample. In this case, reduction of sample input to 50 µl may improve results in downstream assays, particularly in RT-PCR. If using less than 200 µl blood, adjust the sample volume to 200 µl with PBS or 0.9% NaCl.

\* This is not a complete list of suppliers and does not include many important vendors of biological supplies.

## Dried blood spots

Drying blood on filter paper (e.g., Whatman FTA Cards) is an effective form of storage, and samples prepared in this manner are less expensive and safer to transport. A disc (3 mm diameter) punched out from filter paper stained with dried blood contains white blood cells from approximately 5  $\mu$ l whole blood.

## Tissues

When working with tissue samples, mechanical or enzymatic disruption of the tissue structure is the prerequisite for liberation of cells, subsequent release of nucleic acids, and membrane permeability of the material.

Different tissue types can vary widely with regard to texture and rigidity, cell types and content of host nucleic acids and inhibitory substances. In addition, the localization of viral nucleic acids in the tissue may vary depending on tissue type, virus and stage of infection. Additional pretreatments for tissue samples are available at QIAGEN technical service, including a rapid protocol and recommendations for difficult tissues.

Up to 5 mg of fresh or frozen tissue can be used as a starting amount. For higher amounts of tissue it might be necessary to check whether this amount of tissue can still be processed. Furthermore we recommend enabling the user confirmation during the vacuum step to ensure that the liquids have completely passed through the membrane before the protocol proceeds.

## Yields of nucleic acids

For samples containing a low amount of cells (e.g., serum and plasma), the yield of nucleic acids obtained can be below 1  $\mu$ g and is therefore difficult to quantify using a spectrophotometer. In addition, eluates prepared with carrier RNA may contain much more carrier RNA than target nucleic acids. The QIAamp 96 Virus 96 QIAcube HT Kit recovers total nucleic acids. Therefore, cellular DNA and RNA and circulating cell-free nucleic acids

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will be co-purified along with viral RNA and DNA (and bacterial nucleic acid if present) and cannot be distinguished using spectrophotometric measurements. We recommend using quantitative amplification methods such as quantitative real-time PCR or real-time RT-PCR to determine viral nucleic acid yields.

## Using carrier RNA and internal controls

### Carrier RNA

We recommend adding carrier RNA to fluids containing low amounts of cells such as serum, plasma, swab media and wash fluid. This enhances adsorption of viral RNA and DNA to the silica membranes, which is especially important when the target molecules are not abundant. In addition, an excess of carrier RNA reduces the chances of viral RNA degradation in the rare event that RNases are not denatured by the chaotropic salts and detergents in the lysis buffer. Not using carrier RNA may decrease the recovery of viral nucleic acids.

### Internal control

Using the QIAamp 96 Virus QIAcube HT Kit in combination with commercially available amplification systems may require the introduction of an internal control into the purification procedure. Internal control RNA or DNA should be added together with the carrier RNA to the lysis buffer (see Table 2 on page 28). Refer to the assay manufacturer's instructions in order to determine the optimal concentration.

For the standard protocol, it is recommended to add 5  $\mu$ l internal control solution. Using a concentration other than that recommended may reduce amplification efficiency.

## Storing nucleic acids

For short-term storage of up to 24 hours, we recommend storing the purified viral RNA and DNA at 2–8°C. For storage longer than 24 hours, we recommend storing purified nucleic acids at –30 to –15°C, or even –70°C in the case of RNA.

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## Handling RNA

RNases are very stable and active enzymes that generally do not require cofactors to function. Since RNases are difficult to inactivate and only minute amounts are sufficient to destroy RNA, do not use any plasticware or glassware without first eliminating possible RNase contamination. Care should be taken to avoid inadvertently introducing RNases into the RNA sample during or after the purification procedure.

## Preparing RNA

When preparing viral RNA, work quickly during the manual steps of the procedure. If you have not previously worked with RNA, read Appendix A on page 40 before starting. Buffer AVE is RNase-free upon delivery. It contains sodium azide, an antimicrobial agent that prevents growth of RNase-producing organisms. However, as this buffer does not contain any RNase inhibitors, it will not actively inhibit RNases introduced by inappropriate handling. Extreme care should be taken to avoid contamination with RNases when handling Buffer AVE.

## Preparing reagents and instrument

### Carrier RNA stock solution

For use, lyophilized carrier RNA should first be dissolved in Buffer AVE. Add 1550  $\mu$ l Buffer AVE to the tube containing 310  $\mu$ g lyophilized carrier RNA to obtain a stock solution of 0.2  $\mu$ g/ $\mu$ l. Add this solution to Buffer ACL as described in Table 2 on page 28. Unused carrier RNA dissolved in Buffer AVE should be frozen in aliquots at  $-30$  to  $-15^{\circ}\text{C}$ . Aliquots of carrier RNA should not be subjected to more than 3 freeze-thaw cycles.

## QIAGEN Proteinase K

The QIAamp 96 Virus QIAcube HT Kit contains ready-to-use QIAGEN Proteinase K supplied in a specially formulated storage buffer. The activity of the QIAGEN Proteinase K solution is 600 mAU/ml.

QIAGEN Proteinase K is stable for at least 1 year after delivery when stored at room temperature (15–25°C). To store for more than 1 year or if ambient temperature often exceeds 25°C, we recommend storing QIAGEN Proteinase K at 2–8°C.

## Buffer ACB

Buffer ACB is supplied as a concentrate. Before using for the first time, add isopropanol (100%) as indicated on the bottle to obtain a working solution. Tick the check box on the bottle label to indicate that isopropanol has been added. Mix well after adding isopropanol.

## Buffer AW1

Buffer AW1 is supplied as a concentrate. Before using for the first time, add ethanol (96–100%) as indicated on the bottle to obtain a working solution. Tick the check box on the bottle label to indicate that ethanol has been added. Mix well after adding ethanol.

## Buffer AW2

Buffer AW2 is supplied as a concentrate. Before using for the first time add ethanol (96–100%) as indicated on the bottle to obtain a working solution. Tick the check box on the bottle label to indicate that ethanol has been added. Mix well after adding ethanol.

## Handling Buffer AVE

Buffer AVE is RNase-free upon delivery. It contains sodium azide, an antimicrobial agent that prevents growth of RNase-producing organisms. However, as this buffer does not contain any RNase-degrading chemicals, it will not actively inhibit RNases introduced by inappropriate handling. For RNA applications, when handling Buffer AVE, avoid

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contamination with RNases. Follow general precautions for working with RNA, such as frequent change of gloves and keeping tubes closed whenever possible.

## TopElute Fluid

TopElute Fluid is used during elution of nucleic acids from the QIAamp membrane. It enables application of a stable and high vacuum and results in equal eluate volumes. In addition, TopElute Fluid eliminates the formation of drops of elution buffer at the outlet nozzles of the QIAamp 96 plates.

TopElute Fluid might be found as a top layer over the elution buffer. It is inert and has no effects on downstream applications.

**Important:** Please ensure that you only take the eluate from below the top layer.

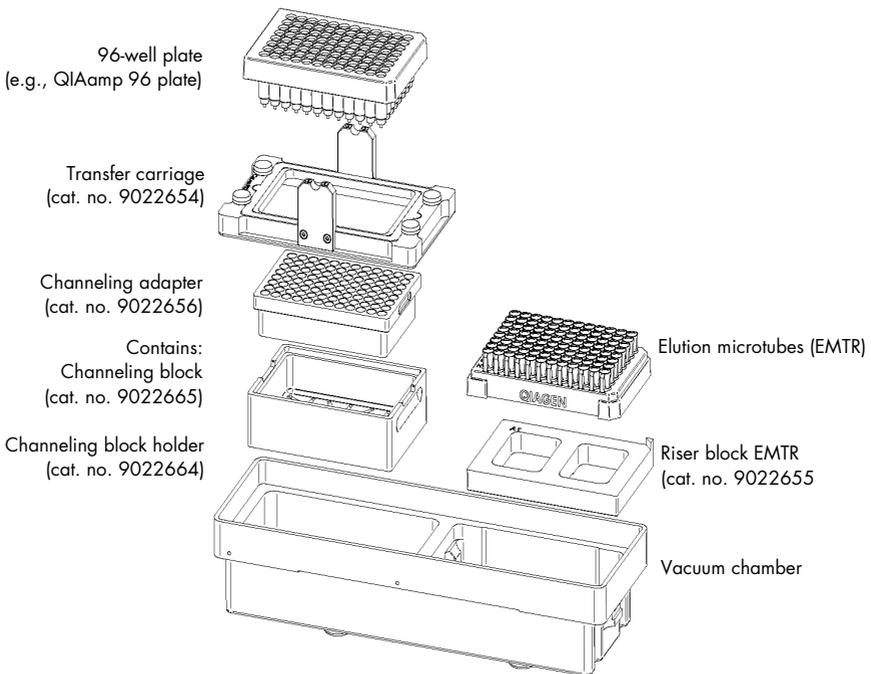
## Assembling the vacuum chamber

All QIAcube HT instruments are delivered with the vacuum chamber components for dedicated QIAcube HT Kits.

**Important:** If you use a QIAextractor instrument, ensure that only parts from the Accessories Pack, QXT (black parts) are used. See "Ordering Information" on page 43.

Figure 1 illustrates the assembly of the vacuum chamber when using QIAamp 96 plates. For further information, please refer to the *QIAcube HT User Manual*.

1. Insert the channeling block holder into the left (waste) chamber of the vacuum chamber.
2. Press firmly on the sides of the channeling block holder to seat it in the chamber, sealing the O-ring on the spigot into the drain.
3. Then, place the channeling block into the channeling block holder.
4. Place the QIAamp 96 plate in the transfer carriage. Load the carriage with the QIAamp 96 plate into the left (waste) chamber of the vacuum chamber.
5. Ensure that the carriage is positioned to the left inside the vacuum chamber. Place the riser block EMTR in the right (elution) chamber of the vacuum chamber with the pin of the riser block EMTR in the top right position.
6. Load an elution microtubes rack (EMTR) into the elution chamber.



**Figure 1. Assembling the vacuum chamber.**

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## Optional features

### Processing of fewer than 96 samples per run

If processing fewer than 96 samples reuse of QIAamp 96 plates, S-Block and EMTR is possible up to three times.

**Note:** We recommend using fresh plasticware for every run. If reusing, take extreme care to prevent cross-contamination.

- Store plates in a way that separates the outlet nozzles under the plate, for example, in the S-Block used in the same run or in a fresh 96-well microtiter plate.
- Cover unused wells of the S-Block and QIAamp 96 plate with a tape sheet at all times.
- Remove unused Elution Microtubes from the EMTR in rows of eight tubes.

### Off-board lysis

For some applications, it may be necessary to lyse samples in a safety cabinet. For some sample types, lysis outside the instrument might be necessary. Please refer to Sample Pretreatment Protocols on pages 30–35. If lysis with Proteinase K is carried out off-board, Proteinase K may be exchanged with Buffer ACL when setting up the worktable.

### Sample data input, data tracking and LIMS connection

Clicking a workspace item displays information about the item in the right-hand pane. For example, click **A1: Reaction** and the **Reaction Data** panel appears in the dialog box in the right-hand pane. Sample descriptions can be imported, inserted manually or inserted using a handheld barcode scanner. See the *QIAcube HT User Manual* for more details. The field **Plate ID** can be used for the unique number that is provided on each EMTR RS plate.

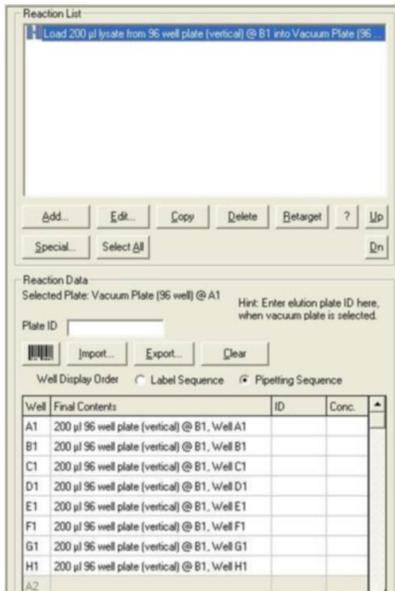


Figure 2: Example for right-hand pane information.

A post-run report is generated for each run and can be used for quality management purposes. It is shown after each run and is automatically saved in the **Reports** subdirectory of the **Data** directory (default location is **C:\Program Files\QIAcubeHT\Data**).

# Protocol: Purification of Viral Nucleic Acids from Diverse Samples

This protocol is for the purification of viral nucleic acids from diverse samples. See Table 1 on page 8 for sample pretreatments.

## Things to do before starting

- Ensure all reagents and samples are equilibrated to room temperature (15–25°C).
- Check for precipitates in reagents. If a reagent contains precipitates, incubate at 37°C with gentle shaking to dissolve precipitates. Avoid vigorous shaking, which causes foaming.
- Check that Buffer ACB, Buffer AW1, Buffer AW2 and carrier RNA have been prepared according to the instructions in “Preparing reagents and instrument” (page 17).
- When working with difficult samples, use a user confirmation step to check if all liquid has passed the membrane. See “Troubleshooting Guide” on page 37 and the *QIAcube HT User Manual* for guidance.
- Ensure that the relevant version of the **QIAamp 96 Virus QIAcube HT.QSP** run file is installed on the instrument.

QIAcube HT protocol files (file extension \*.QSP), which contain all the information required to perform a run on the QIAcube HT instrument, are available from [www.qiagen.com/p/QIAcubeHT](http://www.qiagen.com/p/QIAcubeHT), under the **Resources** tab.

- Ensure that Software version 4.17 or higher is installed. This is mandatory to process QIAamp 96 plates on the QIAcube HT and QIAextractor.
- Ensure that you are familiar with operating the instrument. Refer to the *QIAcube HT User Manual* for operating instructions.
- If the volume of the samples is less than 200 µl, add PBS or 0.9% NaCl to a final volume of 200 µl.

- Prepare a mixture of Buffer ACL, carrier RNA and internal control (if applicable) according to Table 2 on page 28.

## Procedure

1. Place the tip discard chute on the worktable so that the chute is over the tip disposal box. Ensure that the tip discard chute is open and unblocked. Remove the UV protective cap from the tip chute.

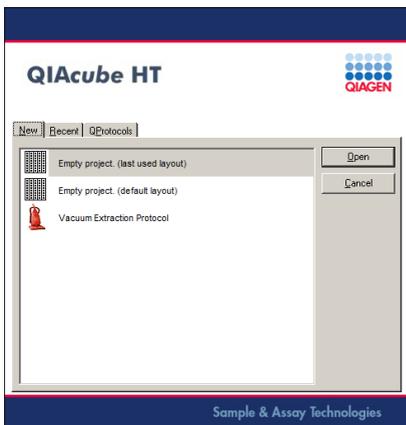
Ensure that the tip disposal box is empty and that the opening is aligned with the tip discard chute.

2. Switch on the instrument. The switch is located at the back of the instrument, on the lower left

3. Launch the QIAcube HT Software.

**Note:** If the QIAcube HT Software is already open, click  in the toolbar.

4. The following screen appears.



5. Select the **QProtocols** tab.

All Q Protocols that are saved in the appropriate **QProtocols** folder will be listed.

6. To open the run file, left-click on the Q Protocol **QIAamp 96 Virus QIAcube HT** to select it and then click **Open**.

Alternatively, double-click on the Q Protocol.

7. A **Protocol Description** of the selected Q Protocol will be displayed and the  icon will appear in the toolbar.

8. Check that the Q Protocol meets your requirements, and then click **Close**.

**Note:** To view the Q Protocol information box again, click on the  icon in the toolbar.

9. Click  in the toolbar.

The **Configuration (1)** step of the **Vacuum extraction** wizard opens. This wizard displays protocol parameters. For information about adjusting the parameters, see the *QIAcube HT User Manual*.

10. Select the appropriate number of samples arranged in columns in the 96-well plate.

Ensure the **Turn the HEPA filter on automatically** option is checked, and click **Jump to End**.

Reagent and consumable lot numbers can be entered in the **Configuration (1)** window for tracking.

The **Jump to End** button is located at the bottom left of the **Configuration (1)** window.

The **Wizard Summary** window opens. The information in this window can be printed for documentation purposes.

11. Confirm the protocol by clicking **Finish**. The wizard closes.

The QIAcube HT Software calculates the reagent volumes and the number of tips required to complete the protocol. These values are displayed with the worktable layout in the QIAcube HT workspace. For detailed information, see the *QIAcube HT User Manual*.

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12. Ensure that there are sufficient numbers of tips for at least all steps until and including lysate transfer, that tip boxes are placed in the indicated positions, and that the lids have been removed from the tip boxes.

Check that the number and position of available and unused tips is the same on the instrument worktable and in the software workspace.

If more tips are required, you will be prompted to replace empty tip racks with new tip racks during the run. Information about the approximate time for refill will be given in the pre run check. For more information, see the *QIAcube HT User Manual*.

In the software workspace, click on a tip in any tip position to open the **Tip info** preview.

13. Prepare the vacuum chamber as described in "Assembling the vacuum chamber", page 19. See the *QIAcube HT User Manual* for more information.

**Note:** If fewer than 12 columns (96 wells) are to be processed, seal the unused columns of the QIAamp 96 plate with adhesive tape (supplied). Unused wells must be sealed to ensure proper vacuum operation.

**Note:** Trim any excess tape.

**Note:** When reusing QIAamp 96 plate, S-Block or elution plate, take care to avoid cross-contamination.

**Note:** Be sure the QIAamp 96 plate is aligned to the left in the carriage and that the carriage is positioned to the left inside the vacuum chamber.

14. Add 200 µl sample to the selected S-Block wells. Place the S-Block in the B1 position of the QIAcube HT worktable.

**Note:** Samples are processed in columns of 8 wells. We recommend covering unused wells during pipetting for subsequent reuse of the S-Block.

**Note:** If fewer than 8 per columns samples are to be processed, unused wells in the column must contain water or buffer. The volume added to unused wells should be the same as the sample volume to avoid foaming.

15. Place clean reagent troughs in the indicated positions.

16. Transfer the indicated volumes of all reagents, except Buffer ACL mixture, into the corresponding reagent troughs, close the lids, and place them on the indicated positions on the worktable.

**Important:** If processing fewer than 96 samples, pipette one eighth of the required volume of Proteinase K into each well of the reagent trough to avoid uneven distribution of the Proteinase K during the protocol run.

The screenshot displays a software interface for a laboratory protocol. At the top, there is a menu bar with 'File', 'Control', 'Wizards', 'Options', and 'Help'. Below the menu bar is a toolbar with various icons. The main area shows a 96-well plate layout (A1-H12) and several reagent troughs with their respective volumes. The troughs are labeled R1, B1, C1, B2, and C2. R1 contains AW2 (77 ml) and AW1 (60.6 ml). B1 is a 2.2ml QIAGEN S-Block. C1 contains Proteinase K (2.24 ml), ACL Mix (16.36 ml), AVE (10.6 ml), and TopVue Fluid (5.88 ml). B2 and C2 are 200µl OnCor C tip, QIAGEN.

17. Prepare the indicated volume of ACL mixture according to Table 2 on page 28 and mix well.

**IMPORTANT:** Prepare ACL mixture immediately before starting the run.

Table 2. Preparation of Buffer ACL, carrier RNA, and internal control (if applicable)

	Number of samples									
	24	32	40	48	56	64	72	80	88	96
Buffer ACL (ml)	4.5	5.6	6.9	8.0	9.3	10.4	11.5	12.8	13.8	15.0
Carrier RNA (µl)	140	175	215	250	290	325	360	400	430	470
Internal Control (µl)	280	350	430	500	580	650	720	800	860	940

18. Start the run immediately by clicking 

The pre-run check appears.

19. Perform the pre-run check.

Check the state of the worktable items.

Confirm that worktable is setup correctly (instrument does not perform checks for all items). Check the box to the left of the items. A pre-run report can be saved for documentation purposes by clicking .

20. After completing the pre-run check, close the instrument hood and click **OK**.

The **OK** button is disabled until all pre-run check items have been checked.

21. Click **Cancel** when the **Save as** dialog box appears.

**Optional:** Save the run file with a unique file name. See the *QIAcube HT User Manual* for more details.

22. The protocol run begins.

**IMPORTANT:** At the beginning of each run an open circuit test and a plate detection test are performed automatically. If the QIAamp 96 plate in the transfer carriage is improperly aligned to the left side of the vacuum chamber you will be prompted to place it correctly. After adjusting the position, click **Retry** to initiate the tests again.

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23. Cover the elution plate (EMTR) with the lid and remove from the elution chamber, when the protocol is complete. See the *QIAcube HT User Manual* for detailed instructions.

Two liquid phases might be found in the Elution Microtubes. If this is the case, TopElute Fluid will be found as a top layer over the elution buffer. It is inert and has no effect on downstream applications.

**IMPORTANT:** Ensure that you only take the eluate from below the top layer.

## Cleaning the instrument after completing a run

1. Discard racks containing only used tips.

2. Discard leftover reagents.

**Note:** We recommend not reusing reagents in multiple runs. Reagents provided are sufficient for at least 10 runs of 48 samples.

**Note:** Do not clean the trough containing TopElute Fluid with water, but with a dry lint-free cloth only.

3. Discard the S-Block or keep partially used blocks for reuse.

4. Remove the transfer carriage and discard the QIAamp 96 plate or keep partially used QIAamp 96 plates for reuse.

5. Clean the carriage, channeling-block, channeling-block holder and tip chute.

6. With a damp cloth, clean any spilled reagent from the instrument worktable or vacuum chamber.

**Note:** For all further cleaning and maintenance operations, see Section 7 of the *QIAcube HT User Manual*.

7. Turn on the UV lamp to decontaminate the worktable by clicking .

8. See the *QIAcube HT User Manual* for detailed instructions.

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# Sample Pretreatment Protocols

## Pretreatment B1 for Respiratory Samples

This protocol is intended for viscous respiratory samples. Non-viscous respiratory samples do not require pretreatment and can be used directly as starting material in “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, pages 23–29.

### Procedure

1. Liquefy the sample according to either step 1a. or 1b.
  - 1a. Add 1 volume of Sputasol solution to 1 volume of sample, and shake well.  
Place in a 37°C water bath, and incubate with periodic shaking until the sample is completely liquefied.
  - 1b. Mix 1 volume of sample with 1 volume of NAC buffer (10 g N-acetylcysteine per liter of 0.9% NaCl solution). If the sample is very viscous or solid (e.g., when working with lower respiratory samples), try to disrupt it mechanically by pipetting up and down. Incubate at room temperature (15–25°C) for 30 min with constant shaking.  
For easier pipetting, it may be necessary to cut off the end of the pipette tip.  
If the sample is solid, the incubation time needs to be increased to completely liquefy the sample.
2. Centrifuge the liquefied sample to pellet debris, and transfer the clear supernatant to a clean tube.
3. Use 200 µl lysate as starting material and proceed directly with “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, page 23.

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## Pretreatment B2 for Liquid Transport Media

Most transport media, such as PreservCyt® solution, require no pretreatment and can be used directly as starting material in “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, pages 23–29. For some media such as SurePath® solution, a pretreatment prior to nucleic acid purification can improve extraction efficiency.

### Procedure

1. Add 400 µl media to 120 µl Buffer ATL and 30 µl proteinase K solution.  
The Buffer ATL and proteinase K solution supplied with the kit might not be sufficient for all users. If necessary, please order separately.
2. Mix the solution carefully.
3. Use 200 µl lysate as starting material and proceed directly with “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, page 23.

**Note:** Proteinase K can be replaced by Buffer ACL on the QIAcube HT worktable.

## Pretreatment B3 for Urine

### Procedure

1. Add 400 µl urine to 120 µl Buffer ATL and 30 µl proteinase K solution.  
The Buffer ATL and proteinase K solution supplied with the kit might not be sufficient for all users. If necessary, please order separately. See “Ordering Information” on page 43.
2. Mix the solution carefully.
3. Use 200 µl lysate as starting material and proceed directly with “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, page 23.

**Note:** Proteinase K can be replaced by Buffer ACL on the QIAcube HT worktable.

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## Pretreatment B4 for Dried Blood Spots

### Things to do before starting

- Read “Dried blood spots” on page 15.

### Procedure

1. Add dried blot spots (e.g., from Whatman FTA Cards) to 500 µl Buffer ATL and 30 µl proteinase K solution.  
The Buffer ATL and proteinase K solution supplied with the kit might not be sufficient for all users. If necessary, please order separately. See “Ordering Information” on page 43.
2. Incubate at 56°C for 1 h with constant agitation.
3. Centrifuge briefly, and transfer 500 µl supernatant to a clean tube.
4. Use 200 µl lysate as starting material and proceed directly with “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, page 23.

**Note:** Proteinase K can be replaced by Buffer ACL on the QIAcube HT worktable.

## Pretreatment B5 for Swabs and Buccal Cells

### Things to do before starting

- Read “Swabs” on page 13.

### Procedure

1. Add the swab to 800 µl Buffer ATL and 30 µl proteinase K solution.  
The Buffer ATL and proteinase K solution supplied with the kit might not be sufficient for all users. If necessary, please order separately. See “Ordering Information” on page 43.
2. Incubate at 56°C for 1 h with constant agitation.
3. Remove the swab from the tube, and centrifuge the tube to collect condensation.

4. Use 200 µl lysate as starting material and proceed directly with "Protocol: Purification of Viral Nucleic Acids from Diverse Samples", page 23.
5. **Note:** Proteinase K can be replaced by Buffer ACL on the QIAcube HT worktable.

## Pretreatment F1 for Stool Suspensions

### Procedure

1. Mix 1 volume of stool with 9 volumes of 0.9 % NaCl.
2. Vortex vigorously to prepare a homogeneous suspension.
3. Centrifuge the suspension to pellet debris, and transfer the clear supernatant to a clean tube.

**Note:** Depending on the bacteria species and the speed of centrifugation, bacteria will be present in the pellet.

4. Use 200 µl lysate as starting material and proceed directly with "Protocol: Purification of Viral Nucleic Acids from Diverse Samples", page 23.

## Pretreatment T1 Mechanical Disruption of Tissue

This pretreatment is for the extraction of viral RNA and viral DNA from most types of tissue.

### Important point before starting

- Stainless steel beads must be ordered separately (see "Ordering Information" on page 43).

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## Procedure

1. Place up to 5 mg tissue in 2 ml microcentrifuge tubes each containing 1 stainless steel bead (5 mm mean diameter).

**Note:** Up to 20 mg tissue can be processed. We recommend starting with 5 mg and increasing the amount if needed. For tissues with a very high number of cells for a given mass of tissue (e.g., spleen), a reduced amount of starting material (5–10 mg) should be used.

If working with fibrous tissue, cutting the tissue into smaller pieces before starting disruption will improve disruption efficiency.

2. Add 300  $\mu$ l PBS or 0.9% NaCl solution to each tube.
3. Place the tubes in the TissueLyser II Adapter Set.
4. Operate the TissueLyser II for 2 min at 25 Hz.

**Optional:** If working with fiber-rich tissue, disassemble the adapter set, rotate the rack of tubes so that the tubes nearest to the TissueLyser II are now outermost, and reassemble the adapter set. Operate the TissueLyser II for a further 2 min at 25 Hz.

5. Disassemble the adapter set. Centrifuge the samples at 14,000  $\times$   $g$  for 2 min at room temperature (15–25°C).
6. Use 200  $\mu$ l of the supernatant from step 5 as the starting material for “Protocol: Purification of Viral Nucleic Acids from Diverse Samples”, page 23.
7. For fiber-rich tissues, complete disruption may not always be possible. Ensure that no solid particles are transferred to the purification protocol.

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## Pretreatment T2 for Enzymatic Digestion of Tissue

This pretreatment is for the extraction of viral DNA from most types of tissue. It is not suitable for viral RNA because the lysis conditions do not sufficiently conserve RNA integrity.

### Things to do before starting

- Check Buffer ATL for precipitate upon storage. If it contains precipitates, warm to 56°C to dissolve precipitates.
- Heat a thermomixer block, shaking water bath or rocking platform to 56°C for use in step 3 of the pretreatment protocol.

### Procedure

1. Cut up to 5 mg tissue into small pieces and place in a 2 ml microcentrifuge tube. Add 180 µl Buffer ATL.

**Note:** Up to 20 mg tissue can be processed. We recommend starting with 5 mg and increasing the amount if needed. For tissues with a very high number of cells for a given mass of tissue (e.g., spleen), a reduced amount of starting material (5–10 mg) should be used.

We recommend cutting the tissue into small pieces for efficient lysis.

2. Add 20 µl QIAGEN Proteinase K. Close the cap and mix thoroughly by vortexing. Briefly centrifuge the tube to collect any solution from the cap.
3. Incubate at 56°C with constant agitation until the tissue is completely lysed.

Lysis time varies depending on the type of tissue processed. Lysis is usually complete in 1–3 h. If more convenient, overnight lysis is possible but should be evaluated for specific sample types.

After incubation, the lysate may appear viscous, but should not be gelatinous. If a substantial gelatinous pellet remains after incubation and vortexing, extend incubation time at 56°C for the QIAGEN Proteinase K digest and/or increase the amount of

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QIAGEN Proteinase K to 40  $\mu$ l. Reduce the amount of starting material in future preparations of this tissue type.

If no thermomixer, shaking water bath or rocking platform is available, incubate in a heating block or water bath and vortex occasionally during incubation to disperse the sample.

4. Optional: If solid tissue or debris remains in the tubes after lysis, add 50  $\mu$ l Buffer ATL. Mix by vortexing and centrifuge at 6,000  $\times$  g (8,000 rpm) for 1 min. Use 200  $\mu$ l of the supernatant in the next step.
5. Use 200  $\mu$ l lysate as starting material and proceed directly with "Protocol: Purification of Viral Nucleic Acids from Diverse Samples", page 23.

**Note:** Proteinase K can be replaced by Buffer ACL on the QIAcube HT worktable.

# 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](http://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/or protocols in this handbook or sample and assay technologies (for contact information, visit [www.qiagen.com](http://www.qiagen.com)).

## Comments and suggestions

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### Instrument issues

- |  |  |
|--|--|
| a) Recovery in case of instrument failure or user interruption | The QIAcube HT interrupts a run upon opening of the hood. The run will proceed normally once the hood is closed. To ensure process safety, this incident is reported in the post-run report.   |
| b) Instrument failure/cancelled run                            | It is possible to restart the protocol from the last successful step. The post-run report indicates the step where the error occurred. It is often possible to delete all steps before the indicated step in the right-hand pane and to restart the run from this point. Be sure that all parts and buffers are in the correct position.   |
| c) Blocked membranes   | <p>When processing samples that might potentially block the membrane, we recommend using a user confirmation step after addition of the first wash buffer (AW1).</p> <p>If liquid is still visible, remove 500 µl using a pipet. Then scrape the surface of the membrane with a fresh pipet tip in order to relocate any solid particles that may block the membrane. Take care not to damage the membrane. If there is still no liquid flow, pipet all liquid from the well and proceed with the run.</p> <p>After the instrument has added Buffer AW2, open the hood to pause the run. Check if the well is still blocked. If so, remove all liquid using a pipet and mark well as invalid.</p> <p>Do not perforate the membrane. Uncovered perforated wells will disturb vacuum integrity during elution across the whole plate.</p> <p>Proceed the run. No buffer will float over from the blocked well into other wells from this step on.</p> <p>Next time, use less sample (tissue) and prolong the digestion step.</p> |

## Comments and suggestions

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### Little or no viral DNA or RNA in the eluate

- |  |  |
|--|--|
| a) Buffer ACB prepared incorrectly                                 | Check that Buffer ACB concentrate was diluted with the correct volume of isopropanol, as indicated on the bottle. Use 100% isopropanol. Repeat the purification protocol with new samples.   |
| b) Buffer AW1 or Buffer AW2 prepared incorrectly                   | Check that Buffer AW1 or Buffer AW2 concentrate was diluted with the correct volume of ethanol, as indicated on the bottle. Use 96–100% ethanol. Do not use denatured alcohol, which contains other substances such as methanol or methylethylketone. Repeat the purification protocol with new samples.   |
| c) Insufficient sample lysis                                       | QIAGEN Proteinase K was stored at elevated temperatures for too long. Repeat the purification procedure using new samples and fresh QIAGEN Proteinase K (see storage recommendations on page 5).<br><br>For some DNA viruses and bacteria, heated lysis may improve lysis efficiency. For this purpose an off-board-lysis protocol is available. |
| d) Carrier RNA not added to Buffer ACL mix or degraded carrier RNA | Please refer to the recommendations for preparation, storage and addition of carrier RNA.  |
| e) RNase contamination in Buffer AVE                               | Take care not to introduce RNases, which can degrade viral RNA. This may occur if tubes containing Buffer AVE are opened repeatedly. In case of RNase contamination, replace the open vial of Buffer AVE with a new vial. Repeat the purification procedure with new samples.  |
| f) Nucleic acids in samples already degraded prior to purification | Samples were frozen and thawed more than once or stored at room temperature (15–25°C) for too long. Always use fresh samples or samples thawed only once. Repeat the purification protocol with new samples.   |

### DNA or RNA does not perform well in downstream applications

- |  |  |
|--|--|
| a) Little or no DNA or RNA in the eluate           | See “Little or no viral DNA or RNA in the eluate”, above.  |
| b) Too much eluate in the amplification reaction   | Some sample types may contain high amounts of background nucleic acids (e.g. whole blood, tissue) or PCR inhibiting substances (stool). High amounts of background nucleic acids may inhibit amplification reactions, and removal of inhibitors may not be complete without additional treatment. Reduce the amount of sample input and/or the amount of eluate added to the amplification reaction. |
| c) Too much background nucleic acids in the eluate | Determine the maximum amount of carrier RNA suitable for the amplification reaction.<br><br>In RT-PCR, a low DNA background is preferable. Use less eluate or use DNase during nucleic acid extraction. An additional protocol is available from QIAGEN Technical Services.  |

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### Comments and suggestions

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- d) Performance of purified nucleic acids in assays varies with aging of reconstituted wash buffers      Salt and ethanol components of Buffer AW1 or Buffer AW2 may have separated out after being left for a long period between preparations. Always mix buffers thoroughly before each preparation.
- e) Residual ethanol in the eluate      Perform the drying step at 25 kPa for 5 minutes.

### Precipitate in buffers

- a) Precipitate in Buffer ACL or Buffer ACB      Precipitate may form after storage at low temperature or prolonged storage. To dissolve precipitate, incubate Buffer ACL or ACB for 30 min at 37°C with occasional shaking.

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# Appendix A: General Remarks on Handling RNA

## Handling RNA

Ribonucleases (RNases) are very stable and active enzymes that generally do not require cofactors to function. Since RNases are difficult to inactivate and even minute amounts are sufficient to destroy RNA, do not use any plasticware or glassware without first eliminating possible RNase contamination. Great care should be taken to avoid inadvertently introducing RNases into the RNA sample during or after the purification procedure. To create and maintain an RNase-free environment, the following precautions must be taken during pretreatment and use of disposable and nondisposable vessels and solutions while working with RNA.

## General handling

Proper microbiological, aseptic technique should always be used when working with RNA. Hands and dust particles may carry bacteria and molds and are the most common sources of RNase contamination. Always wear latex or vinyl gloves while handling reagents and RNA samples to prevent RNase contamination from the surface of the skin or from dusty laboratory equipment. Change gloves frequently and keep tubes closed whenever possible. Keep purified RNA on ice when aliquots are pipetted for downstream applications.

To remove RNase contamination from bench surfaces, nondisposable plasticware and laboratory equipment (e.g., pipets and electrophoresis tanks), use of RNaseKiller (cat. no. 2500080) from 5 PRIME ([www.5prime.com](http://www.5prime.com)) is recommended. RNase contamination can alternatively be removed using general laboratory reagents. To decontaminate plasticware, rinse with 0.1 M NaOH, 1 mM EDTA\* followed by RNase-free water (see "Solutions", page

\* 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.

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41), or rinse with chloroform\* if the plasticware is chloroform-resistant. To decontaminate electrophoresis tanks, clean with detergent (e.g., 0.5% SDS),\* rinse with RNase-free water, rinse with ethanol (if the tanks are ethanol-resistant), and allow to dry.

## Disposable plasticware

The use of sterile, disposable polypropylene tubes is recommended throughout the procedure. These tubes are generally RNase-free and do not require pretreatment to inactivate RNases.

## Glassware

Glassware should be treated before use to ensure that it is RNase-free. Glassware used for RNA work should be cleaned with a detergent\*, thoroughly rinsed and oven baked at 240°C for at least 4 hours (overnight, if more convenient) before use. Autoclaving alone will not fully inactivate many RNases. Alternatively, glassware can be treated with DEPC\* (diethyl pyrocarbonate), as described in “Solutions” below.

## Solutions

Solutions should be purchased RNase-free or treated with 0.1% DEPC. We recommend purchasing RNase-free water. This is because, if trace amounts of DEPC remain after autoclaving buffer, purine residues in RNA might be modified by carbethoxylation and performance of enzymatic reactions such as PCR may be negatively affected. Therefore, residual DEPC must always be eliminated from solutions or vessels by autoclaving or heating to 100°C for at least 15 minutes.

DEPC is a strong, but not absolute, inhibitor of RNases. It is commonly used at a concentration of 0.1% to inactivate RNases on glass or plasticware or to create RNase-free solutions. DEPC inactivates RNases by covalent modification. Add 0.1 ml DEPC to 100 ml of

\* 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.

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the solution to be treated and shake vigorously to bring the DEPC into solution. Let the solution incubate for 12 hours at 37°C. Autoclave for 15 minutes to remove any trace of DEPC. DEPC will react with primary amines and cannot be used directly to treat Tris\*, buffers. DEPC is highly unstable in the presence of Tris buffers and decomposes rapidly into ethanol and CO<sub>2</sub>. When preparing Tris buffers, use RNase-free water to dissolve Tris to make the appropriate buffer.

\* 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.

# Ordering Information

Product	Contents	Cat. no.
QIAamp 96 Virus QIAcube HT Kit	For 480 preps: QIAamp plates, QIAGEN Proteinase K, Carrier RNA, Buffers	57731
QIAcube HT Plasticware	For 480 preps: 5 S-Blocks, 5 EMTR RS, 2 x 50 Caps for EMTR, 9 x 96 Filter-Tips OnCor C, TapePad	950067
Elution Microtubes RS	24 x 96 Elution Microtubes, racks of 96; includes cap strips	120008
S-Blocks	24 x 96-well blocks with 2.2 ml wells	19585
Buffer ATL (200 ml)	200 ml Tissue Lysis Buffer for 1000 preps	19076
Buffer ASL	4 x 140 ml Stool Lysis Buffer	19082
<b>TissueLyser</b>		
TissueLyser II	Bead mill, 100-120/220-240 V, 50/60 Hz; requires the TissueLyser Adapter Set 2 x 24 or TissueLyser Adapter Set 2 x 96*	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
TissueLyser Adapter Set 2 x 96	2 sets of adapter plates for use with Collection Microtubes (racked) on the TissueLyser II	69984
TissueLyser LT	Compact bead mill, 100-240 V AC, 50–60 Hz; requires the TissueLyser LT Adapter, 12-Tube†	85600

\* The TissueLyser II must be used in combination with the TissueLyser Adapter Set 2 x 24 or TissueLyser Adapter Set 2 x 96.

† The TissueLyser LT must be used in combination with the TissueLyser LT Adapter, 12-Tube.

<b>Product</b>	<b>Contents</b>	<b>Cat. no.</b>
TissueLyser LT Adapter, 12-Tube	Adapter for disruption of up to 12 samples in 2 ml microcentrifuge tubes on the TissueLyser LT	69980
Pathogen Lysis Tubes L	50 Pathogen Lysis Tubes and 1 vial of Reagent DX	19092
Pathogen Lysis Tubes S	50 Pathogen Lysis Tubes and 1 vial of Reagent DX	19091
Stainless Steel Beads, 5 mm (200)	200 stainless steel beads (5 mm diameter), suitable for use with TissueLyser systems	69989
<b>QIAcube HT instrument</b>		
QIAcube HT System	Robotic workstation with UV lamp, HEPA filter, laptop, QIAcube HT operating software, start-up pack, installation and training, 1-year warranty on parts and labor	9001793
Accessories Pack, QXT	Upgrade kit with adapters for using dedicated QIAcube HT Kits on the QIAxtractor; includes Transfer Carriage (9022654), Riser Block EMTR (9022655) and Channeling Adapter (9022656)	9022649
<b>Accessories</b>		
Collection Microtubes (racked)	(10 x 96) nonsterile polypropylene tubes; 960 in racks of 96	19560
Collection Microtube Caps	For capping collection microtubes and round-well blocks; 960 in strips of 8	19566
Sterile Foam-Tipped Swabs (100)	100 Sterile Foam-Tipped Swabs	159340

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For a complete list of accessories, visit [www.qiagen.com/p/QIAcubeHT](http://www.qiagen.com/p/QIAcubeHT).

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at [www.qiagen.com](http://www.qiagen.com) or can be requested from QIAGEN Technical Services or your local distributor.

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## Notes

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