

For general laboratory use.



High Pure Viral Nucleic Acid Large Volume Kit



Version 06

Content version: July 2011

For isolation of viral nucleic acids for PCR and RT-PCR

Cat. No. 05 114 403 001

Kit for 40 isolations

Store the kit at +15 to +25°C

- 🕒 If properly stored, all kit components are stable through the expiration date printed on the label.

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1. What this Product Does

Number of Tests The kit is designed for 40 isolations.

Kit Contents All solutions are clear except Vial 1 Binding buffer is clear to slightly turbid, and colorless to slightly yellowish viscous solution. Do not use them when precipitates have formed. Warm the solutions at +15 to +25°C or in a +37°C water bath until the precipitates have dissolved.

Vial/Cap	Label	Contents / Function
1 green	Binding Buffer	6 × 25 ml [6 M guanidine-HCl, 10 mM Tris-HCl, 10 mM urea, 20% Triton X-100 (w/v), pH 4.4 (+25°C)].
2	Poly(A)	Lyophilizate 2 mg poly(A) carrier RNA for binding of RNA
3 pink	Proteinase K	Lyophilizate 2 × 100 mg for the digestion of proteins
4a black	Inhibitor Removal Buffer	33 ml, add 20 ml absolute ethanol [5 M guanidine-HCl, 20 mM Tris-HCl, pH 6.6 (+25°C) final concentration after addition of ethanol]
4 blue	Wash Buffer	10 ml, add 40 ml absolute ethanol each case [20 mM NaCl, 2 mM Tris-HCl, pH 7.5 (+25°C) final concentrations after addition of ethanol]
5 colorless	Elution Buffer	30 ml Water, PCR Grade
6	High Pure Extender Assembly	8 bags, 5 pieces each in a single zip pack
7	Collection Tubes	2 bags with 50 polypropylene tubes (2 ml)

Storage and Stability

Store the High Pure Viral Nucleic Acid Large Volume Kit components at +15 to +25°C. If properly stored, all kit components are stable until the expiration date printed on the label.

⚠ Improper storage at +2 to +8°C (refrigerator) or –15 to –25°C (freezer) will adversely impact nucleic acid purification when precipitates form in the solutions.

This kit is shipped at +15 to +25°C.

Store reconstituted poly(A) carrier RNA solution in aliquots. Aliquots stored at –15 to –25°C are stable for 12 months.

Store reconstituted Proteinase K in aliquots. Aliquots stored at -15 to -25°C are stable for 12 months.

**Additional
Equipment and
Reagents
Required**

- Absolute ethanol
- Standard tabletop centrifuge with swing-bucket rotor capable of $5,000 \times g$ centrifugal force for 50 ml polypropylene tubes
- Standard tabletop microcentrifuge capable of $13,000 \times g$ centrifugal force (*e.g.*, Eppendorf 5415C or equivalent)
- Microcentrifuge tubes, 1.5 ml, sterile

Application

The High Pure Viral Nucleic Acid Large Volume Kit is designed for the purification of viral nucleic acids from up to 2.5 ml of mammalian serum, plasma or whole blood.

When using whole blood, total nucleic acids are purified, including viral nucleic acids. The purified viral nucleic acids are applied in PCR or RT-PCR directly after elution in nuclease-free water.

Preparation Time

Total time	Approximately 25 min
Hands-on time	10 min

2. How to Use this Product

2.1 Before you Begin

Precautions

- ⚠ Binding Buffer and Inhibitor Removal buffer contain guanidine hydrochloride which is an irritant. Always wear gloves and follow standard safety precautions to minimize contact when handling.
- ⚠ Do not let these buffers touch your skin, eyes, or mucous membranes. If contact does occur, wash the affected area immediately with large amounts of water; otherwise, the reagent may cause burns. If you spill the reagent, dilute the spill with water before wiping it up.
- ⚠ Never store or use the Binding Buffer near human or animal food.
- ⚠ Always wear gloves and follow standard safety precautions when handling these buffers.

Handling Requirements

- Exercise the normal precautions required for handling all laboratory reagents.
- Do not pool reagents from different lots.
- Immediately after usage, close all bottles in order to avoid leakage, varying buffer-concentrations or buffer conditions. After first opening store all bottles in an upright position.
- Do not use a kit after its expiration date.
- Avoid contact of the Binding Buffer and Inhibitor Removal Buffer with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with large amount of water. Burns can occur if left untreated. If the reagent spills, dilute with water before wiping dry.
- Do not use any modified ethanol.
- Use only calibrated pipettes.
- Protect the plastic disposables from direct sun light. Do not store the High Pure Extender Assembly near a window.

Laboratory Procedures

- All human sourced material and all resulting waste should be considered potentially infectious. Thoroughly clean and disinfect all work surfaces with disinfectants recommended by the local authorities.
- Do not eat, drink or smoke in the laboratory work area.
- Do not pipette by mouth.
- Wear protective disposable gloves, laboratory coats and eye protection when handling specimens and kit reagents.
- Avoid microbial and nuclease contamination of reagents when removing aliquots from reagent bottles.
- The use of sterile disposable pipettes is recommended.
- Wash hands thoroughly after handling samples and test reagents.

Waste Handling

- Dispose unused reagents and waste in accordance with country, federal state, and local regulations.
- Material Safety Data Sheets (MSDS) are available upon request from the local Roche office.

Sample Material Purification of viral nucleic acids from up to 2.5 ml

- serum
- plasma
- whole blood

⚠ Centrifuge samples containing precipitates before purification.

Preparation of Working Solutions

Beside the ready-to-use solutions supplied with this kit, you will need to prepare the following working solution:

Content	Reconstitution/ Preparation	Storage and Stability	For use in
Proteinase K (Vial 3; pink cap)	Dissolve Proteinase K in 5.5 ml Elution Buffer and mix thoroughly. Prepare 130 µl aliquots.	Store aliquots at –15 to –25°C, stable for 12 months	Protocol Step 1: Cell lysis
poly(A) carrier RNA (Vial 2)	Dissolve poly(A) carrier RNA (vial 2) in 0.65 ml Elution Buffer (vial 5). Prepare 15 µl aliquots.	Store aliquots at –15 to –25°C, stable for 12 months	For the preparation of the working solution
	Working solution: Thaw one vial with 15 µl poly(A) carrier RNA and mix thoroughly with 0.5 ml to 2.5 ml Binding Buffer (vial 1) according to Tab. 1 of the isolation protocol (section 2.4).	⚠ Always prepare freshly before use! Do not store!	Protocol step 1
Inhibitor Removal Buffer (Vial 4a; black cap)	Add 20 ml absolute ethanol to Inhibitor Removal Buffer and mix well. ⚠ Label and date bottle accordingly after adding ethanol.	Store at +15 to +25°C. Stable until the expiration date printed on kit label.	Protocol step 6: To remove PCR inhibitors
Wash Buffer (Vial 4; blue cap)	Add 40 ml absolute ethanol to each Wash Buffer and mix well. ⚠ Label and date bottle accordingly after adding ethanol.	Store at +15 to +25°C. Stable until the expiration date printed on kit label.	Protocol step 8 and 9: Removal of residual impurities

Controls

⚠ It is the user's own responsibility to apply an appropriate control concept.

2.2 Use of the High Pure Extender Assembly

The High Pure Extender Assembly is delivered in single zip-bags. Five High Pure Extender Assemblies are additionally packed in labeled zip-bags. Each High Pure Extender is assembled in a 50 ml polypropylene Tube. The High Pure Extender Assembly is designed for use with table-top centrifuges and swing-bucket rotors with $4,000 \times g$ force applicable.

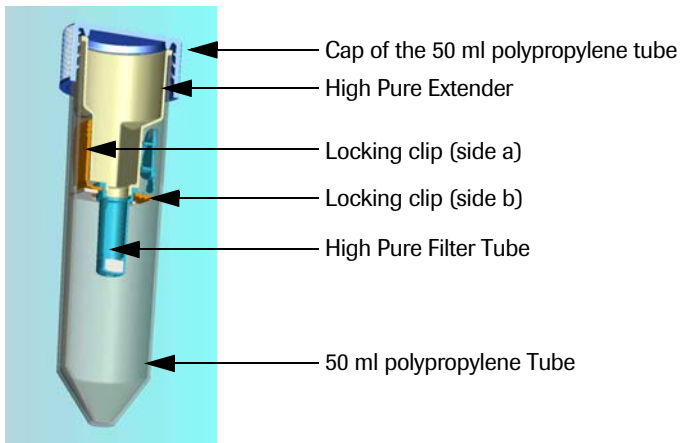


Fig. 1: High Pure Extender Assembly

Remove the High Pure Extender Assembly from the zip-bags prior to use. In order to load the sample onto the Assembly unscrew the cap of the 50 ml polypropylene tube. After sample loading, close the High Pure Extender Assembly with the 50 ml polypropylene tube cap.

After the first centrifugation step, the sample has passed through the High Pure Extender Assembly and is collected at the bottom of the 50 ml polypropylene tube. Nucleic acids are bound to the silica fleece at the bottom of the High Pure filter tube. For further processing remove the High Pure Extender Assembly from the 50 ml polypropylene tube. Discard the tube containing the flow-through. Remove the High Pure Filter Tube from the High Pure Extender Assembly (see Figures 2 to 6).

Disassembly of the High Pure Extender Assembly

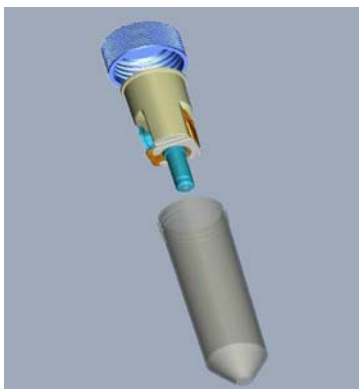


Fig. 2: Unscrew the 50 ml polypropylene tube and remove the High Pure Extender Assembly from the 50 ml polypropylene tube.



Fig. 3: Place the High Pure Extender in a new collection tube, which is placed securely in a tube rack on the bench.

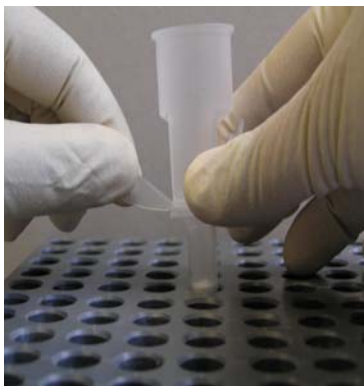


Fig. 4: Secure the High Pure Extender Assembly with one hand while grasping the locking clip (side a) of the High Pure Filter Tube cap on the opposite side with the other hand. Remove the first part of the locking clip (side a) by screwing the clip in either direction.



Fig. 5: Rotate the High Pure Extender Assembly. Remove the second part of the locking clip (side b) by pulling the locking clip away from the Extender Assembly.

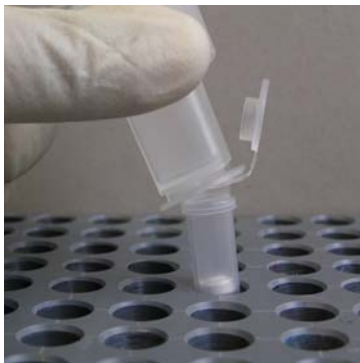
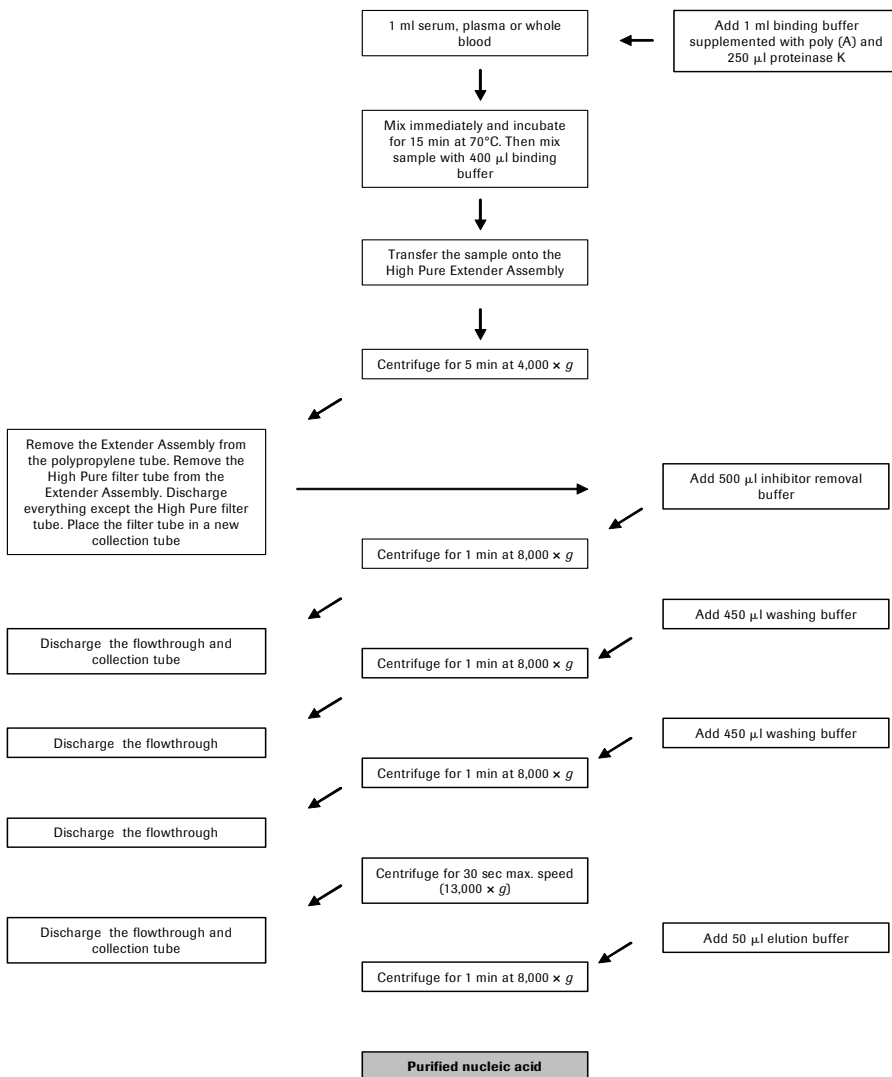


Fig. 6: Remove the High Pure Extender from the High Pure Filter Tube by tilting the High Pure Extender away from the High Pure Filter Tube toward the side without the cap.

- ⚠ We do not recommend the use of fixed-angle rotors in combination with the High Pure Extender Assembly. However, if you decide to use a centrifuge with fixed-angle rotors, do not centrifuge High Pure Extender Assembly above $3,000 \times g$. The use of fixed-angle rotors results in incomplete flow of the liquid through the Assembly. The remaining sample solution therefore stays within the High Pure spin column filter tube. Remove this remaining liquid by an additional spin of the High Pure Filter Tube in a bench-top centrifuge before the first washing step. For this additional spin remove the High Pure Spin Column from the High Pure Extender Assembly (according to Figure 2 to 6).

2.3 Experimental Overview



2.4 Isolation Protocol

Procedure for Preparing Nucleic Acids from 1 ml Samples of Serum, Plasma or Whole Blood

- ① To a nuclease-free 15 ml tube
 - Add 1 ml serum, plasma or whole blood
 - Add 1 ml working solution, freshly prepared, [carrier RNA-supplemented Binding Buffer]
 - Add 250 μ l Proteinase K solution, and mix immediately.
 - Incubate for 15 min at +70°C.
- ② Add 400 μ l Binding Buffer and mix.
- ③ To transfer the sample to a High Pure Extender Assembly:
 - Pipet entire sample into the upper reservoir of the High Pure Extender Assembly.
- ④
 - Insert the entire High Pure Filter Tube assembly into a standard tabletop centrifuge with a swing-bucket rotor.
 - Centrifuge 5 min at 4,000 $\times g$.
- ⑤ After centrifugation:
 - Remove the Filter Tube from the High Pure Extender Assembly, discard the flow-through liquid, and the 50 ml tube.
 - Combine the Filter Tube with a new Collection Tube.
- ⑥ After combining the Filter Tube with a new Collection Tube.
 - Add 500 μ l Inhibitor Removal Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 $\times g$.
- ⑦ After centrifugation:
 - Remove the Filter Tube from the Collection Tube, .
 - Combine the Filter Tube with a new Collection Tube.
 - Discard the Collection Tube including the flow-through liquid.
- ⑧ After removal of inhibitors:
 - Add 450 μ l Wash Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 $\times g$ and discard the flow-through.
- ⑨ After the first wash and centrifugation:
 - Remove the Filter Tube from the Collection Tube, discard the flow-through liquid.
 - Combine the Filter Tube with a new Collection Tube.
 - Add 450 μ l Wash Buffer to the upper reservoir of the Filter Tube.
 - Centrifuge 1 min at 8,000 $\times g$ and discard the flow-through.
 - Leave the Filter Tube-Collection Tube assembly in the centrifuge and spin it for 30 sec at maximum speed (approximately 13,000 $\times g$) to remove any residual Wash Buffer.
- 🕒 The extra centrifugation time ensures removal of residual Wash Buffer.
- ⑩ Discard the Collection Tube and insert the Filter Tube into a nuclease-free, sterile 1.5 ml microcentrifuge tube.

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To elute the viral nucleic acids:
 - Add 50 µl Elution Buffer to the upper reservoir of the Filter Tube.
 - Incubate for 1 min at room temperature.
 - Centrifuge the tube assembly for 1 min at 8,000 × *g*.
- 12

Either use the eluted nucleic acids directly in PCR (10 – 20 µl DNA eluate) or RT-PCR (3.5 µl viral RNA) or store the eluted viral RNA at –80°C or the viral DNA at +2 to +8°C or at –15 to –25°C for later analysis
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The High Pure Extender Assembly is designed for sample volumes up to 2.5 ml. The sample buffer compositions for different sample volumes are listed in the table below.

Sample Volume	0.5 ml	1 ml	2.5 ml
Binding Buffer	0.5 ml	1 ml	2.5 ml
Poly A	15 µl	15 µl	15 µl
Proteinase K	125 µl	250 µl	250 µl
Binding Buffer (protocol step 2)	0.2 ml	0.4 ml	1 ml

Tab. 1: Sample buffer compositions for different sample volumes loaded onto the High Pure Extender Assembly

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For isolation of nucleic acids from whole blood use pre-warmed Elution Buffer (+70°C).

3. Results

Sample Materials and Conditions

Validation of the High Pure Viral Nucleic Acid Kit is accomplished with DNA Virus (EBV) and RNA Virus (HAV) samples. Negative human samples (Serum, Citrate Plasma, and EDTA whole blood) were spiked with a dilution series of a virus stock solution prior to the isolation process. Isolation efficiency and quality were analyzed by qPCR and qRT-PCR on the LightCycler® 2.0 Instrument, respectively. Each isolation was performed in triplicate followed by a duplicated analysis on the LightCycler® 2.0 Instrument. Therefore each value is calculated as the mean of 6 CP-values.

Sensitivity and Linearity

In order to demonstrate the sensitivity of the High Pure Viral Nucleic Acid Large Volume Kit, 1 ml Citrated Plasma was spiked with decreasing amounts of HAV viral particles (1×10^5 to 1×10^2). Isolation was performed according to the Instructions for Use of the respective kit followed by quantitative analysis of HAV on the LightCycler® 2.0 Instrument.

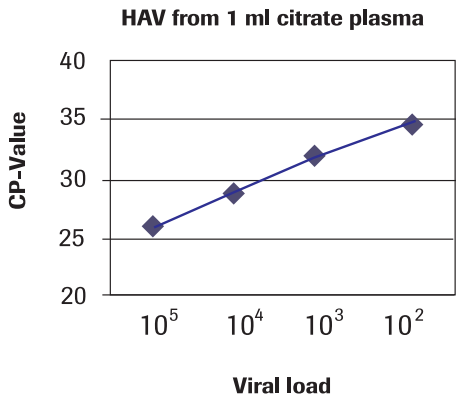


Fig. 7: Crossing Points of a series dilution of HAV particles in human citrate plasma after isolation with the High Pure Viral Nucleic Acid Large Volume Kit and subsequent analysis on the LightCycler® 2.0 Instrument.

As shown in Figure 7 we observe a high sensitivity and good linearity for HAV detection in Citrate Plasma down to 100 copies per 1 ml of sample volume.

Comparison with Other Products

For the comparison with other products, 1 ml of negative human serum was spiked with a dilution series (1×10^6 to 1×10^4) of HAV or EBV virus particles, followed by isolation according to the Instructions for Use of the respective kits.

Isolation efficiency and quality were analyzed by qPCR and qRT-PCR on the LightCycler® 2.0 Instrument utilizing a quantitative analysis of HAV or LightCycler® EBV Quantification Kit (Roche Molecular Diagnostics, Cat. No. 04 846 818 001), respectively.

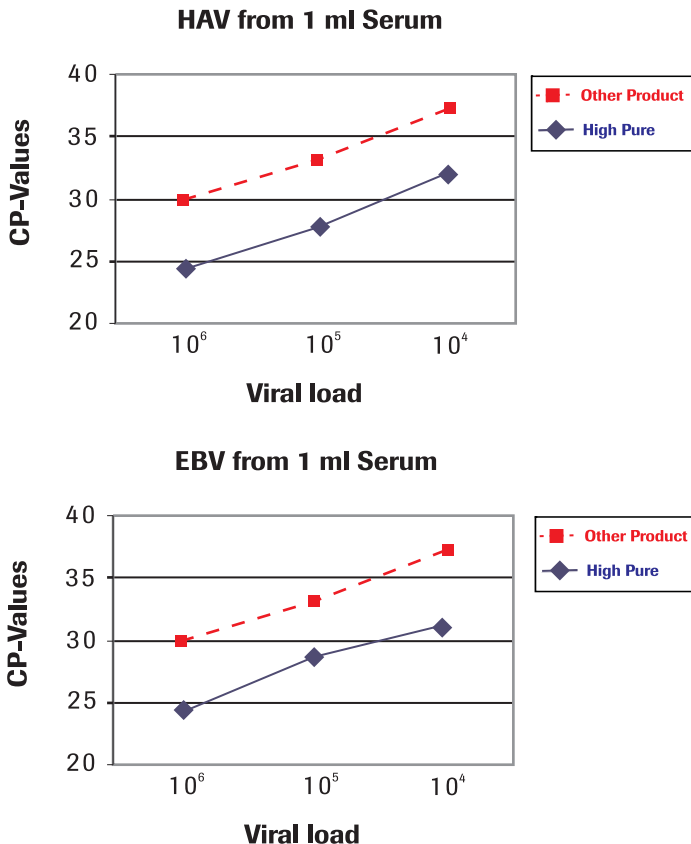



Fig. 8: Crossing Points of a series dilution of HAV and EBV particles in human serum after isolation with the High Pure Viral nucleic Acid Large Volume Kit and subsequent analysis on the LightCycler[®] 2.0 Instrument.

As shown in Figure 8, we observe a higher sensitivity and excellent linearity for HAV and EBV detection in 1 ml human serum compared with another product.

4. Troubleshooting

	Possible Cause	Recommendation
Low nucleic acid yield or purity	Kit stored under non-optimal conditions.	Store kit at +15 to +25°C at all times upon arrival.
	Buffers or other reagents were exposed to conditions that reduced their effectiveness.	<ul style="list-style-type: none"> • Store all buffers at +15 to +25°C. • Close all reagent bottles tightly after each use to preserve pH, stability, and freedom from contamination. • After any lyophilized reagent is constituted, aliquot it and store the aliquot at –15 to –25°C.
	Ethanol not added to Wash Buffer and Inhibitory Removal Buffer	<ul style="list-style-type: none"> • Add absolute ethanol to the buffers before using. • After adding ethanol, mix the buffers well and store at +15 to +25°C. • Always mark Wash Buffer vial and Inhibitory Removal Buffer vial to indicate whether ethanol has been added or not.
	Reagents and samples not completely mixed.	Always mix the sample tube well after addition of each reagent.
Poor elution of nucleic acids with water	Low temperature at Proteinase K digest.	<ul style="list-style-type: none"> • Check temperature during Proteinase K digestion. Heating blocks for 15 ml polypropylene tubes might deliver lower temperatures inside the tube. • Raise temperature of the heating block until +70°C is reached inside the 15 ml tube.
	Water has the wrong pH	If you use your own water or buffer to elute nucleic acids from Filter Tube, be sure it has the same pH as the Elution Buffer supplied in the kit.
Absorbance (A_{260 nm}) reading of product too high	Glass fibers, which might coelute with nucleic acid, scatter light	<ol style="list-style-type: none"> 1. Remove High Pure Filter Tube from tube containing eluted sample and spin sample for 1 min at maximum speed. 2. Transfer supernatant into a new tube without disturbing the glass fibers at the bottom of the original tube.
Low RNA yield	High levels of RNase activity	<ul style="list-style-type: none"> • Be careful to create an RNase-free working environment. • Process starting material immediately or store it at –80°C until it can be processed. • Use eluted RNA directly in downstream procedures or store it immediately at –80°C.

Low RNA yield

Possible Cause	Recommendation
Incomplete Proteinase K digestion	<p>Be sure to dissolve the lyophilized Proteinase K completely, as follows:</p> <ol style="list-style-type: none"> 1. Pipet 5.5 ml of Elution Buffer into the glass vial containing lyophilized Proteinase K. 2. Replace stopper and invert the vial until all the lyophilizate (including any stuck to the rubber stopper) is dissolved. 3. Aliquot the reconstituted enzyme, mark each aliquot with the date of reconstitution, and store at -15 to -25° C. <p> Reconstituted Proteinase K is stable for 12 months when stored properly.</p>

5. Additional Information on this Product

How this Product Works

Isolation of the analyte from serum, plasma or whole blood is required as a pre-requisite for the analysis of viral nucleic acids by the polymerase chain reaction (PCR) or RT-PCR.

Virus lysis is accomplished by incubation of the sample in a special Lysis/Binding buffer in the presence of Proteinase K. Subsequently, nucleic acids bind specifically to the surface of glass fibers in the presence of a chaotropic salt. The binding reaction occurs within seconds due to the disruption of the organized structure of water molecules and the interaction of nucleic acids with the surface of the glass fibers, thereby promoting adsorption to the glass fiber fleece. Since the binding process is specific for nucleic acids, the bound nucleic acids are purified from salts, proteins and other impurities by a washing step and are eluted in low salt buffer or water.

Test Principle

- ① Serum, plasma or whole blood are lysed by incubation with Binding buffer and Proteinase K.
- ② Nucleic acids are bound to the glass fibers pre-packed in the High Pure Filter Tube.
- ③ Bound nucleic acids are washed with a special Inhibitor Removal Buffer to get rid of PCR inhibitory contaminants.
- ④ Washing of bound nucleic acids, purification from salts, proteins and other cellular impurities.
- ⑤ Purified Nucleic Acids are recovered using the Elution Buffer.

Further Reading

- 1 Vogelstein B *et al.* (1979) Preparative and analytical purification of DNA from agarose. *Proc Natl Acad Sci USA* **76** (2), 615-619.
- 2 Mackay IM *et al.* (2003) Molecular Assays for Detection of Human Metapneumovirus. *Journal of Clinical Microbiology* **41**(1),100-105.
- 3 Greenberger S *et al.* (2004) Transcription-controlled gene therapy against tumor angiogenesis. *J Clin Invest.* **113** (7),1017-1024.
- 4 Koidl C *et al.* (2004) Detection of transfusion transmitted virus DNA by real-time PCR. *Journal of Clinical Virology* **29**, 277-281.
- 5 Widschwendter A *et al.* (2004) Analysis of Aberrant DNA Methylation and Human Papillomavirus DNA in Cervicovaginal Specimens to Detect Invasive Cervical Cancer and Its Precursors. *Clinical Cancer Research* **10**, 3396-3400.
- 6 Fred J. DeGraves *et al.* (2003) High-Sensitivity Quantitative PCR Platform. *BioTechniques* **34** (1), 106-115.
- 7 Ying LI *et al.* (2004) Size Separation of Circulatory DNA in Maternal Plasma Permits Ready Detection of Fetal DNA Polymorphism. *Clinical Chemistry* **50**, 1002-1011.

Quality Control

A dilution series of EBV sequence containing plasmids is prepared in 1 ml human plasma, applied to the filter tubes, washed and eluted according to the kit protocol. 5 µl of the eluate is analyzed by an EBV-specific qPCR on the LightCycler® 2.0 Instrument. Down to 10⁴ copies of EBV plasmid DNA are detected.

6. Supplementary Information

6.1 Conventions

Text Conventions To make information consistent and memorable, the following text conventions are used in this document:

Text Convention	Usage
Numbered stages labeled ①, ②, <i>etc.</i>	Stages in a process that usually occur in the order listed
Numbered instructions labeled ①, ②, <i>etc.</i>	Steps in a procedure that must be performed in the order listed
Asterisk *	Denotes a product available from Roche Applied Science.

Symbols

In this document, the following symbols are used to highlight important information:

Symbol	Description
⑨	Information Note: Additional information about the current topic or procedure.
⚠	Important Note: Information critical to the success of the procedure or use of the product.

6.2 Changes to previous versions

- Disclaimer of License deleted

6.3 Ordering Information

Roche Applied Science offers a large selection of reagents and systems for life science research. For a complete overview of related products and manuals, please visit and bookmark our home page, www.roche-applied-science.com, and our Special Interest Sites including:

- Nucleic Acid Isolation and Purification:
<http://www.roche-applied-science.com/napure>
- PCR - Innovative Tools for Amplification:
<http://www.roche-applied-science.com/pcr>

	Product	Pack Size	Cat. No.
Associated Kits	High Pure Viral Nucleic Acid Kit	100 reactions	11 858 874 001
	High Pure Viral Nucleic Acid Buffer Set	100 isolations	12 011 875 001
	High Pure PCR Template Preparation Kit	100 purifications	11 796 828 001
	High Pure Viral RNA Kit	100 purifications	11 858 882 001
LightCycler® Kits for PCR	LightCycler® DNA Master HybProbe	1 kit (96 reactions)	12 015 102 001
		1 kit (480 reactions)	12 158 825 001
	LightCycler® FastStart DNA Master HybProbe	1 kit (96 reactions)	03 003 248 001
		1 kit (480 reactions)	12 239 272 001
	LightCycler® FastStart DNA Master ^{PLUS} HybProbe	1 kit (96 reactions)	03 515 575 001
		1 kit (480 reactions)	03 515 567 001
	LightCycler® FastStart DNA Master ^{PLUS} HybProbe, 100 µl Reactions	1 kit (384 reactions)	03 752 178 001
	LightCycler® DNA Master SYBR Green I	1 kit (96 reactions)	12 015 099 001
		1 kit (480 reactions)	12 158 817 001
	LightCycler® FastStart DNA Master SYBR Green I	1 kit (96 reactions)	03 003 230 001
		1 kit (480 reactions)	12 239 264 001
LightCycler® Kits for RT-PCR	LightCycler® FastStart DNA Master ^{PLUS} SYBR Green I	1 kit (96 reactions)	03 515 869 001
		1 kit (480 reactions)	03 515 885 001
	LightCycler® FastStart DNA Master ^{PLUS} SYBR Green I, 100 µl Reactions	1 kit (384 reactions)	03 752 186 001
	LightCycler® RNA Master HybProbe	1 kit (96 reactions)	03 018 954 001
	LightCycler® RNA Master SYBR Green I	1 kit (96 reactions)	03 064 760 001
	LightCycler® RNA Amplification Kit HybProbe	1 kit (96 reactions)	12 015 145 001

Single Reagents

Product	Pack Size	Cat. No.
LightCycler® RNA Amplification Kit SYBR Green I	1 kit (96 reactions)	12 015 137 001
LightCycler® Control Kit RNA	1 kit (50 reactions)	12 158 841 001
High Pure PCR Product Purification Kit	50 purifications	11 732 668 001
	250 purifications	11 732 676 001
Reverse Transcriptase, M-MuLV	500 U	11 062 603 001
Reverse Transcriptase AMV	500 U	11 495 062 001
	1,000 U	10 109 118 001
Protector RNase Inhibitor	2,000 U	03 335 399 001
	10,000 U	03 335 402 001
Transcriptor Reverse Transcriptase	250 U	03 531 317 001
	500 U	03 531 295 001
	2,000 U	03 531 287 001
Transcriptor First Strand cDNA Synthesis Kit	1 kit (50 reactions)	04 379 012 001
	1 kit (100 reactions)	04 896 866 001
	1 kit (200 reactions)	04 897 030 001

6.4 Trademarks

HIGH PURE, HYBPROBE, FASTSTART, LIGHTCYCLER are trademarks of Roche.

SYBR is a registered trademark of Molecular Probes Inc., Eugene, OR, USA.

All other product names and trademarks are the property of their respective owners.

6.5 Regulatory Disclaimer

For general laboratory use.

Contact and Support

If you have questions or experience problems with this or any Roche Applied Science (RAS) product, please contact our Technical Support staff. Our scientists commit themselves to providing rapid and effective help.

We also want you to contact us if you have suggestions for enhancing RAS product performance or using our products in new or specialized ways. Such customer information has repeatedly proven invaluable to RAS and the world-wide research community.

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site at:**

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