January 2009

QIAvac HiSpeed LS Handbook

For vacuum processing of QIAGEN®

HiSpeed Plasmid Mega/ Giga column

Sample & Assay Technologies

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Components of QIAvac HiSpeed LS

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Elution Rack (for HiSpeed Mega/Giga tips)	4	11
Tool for valve opening/ closing	8	13

Components which are delivered with the HiSpeed Plasmid Mega/Giga EF Kit

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HiSpeed Plasmid Mega/ Giga tip	----	-----
Collection vessel for Eluate of HiSpeed Plasmid Mega/Giga tip	8	13
QIAconcentrator with Extender	9	13
Collection tubes for QIAconcentrator	---	---

Storage

The QIAvac HiSpeed LS is shipped at room temperature (15–25°C), and should be stored dry and clean at room temperature.

Product Use Limitations

The QIAvac HiSpeed LS is intended for general laboratory use. No claim or representation is intended for its use to identify any specific organism or for a specific clinical use (diagnostic, prognostic, therapeutic, or blood banking). It is the user's responsibility to validate the performance of the QIAvac HiSpeed LS for any particular use, since its performance characteristics have not been validated for any specific organism.

Technical Assistance

Lothar Breitkopf

Application Specialist

QIAGEN GmbH

Research & Development

QIAGEN Strasse 1

40724 Hilden

Germany

Tel. +49-2103-29-14672

Fax +49-2103-29-24672

lothar.breitkopf@qiagen.com

Safety Information

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

Introduction

The QIAvac HiSpeed LS is designed for fast and efficient vacuum processing of up to 6 QIAGEN HiSpeed Plasmid Mega EF or HiSpeed Plasmid Giga EF tips in parallel.
The purification of Plasmid DNA is vacuum driven instead of gravity flow, providing greater speed and reduced hands-on time. QIAGEN HiSpeed Plasmid Mega tip or QIAGEN HiSpeed Plasmid Giga tip are connected with a connection adapter to the QIAvac HiSpeed LS manifold.

Principle and procedure

QIAGEN HiSpeed plasmid purification protocols are based on a modified alkaline lysis procedure, followed by vacuum driven purification of plasmid DNA on QIAGEN Anion-Exchange Resin under appropriate low-salt and pH conditions. RNA, proteins, dyes, and low-molecularweight impurities are removed by a medium-salt wash.

Therefore the HiSpeed Plasmid Mega tips or HiSpeed Plasmid Giga tips are connected with a connection adapter to the QIAvac HiSpeed LS. The flow through during equilibration, loading and washing the HiSpeed Plasmid Mega/Giga tips is collected in a suitable waste container system.

After a few rebuilding's the QIAvac HiSpeed LS the Plasmid DNA is ready for the elution of Plasmid DNA under highsalt buffer conditions directly into a collection vessel.

The eluted DNA is then concentrated and desalted by isopropanol precipitation. After addition of isopropanol directly into the collection vessel and a few rebuilding steps of the QIAvac HiSpeed LS the solution is sucked through the membrane of the QIAconcentrator by vacuum.

The QIAconcentrator revolutionizes the isopropanol precipitation step, making it fast, easy, and risk-free. Plasmid DNA eluted from the HiSpeed tip is mixed with isopropanol and applied to the QIAconcentrator using the QIAvac HiSpeed LS provided. The precipitated DNA is trapped in the QIAconcentrator as a thin layer, which allows thorough drying and removal of alcohol by centrifugation after alcohol wash step. The DNA is then simply eluted from the QIAconcentrator with e.g. Buffer TE into a fresh tube also by centrifugation.

QIAGEN HiSpeed Plasmid Mega/Giga Kit allow very fast large-scale plasmid preparations using the vacuum system without time consuming gravity flow procedures or centrifugation, pellet drying and resuspension procedures after isopropanol precipitation. The highly concentrated DNA is ready for immediate use.

Important points before starting

Please take a few moments to read this handbook carefully before beginning the DNA preparation.

- When working with chemicals always wear a suitable lab coat, disposable gloves and protective goggles
- Always wear protective goggles when working with the system under vacuum.
- Important Note: Be careful that the QIAGEN-tips don't run dry during purification. This will lead to significantly decreased flow rates until the air is flushed out of the resin again.
- Check regularly the liquid level of the waste container and empty the container before it is completely filled.
- The QIAvac HiSpeed LS manifolds are not resistant to ethanol, methanol or other organic solvents. Do not bring solvents into contact with QIAvac HiSpeed LS. If solvents are spilled on the unit, rinse thoroughly with distilled water. Do not incubate acrylic components in alcohol containing reagents. The QIAvac HiSpeed LS should be cleaned with water or laboratory detergent after use. Ethanol should not be used
- Do not use cleaning material that contain abrasives

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Equipment and Reagents to be supplied by User

- Vacuum pump (e.g., QIAGEN Vacuum Pump)
- Waste system, container and tubings for collecting of liquid waste

Assembling the QIAvac HiSpeed LS

Overview of QIAvac HiSpeed LS

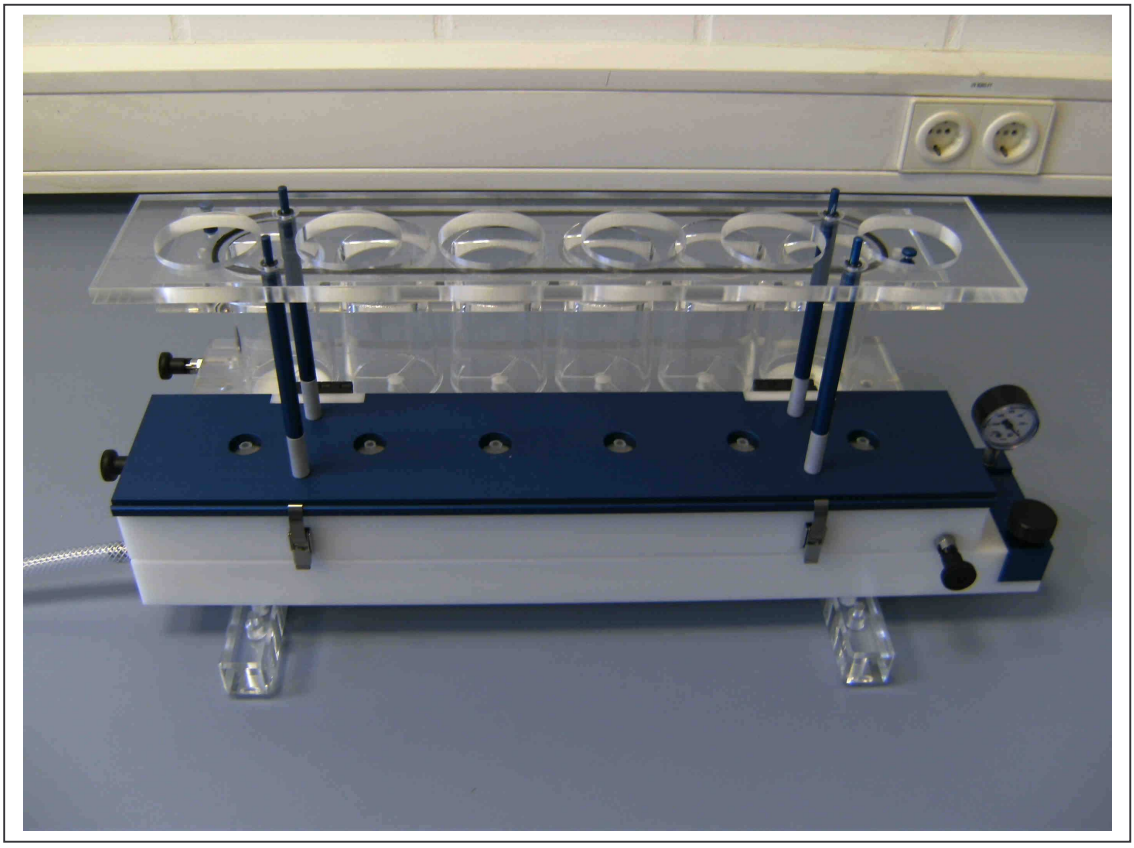


Figure 1

Detailed description of QIAvac HiSpeed LS components:

Component 1: basic wash station

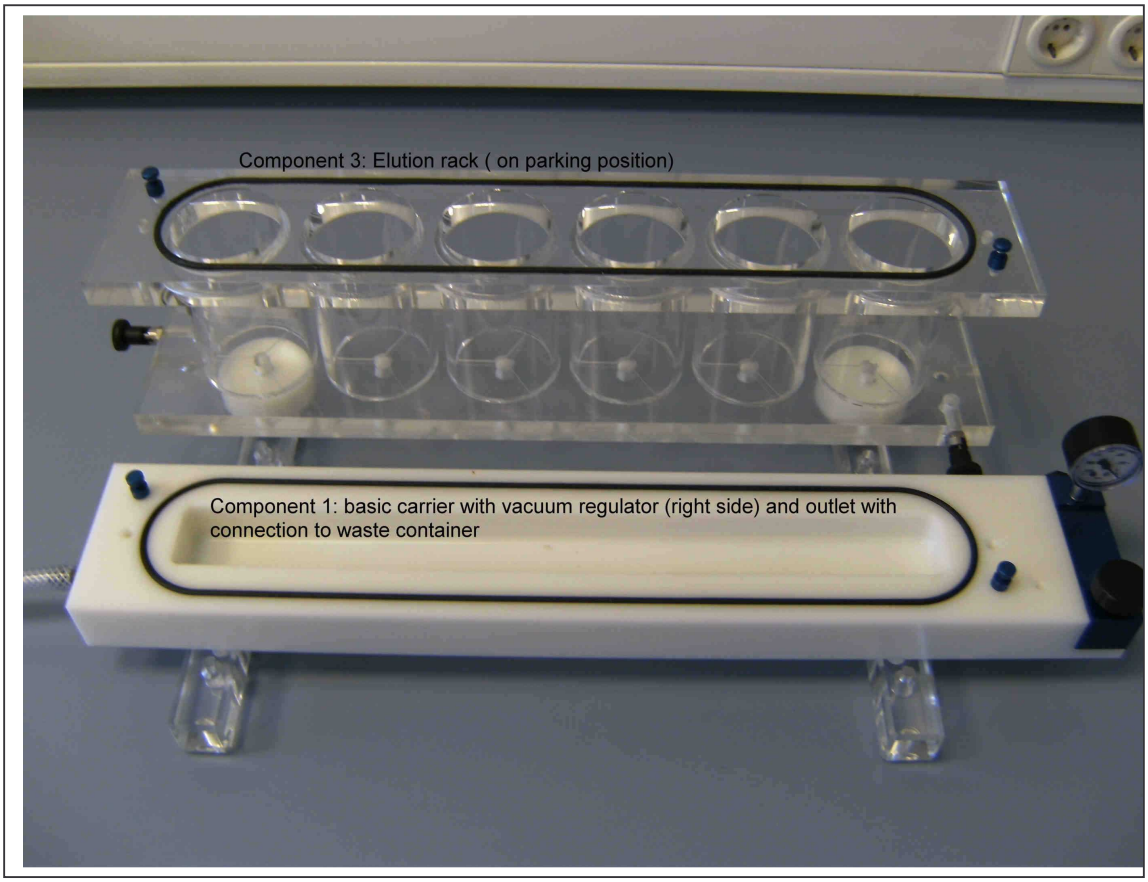


Figure 2

Component 2: column holder

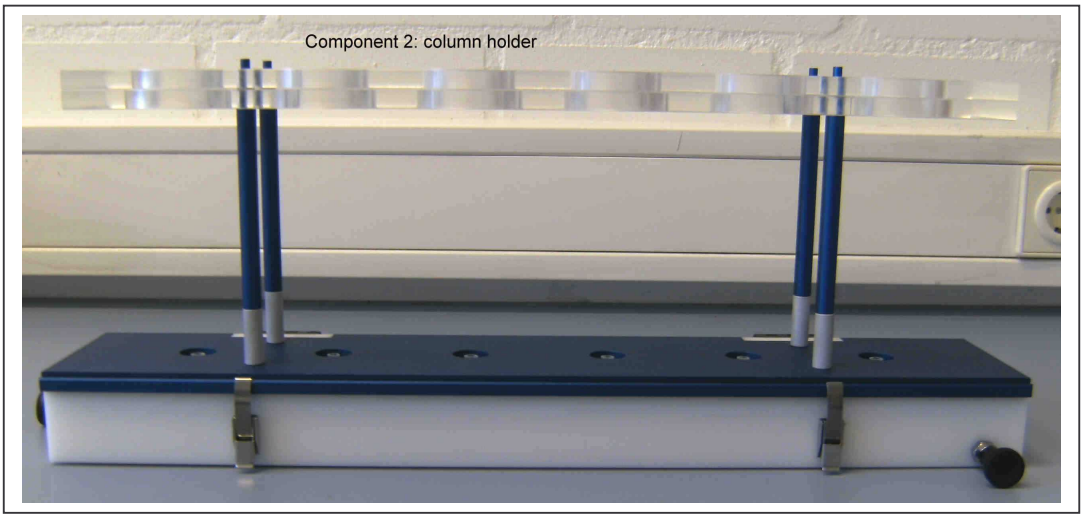


Figure 3

Component 3: Elution rack

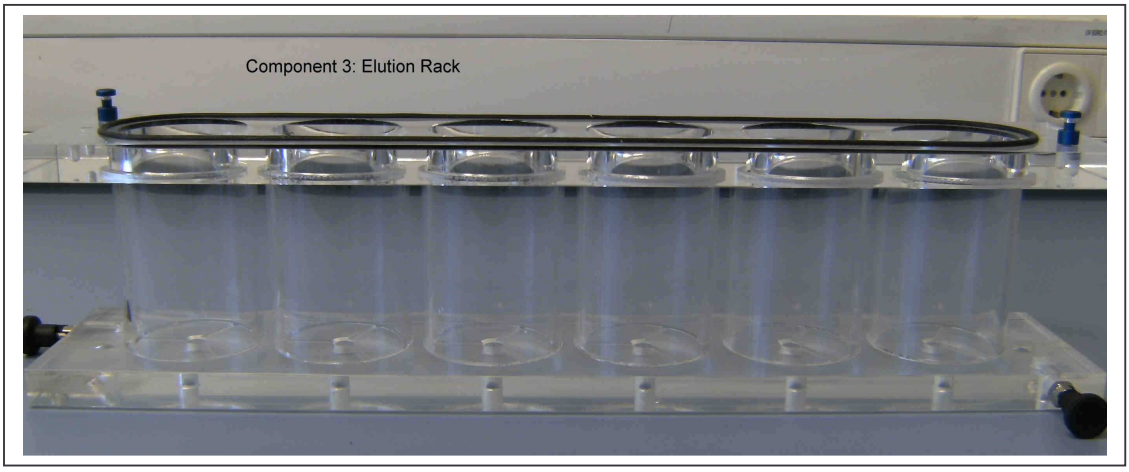


Figure 4

Component 4: connection adapter

Figure 5

Component 5: Tool

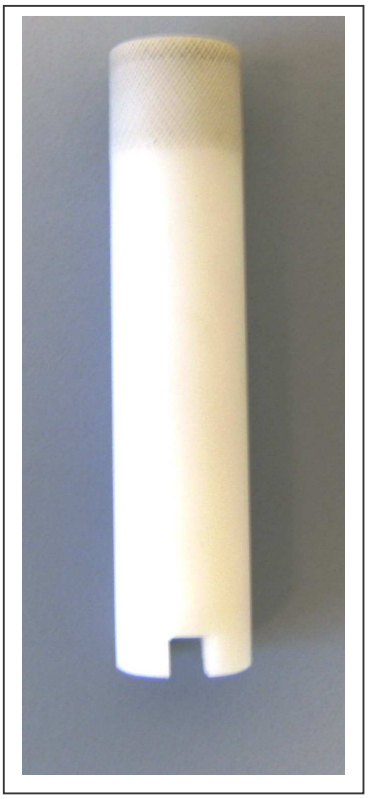


Figure 6

Open/close valve with tool

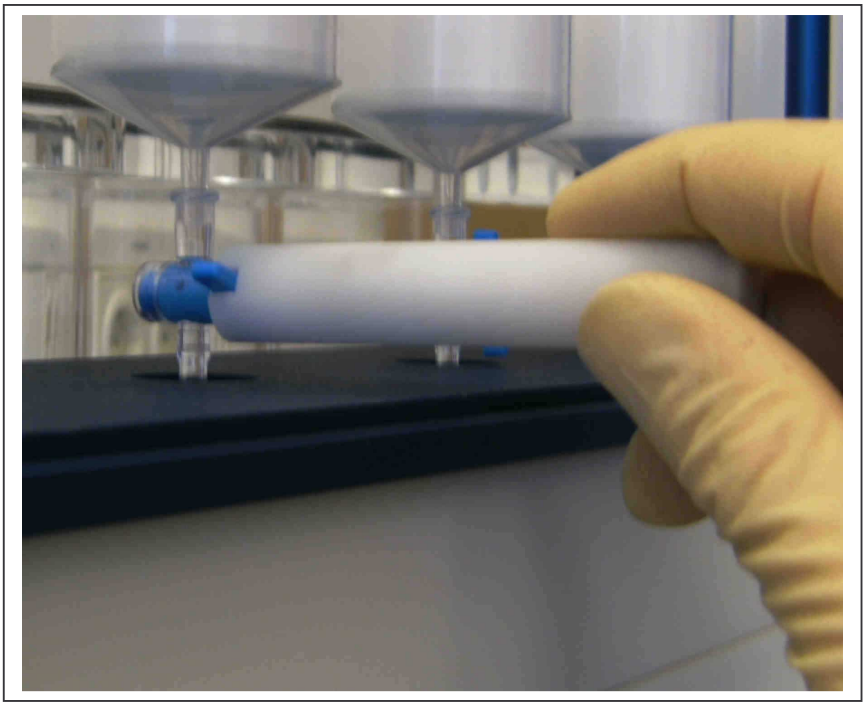


Figure 7

Component 6: Collection vessel To collect Eluate from HiSpeed Plasmid Mega/Giga tip

Figure 8

Component 7: QIAconcentrator with extender

Figure 9

Protocol: Processing QIAGEN HiSpeed Plasmid Mega/ Giga Column on the QIAvac HiSpeed LS

Procedure of assembling the QIAvac HiSpeed LS

1. Place Component 1 on a flat, planar surface and connect it with a waste container and vacuum pump to build up a vacuum system. For connection use suitable tubings that fits onto the tubing connection on the left side of component 1. The waste container is connected with component 1 and a vacuum source. (see Figure 10 as an example for a waste system with two containers)
2. Place component 2 (column holder) onto component 1 (basic carrier) and fix it with the snap-on connection, therefore please press slightly onto the upper component 2
3. Open the lid of component 2 and exchange the blind plugs by the number of connection adapters you will use for the purification. The upper part (blue valve) of the connection adapter has to be removed before closing the lid. After closing the lid the removed valves are plugged into the connection adapter again. Positions which are not needed are closed by a blind plug.
4. Insert the QIAGEN HiSpeed Mega/Giga column into the luer slot of the valve on the inlet side of the connection adapter.
5. The QIAvac HiSpeed LS is now ready for the Equilibration, Binding and wash step according to the protocol in the QIAGEN HiSpeed Plasmid Mega/Giga EF Kit Handbook.

Example for installation of the QIAvac HiSpeed LS

QIAvac HiSpeed LS connected with Waste Container and Vacuum pump (has to be supplied by user)

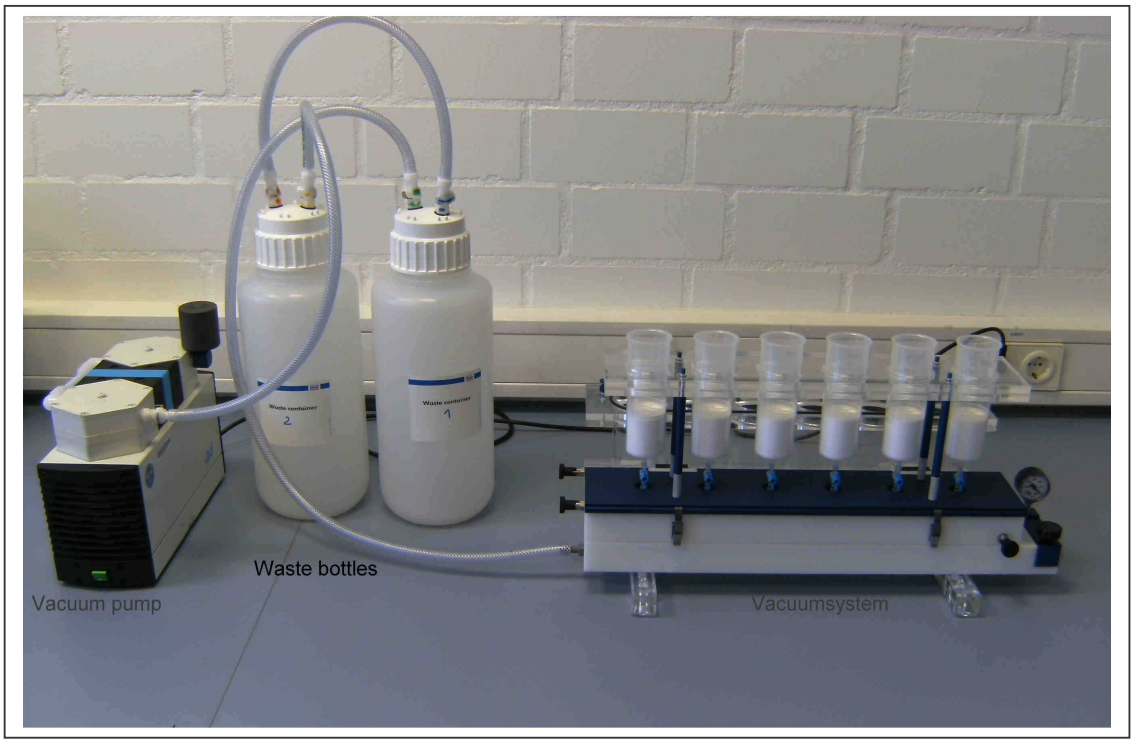


Figure 10

Ordering information for Waste system accessories

The following components may be used:

- 5 liter waste container (e.g., Nalgene Heavy Duty Vacuum Bottle, cat. no. 2126-5000)
- 10 liter waste container (e.g., Nalgene Heavy Duty Vacuum Carboy [PP], cat. no. 2226-0020)
- 20 liter waste container (e.g., Nalgene Heavy Duty Vacuum Carboy [PP], cat. no. 2226-0050)
- Quick filling/venting closure (e.g., Nalgene, cat. no. 2158-0021)
- Vacuum tubing (e.g., Nalgene 180 clear plastic vacuum tubing, cat. no. 8000-0065 [50 ft. per case])
- Vacuum tubing (e.g., Nalgene 180 clear plastic vacuum tubing, cat. no. 8000-0065 [10 ft. per case])

Detailed description of QIAGEN HiSpeed Plasmid column assembling

The connection adapter are placed into the holder in component 2 (figure 12) positions which are not used are prepared with blind plugs.

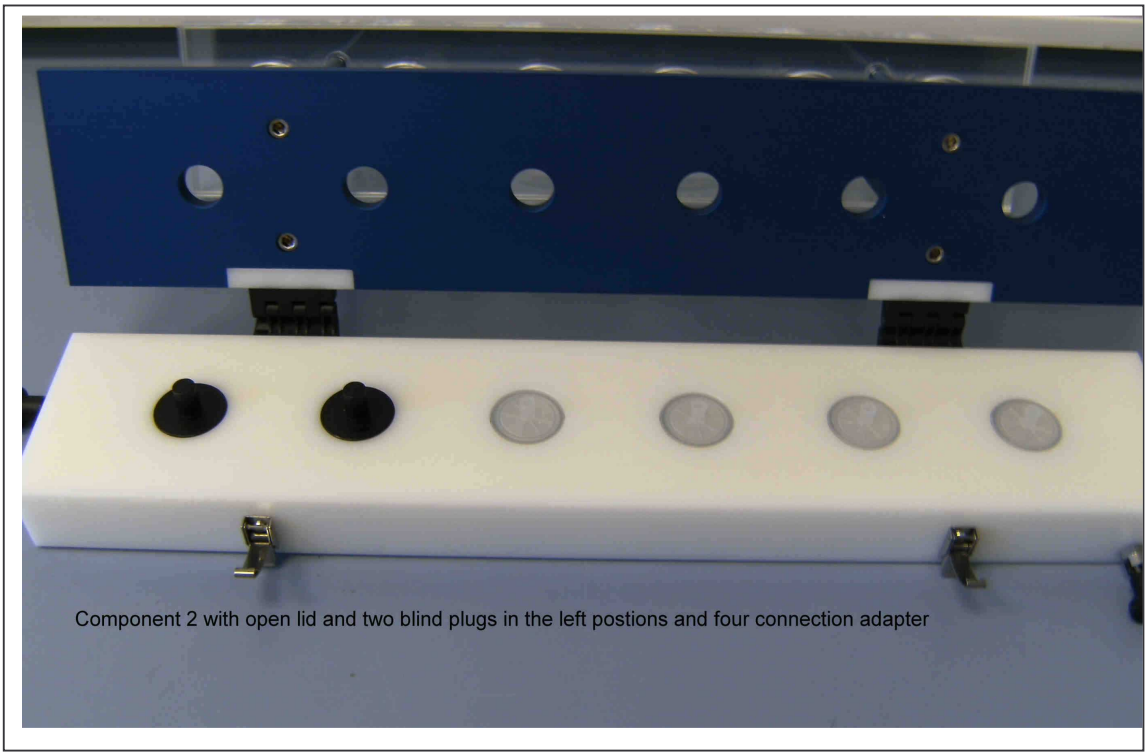


Figure 11

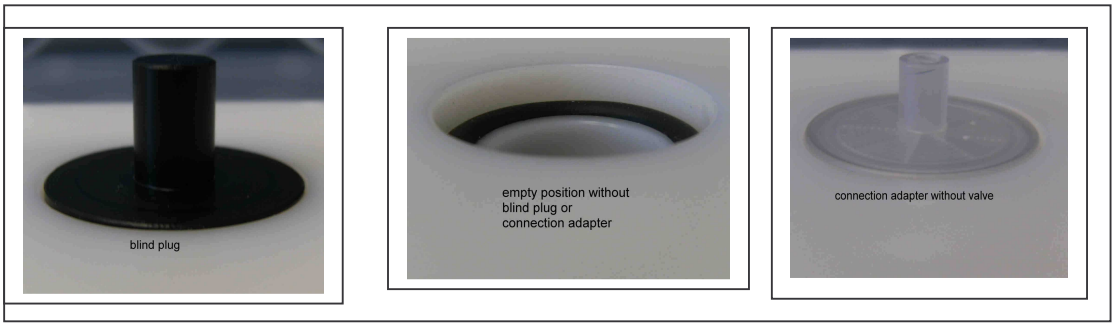


Figure 12

Assembling of valves into the connection adapter

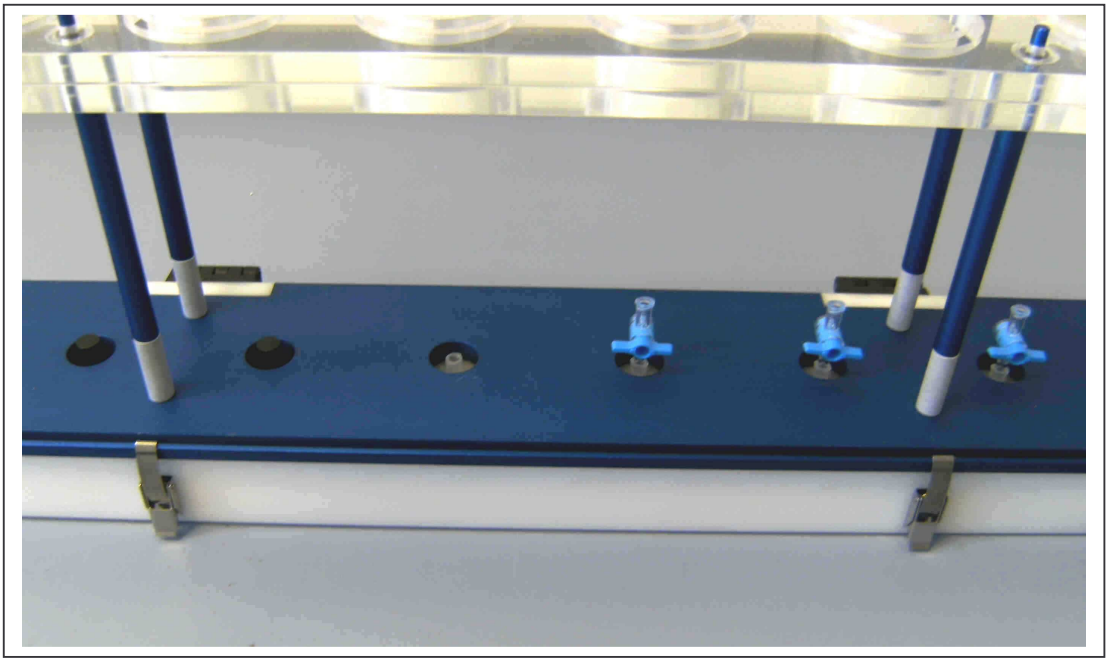


Figure 13

The lid is closed and fixed with both stainless steel cramps. The blue valves are plugged into the connection adapter (see three positions on right side). On the left side two positions are closed with blind plugs, in the middle position only the connecting adapter is fixed without valve.

Assembling of QIAGEN HiSpeed Plasmid Giga tips

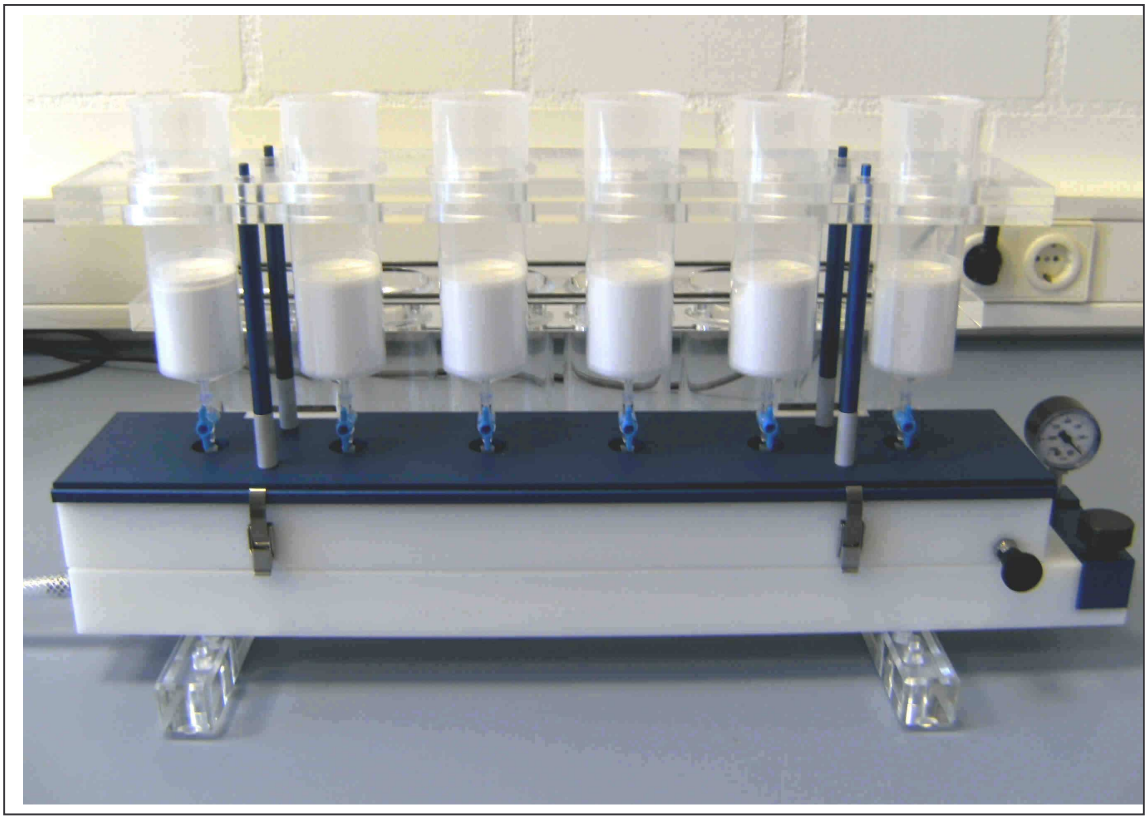


Figure 14

QIAvac HiSpeed LS ready to use for first purification steps with HiSpeed Plasmid Giga tips

See according pages of the HiSpeed Plasmid Mega/Giga EF Handbook

Protocol: Processing QIAGEN HiSpeed Plasmid Mega/ Giga tips on the QIAvac HiSpeed LS

Rebuilding of QIAvac HiSpeed LS for Elution of QIAGEN HiSpeed Plasmid Mega/Giga tips

1. After releasing the vacuum after wash step with Buffer QC insert the according number of collection vessel in each holder of component 3 (elution rack).(see figure 15)
2. Open the snap-on connection between component 1 and component 2 and place component 2 onto component 3. Fix both snap-on connections between component 2 and 3.
3. Remove the connected component 2 and component 3 from the parking position and place it onto component 1(see Figure 15). Connect component 1 with component 3 with both snap-on connections (see figure 16)
4. The system is now ready for the elution procedure according to the protocol in the HiSpeed Plasmid Mega/Giga Vacuum Kit Handbook.

Component 3 (Elution rack, fixed on basic carrier) assembled with 3 elution bottles.

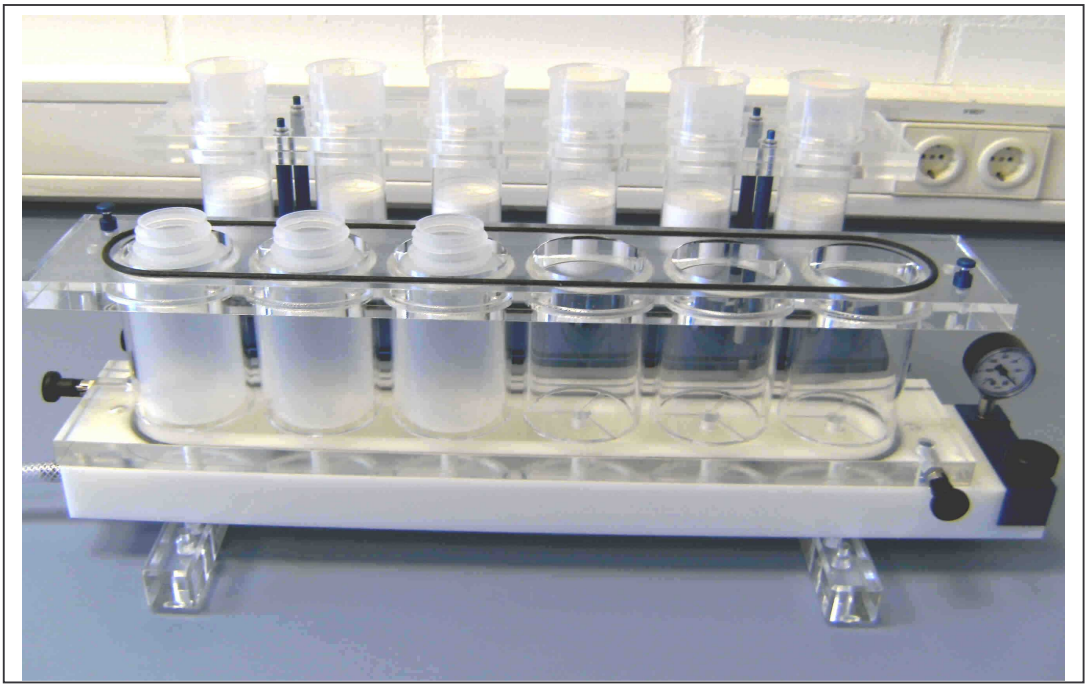


Figure 15

Component 2 connected with component 3

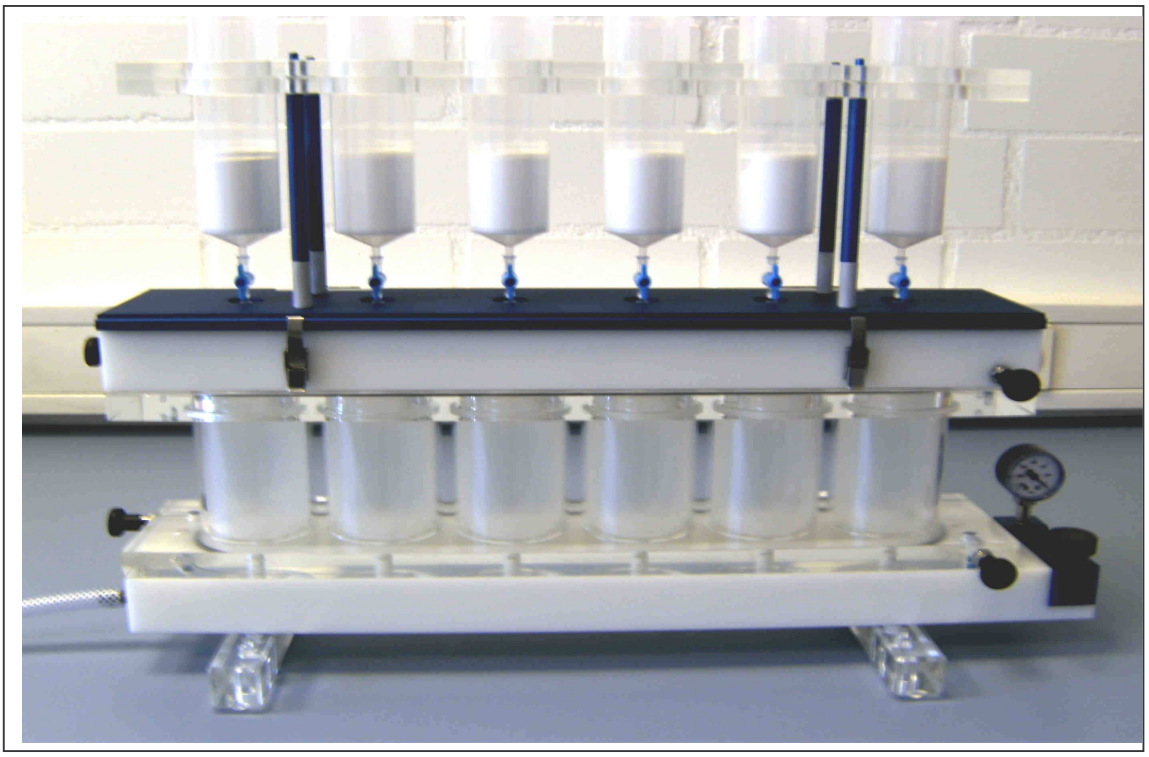


Figure 16 QIAvac HiSpeed LS ready for elution of QIAGEN HiSpeed Plasmid Giga column

Protocol: Processing QIAGEN concentrator on the QIAvac HiSpeed LS

Procedure for Plasmid DNA concentration with QIAconcentrator Mega/Giga

1. After elution of plasmid DNA open the snap-on connection between component 1 and component 3. Place component 3 with the still connected component 2 on the parking position. Open the snap-on connection between component 3 and component 2 and replace then component 2 onto component 1. Connect both components with the snap-on connections.
2. The Eluates in the collection vessels in component 3 are now ready for isopropanol precipitation according to the protocol in the HiSpeed Plasmid Mega/Giga EF Kit Handbook
3. The HiSpeed Plasmid Mega/ Giga tips are disconnected from component 6 (blue Valve) in component 1and discarded.
4. Remove the upper holder for the HiSpeed Plasmid Mega/Giga tips from the lower one which is tightly connected.
5. Prepare the Extender unit with QIAconcentrator (see figure 7, page 12) by plugging together and connect the outlet of the QIAconcentrator with the Inlet of the connection adapter (blue valve).
6. The system is now ready for the concentration procedure according to the protocol in the HiSpeed Plasmid Mega/Giga EF Kit Handbook.

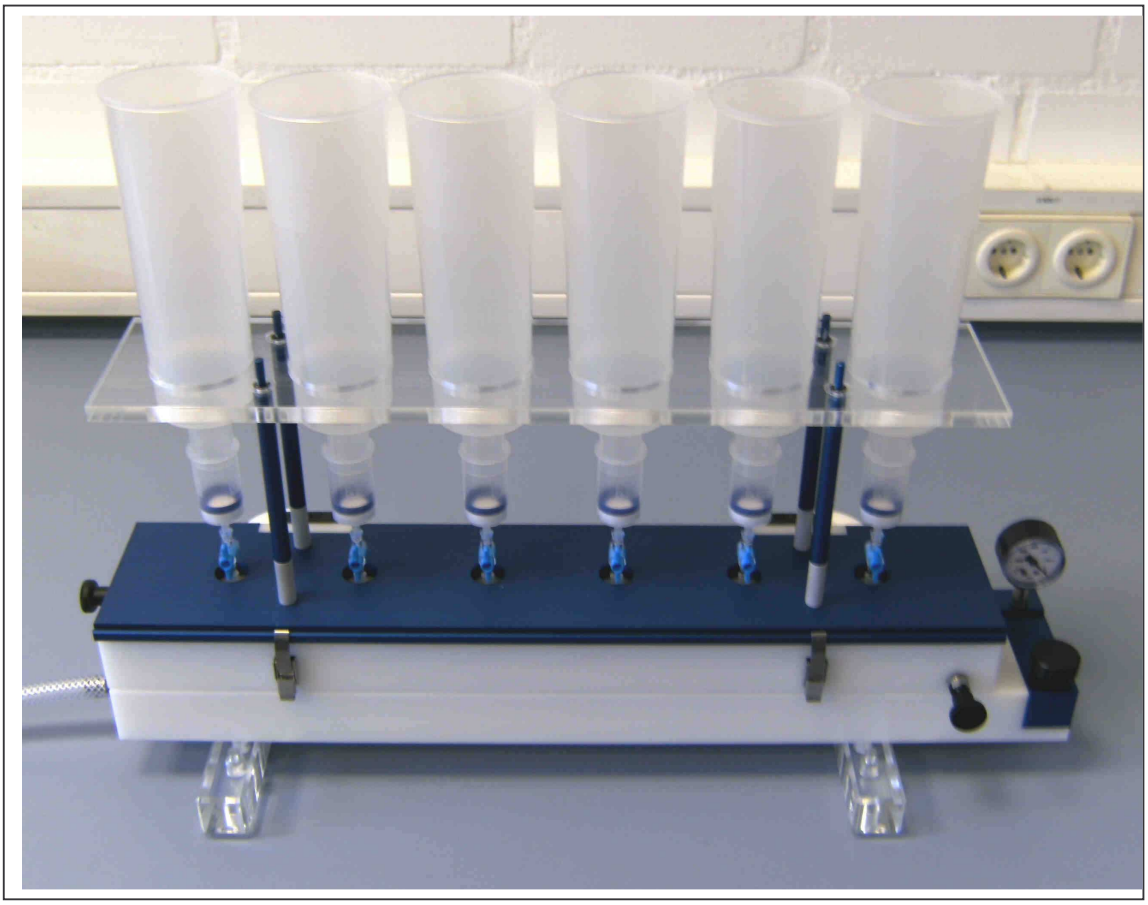


Figure 17

QIAconcentrator Giga ready for plasmid DNA concentration procedure on QIAvac HiSpeed LS

Protocol: Processing QIAGEN concentrator

Procedure for air drying and elution of Plasmid DNA from QIAGEN concentrator Mega/Giga

1. The extender are disconnected from the QIAconcentrators, removed and discarded. Disconnect the QIAconcentrator from the QIAvac HiSpeed LS and place it into a 50ml centrifugation tubes according to the protocol in the HiSpeed Mega/Giga Handbook
2. Air drying of the membrane and elution of Plasmid DNA are done by centrifugation according to the protocol in the HiSpeed Plasmid Mega/Giga EF Kit Handbook.

Appendix A

Table 1: Waste Volumes

	HiSpeed Plasmid Mega Kit		HiSpeed Plasmid Giga Kit	
	1 Prep	**5 Preps**	**1 Prep**	**5 Preps**
Equilibration	35ml	175ml	75ml	375ml
Lysate	135ml	675ml	330ml	1650ml
Wash Volumes	150ml	750ml	300ml	1500ml
Isopropanol - Precipitation wash	65ml	325ml	185ml	925ml
EtOH wash	10ml	60ml	10ml	60ml
Total	**400ml**	**2000ml**	**900ml**	**4500ml**

Table 2: Volume Capacity

To protect the vacuum source it is recommended to use a waste system of two containers. One container should be filled with liquid waste the other one is an overflow trap for vacuum source protection.

Setup A can be also used with a 10 liter container for liquid waste collection and a 5 liter container an overflow trap.

Setup	Assembly	Volume capacity
A	QIAvac HiSpeed LS + 2 x 5 liter container	5 liter
B	QIAvac HiSpeed LS + 10 liter container	8 liter
C	QIAvac HiSpeed LS + 20 liter container	18 liter

Ordering information for Waste system accessories

The following components may be used:

- 5 liter waste container (e.g., Nalgene Heavy Duty Vacuum Bottle, cat. no. 2126-5000)
- 10 liter waste container (e.g., Nalgene Heavy Duty Vacuum Carboy [PP], cat. no. 2226-0020)
- 20 liter waste container (e.g., Nalgene Heavy Duty Vacuum Carboy [PP],

cat. no. 2226-0050)

- Quick filling/venting closure (e.g., Nalgene, cat. no. 2158-0021)
- Vacuum tubing (e.g., Nalgene 180 clear plastic vacuum tubing, cat. no. 8000-0065 [50 ft. per case])
- Vacuum tubing (e.g., Nalgene 180 clear plastic vacuum tubing, cat. no. 8000-0065 [10 ft. per case])

Appendix B: Vacuum regulator

The Vacuum Regulator measures the pressure difference between the inside and outside of a vacuum system in millibars* (Figure 7). Use of the Vacuum Regulator makes it easy to monitor the pressure generated by the vacuum source, ensuring that it is sufficient for the appropriate QIAGEN purification chemistry.

Vacuum Regulator

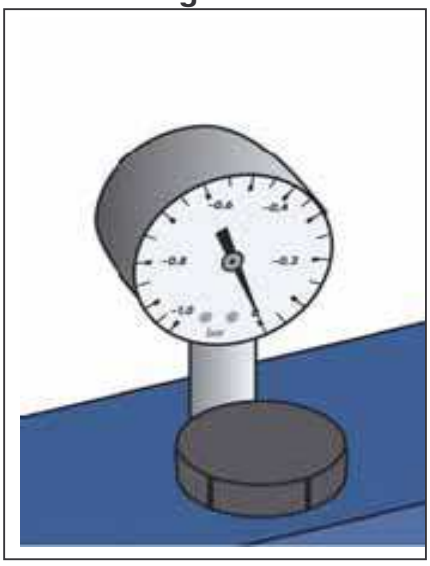


Figure Schematic diagram of the Vacuum Regulator.
* Note: the design of the scale is subject to change. The vacuum pressure may be indicated in millibars or in bars (as seen in the figure).

Table 4. Conversion Table for Pressure Units

To convert from millibars (mbar) to: Multiply by	
Millimeters of mercury (mm Hg)	0.75
Kilopascals (kPa)	0.1
Inches of mercury (inch Hg)	0.0295
Torrs (Torr)	0.75
Atmospheres (atm)	0.000987
Pounds per square inch (psi)	0.0145

Apendix C: Cleaning and Decontaminating the QIAvac HiSpeed LS

The QIAvac HiSpeed LS should be regularly cleaned to maintain optimum performance. The QIAvac HiSpeed LS must also be decontaminated before removal from the laboratory.

- When working with chemicals always wear a suitable lab coat, disposable gloves and protective goggles. For more information, consult the appropriate material safety data sheets (MSDSs), available from the product supplier.
- Do not use cleaning materials that contain abrasives
- The acrylic glass parts of the QIAvac HiSpeed LS are not resistant to ethanol, methanol or other organic solvents. Do not bring solvents into contact with the acrylic glass parts. If solvents are spilled on the unit, rinse thoroughly with distilled water. Do not incubate acrylic glass components in alcohol containg reagents. The acrylic glass parts of the QIAvac HiSpeed LS should be cleaned with water or laboratory detergent after use. Ethanol should not be used
- The parts which are built out of the white plastic should also be cleaned with standard laboratory detergents. A disinfection of the base unit where the liquid waste is collected and removed should be done with standard laboratory disinfectant. It has to be avoiding that acrylic glass parts of the QIAvac HiSpeed LS get in contact with disinfectant.

Appendix D: Handling and Maintenance of the QIAvac HiSpeed LS 24

The following guidelines should be followed when working with the QIAvac HiSpeed LS.

 Always place the QIAvac HiSpeed LS on a secure bench top or work area. If dropped, the QIAvac HiSpeed manifold may crack.

 Always store the QIAvac HiSpeed LS clean and dry. For cleaning procedures see

"Cleaning and Decontaminating the QIAvac HiSpeed", page 25).

 Always use caution and wear safety glasses when working near a vacuum manifold under pressure.

 Contact QIAGEN Technical Services or your local distributor for information concerning spare or replacement parts.

Troubleshooting

This troubleshooting guide may be helpful in solving problems that may arise.

	Comments and suggestions
Low or no vacuum	
General	No vacuum may be causes by a number of factors. Make sure that the vacuum source is running. If yes check that the vacuum regulator is closed.
	Check that all components are correctly assembled
	Check that all tubings to the waste container are correctly connected and the lids of waste container are tighten
	Check that all gaskets of the QIAvac HiSpeed LS are present and in the right position
	Check that all position of component 2 are either closed by blind plugs or connection adapter are assembled
	Check that all VacValves are closed.

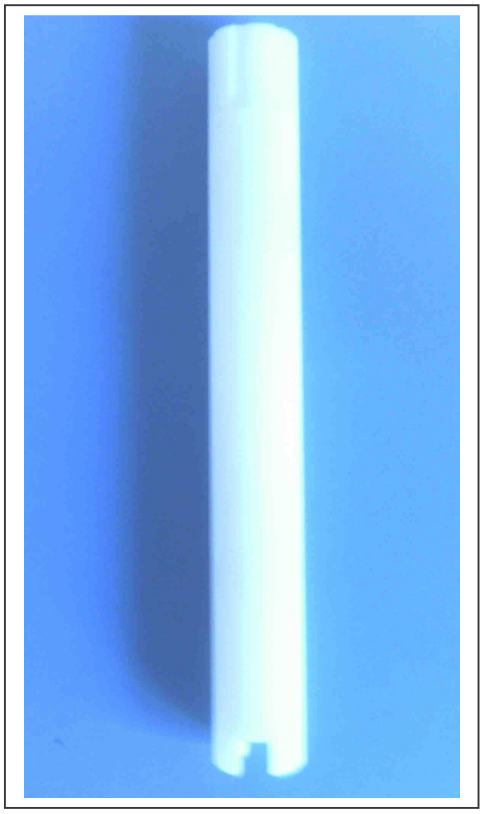
Spare Parts

The following spare parts are available; please also see the figures below

Please inquire the spare parts with the corresponding Material number.

Material number	Description of spare part
1057001	Screwdriver for valves
1057003	Filter dummy
1057004	Bolt Blue
1057005	Locking Pin, steel
1057007	Cramp
1057008	Gasket kit
1057009	Gauge
1057010	Tube clip 6 mm

Screwdriver for valves
(Mat. No.: 1057001)



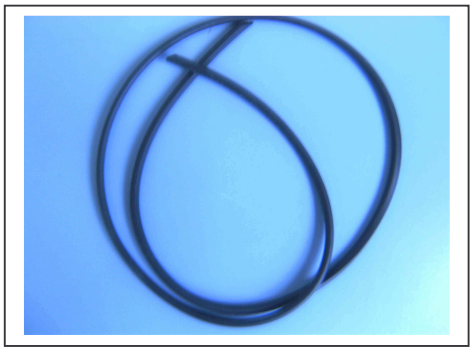
Filter dummy (Mat. No.:1057003)

Bolt Blue (Mat. No.: 1057004)

Locking Pin, steel (Mat. No.: 1057005)

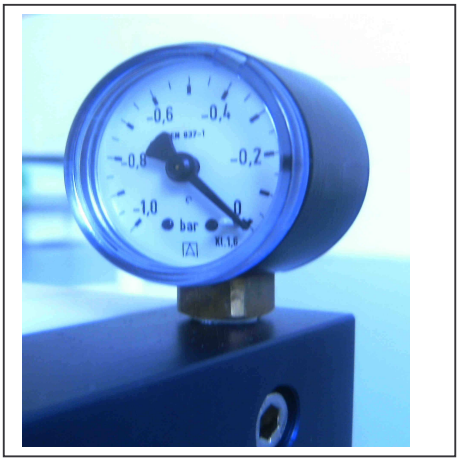
Cramp (Mat. No.:1057007)

Gasket Kit (Mat.No.: 1057008)



For replacing a damaged gasket a suitable length has to be cut off from the Gasket Kit and placed into the corresponding groove.

Gauge (Mat.No.: 1057009)



Tupe Dip 6mm (Mat. No. 1057010)

Ordering Information

Product	Contents	Cat. no.
QIAvac HiSpeed LS	Vacuum device for parallel processing of up to 6 HiSpeed Mega or Giga Columns	Inquire
Accessories		
HiSpeed Plasmid Mega EF Kit (5)	For 5 preps: 5 HiSpeed Mega Columns, 5 QIAconcentrators, Collection vessels, Extender , QIAfilter Mega Giga Cartridges Buffers, connection adapter	Inquire
HiSpeed Plasmid Giga EF Kit (5)	For 5 preps: 5 HiSpeed Giga Columns, 5 QIAconcentrators, Collection vessels, Extender , QIAfilter Mega Giga Cartridges Buffers, connection adapter	Inquire
Vacuum Pump (230V/50Hz)	Universal vacuum pump	84020
Spare Parts for QIAvac HiSpeed LS	See page 27 to 29	Inquire
Related products		
QIAGEN HiSpeed Plasmid Kits — For ultrafast purification of up to 750µg transfection- grade plasmid or cosmid DNA		
QIAGEN HiSpeed Plasmid Midi Kit (25)*	For 25 plasmid midipreps	12643
QIAGEN HiSpeed Plasmid Maxi Kit (10)*	For 10 plasmid maxipreps	12662
QIAGEN HiSpeed Plasmid Maxi Kit (25)	For 25 plasmid maxipreps	12663

www.qiagen.com

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Austria ■ Orders 0800/28-10-10 ■ Fax 0800/28-10-19 ■ Technical 0800/28-10-11

Belgium ■ Orders 0800-79612 ■ Fax 0800-79611 ■ Technical 0800-79556

Canada ■ Orders 800-572-9613 ■ Fax 800-713-5951 ■ Technical 800-DNA-PREP (800-362-7737)

China ■ Orders 021-51345678 ■ Fax 021-51342500 ■ Technical 021-51345678

Denmark ■ Orders 80-885945 ■ Fax 80-885944 ■ Technical 80-885942

Finland ■ Orders 0800-914416 ■ Fax 0800-914415 ■ Technical 0800-914413

France ■ Orders 01-60-920-926 ■ Fax 01-60-920-925 ■ Technical 01-60-920-930 ■ Offers 01-60-920-928

Germany ■ Orders 02103-29-12000 ■ Fax 02103-29-22000 ■ Technical 02103-29-12400

Ireland ■ Orders 1800-555-049 ■ Fax 1800-555-048 ■ Technical 1800-555-061

Italy ■ Orders 02-33430411 ■ Fax 02-33430426 ■ Technical 800-787980

Japan ■ Telephone 03-5547-0811 ■ Fax 03-5547-0818 ■ Technical 03-5547-0811

Luxembourg ■ Orders 8002-2076 ■ Fax 8002-2073 ■ Technical 8002-2067

The Netherlands ■ Orders 0800-0229592 ■ Fax 0800-0229593 ■ Technical 0800-0229602

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