A&A BIOTECHNOLOGY
innovating life science

Manual

MagnifiQ™ 96 Pathogen instant kit

Kit for automated, magnetic isolation of DNA and RNA of pathogens in the 96 samples per plate format. Contains ready-to-use, reagent-filled plates and all necessary consumables.

REF	size	compatible devices *
607A-96V-960	960 isolations	Auto-Pure 96

*** Compatible devices**
The kit has been tested with ThermoFisher Scientific KingFisher Flex and Allsheng Auto Pure 96 devices. This does not preclude it from working with other devices. If your device is not listed, please contact us at info@aabiot.com.

For in vitro diagnostics use.

CE IVD

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Guarantee
A&A Biotechnology provides a guarantee on this product.
The company does not guarantee the correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

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Advantages

- Requires only a few minutes of manual work while adding the samples. The rest of the procedure is carried out in an automatic extraction device.
- It does not require the initial preparation of buffers. Just add prepared samples to the plate and after about 30 min you get extracted material.
- Universal extraction of both DNA and RNA.

Sample types

sample type	sample size
Animal tissue	up to 20 mg
Blood (fresh or frozen, plasma, serum), body fluids	up to 200 µl
Swab	1 pc

Specification

protocol time	~ 30 min
elution volume	100 µl
elution solution	Tris buffer (10 mM, pH 8.5)
binding capacity	30 µg DNA/RNA
downstream applications	qPCR, RT-qPCR, sequencing

Description

MagnifiQ™ 96 Pathogen instant kit is designed for RNA and DNA isolation from viruses and Gram(-) bacteria.
The isolated material is perfect for further analyzes and tests by qPCR and RT-PCR methods and for sequencing.

The **MagnifiQ™** product series is based on the automated isolation of nucleic acids with use of magnetic beads.
This method significantly shortens working time and reduces risk of mistakes in comparison to manual methods.

Contents

960 isolations

component	quantity	cat #	storage
CP - comb plate	1 pc	K-P96V22C	15–25 °C
SP - sample plate	10 pcs	K-P96V22SA	15–25 °C
WP 1 - wash 1 plate	10 pcs	K-P96V22W1A	15–25 °C
WP 2-3 - wash 2-3 plate	20 pcs	K-P96V22W23A	15–25 °C
BP - beads plate	10 pcs	K-P96V22BA	15–25 °C
EP - elution plate	10 pcs	K-P96V05EA	15–25 °C
Proteinase K	22 ml	K-PRK-22	2–8 °C*
LTE 2X buffer	210 ml	K-LTE2X-210	15–25 °C
tip comb 96	5 x 2 pcs	K-C96V-2	15–25 °C
protective film	10 pcs	K-MQF-10	15–25 °C

* Proteinase K can be stored at 15-25 °C for up to 12 months.

Additional equipment and reagents

Necessary

- pipette
- pipette tips
- 96 deep-well plates 2.2 ml (sample lysis)

Optional

- sterile water, Tris buffer, PBS buffer
- vortex
- 8-channel pipette
- 1.5 ml Eppendorf tubes (sample lysis)

Important notes

The following material preparation protocols apply to the procedure carried out in a 96 deep-well plate. If the material preparation is to be carried out in 1.5 ml Eppendorf tubes see the Additional Information.

Material preparation

Animal tissue

1. Homogenize up to **20 mg** of animal tissue in the PBS or Tris buffer.
2. Centrifuge the sample for **1 min** at **500 RPM**.
3. Transfer **200 µl** of the supernatant to the well of the 96 deep-well plate (not included).
4. Add **200 µl** of **LTE 2X** buffer and **20 µl** of **Proteinase K.**
   Mix the contents of the wells by pipetting.
5. Follow point 1. of the Extraction Protocol.

Blood (fresh or frozen, plasma, serum), body fluids

1. Transfer **200 µl** the sample to the 96 deep-well plate (not included).
2. Add **200 µl** of **LTE 2X** and **20 µl** of **Proteinase K** to the wells.
   Mix the contents of the wells by pipetting.
3. Seal the plate with a protective film and incubate for **20 min** at **55 °C** with mixing **1600 RPM.**
4. Centrifuge for **1 min** at **1000 x g**.
5. **Attention.** In the Extraction Protocol, use the supernatant as the sample.

   Follow point 1. of the Extraction Protocol.

Swabs with transport medium

No additional material preparation is required.

1. Follow point 1. of the Extraction Protocol.

Dry swabs

1. Break or cut off part of the swab with the collected sample and place it in the wells of 96 deep-well plate (not included).
2. Add **500 µl** of sterile water, **Tris** buffer or PBS buffer.

   Note. Part of the swab with the sample should be completely immersed in the buffer.
3. Leave at room temperature for **10 min**.
4. Mix the contents of the wells by pipetting.
5. Transfer **200 µl** of supernatant to the new 96 deep-well plate (not included).
6. Add **200 µl** of **LTE 2X** buffer and **20 µl** of **Proteinase K.**
7. Follow point 1. of the Extraction Protocol.

Protocol

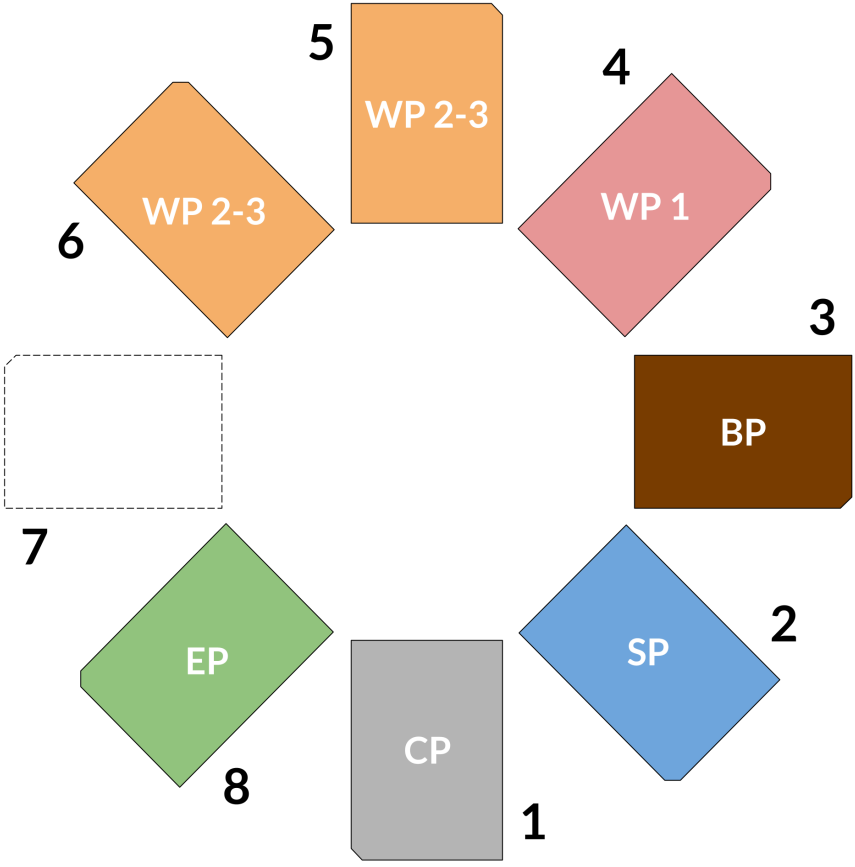
Protocol files

device	protocol name	protocol file	installation
Auto-Pure 96	MQ-UNI-96	aabiot.com/protocols/magnifiq/96/MQ-UNI-96.txt	1. Create folder "items" on a USB drive and copy the protocol file to it. 2. Insert the USB drive into a USB slot in the device. 3. On a device screen, go to Settings > Im.&Export > Import. 4. Select the protocol and tap "Import."

Extraction protocol

Before the isolation procedure, all plates should be centrifuged for **1 min** at **1000 RPM**. Centrifuge to remove remaining solution from the top of the protective foil.

1. Remove the foil from the **SP** plate.
2. Add **400 µl** of samples to the wells of the **SP** plate.
3. Place **96 tip comb** into the **CP** plate.
4. Remove the adhesive foil from the rest of the plates. Place the plates on the working table of the extraction device according to the scheme:



6. Run the protocol on your device.

7. After the program is over, remove the **EP** plate from the extraction device and seal it with **protective film**.

   Note. For longer storage of extracted material, transfer it from the plate to appropriate tubes and store at 4 °C for DNA or -70 °C for RNA.

8. Discard remaining plates except the **CP** plate, which can be reused.

Additional information

Preparation of material in 1.5 ml Eppendorf tubes

Lysis of the material in 1.5 ml Eppendorf tubes should be carried out according to the respective procedure for 96 deep-well plate in the Material Preparation section. The following change should be made:

- Incubation parameters
  Lower the incubation temperature by **5 °C** and shorten the time by **10 min**.

Troubleshooting

Problem	Possible cause	Suggested solution
No nucleic acid at all.	Material was not processed.	Please ensure the sample lysate was added to the sample plate. Inspect the volumes of the respective sample plate wells. In case of uncertainty repeat the procedure with new sample material.
	Automated extraction protocol was not started.	Please check the position of the beads in the plate. If the beads were not transferred from their original position START the protocol for nucleic acid extraction.
Low RNA yield.	Initial sample material was partially degraded.	Please repeat the isolation with the same and different sample type to double check the sample quality.
	Elution well was contaminated with exogenous RNAse.	Please repeat the isolation with the fresh portion of elution reagent to exclude the RNAse contamination.
Low DNA yield	Extraction of DNA during sample lysis was not sufficient.	To obtain higher yields of DNA, the incubation time in lysis buffer can be prolonged (usually up to 20 min).
		For sample that the lysis is supported by Proteinase K digestion please ensure the Proteinase K was added or increase the volume of enzyme up to 40 µl per sample.
	Sample contains too much RNA.	Add 10–20 µl RNase A solution to the lysis buffer before heat incubation. If this is not successful, add the enzyme to the cleared lysate and incubate for 30 min at 37 °C.
	Suboptimal elution.	The DNA can be either eluted in higher or lower volumes (40- 100 µL) or by prolonging the elution step up. Check your automated extractor programme and if needed decrease the elution volume in the elution plate/ row. Note that it is advantageous to perform the elution pre-heated to 65-70 °C. Check your automated extractor programme.
Degraded DNA	Sample was contaminated with DNAse.	Check working area and pipettes for DNAse content. Use cleaning product to remove any enzymatic activity contamination.
	Sample dependent problem.	Highly processed samples may be responsible for impossibility to extract high molecular weight DNA.
Low DNA / RNA quality	Sample contains DNA-degrading contaminants (e.g., phenolic compounds, metabolites)	Investigate if repeating the wash 1 or EtOH 80% wash would improve the quality of eluate.

Safety information

DANGER

Proteinase K

H315 Causes skin irritation.
H319 Causes serious eye irritation.
H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H335 May cause respiratory irritation.
P261 Avoid breathing dust.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P342+P311 If experiencing respiratory symptoms call a Poison Center or doctor/physician.

WARNING

LTE 2X

H302 Harmful if swallowed.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

DANGER

WP 1 plate, WP 2-3 plate

H225 Highly flammable liquid and vapor.
H302 Harmful if swallowed.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
H336 May cause drowsiness or dizziness.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P261 Avoid breathing dust/fume/gas/mist/vapours/ spray.
P280 Wear protective gloves/protective clothing/eye protection/face protection/hearing protection.
P301+P312+P330 If swallowed: Call a poison center/doctor/ if you feel unwell.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P337+P313 If eye irritation persists: Get medical advice/ attention.

DANGER

SP plate

H302+H312+H332 Harmful if swallowed, in contact with skin or if inhaled.
H314 Causes severe skin burns and eye damage.
H412 Harmful to aquatic life with long lasting effects.
P273 Avoid release to the environment.
P280 Wear protective gloves/protective clothing/eye protection/face protection/hearing protection.
P301+P312+P330 If swallowed: Call a poison center/doctor/ if you feel unwell.
P303+P361+P353 If on skin (or hair): Take off immediately all contaminated clothing. Rinse skin with water or shower.
P304+P340 If inhaled: Remove person to fresh air and keep comfortable for breathing.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

Explanation of symbols used

symbol	symbol meaning	symbol	symbol meaning
IVD	Indicates the *In vitro* diagnostics medical device	REF	Indicates the manufacturer's catalogue number
	Indicates the medical device manufacturer		Indicates the need for the user to consult the instructions for use
LOT	Indicates the manufacturer's batch code or lot can be identified		Indicates the need for the user to consult the instructions for use for important information such as warnings and cautions
	Indicates the date after which the medical device is not to be used		Indicates the temperature limits to which the medial device can be safely exposed

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