A&A BIOTECHNOLOGY
innovating life science

*Manual*

# MagnifiQ™ 16 Genomic DNA instant kit

Kit for automated, magnetic isolation of genomic DNA in the 16 samples per plate format. Contains ready-to-use, reagent-filled plates and all necessary consumables.

| catalog # | size | compatible devices * |
|---|---|---|
| 604A-16U-64 | 64 isolations | Auto-Pure 32A |
| 604A-16V-64 | 64 isolations | Auto-Pure Mini<br>Auto-Pure S32 |
| 604A-16U-256 | 256 isolations | Auto-Pure 32A |
| 604A-16V-256 | 256 isolations | Auto-Pure Mini<br>Auto-Pure S32 |

*** Compatible devices**
The kit has been tested with specific Allsheng brand isolation 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 research use only.

**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

# Table of Contents

# Advantages

- MagnifiQ™ 16 Genomic DNA instant kit does not require initial preparation of buffers. Just add samples to the plate and get extracted material within approximately half an hour.
- It enables isolation of different samples with universal kit and automated extraction programme.

# Sample type

| | sample size |
|---|---|
| Bacteria G-, G+ (cultures) | up to 2 x $10^8$ |
| Yeast (cultures) | up to 1 ml |
| Cell cultures | up to 1 x $10^6$ |
| Blood fresh or frozen, serum, plasma | up to 200 µl |
| Animal tissue | up to 20 mg |
| Swab | 1 pc |
| Feces, environmental samples (soil, activated sediment, compost) | 20 - 50 mg |
| Feces, environmental samples (soil, activated sediment, compost) stored in conservation solution | 250 - 500 µl |

# Specification

| | |
|---|---|
| **protocol time** | ~ 30 min. |
| **elution volume** | 100 µl [1] |
| **elution solution** | Tris buffer |
| **binding capacity** | 30 µg DNA |
| **downstream applications** | qPCR, RT-qPCR, sequencing |

[1] The elution volume prepared on the plate is 100 µl. To obtain a smaller elution volume, subtract the appropriate amount of elution solution from the wells of columns 6 and 12 on the XP-GD plate. Attention! Do not reduce the elution volume below 50 µl. To obtain a larger elution volume, add the appropriate amount of elution solution from the wells of columns 6 and 12 on the XP-GD plate. Attention! Do not increase the elution volume above 300 µl.

# Description

**MagnifiQ™ 16 Genomic DNA instant kit** is designed for DNA isolation from various types of biological materials. 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 mistake in comparison to manual methods.

# Contents

| | 604A-16U-64 | | 604A-16U-256 | | |
|---|---|---|---|---|---|
| **component** | **quantity** | **cat #** | **quantity** | **cat #** | **storage** |
| **XP-GD** - extraction plate | 4 pcs | K-P96U22XGD | 16 pcs | K-P96U22XGD | 15–25 °C |
| **Proteinase K** | 2 x 1.5 ml | K-PRK-15A | 12 ml | K-PRK-12 | 15–25 °C |
| **Tris** buffer | 30 ml | K-TRIS-30 | 115 ml | K-TRIS-115 | 2–8 °C* |
| **LTE 2X** buffer | 15 ml | K-LTE2X-15 | 55 ml | K-LTE2X-55 | 15–25 °C |
| **LSDE** buffer | 35 ml | K-LSDE-35 | 140 ml | K-LSDE-140 | 15–25 °C |
| **tip comb 8** | 8 pcs | K-C8U-8 | 32 pcs | K-C8U-32 | 15–25 °C |
| **protective film** | 4 pcs | K-MQF-4 | 16 pcs | K-MQF-16 | 15–25 °C |

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

| | 604A-16V-64 | | 604A-16V-256 | | |
|---|---|---|---|---|---|
| **component** | **quantity** | **cat #** | **quantity** | **cat #** | **storage** |
| **XP-GD** - extraction plate | 4 pcs | K-P96V22XGD | 16 pcs | K-P96V22XGD | 15–25 °C |
| **Proteinase K** | 2 x 1.5 ml | K-PRK-15A | 12 ml | K-PRK-12 | 2–8 °C* |
| **Tris** buffer | 30 ml | K-TRIS-30 | 115 ml | K-TRIS-115 | 15–25 °C |
| **LTE 2X** buffer | 15 ml | K-LTE2X-15 | 55 ml | K-LTE2X-55 | 15–25 °C |
| **LSDE** buffer | 35 ml | K-LSDE-35 | 140 ml | K-LSDE-140 | 15–25 °C |
| **tip comb 8** | 8 pcs | K-C8U-8 | 32 pcs | K-C8U-32 | 15–25 °C |
| **protective film** | 4 pcs | K-MQF-4 | 16 pcs | K-MQF-16 | 15–25 °C |

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

# Additional equipment and reagents

## Necessary

- 1.5 ml Eppendorf tubes (sample lysis)
- automated pipette
- pipette tips

## Optional

- RNAse (10 µl per sample), cat # 1006-10
- 2.2 ml plates (sample lysis)
- swing-out rotor centrifuge for 96-well plates
- protective film (lysis in a 96 deep-well plate)
- thermoblock
- vortex

# Important notes

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

# Material preparation

## Bacteria G-, G+ (cultures)

### Additional reagents you will need:

**Bacteria lysis kit** (cat. # 604BK-50, 604BK-100)

- **BacBreaker** bacteria lysis enzyme mix (20 µl per sample)
- **BS** suspension buffer (200 µl per sample)

**Option:**

- **Lysostaphin** (5 µl per sample), cat # 1007-3; For *Staphylococcus aureus* we recommend using lysostaphin.

1. Transfer the bacterial culture sample containing **2 x $10^8$** bacteria to the 1.5 ml Eppendorf tube (not included).
   Centrifuge for **3 min** at **10 000 RPM**. Discard the supernatant.
2. Suspend the bacterial pellet in **200 µl** of **BS** buffer.
3. Add **20 µl** of **BacBreaker** enzyme mix.

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10).

   **Attention.** For lysis of *Staphylococcus* bacteria, add **5 µl** of **lysostaphin**.
4. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C**.

   **Attention.** For lysis of *Staphylococcus* bacteria with lysostaphin, mix and incubate for 10 min at 37 °C.
5. Add **200 µl** of **LTE 2X** and **20 µl** of **Proteinase K**.

6. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C** with shaking.

   **Note**. If an automatic continuous shaking is not available, mix the samples by inverting the tubes a few times.

7. Centrifuge the sample for **2 min** at **10 000 RPM**.

8. **Attention.** In the isolation protocol, use the supernatant as the sample.

   Follow point 1. of the protocol.

## Yeast (cultures)

### Additional reagents you will need:

**Yeast lysis kit** (cat. # 604YK-50, 604YK-100)

- **Lyticase** (10 µl per sample)
- **DTT RTU** (10 µl 1M solution per sample)
- **BS** suspension buffer (200 µl per sample)

Prepare **1M DTT** solution. Add 1 ml of sterile water (not included) to a vial containing DTT powder to obtain 1M DTT solution. Mix or vortex until complete dissolution of DTT powder. Store a clear solution at -20 °C.

1. Transfer **1 ml** of yeast culture to 1.5 ml Eppendorf tube (not included).
   Centrifuge for **3 min** at **10 000 RPM**. Discard the supernatant.

2. Suspend the yeast pellet in **200 µl** of **BS** buffer.

3. Add **10 µl** of **lyticase** and **10 µl 1M DTT.**

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10).

4. Vortex the sample for **10 s** and incubate for **15 min** at **37 °C**.

5. Add **200 µl** of **LTE 2X** and **20 µl** of **Proteinase K**.

6. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C** with shaking.
   **Note**. If an automatic continuous shaking is not available, mix the samples by inverting the tubes a few times.

7. Centrifuge the sample for **2 min** at **10 000 RPM**.

8. **Attention.** In the isolation protocol, use the supernatant as the sample.
   Follow point 1. of the protocol.

## Cell cultures

1. Transfer the cell culture sample containing **1 x $10^6$** cells to 1.5 ml Eppendorf tube (not included). Centrifuge for **3 min** at **10 000 RPM**. Discard the supernatant.
2. Suspend the cell pellet in **200 µl** of **Tris** buffer.
3. Add **200 µl** of **LTE 2X** and **20 µl** of **Proteinase K**.

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10).
4. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C** with shaking.

   **Note**. If an automatic continuous shaking is not available, mix the samples by inverting the tubes a few times.
5. Follow point 1. of the protocol.

## Blood: fresh or frozen, plasma, serum

1. Transfer **200 µl** the sample to 1.5 ml Eppendorf tube (not included).
2. Add **200 µl** of **LTE 2X** and **20 µl** of **Proteinase K**.
3. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C** with shaking.

   **Note**. If automatic shaking is not available, mix the samples by inverting the tubes a few times.
4. Centrifuge for **20 s** at **10 000 RPM.**

   **Note.** Centrifuge to remove remaining material from lids of the tubes and deposit non-lysed material at the bottom of the tube.
5. **Attention.** In the isolation protocol, use the supernatant as the sample.

   Follow point 1. of the protocol.

## Animal tissue

1. Transfer **up to 20 mg** of fragmented animal tissue to 1.5 ml Eppendorf tube (not included).

   **Note.** The tissue should be fragmented by cutting into pieces or homogenization.

2. Add **400 µl** of **LSDE** buffer and **40 µl** of **Proteinase K.**

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10).

3. Vortex the sample for **10 s** and incubate until complete lysis at **50 °C** with shaking.

   **Note**. If an automatic continuous shaking is not available, mix the samples by inverting the tubes a few times.

   **Information.** The lysis step can last from 1 to 12 hours. For maximum efficiency, lysis should be carried out until the tissue is completely dissolved in the lysis solution.

4. Centrifuge the sample for **2 min** at **10 000 RPM**.

5. **Attention.** In the isolation protocol, use the supernatant as the sample.

   Follow point 1. of the protocol.

## Swabs with transport medium

No additional material preparation is required.

## Dry swabs

1. Break or cut off part of the swab with the collected sample and place it in a 1.5 ml Eppendorf tube (not included).

   **Note.** The portion of the swab with the collected sample should fit completely into the tube.

2. Add **500 µl** of **LSDE** buffer and **20 µl** of **Proteinase K.**

   **Note.** Part of the swab with the sample should be completely immersed in the buffer.

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10).

3. Vortex the sample for **10 s** and incubate for **10 min** at **50 °C** with shaking.

   **Note**. If an automatic continuous shaking is not available, mix the samples by inverting the tubes a few times.

4. **Attention.** For the isolation process, take the entire volume of the sample, but not more than 400 µl.

   Follow point 1. of the protocol.

# Feces, environmental samples (soil, activated sediment, compost)

## Additional reagents you will need:

**Microbiome lysis kit (**cat. # 604MK-50, 604MK-100**)**

- **Bead-beat tubes** (2 ml screwed Bead-beat tubes containing beads mix)
- **L3P** precipitation solution (100 µl per sample)
- **LSDE** buffer (additional 500 µl per sample)
- **antifoam** (10 µl per sample)

Before starting the process, mix the **LSDE** buffer with **antifoam**. Prepare the **LSDE-antifoam** mix by combining **1 ml** of **LSDE** buffer with **10 µl** of **antifoam** per sample. Prepare a volume sufficient for the number of isolated samples with a 10% excess. Mix well before use.

1. Transfer **20-50 mg** of sample to the 2 ml screwed Bead-beat tube containing beads mix.
   Add **1 ml** of **LSDE-antifoam** buffer.

2. **Option A:** Bead Beating. Use the suitable tube bead beater with the following programme:
   **3 x run of 20 s** at **maximum force** with **2 min** cool down rest.

   **Option B:** Place the tubes in the incubator with mixing function (e.g. Thermomixer) and vortex at **2000 RPM** for **30 min** at **room temperature**.

3. Centrifuge for **5 min** at **10 000 RPM**.

4. Transfer **500 µl** of supernatant to a new 1.5 ml Eppendorf tube (not included).

5. Add **20 µl** of **Proteinase K.**

6. Vortex the sample for **10 s** and incubate for **15 min** at **50 °C** with shaking **1400 RPM**.

   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10) and incubate for **10 min** at **37 °C** with shaking at **1400 RPM.**

7. Add **100 µl** of **L3P** precipitation solution. Close the tube and mix whole content by inverting the tube.

8. Place on ice for **3 min.**

9. Centrifuge the sample for **5 min** at **10 000 RPM**.

10. **Attention.** In the isolation protocol, use the supernatant as the sample.

    Follow point 1. of the protocol.

# Feces, environmental samples (soil, activated sediment, compost) stored in conservation solution

## Additional reagents you will need:

For samples stored in the **StoolSave™ DNA Protection kit (cat. # 006-10)**:

- **Bead-beat tubes** (2 ml screwed Bead-beat tubes containing beads mix), cat. # K-PKCM-50
- **L3P** precipitation solution (100 µl per sample), cat. # K-L3P-60

For samples stored in another preservation solution:

**Microbiome lysis kit (**cat. # 604MK-50, 604MK-100**)**

- **Bead-beat tubes** (2 ml screwed Bead-beat tubes containing beads mix)
- **L3P** precipitation solution (100 µl per sample)
- **LSDE** buffer (additional 250 µl per sample)
- **antifoam** (10 µl per sample)

1. **Samples stored in conservation solution StoolSave™ DNA Protection kit**:
   Transfer **500 µl** of sample suspended in conservation/ transportation solution to the 2 ml screwed Bead-beat tube containing zirconia beads.
   Add **500 µl** of **LSDE** buffer.

   **Samples stored in another preservation solution:**
   Before starting the process, mix the **LSDE** buffer with **antifoam**. Prepare the **LSDE-antifoam** mix by combining **750 µl** of **LSDE** buffer with **10 µl** of **antifoam** per sample. Prepare a volume sufficient for the number of isolated samples with a 10% excess. Mix well before use.

   Transfer **250 µl** of sample suspended in conservation/ transportation solution to the 2 ml screwed Bead-beat tube containing zirconia beads.
   Add **750 µl** of **LSDE-antifoam** buffer.

2. **Option A:** Bead Beating. Use the suitable tube bead beater with the following programme:
   **3 x run of 20 sec** at **maximum force** with **1 min** cool down.

   **Option B:** Place the tubes in the incubator with mixing function (e.g. Thermomixer) and vortex at **2000 RPM** for **30 min** at **room temperature**.

3. Centrifuge for **5 min** at **10 000 RPM**.

4. Transfer **500 µl** of supernatant to a new 1.5 ml Eppendorf tube (not included).

5. Add **20 µl** of **Proteinase K.**

6. Vortex the sample for **10 s** and incubate for **15 min** at **50 °C** with shaking **1400 RPM**.
   **Optional RNA removal.** Add **10 µl** of **RNAse** (cat # 1006-10) and incubate for **10 min** at **37 °C** with shaking at **1400 RPM.**

7. Add **100 µl** of **L3P** precipitation solution. Close the tube and mix whole content by inverting the tube.

8. Place on ice for **3 min.**

9. Centrifuge the sample for **5 min** at **10 000 RPM**.

10. **Attention.** In the isolation protocol, use the supernatant as the sample.

    Follow point 1. of the protocol.

# Protocol

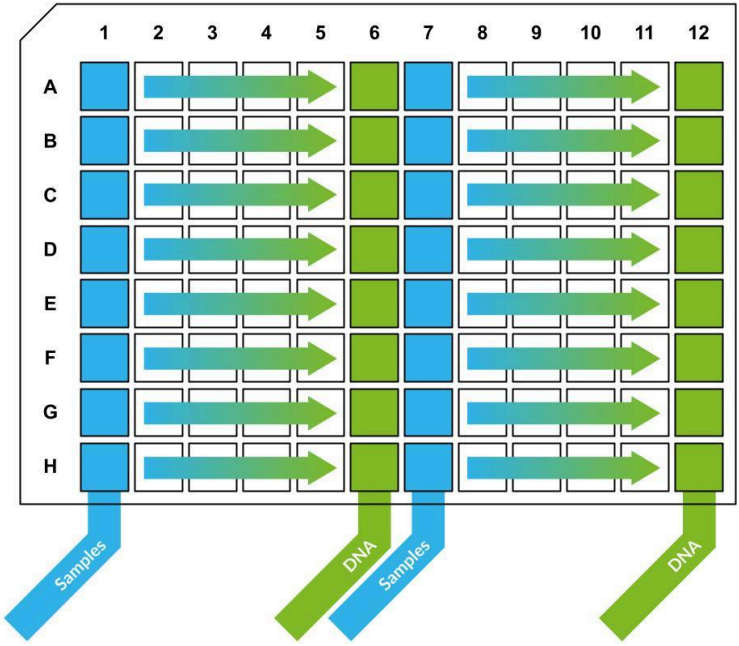
## Protocol files

| device | protocol name | protocol file | installation |
|---|---|---|---|
| Auto-Pure Mini | MQ-UND-MI | aabiot.com/protocols/magnifiq/MI/MQ-UND-MI.txt | 1. Create folder "items" on a USB drive and copy the protocol file to it.<br>2. Insert the USB drive into a USB slot in the device.<br>3. On a device screen, go to Settings > System > Transfer >Import.<br>4. Select the protocol and tap "Import". |
| Auto-Pure Mini (QR code) | MQ-UND-MI |  | 1. On a device screen, go to Run > ☰ > ⌜⌝<br>2. Scan the QR code with the device's scanner. |
| Auto-Pure 32A | MQ-UND-32A | aabiot.com/protocols/magnifiq/32A/MQ-UND-32A.txt | 1. Create folder "items" on a USB drive and copy the protocol file to it.<br>2. Insert the USB drive into a USB slot in the device.<br>3. On a device screen, go to Settings > Im.&Export > Import.<br>4. Select the protocol and tap "Import." |
| Auto-Pure S32 | MQ_UND_S32 | aabiot.com/protocols/magnifiq/S32/MQ_UND_S32.txt | 1. Create folder "im_export_protocols" on a USB drive and copy the protocol file to it.<br>2. Insert the USB drive into a USB slot in the device.<br>3. On a device screen, go to Protocols >Import.<br>4. Select the protocol and tap "Import". |

# Extraction protocol

1. Carefully centrifuge the **XP-GD** plate for **1 min** at **2000 RPM.**
2. Gently remove the foil from the **XP-GD** plate.
3. Add **400 µl** of samples to the wells in columns **1** and **7** of the **XP-GD** plate.
4. Place one or two **XP-GD** plates in the extraction device.
5. Place the appropriate number of t**ip combs 8** in the extraction device.
6. Run the protocol on your device.
7. After the program is over, remove the combs and then remove **XP-GD** plate from the extraction device and seal it with **protective film**. The extracted DNA is located in columns **6** and **12**.

**Note.** For longer storage of extracted material, transfer it from the plate to appropriate tubes and store at 4 °C.



# Additional information

## Preparation of material in a 96 deep-well plate

Not applicable: feces, environmental samples and FFPE samples.

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

- Incubation parameters
  Increase the incubation temperature by **5 °C** and extend the time by **10 min** with a minimum of **1000 RPM** continuous shaking speed. Secure tight sealing to prevent any well-to-well cross contamination!
- Centrifugation parameters
  Centrifuge the plate for **5 min** at **1 000 x g**.

# Safety information

DANGER

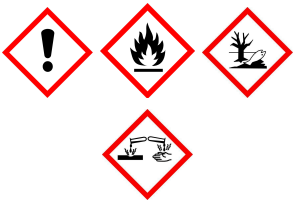
### Proteinase K

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

### XP-GD - extraction plate

H225 Highly flammable liquid and vapor.
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.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
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.

A&A BIOTECHNOLOGY

innovating life science

A&A Biotechnology, ul. Strzelca 40, 80-299 Gdańsk, Poland
phone +48 883 323 761, +48 600 776 268
info@aabiot.com, www.aabiot.com

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