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Technical Note

MagSi-NA Pathogens and PurePrep 32 Nucleic Acid Purification System | OneStep Lysis/Binding protocol

Product Description

MagSi-NA Pathogens allows fast and cost-effective extraction of total nucleic acids from various samples like serum/plasma or swab washes. This total nucleic acid purification kit is optimized to extract pathogen DNA and RNA from samples with the highest purity and delivering nucleic acids which is suitable for qPCR based analysis. The kit includes ready-to-use buffers, Proteinase K, Poly-A-RNA and magnetic particles. The kit can be easily automated on the PurePrep 32 Nucleic Acid Purification System with the suitable consumables.

The PurePrep 32 instrument can process up to 32 samples in a single run. It uses movable magnetic rods that collect and transfer magnetic particles across microplates eliminating the need for multiple pipette tips. Carefully designed rod covers prevent cross-contamination and allow for reproducible and efficient sample mixing and magnetic particle resuspension.

Protocol information

The current technical note describes a convenient combined one step lysis and binding procedure which reduces the manual hands on time and avoids the user invention after the lysis step.

First, 211 µL of the lysis working solution per sample (premix consisting of: 200 µL Lysis Buffer PA1, 10 µL Proteinase K and 1 µL Poly-A-RNA) is added to the pre-dispensed sample into the processing plate of the PurePrep 32 instrument. Secondly, a mix consisting of 20 µL MagSi-PA VII magnetic beads and 400 µL of Binding Buffer U1 is added into the processing plate. Following the combined lysis and binding step, three washing steps of the magnetic beads are performed. Finally the purified DNA/RNA is eluted from the magnetic beads and can be used directly for down-stream qPCR analysis. The MagSi-NA Pathogens magnetic beads are optimized for extremely fast separation times even from sample lysates with a high viscosity. The purification time per 32 samples is approximately 25 minutes.

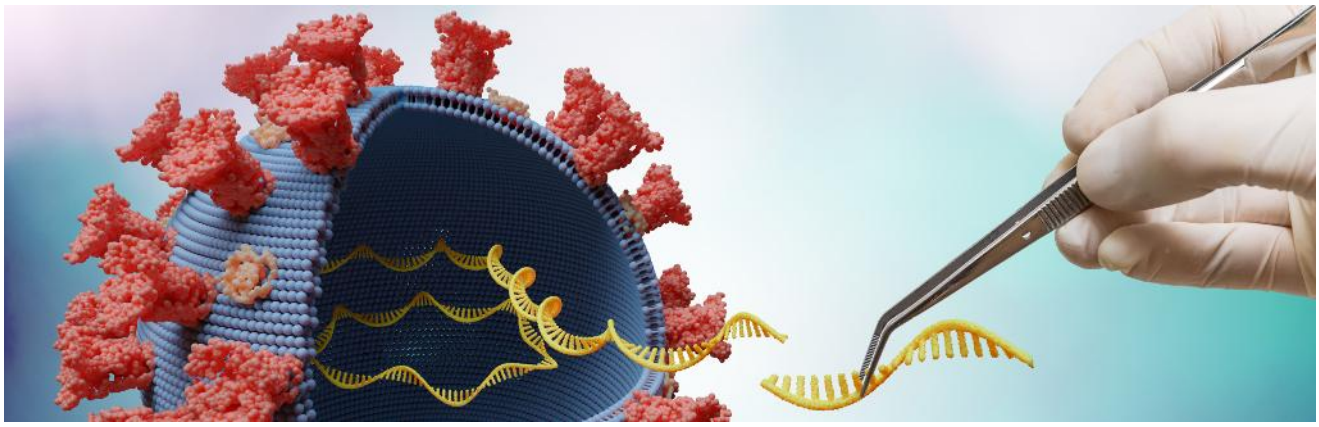

Table 1: Required reagents and equipment

Product	Art. No.	Required number per run
MagSi-NA Pathogens (96 preps)[§]	MDKT00210096	-
MagSi-NA Pathogens (10x96 preps)[§]	MDKT00210960	-
PurePrep 32 Nucleic Acid Purification Instrument	AS00002	-
PurePrep 16/32 deep-well plates	MDPL00300050	1 or 2
PurePrep 16/32 tip-combs	MDPL00310200	1 or 2

[§]bulk quantities of the kit available on request

User notes

- PurePrep 32 protocol files are available on request (email: info@magtivio.com)
- Protocol files are previously imported on the instrument but can also be easily imported to the instrument via USB drive
- For tips and advice on how to adapt the instrument protocol please email info@magtivio.com
- For further information about the MagSi-NA Pathogens kit, please refer to the Product Manual
- MagSi-NA Pathogens is optimized for total NA extraction from serum / plasma, swabs and other suitable sample materials

Importing the instrument protocol (if needed)

To save the MagSi-NA Pathogens protocol to your PurePrep 32 Nucleic Acid Purification System:

1. Plug in the USB drive
2. Switch on the instrument
3. From the main menu select "Settings"
4. Select "Im.&export", and "Import"
5. Select the file to be imported from the list or select all files
6. Select "Import", file(s) will be uploaded to the instrument now
7. Select "Back" two times to return to the main menu
8. Select "Manage Prog."
9. Select the protocol to create a shortcut

Filling the extraction plates

1. Prepare a Lysis Working Solution by adding Proteinase K and Poly-A-RNA to Lysis Buffer PA1 as following:
    - Per 200 µL Lysis Buffer PA1, add 10 µL Proteinase K (20 mg/mL), 1 µL Poly-A-RNA
    - Prepare a little more Lysis Working Solution than needed due to loss during pipetting (e.g. for 32 extractions prepare solution for 34 extractions.
2. Prepare Magnetic Beads / Binding Mix as following:
    - Per 400 µL Binding Buffer U1 add 20 µL of MagSi-PA VII beads
    - Prepare a little more Magnetic Beads / Binding Mix than needed due to loss during pipetting (e.g. for 32 extractions prepare solution for 34 extractions

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Table 2: Plate filling instructions for PurePrep 32 Nucleic Acid Purification System and MagSi-NA Pathogens protocol

Column	Component	Volume
1 and 7	Sample Lysis Working Solution MagSi-PA VII / Binding Buffer U1 mixture	200 µL 211 µL 420 µL
2 and 8	Wash Buffer I for 1st wash step	800 µL
3 and 9	Wash Buffer I for 2nd wash step	800 µL
4 and 10	Wash Buffer II for 3rd wash step	800 µL
6 and 12	Elution Buffer	100 µL

3. Continue by filling the plates as described in Table 2, and steps 4 to 6:
    - Column 1 and 7 (Sample, Lysis Working Solution, MagSi-PA VII / Binding Buffer U1)
    - Column 2,3 and 8,9 (2 columns each plate, both with Wash Buffer I)
    - Column 4 and 10 (Wash Buffer II)
    - Column 6 and 12 (Elution Buffer)
4. Add 200 µL sample to the sample columns
5. Add 211 µL Lysis Working Solution to the sample columns
6. Add 420 µL Magnetic Beads / Binding Mix to the sample columns

    Important note:

    Mix very well prior to adding to avoid sedimentation of the beads within the dispensing step. Steps 4 and 5 can be exchanged

Executing the protocol

1. Load the plates to the PurePrep 32 instrument.
2. Make sure that the plates are loaded in the correct orientation (especially when using partially filled plates). Place the A1 well of each plate to the A1 mark on the instruments deck. Make sure that the plates are inserted tightly.
3. Insert the tip combs.
4. Press on the Tab "Run Prog.", select the shortcut icon for the protocol and press Run to start the protocol
5. At the end of the run remove the tip-combs and plates from the instrument.

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