

For research use only. Not for use in diagnostic procedures.

Prepito Viral DNA/RNA300 Kit

Simultaneous isolation of viral DNA and RNA from Plasma, Serum, Naso- or Oropharyngeal Swabs, BAL and Sputum

Kit Components

Magnetic Beads	Poly(A) RNA
Lysis Buffer	Poly(A) RNA Buffer
Binding Buffer	Proteinase K
Wash Buffer 3	Deep Well Plates
Wash Buffer 4	Disposable Tips
Elution Buffer	

Completion time: approximately 70 minutes

Equipment and Other Material to be provided by the User

- RNase-free water (for reconstitution of **Proteinase K**)
- disposable gloves
- pipette and pipette tips with aerosol barrier

Ensure that all used material is RNase free.

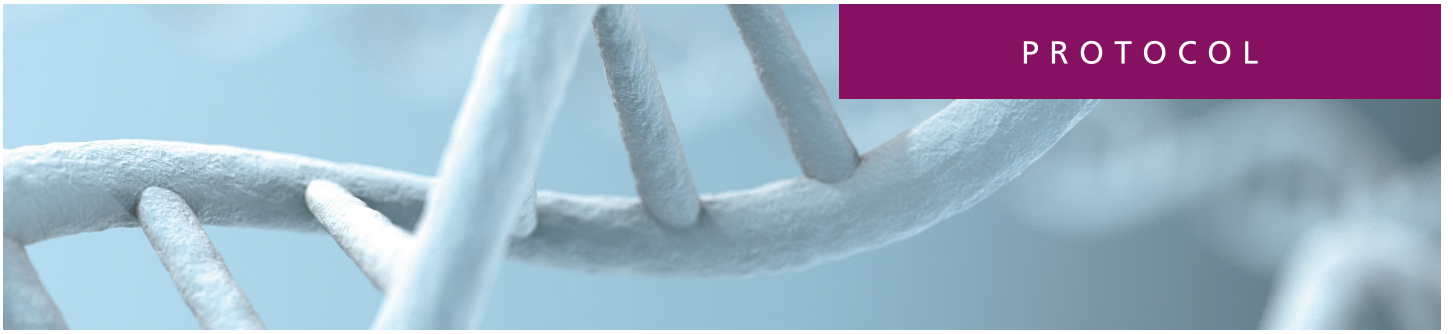
Sample Material

The **Prepito Viral DNA/RNA300 Kit** can be used for plasma and serum, but also for different kinds of respiratory swabs and samples. Plasma, serum and transport media from naso- or oropharyngeal swabs can be used directly in aliquots of 300 µl per isolation. Sample material from dried swabs, bronchoalveolar lavage (BAL) and sputum has to be liquefied before use.

Any further questions?

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Storage Conditions and Safety Information

Expiry dates are stated on the box and on each single component of the kit. Do not use any component of the kit beyond the expiry date. All kit components can be stored at room temperature. The kit buffers contain irritant substances. Take appropriate laboratory safety measures and wear gloves when handling.

Lysis Buffer and **Poly(A) RNA Buffer** have to be stored in the dark. **Lysis Buffer** may form a precipitate upon storage. If necessary, warm to 55 °C to dissolve (after heating the **Lysis Buffer** can directly be used in the isolation process without the need of cooling down). Precipitates in the **Poly(A) RNA Buffer** can be dissolved at room temperature.

After dissolving, **Proteinase K** solution and **Poly(A) RNA** solution have to be stored at 2 – 8 °C. The solutions can be used for 4 weeks. For long term storage, we recommend aliquoting the **Proteinase K** solution and the **Poly(A) RNA** solution and storing at – 20 °C.

General Remarks

The **Elution Buffer** included in this kit is 10 mM Tris-HCl pH 8.0. TE buffer pH 8.0 can also be used without any protocol adjustments. RNase free water pH 8.0 may also be used, but the yield could be slightly decreased.

The **Magnetic Bead** suspension should be mixed vigorously before dispensing, otherwise the suspension is not homogenous and the DNA/RNA yield could be reduced.

Any further questions?

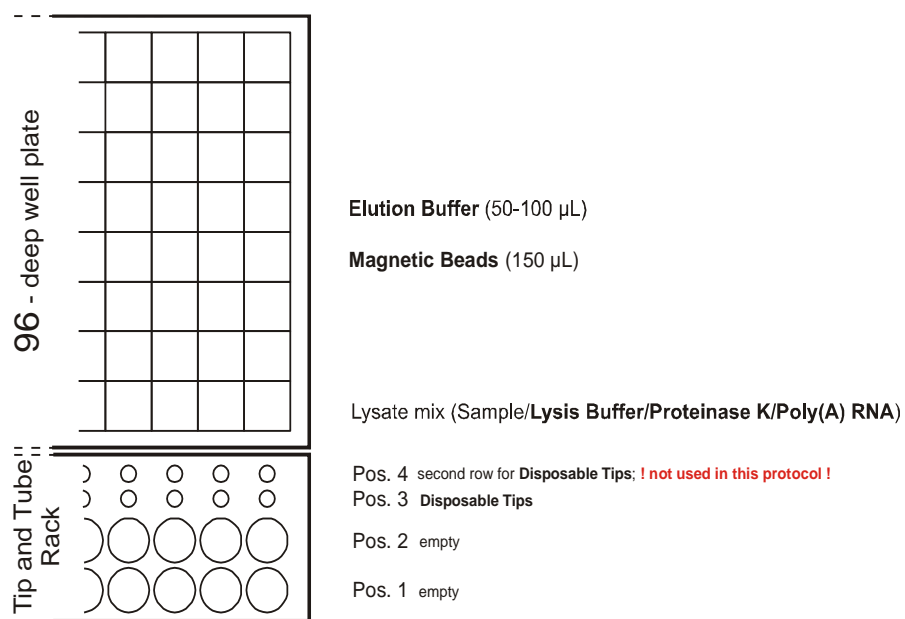
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Positioning Procedure

See “Protocol Steps” for detailed Information.



Before You Start

- Check all kit components for integrity. In case of damages contact your supplier.
- Connect the tubes according to their numbering to the respective counterparts at the **chemagic 8-Pack**. Remove the lids from the individual buffer bottles in the **chemagic 8-Pack** and pierce the septum with the spike placed at the end of each tube. Place the **chemagic 8-Pack** upside down on the reagent holder and use the manual priming function for a complete filling of the dispensing system.
- Dissolve the lyophilized **Proteinase K** in RNase-free water (see instruction on the tube) and **Poly(A) RNA** in 440 μ L **Poly(A) RNA Buffer** per tube.

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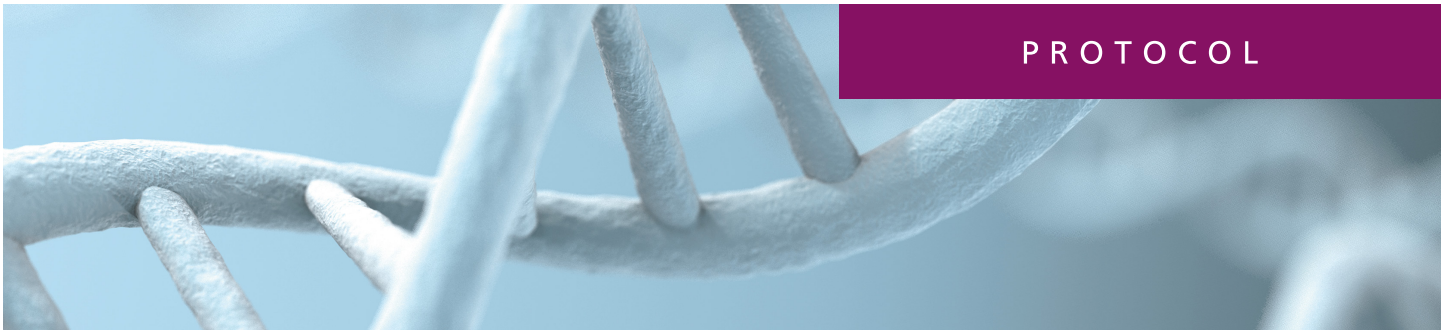
Protocol Steps (chemagic Prepito serial numbers 1 – 99)

1. Switch on the **chemagic Prepito** and wait until the self test is finished.
 2. Press [**change protocol**].
 3. Select the **Prepito Viral DNA/RNA300 Kit** protocol by pressing [**Viral300 no tubes**].
 4. Press [**continue**].
 5. Enter the access code [**2366**] for authorization and confirm by pressing [**enter**].
 6. Confirm the selection of the correct protocol by pressing [**enter**].
 7. Read the protocol information in the appearing information screen. Confirm by pressing [**continue**].
 8. Select the sample positions and confirm by pressing [**continue**].
 9. Enter the kit barcode with the barcode scanner and confirm by pressing [**ok**].
 10. For the registration of the samples and elution tubes press [**yes**] and follow the instructions on the touch screen panel to enter the according barcodes.
 11. Prepare the **chemagic Tip & Tube Rack** with the required material. Place one **Disposable Tip** (position 3) for each sample into the positions according to the sample positions.
 12. Add 50 – 100 μ L **Elution Buffer** for each sample into the position of the Deep Well (DWP) defined as **Elution Buffer** wells (Pos. “**Elution Buffer**”, see section above “Positioning Procedure”).
 13. Add 150 μ L **Magnetic Beads** for each sample into the position of the Deep Well (DWP) defined as **Magnetic Bead** wells (Pos. “**Magnetic Beads**”, see section above “Positioning Procedure”).
- !** *Shake the vessel with the Magnetic Beads vigorously until all Magnetic Beads are completely resuspended. An incomplete resuspension of the Magnetic Beads can result in a decreased yield of extracted nucleic acids.*
14. Add 4 μ L **Poly(A) RNA** solution and 10 μ L **Proteinase K** solution into the sample position of the Deep Well (DWP) defined as sample wells (Pos. “**Lysate mix**”, see section above “Positioning Procedure”).

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15. Add 300 μ L sample material and 300 μ L **Lysis Buffer** into the well filled with **Poly(A) RNA** and **Proteinase K** solutions.
16. An information screen indicates the previously selected sample positions. Ensure that the sample positions in the DWP correspond to the selected positions. Place the DWP on its default position on the tracking system and press [**continue**].
17. Place the **chemagic Tip & Tube Rack** on its default position on the tracking system. Check for accurate fit of the DWP and the **chemagic Tip & Tube Rack** and lock both by closing the safety latch.
18. Close the front door and immediately start the automated isolation process by pressing [**start**].

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Protocol Steps (chemagic Prepito serial numbers 100 and later)

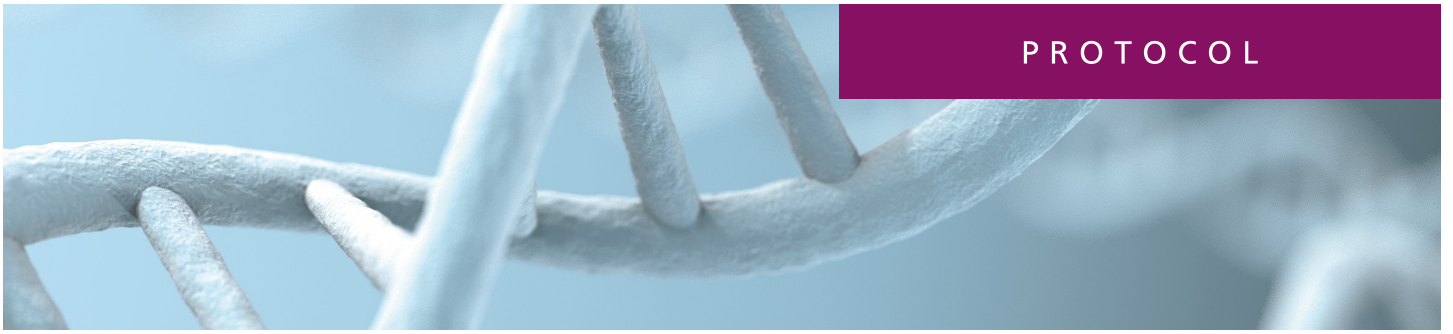
1. Switch on the **chemagic Prepito** and wait until the self test is finished.
2. Press [**Change Protocol**].
3. Press [**Serum/Plasma**] in the Select Protocol Group window.
4. Select the **Prepito Viral DNA/RNA300 Kit** protocol by pressing [**Viral300 no tubes**] and confirm by pressing [**OK**].
5. Confirm the protocol selection in the Select Protocol Group window by pressing [**OK**].
6. Enter the 4 digit access code [**2366**] for authorization and confirm by pressing [**Enter**].
7. Press [**Start Process**].
8. Read the protocol information in the appearing information screen and confirm by pressing [**Continue**].
9. Select the sample positions and confirm by pressing [**OK**].
10. Enter the kit barcode with the barcode scanner and confirm by pressing [**OK**].
11. For the registration of the samples and elution tubes press [**Yes**] and follow the instructions on the touch screen panel to enter the according barcodes.
12. Prepare the **chemagic Tip & Tube Rack** with the required material. Place one **Disposable Tip** (position 3) for each sample into the positions according to the sample positions.
13. Add 50 – 100 μ L **Elution Buffer** for each sample into the position of the Deep Well (DWP) defined as **Elution Buffer** wells (Pos. “**Elution Buffer**”, see section above “Positioning Procedure”).
14. Add 150 μ L **Magnetic Beads** for each sample into the position of the Deep Well (DWP) defined as **Magnetic Bead** wells (Pos. “**Magnetic Beads**”, see section above “Positioning Procedure”).

! *Shake the vessel with the Magnetic Beads vigorously until all Magnetic Beads are completely resuspended. An incomplete resuspension of the Magnetic Beads can result in a decreased yield of extracted nucleic acids.*

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15. Add 4 μL **Poly(A) RNA** solution and 10 μL **Proteinase K** solution into the sample position of the Deep Well (DWP) defined as sample wells (Pos. “Lysate mix”, see section above “Positioning Procedure”).
16. Add 300 μL sample material and 300 μL **Lysis Buffer** into the well filled with **Poly(A) RNA** and **Proteinase K** solutions.
17. An information screen indicates the previously selected sample positions. Ensure that the sample positions in the DWP correspond to the selected positions. Place the DWP on its default position on the tracking system and press [**Continue**].
18. Place the **chemagic Tip & Tube Rack** on its default position on the tracking system. Check for accurate fit of the DWP and the **chemagic Tip & Tube Rack** and lock both by closing the safety latch.
19. Close the front door and immediately start the automated isolation process by pressing [**Start**].

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