PI-BIOTECH
genética avançada

PI-CLEAR
nucleic acid purification kit

Pi-Clear nucleic acid purification kit - DNA/RNA viral

ATTENTION: Before starting the assay, read the item "Special Care" in "User Guide".

Spin Column – PB1	50 units (spin column + collection tube)
Proteinase K - PK	300 µL
Carrier - CR	350 µg (freeze dried reagent. Resuspend in 350 µL of PB3)
Lysis Buffer – TL	15 mL
Wash Buffer 1 – TP1	50 mL
Wash Buffer 2 – TP2	**OBS:** 48 mL of Ethanol 96-100 % must be added Final Volume: 60 mL
Recovery tube (1,5 mL) – PB2	50 units
Nuclease-free Water – PB3	15 mL

Storage: PB1, TL, TP1, TP2, PB2 and PB3 must be stored at room temperature (15 - 25 °C); PK and CR (after resuspension in PB3) must be stored at -20 °C.

Required materials: Ethanol 96-100% and extra DNase/RNase-free microtubes.
Validated procedure: sputum, liquor, swab, semen, whole blood, blood plasma and blood serum.

Procedure

A)Sample Lysis

1. Add 200 µL of sample into a clean, sterile and DNase/RNase free microtube;
   Obs: prepare sample as described in “Instruction for Use”;
2. Add 5 µL Proteinase K (PK) and vortex;
3. Add 200 µL Lysis Buffer (TL) and vortex.

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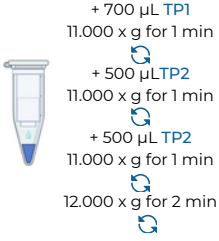
Nucleic acid Extraction and Purification

B) Binding

4. Add 5,6 µL Carrier (CR) and vortex;
5. Incubate at room temperature for 3 minutes;
6. Add 200 µL Ethanol (96-100 %) and vortex;
7. Incubate at room temperature for 5 minutes.
8. Transfer up total volume of lysed sample to spin column with the collection tube (PB1);
9. Centrifuge at 4.000 xg for 3 minutes;
10. Discard the flow-through.

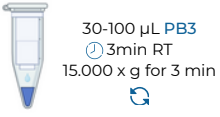
C)Washing

11. Add 700 µL Wash Buffer 1 (TP1);
12. Centrifuge at 11.000 x g for 1 minute;
13. Discard the flow-through;
14. Add 500 µL Wash Buffer(TP2);
15. Centrifuge at 11.000 x g for 1 minute;
16. Discard the flow-through;
17. Repeat steps 14-16 once;
18. Centrifuge at 12.000 x g for 2 minutes to dry the membrane with bound DNA/RNA;
19. Discard the collection tube and insert the spin column into a recovery tube (PB2).



D) Elution

20. Add 30-100 µL Nuclease-free Water (PB3) to the center of spin column;
21. Incubate at room temperature for 3 minutes;
22. Centrifuge at 15.000 x g for 3 minutes and discard the spin column;
23. Store your purified DNA/RNA at -20 °C. For long-term storage, keep the purified DNA/RNA at -70 °C.



Acess the complete “User Guide”