

DENAzist PCR-Max-Clean Kit

Cat. No.: S-1060-25; 25 applications

Cat. No.: S-1060-50; 50 applications



MANUAL

Description: DENAzist PCR-Max-Clean kit is suitable for purification of nucleic acids from PCR, restriction enzyme digestion, or other enzymatic reactions. The recovery process efficiently removes nucleotides, enzymes, salts, and small fragments of nucleic acid (less than 50 base). Purified nucleic acids can be used in a variety of applications including PCR and qPCR, Sanger sequencing, plasmid subcloning, *in vitro* transcription and transfection.

VERSION: Nov. 2024

Important instructions before first use

For 25 application kit, add 12 ml ethanol (96%-100%) to the container labeled PC2.

For 50 application kit, add 24 ml ethanol (96%-100%) to the container labeled PC2.

Label the container to record that ethanol has been added before first use.

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PROTOCOL for recovery of nucleic acids from enzymatic reactions

Before the first use, make sure that ethanol has been added to the container labeled PC2.

1. Measure the volume of the enzymatic reaction and adjust it to 100 μ l with **PC1 buffer**. If the reaction volume is 100 μ l or more, no volume adjustment is needed. This volume is referred to as the "initial volume."
2. Add 3 volumes of the initial volume from the **PC1 buffer** and mix.
3. Add a volume of **sodium acetate** solution equal to 1/20th of the combined initial and PC1 volumes, then mix well. For example, to 100 μ l of the initial volume plus 300 μ l of buffer PC1, add 20 μ l of sodium acetate solution.
4. Add a volume of **ethanol** equal to 1/2 of the combined initial and PC1 volumes, then mix well. For example, to 100 μ l of the initial plus 300 μ l of PC1 volumes, add 200 μ l of ethanol.

-----DNA BINDING TO THE COLUMN-----

5. Mount a spin column onto a collection tube (both are included in the kit). Using a sampler (pipettor), transfer the entire mixture to the upper reservoir of the spin column.
6. Centrifuge the spin column at 8,000 rpm for 2 minutes. Discard the flow-through accumulated in the collection tube.

-----WASHING-----

7. Remount the spin column onto the collection tube. Add 700 μ l from **PC2 buffer** onto the spin column.
8. Centrifuge the spin column and its collection tube at 8,000 rpm for 1 minute. Discard the flow-through.
9. Without adding any solution, centrifuge the tube

once more at top speed (13,000-14,000 rpm) for 3 minutes.

-----ELUTION-----

10. Separate the spin column from its collection tube and place it into a new 1.5 ml microfuge (Eppendorf) tube. Add 50 μ l of **PC3** solution to the center of the spin column. Leave the spin column (mounted on the microfuge tube) for 5 minutes. Centrifuge the spin column at 13,000 rpm for 2 minutes.
11. Discard the spin column. The eluted solution at the bottom of the microfuge tube will contain the purified nucleic acid. Store the sample in a -20 °C freezer until further downstream processes. ▲

Kit components	25 app.	50 app.
PC1 buffer	10 ml	20 ml
PC2 buffer	9 ml	18 ml
PC3 buffer	3 ml	6 ml
Na Acetate solution	0.7 ml	1.4 ml
Spin column	25	50

Store all components at room temperature.
Expiry date: 18 month from the date of manufacture

Notes: Research Use Only

This product insert declares that this product has been analyzed and passed the quality control tests at the time of manufacture.