PCR Clean-up Kit

Cat. No.: S-1060; 25 applications
Cat. No.: S-1060-1; 50 applications

DENA ZIST ASIA
MOLECULAR BIOLOGY PRODUCTS
www.denazist.ir

Manual

Description: DENAzist PCR Clean-up kit is suitable for purification of nucleic acids from PCR, restriction enzyme digestion, or other enzymatic reactions. The recovery process 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 2021

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.

PROTOCOL for recovery of nucleic acids from gel

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

1. Transfer a small volume of PC3 buffer into a microfuge tube and place the tube in a 60 °C water bath. This will be used for elution.
2. Measure the volume of the enzymatic reaction. Adjust the volume to 100 µl with sterile distilled water. If the reaction volume is 100 µl or more, there is no need for volume adjustment. The volume at this stage is called the "initial volume".
3. Add equal to the initial volume from **PC1** buffer and mix. Add 10 µl from **Na acetate** solution and mix.
4. Add equal to the initial volume from 96-100% ethanol. Mix by pipetting.

-------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 whole mix to the upper reservoir of the spin column.
6. Centrifugate 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 µl from **PC2** buffer onto the spin column. Let the tube stand at room temperature for 2 minutes.
8. Centrifuge the spin column and its collection tube at 10,000 rpm for 1 minute. Discard the flow-through.
9. Without adding any solution, centrifuge the tube one more time at top speed (13,000 -14,000 rpm) for 3 minutes.

-------ELUTION-------

10. Separate the spin column from its collecting tube and place it into a new 1.5 ml microfuge (Eppendorf) tube. Add 40-100 µl from pre-warmed (to 60 °C) **PC3** solution onto the center of spin column. Leave the spin column (mounted on the microfuge tube) for 5 minutes at 37 °C. Centrifuge the spin column at 10,000 rpm for 2 minutes.

Important: The volume for elution will determine the final concentration. If higher concentration of DNA is needed, the volume for elution could be 40-50 µl.

11. Return the eluted solution back on to the center of spin column. Leave the spin column mounted on the microfuge tube for 4 min at room temperature. Centrifuge the spin column mounted on the microfuge tube at 13,000 rpm for 2 minutes.
12. Discard the spin column. The eluted solution at the bottom of the microfuge tube will contain the purified nucleic acid. Until further downstream processes store the sample in -20 °C freezer. ◢

Kit components:

Buffers: PC1, PC2,PC3
Na acetate
Spin Columns

Store all components at room temperature

Notes: Research Use Only
This product insert declares that this products has been analyzed and passed the quality control tests at the time of manufacture.