A&A BIOTECHNOLOGY
innovating life science

Manual

Clean-Up 96-well

Kit for DNA cleanup after PCR and other enzymatic reactions using restriction enzymes, ligase, kinase, etc.
Format: 96-well plates.

catalog #	size
021-192	192 isolations

For research use only.

Guarantee
A&A Biotechnology provides a guarantee on this product.
The company does not guarantee the correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

Table of Contents

Contents

component	192 isolations	storage
P96 purification plate	2 pcs.	room temp.
E96 elution plate	2 pcs	room temp.
R96 receiving plate	2 pcs	room temp.
Protective film	2 pcs	room temp.
Reservoirs	3 pcs	room temp.
A1 wash solution	190 ml	room temp.
GI binding solution	105 ml	room temp.
Sodium acetate (3M, pH 5.5)	3 ml	room temp.
TE buffer	12 ml	room temp.

Binding capacity of minicolumn: up to 20 µg of DNA

Additional equipment and reagents

Necessary

- 1.5 ml sterile Eppendorf tubes
- Centrifuge with swing-out rotor for 96-well plates
- Self-adhesive foil / protective film

Optional

- Sterile water (cat.# 003-075, 003-25)
- Vorex

Comments

- Binding capacity of minicolumn: up to 20 µg of DNA / Minimum binding capacity of minicolumn: 2 µg DNA

  If DNA is below 2 µg we recommend using a Clean-Up Concentrator Kit (cat.# 021-50C, 021-250C)
- DNA fragments range: 100-10 000 bp
- Typical DNA recovery: 60-90%
- Elution volume: 30-50 µl

Isolation protocol

1. Mix DNA sample (up to 100 µl) with **5 volumes** of **GI** binding solution. Mix the sample by inverting the tube or vortexing.

   GI binding solution contains the color pH indicator. Upon mixing the DNA sample with GI binding solution, the yellow color of the mixture indicates an optimal pH for DNA binding.

   If the mixture color turns pink the pH of the solution is too high. In such conditions DNA binds ineffectively to the silica membranes and may be lost.

   Too high pH can be corrected by adding 1-10 µl of 3M sodium acetate solution (pH 5.5) (included) and mix. Purification can be continued after reaching a yellow color.

    optimal condition pH ≤7.2

    too high pH

2. Assemble the **P96** purification plate with the **R96** receiving plate.

3. Apply the sample onto the well of the **P96** purification plate.
   Stick the **protective film** on the **P96** purification plate.

4. Transfer the assembled plates to the swing-out rotor.

   Note: If an odd number of plates use the counter-plate for centrifugation.

   Centrifuge for **1 min** at **2 000 x g**.

5. Carefully separate the plates. Discard the filtrate from the **R96** receiving plate.
   Assemble the **P96** purification plate with the **R96** receiving plate.
   Remove the **protective film** from the **P96** purification plate.

6. Apply **600 µl** of **A1** wash solution onto each well of the **P96** purification plate.
   Stick the **protective film** on the **P96** purification plate (not included).

7. Transfer the assembled plates to the swing-out rotor.

   Note: If an odd number of plates use the counter-plate for centrifugation.

   Centrifuge for **2 min** at **2 000 x g**.

8. Carefully separate the plates. Discard the filtrate from the **R96** receiving plate.
   Assemble the **P96** purification plate with the **R96** receiving plate again.
   Remove the protective film from the **P96** purification plate.

9. Apply **300 µl** of **A1** wash solution onto each well of the **P96** purification plate.

Stick the **protective film** on the **P96** purification plate (not included).

10. Transfer the assembled plates to the swing-out rotor.

Note: If an odd number of plates use the counter-plate for centrifugation.

Centrifuge for **10 min** at **2 000 x g**.

11. Carefully separate the plates. Remove the **R96** receiving plate.
Assemble the **P96** purification plate with a **E96** elution plate.
Remove the protective film from the **P96** purification plate.

12. Apply **50 µl** of **TE** buffer or sterile water (not included) onto each well of the **P96** purification plate.

Applying elution liquid (TE buffer or sterile water) onto the minicolumn be sure that liquid is applied directly onto the resin. If some of the liquid stays on the column wall the elution will be less effective.
Elution in a smaller volume is less efficient, but the extracted DNA has a higher concentration. Elution in 50 µl volume is more efficient, but DNA has a lower concentration.

Stick the **protective film** on the **P96** purification plate (not included).

Keep for **3 min** at **room temp.**

13. Transfer the assembled plates to the swing-out rotor.

Note: If an odd number of plates use the counter-plate for centrifugation.

Centrifuge for **3 min** at **2 000 x g**.

14. Carefully separate the plates. Remove the **P96** purification plate.
Stick the **protective film** on the **E96** elution plate.

Store the purified samples at 4 °C until later use.

Safety information

WARNING

GI binding solution

H302 Harmful if swallowed.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

DANGER

A1 wash solution

H225 Highly flammable liquid and vapor.
H319 Causes serious eye irritation.
H336 May cause drowsiness or dizziness.
P210 Keep away from heat, sparks, open flames, hot surfaces. No smoking.
P261 Avoid breathing vapors.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

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