

Manual

Clean-Up

Kit for DNA cleanup after PCR and other enzymatic reactions using restriction enzymes, ligase, kinase, etc.

catalog #	size
021-50	50 isolations
021-250	250 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



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Contents

component	021-50	021-250	storage
Minicolumns	50 pcs	250 pcs	room temp.
GI binding solution	45 ml	210 ml	room temp.
A1 wash solution	50 ml	250 ml	room temp.
Sodium acetate (3M, pH 5.5)	500 µl	3 ml	room temp.
TE buffer	5 ml	16 ml	room temp.

Additional equipment and reagents

Necessary

- 1.5 ml sterile Eppendorf tubes
- Vortex
- Microcentrifuge

Additional

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

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 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 samples (up to 150 μ l) with 5 volumes of GI binding solution. Mix the samples by inverting the tubes or vortexing
	GI binding solution contains the color pH indicator. Upon mixing the DNA sample with GI binding solution, 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
2.	Briefly centrifuge the samples to remove the leftovers of solution from the tube walls and caps.
3.	Apply samples onto the minicolumns.
4.	Centrifuge for 30 s at 10 000-15 000 RPM .
5.	Remove the minicolumns, discard the filtrate. Place the minicolumns to the same tubes.
6.	Add 600 μl of A1 wash solution.
7.	Centrifuge for 30 s at 10 000-15 000 RPM .
8.	Remove the minicolumns, discard the filtrate. Place the minicolumns to the same tubes.
9.	Add 300 μl of A1 wash solution.
10.	Centrifuge for 1 min at 10 000-15 000 RPM .
11.	Remove the minicolumns, discard the filtrate. Place the minicolumns to the same tubes.
12.	Centrifuge for 1 min at 10 000-15 000 RPM .
13.	Transfer the minicolumns to new 1.5 ml tubes (not included).

14. Add 50 µl of TE buffer or sterile water (not included) directly onto the minicolumn resin.

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.

15.	Incubate for 3 min at room temp .
16.	Centrifuge for 1 min at 10 000-15 000 RPM .

17. Remove the minicolumns and store the tubes with purified DNA at 4-8°C until later use.

Safety information

\wedge	GI binding solution
	H302 Harmful if swallowed.
	H315 Causes skin irritation. H319 Causes serious eye irritation.
WARNING	P305 P305 P338 If in eyes: "Insecautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
• •	A1 wash solution
	H225 Highly flammable liquid and vapor.
< { X X X X X X X X	H319 Causes serious eye irritation.
	H336 May cause drowsiness or dizziness.
	P210 Keep away from heat, sparks, open flames, hot surfaces. No smoking.
DANGER	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,

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