

SpinColumn PCR Purification kit

I. DESCRIPTION

The SpinColumn is a reagent kit from ShineGene. It contains all the necessary reagents, mini-columns, and collection tubes for the fast and reliable extraction of DNA from PCR reactions and for DNA cleanup in enzymatic reactions. In only five minutes, ColumnClean can recover up to 20 µg of single- or double-stranded DNA product. DNA molecules ranging in size from 70 to 10,000 base pairs are adsorbed onto the Column while impurities such as enzymes and proteins, small DNA fragments, dyes, salts, nucleotides and short oligonucleotides (such as primers and probes) are washed away.

Eluted in a small volume of low-salt buffer with complete removal of contaminants and inhibitors, the purified DNA is immediately ready for downstream applications such as PCR, transformation, restriction enzyme digestion, cloning, sequencing, *in vitro* translation and transfection.

II. KIT CONTENTS

Kit contains enough supplies for 50/100 preps.

Components	ZN00201(50Preps)(EUR50.00)	ZN00202(100Preps)(EUR80.00)
Binding Buffer I	20ml	40ml
Wash Solution (A)	12ml	24ml
Elution Buffer(B)	5ml	10ml
Column	50	100
2ml Collection Column	50	100
Protocol	1	1

(A) Before use, add 48ml of 100% of ethanol to 12ml Wash Solution for ZN00201, or add 96ml of 100% ethanol to 24ml Wash Solution for ZN00202. For other volumes of wash solution, simply add enough ethanol to make a 4:1 ratio (volume of added ethanol: volume of Wash Solution = 4:1).

(B) Elution Buffer is 2mM Tris-HCl pH 8.0~8.5. Although TE buffer pH 8.0 or water may be substituted, the resulting yields may be up to 20% lower.

III. APPLICATIONS

The Kit enables the fast purification of high-quality DNA products from PCR reactions and enzymatic reactions. The extracted DNA is immediately ready for further downstream applications such as the following:

- PCR and cloning
- Restriction enzyme digestion
- Transformation
- Sequencing
- *In vitro* translation

IV. KEY FEATURES

- Easy to perform: ColumnClean's simple and rapid procedure purifies DNA in five minutes.
- High capacity: Each column has a capacity of 20 µg DNA.
- High purity: The kit completely removes contaminants and inhibitors.
- Reproducible yields: Recovery is typically between 90% and 98%, reproducible every time.

V. STORAGE

- This kit should be stored dry at room temperature. So stored, the kit is stable for 12 months.

VI. Protocol for Purification of PCR* Products

1. Transfer PCR* reaction mixture to a 1.5ml microfuge tube and add 3 volumes of Binding Buffer I.
2. Transfer the above mixture solution to the column and let stand at room temperature for 2 minutes. Centrifuge at 10,000 rpm for 2 minutes.
3. Remove the flow-through in the tube. Add 500ul of Wash Solution to the column and centrifuge at 10,000 rpm for 2 minutes.
4. Repeat washing procedure in step 3. Spin at 10,000 rpm for an additional minute to remove any residual Wash Solution.
5. Transfer the column into a clean 1.5ml microfuge tube and add 30-50ul of Elution Buffer. Incubate at room temperature for 2 minutes. Centrifuge at 10,000 rpm for 2 minutes to elute the DNA.

Note: It is extremely important to add the Elution Buffer to the center of the column. Incubating the column at higher temperatures (37° to 50°C) may slightly increase the yield. Pre-warming the Elution Buffer at 55° to 80°C may also slightly increase elution efficiency. If a higher DNA concentration is desirable, 20ul (or less) of elution buffer can be used to elute the DNA. It is critical that the elution buffer be applied directly in the center of the column. (To recover maximum amount of DNA it is recommended to repeat the elution step.).

6. Store the purified DNA at -20°C.

Note:

1. If PCR* reaction mixture contains seriously non-specific amplified DNA fragments, use of the DNA

Gel Extraction Kit is recommended.

2. This kit can not remove the template and primers with chain length longer than 50-mer.

* The Polymerase Chain Reaction (PCR*) is covered by patents owned by Hoffman-La Roche Inc.