

# PrimePrep™ Gel Purification Kit

## Introduction

PrimePrep™ Gel Purification Kit offer simple, rapid and cost-effective method for purification from agarose gel in TAE or TBE buffer system.

The purified DNA can be directly used in ligation, sequencing and other downstream applications.

## Kit Components

Reagents \ Cat. No.	K-8000 (50 prep.)	K-8001 (200 prep.)
Spin column	50 ea	50ea x 4
Buffer Gel-B	60 ml (30 ml x 2)	240 ml (80 ml x 3)
Buffer PW	10 ml	30 ml (15 ml x 2)
Buffer PE	10 ml	20 ml

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## Before you begin

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► **Add ethanol to Buffer PW before use.**

→ Add 40 ml (K-8001: 60 ml) of absolute ethanol before use.

► **Isopropanol (100%) and a heating block or water bath at 50°C are required.**

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## Experimental Protocol

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- 1. Excise the DNA fragment from the agarose gel with a sharp scalpel or razor blade.**  
Minimize gel volume by cutting gel slice as small as possible.
- 2. Weigh the gel slice in a microcentrifuge tube. Add 3 volumes of Buffer Gel-B to 1 volume of gel.**  
If the 100 mg of agarose gel slice, add 300  $\mu\text{l}$  of Buffer Gel-B.  
If more than 1.5% agarose gel, add 6 volumes of Buffer Gel-B.
- 3. Incubate at 50°C for 10 min (or until the gel slice has completely dissolved). Invert the tube every 2-3 min to help dissolve gel.**
- 4. Add 1 gel volume isopropanol to the sample and mix immediately by using pipett or inverting.**
- 5. Transfer the sample to the Spin column.**
- 6. Centrifuge at 13,000 rpm for 1 min.**  
Discard the flow-through and re-insert the Spin column to the collection tube.  
For sample volumes  $> 800 \mu\text{l}$ , load and spin again.
- 7. (Optional step) Add 500  $\mu\text{l}$  of Buffer Gel-B to the Spin column and centrifuge at 13,000 rpm for 1 min. Discard flow-through and re-insert the Spin column to the collection tube.**  
If DNA will subsequently be used for sequencing, in vitro transcription, or microinjection.
- 8. Add 700  $\mu\text{l}$  Buffer PW to Spin column and centrifuge at 13,000 rpm for 30 sec. Discard flow-through and re-inserting the Spin column to the collection tube.**
- 9. Centrifuge once more at 13,000 rpm for 1 min to remove residual wash buffer.**
- 10. Place the spin column into a clean 1.5 ml microcentrifuge tube.**  
The clean 1.5 ml microcentrifuge tube is not provided.
- 11. To elute DNA, add 50  $\mu\text{l}$  of Buffer PE or deionized distilled water to the center of the membrane in the Spin column, let the column stand for 1 min, and then centrifuge at 13,000 rpm for 1 min.**  
For increased DNA concentration, add 30  $\mu\text{l}$  Buffer PE to the center of the membrane in the Spin column, let the column stand for 1 min, and then centrifuge for 1 min.