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PrimeWay Gel Extraction Kit/ PCR Purification Kit II (KIT-9051)

Molecular Biology Kit

PrimeWay Gel Extraction/ PCR Purification Kit II

Product No: KIT-9051

PrimeWay Gel Extraction/PCR Purification Kit II offers two applications in one kit. It is a simple procedure and uses a silica-based spin column to perform gel extraction or purify the DNA fragments from 65 bp to 15 kb within 16 minutes with a high recovery rate. This kit enables removal of primers, dNTPs, enzymes, salts, and short PCR products (< 65 bp), making it suitable for downstream processes such as DNA sequencing, PCR, in-vitro transcription, restriction mapping, cloning, and labelling applications.

For Research Use Only. Not for use in Diagnostic Procedures.

Kit Contents

No	Product	KIT-9051-10 10 preps	KIT-9051-50 50 preps	KIT-9051-250 250 preps
1	BD Buffer	7 mL	35 mL	175 mL
2	Wash Buffer W1	2 mL	10 mL	45 mL
3	Wash Buffer W2	2 mL	9 mL	45 mL
4	Elution Buffer	1.5 mL	6 mL	30 mL
5	PrimeWay Gel/PCR II Column	10 pcs	50 pcs	5 x 50 pcs

Storage

This kit should be stored at room temperature (21–25 °C).

Product Specification

	PCR Product Purification	Gel Extraction
Binding capacity	20 µg	20 µg
Sample	≤ 100 µL PCR product	≤ 300 mg gel size
DNA fragment size	65 bp–15 kb	65 bp–15 kb
Recovery	Up to 100%	Up to 99%
Elution	30-50 µL	30-50 µL
Duration	~6 minutes	~16 minutes

Materials Supplied by Users

- ✓ Ethanol (96–100%)
- ✓ Centrifuge, at speed of 11,000–18,000 × g
- ✓ Vortex mixer
- ✓ Water Bath/ Dry Bath at 55 °C (Gel Extraction only)
- ✓ 1.5 mL microcentrifuge tubes

Precautions for Users

- ✓ Some buffers in this kit contain irritants. Handle with care and avoid skin contact. In case of contact, wash the affected area thoroughly with water and seek medical attention.
- ✓ Always wear a lab coat, disposable gloves and a surgical mask.

Before Start

- ✓ Set the water bath/ dry bath to 55 °C.
- ✓ Add ethanol (96–100%) to the Wash Buffer as follows:

P/No	Ethanol to be added	
	Wash Buffer W1	Wash Buffer W2
KIT-9051-10	8 mL	8 mL
KIT-9051-50	40 mL	36 mL
KIT-9051-250	180 mL	180 mL

Protocol – Gel Extraction

Sample	1. Excise the gel slice using a clean scalpel. (a) 1% gel: up to 300mg slice (b) 2% gel: up to 200mg slice 2. Transfer the gel slice into a new 1.5 mL microcentrifuge tube. 3. Add **500 µL BD Buffer** to the gel and vortex to mix. 4. Incubate the mixture at 55 °C for 10 minutes until the gel slice is completely dissolved. 5. Vortex to mix, then spin down briefly before proceeding to the next step.
Binding	6. Transfer **up to 700µL** of gel mixture into **PrimeWay Gel/PCR II Column.** ***Note**: If the gel mixture > 700 µL, repeat this step by loading the remaining volume after Step 7.* 7. Centrifuge at 11,000 × *g* for 30 seconds. Discard the flow-through and place the **PrimeWay Gel/PCR II Column** back into collection tube.
Washing	8. Add **700 µL Wash Buffer W1** to the **PrimeWay Gel/PCR II Column.** ***Important:** Ensure that ethanol has been added to the buffer before first use.*

Washing	9. Centrifuge at 11,000 × *g* for 30 seconds. Discard the flow-through and place the **PrimeWay Gel/PCR II Column** back into collection tube. 10. Add **500 µL Wash Buffer W2** to the **PrimeWay Gel/PCR II Column.** ***Important:*** *Ensure that ethanol has been added to the buffer before first use.* 11. Centrifuge at 11,000 × *g* for 30 seconds. Discard the flow-through and place the **PrimeWay Gel/PCR II Column** back into collection tube.
Drying	12. Dry the column by centrifuging at maximum speed (~16,700 x *g*) for 1 minute. ***Note****: Ensure that all Wash Buffer is completely removed, as any residual liquid can inhibit downstream applications.*
Elution	13. Transfer the **PrimeWay Gel/PCR II Column** into a new 1.5 mL microcentrifuge tube. 14. Add **30-50 µL Elution Buffer** to the center of the **PrimeWay Gel/PCR II Column**. *(**Optional**: Preheat Elution Buffer at 70°C before adding to the column for improved yield.)* 15. Let the column stand for 1 minute. 16. To elute the DNA, centrifuge at maximum speed (~16,700 x *g*) for 1 minute. Store the DNA at -20 ˚C.

Protocol – PCR Purification

Sample	1. Transfer **up to 100 µL PCR product** into a new 1.5 mL microcentrifuge tube. ***Note**: If >100 µL PCR product, divide it into multiple tubes for processing.* 2. Add **4 volumes of BD Buffer** to 1 volume of PCR product and vortex to mix. *Example:* • *For 25 µL PCR product, add 100 µL of BD Buffer* • *For 50 µL PCR product, add 200 µL of BD Buffer* ***Note**: For improved recovery of small fragments (< 100bp), add 1 volume BD Buffer and 1 volume absolute ethanol to 1 volume of PCR product.*
Binding	3. Transfer the PCR mixture into the **PrimeWay Gel/PCR II Column.** 4. Centrifuge at 11,000 × *g* for 30 seconds. Discard the flow-through and place the **PrimeWay Gel/PCR II Column** back into collection tube.
Washing	5. Add **750 µL Wash Buffer W2** into the **PrimeWay Gel/PCR II Column.** ***Important:** Ensure that ethanol has been added to the buffer before first use.* 6. Centrifuge at 11,000 × *g* for 30 seconds. Discard the flow-through and place the **PrimeWay Gel/PCR II Column** back to its collection tube.

Drying	7. Dry the column by centrifuging at maximum speed (~16,700 x *g*) for 1 minute. ***Note**: Ensure that all Wash Buffer is completely removed, as any residual liquid can inhibit downstream applications.*
Elution	8. Transfer the **PrimeWay Gel/PCR II Column** into a new 1.5 mL microcentrifuge tube. 9. Add 3**0-50 µL Elution Buffer** to the center of the **PrimeWay Gel/PCR II Column**. 10. Let the column stand for 1 minute. 11. To elute the DNA, centrifuge at maximum speed (~16,700 x *g*) for 1 minute. Store the DNA at -20 ˚C.

Troubleshooting Guidelines**

Problems	Possible Reason	Recommended Action
Low DNA recovery	Reagents not prepared properly	Make sure ethanol (96–100%) has been added to the Wash Buffer. Mix well before use.
	Incomplete dissolution of the gel slice	• Increase the incubation time. • If gel slice is too large, divide it into multiple tubes.
	Inefficient DNA elution	Ensure that the Elution Buffer is added to the center of the column membrane and is completely absorbed before centrifugation.
	Incorrect centrifugation force	Perform the DNA binding and washing steps at a centrifugation speed of 11,000 x g to achieve maximum DNA recovery.
Low DNA yield	High elution volume	Adjust the elution volume to a minimum of 12 µL to achieve a concentrated DNA recovery with approximately 50% efficiency. A second elution in a separate tube can be performed to recover any remaining DNA.
Problem in the downstream processes	Presence of salt residue during PCR Purification	Performing two washes may improve purity. Follow steps 5 to 13 of the gel extraction protocol after sample preparation.

Please contact us at https://base-asia.com/contact/ for more information.