

PurElute™ GX DNA Gel Extraction & Cleanup Kit

Product	Catalog #	Purifications
PurElute™ GX DNA Gel Extraction and Cleanup Kit	81002	5
	81004	50
	81006	250

Description

The PurElute™ GX DNA Gel Extraction & Cleanup Kit is designed for the easy extraction and purification of dsDNA from solutions or agarose gel in TAE or TBE buffers. A Binding Buffer containing chaotropic salt and reinforced buffer dissolves agarose gel and facilitates the capture of DNA fragments ≥ 70 bp in a spin column. The column is washed and the purified DNA is eluted in 15-50 µl of Elution Buffer or water.

Benefits

- **High purity DNA** – Eliminate primers, primer-dimers, adapters, dNTPs, salts, DNA labels, and enzymes
- **High recovery** – 75-95% recovery, depending on fragment size and elution volume
- **Rapid protocol** – Less than 20 minutes
- **Convenient processing options** – Centrifuge or vacuum
- **User-Friendly** – Simple spin column format
- **Purifies almost any fragment size** – 70bp - 20kb
- **Binding capacity** – Up to 5µg of DNA

Kit Components	81002	81004	81006
GX Spin Column	5	50	5×50
GX Binding Buffer	6 ml	60 ml	3×100 ml
GX Wash Buffer	1 ml	12 ml	2×25 ml
GX Elution Buffer	1 ml	15 ml	15 ml

Equipment and Materials Required

1. Agarose gel electrophoresis apparatus, UV illuminator
2. Microcentrifuge
3. 50-55°C water bath or heating block
4. 1.5-ml microcentrifuge tubes
5. Ethanol
6. 3M Sodium acetate (pH 4.5 – 5.2)*
7. TE Buffer (pH 8)*

*May be needed for pH or volume adjustment

Storage Condition

All kit components can be stored at room temperature for up to 1 year.

Quality Control

PurElute™ GX DNA Gel Extraction & Cleanup Kit is tested for the extraction and purification of EcoRI digested pUC19 DNA from both the agarose gel and the digestion reaction. Purified products are evaluated for recovery efficiency with agarose gel electrophoresis.

Before Starting

Add 4 volumes of ethanol (96 -100%) into the bottle of Wash Buffer and mix well. The volume of ethanol is indicated on the bottle label. Label the bottle “ethanol added”.

Centrifugation Protocol

Binding Mixture Preparation

Extraction from agarose gels:

- 1) Visualize DNA bands using a long wavelength UV source. Cut out the band containing the desired fragment and carefully trim away excess gel.
- 2) Weigh the gel slice.
- 3) Add 300 μ l of Binding Buffer for every 100 mg of the gel (3:1).
- 4) Incubate in a 50-55°C water bath or heating block for 10 minutes or until the gel has dissolved.
 - Mix every 2-3 minutes by inverting the tube several times.
- 5) After the gel has dissolved the color of the mixture should remain yellow.
 - If the mixture is orange to violet, add 10 μ l aliquots of 3 M sodium acetate (pH 4.5 - 5.2) until the color changes to yellow.

DNA Cleanup from solutions:

- 1) Add 300 μ l of Binding Buffer for every 100 μ l of sample (3:1); vortex briefly to mix.
 - For samples with a volume <100 μ l, add TE Buffer (pH 8) to bring the total volume to 100 μ l.
- 2) After addition of the Binding Buffer the color of the mixture should remain yellow.
 - If the mixture is orange to violet, add 10 μ l aliquots of 3 M sodium acetate (pH 4.5 - 5.2) until the color changes to yellow.

DNA Capture

- 1) Load up to 750 μ l of the binding mixture into a spin column.
- 2) Centrifuge at 16,000 \times g for 1 minute.
- 3) Discard the flow through.
- 4) Repeat steps 1 through 3 until all of the mixture has been processed.

Column Wash

- 1) Add 750 μ l of Wash Buffer into the spin column.
- 2) Centrifuge at 16,000 \times g for 1 minute.
- 3) Discard the flow through.
- 4) Centrifuge at 16,000 \times g for 1 minute.

DNA Elution

- 1) Transfer the spin column into a clean microcentrifuge tube.
- 2) Add 15-50 μ l of Elution Buffer to the center of the membrane in the spin column.
- 3) Allow the column to sit for 1 minute at room temperature, then centrifuge at 16,000 \times g for 1 minute.
- 4) Discard the spin column. Mix the purified sample well by pipeting or brief vortexing. Store at -20°C.

Vacuum Protocol

Binding Mixture Preparation

Follow the same procedure from the centrifugation protocol.

DNA Capture

- 1) Connect a spin column to a vacuum manifold.
- 2) Load up to 750 μ l of the binding mixture into a column.
- 3) Turn on vacuum and allow the binding mixture to flow through the column completely.
- 4) Repeat steps 2 and 3 until all of the mixture has been processed.
- 5) Switch off vacuum.

Column Wash

- 1) Add 750 μ l of Wash Buffer into the spin column.
- 2) Turn on vacuum and allow the Wash Buffer to flow through the column completely.
- 3) Switch off vacuum and transfer the spin column into the receiving tube.
- 4) Centrifuge at 16,000 \times g for 1 minute.

DNA Elution

- 1) Transfer the spin column into a clean microcentrifuge tube.
- 2) Add 15-50 μ l of Elution Buffer to the center of the membrane in the spin column.
- 3) Allow the column to sit for 1 minute, then centrifuge at 16,000 \times g for 1 minute.
- 4) Discard the spin column. Mix the purified sample well by pipeting or brief vortexing. Store at -20°C.

Notes

- 1) Use agarose gel electrophoresis analysis along with a standard of known concentration or fluorescence dsDNA quantitation assay to determine DNA concentration of the purified products.
- 2) Visit <http://www.edgebio.com> for FAQ and Troubleshooting guide information.