PureLink[™] Quick Gel Extraction Kit USER GUIDE

For purifying DNA fragments from agarose gels

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D	28 March 2025	 Updated to the current document template, with associated updates to the limited license information, warranty, trademarks, and logos. The contents and storage table was updated to include additional instructions for storage of purification columns. 	
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Product information

Product description

The PureLink[™] Quick Gel Extraction Kit allows rapid and efficient purification of DNA fragments from TAE or TBE agarose gels of various percentages. DNA can be extracted and purified from agarose gels with different melting points in ~30 minutes using PureLink[™] silica membrane-based Quick Gel Extraction Columns by either centrifugation or vacuum protocols.

Note: The PureLink[™] Quick Gel Extraction Kit is **not** designed to purify supercoiled plasmid DNA or genomic DNA from agarose gels. **Only linear DNA fragments can be purified from gels using these kits.**

Features

The PureLink[™] Quick Gel Extraction Kit offers the following advantages:

- Purify DNA fragments from TAE and TBE agarose gels of various percentages and melting points.
- Complete the procedure in \sim 30 minutes.
- Easily purify DNA fragments from 40 bp to 10 kb from gels.
- Obtain high recovery of DNA fragments.
- Bind and purify up to 15 µg DNA with one column.
- Purify DNA fragments that are high-quality and show reliable performance in PCR, restriction enzyme digestion, cloning, and labeling.
- The purified DNA is suitable for various downstream applications, including:
 - DNA sequencing
 - Cloning
 - Restriction enzyme digestion
 - PCR reactions
 - Labeling



Kit specifications

Parmeter	Specification
Starting Material	≤400 mg agarose gel slice
Binding Capacity	\sim 15 µg DNA per column
Column Reservoir Capacity	850 μL
Wash Tube Capacity	2.0 mL
Elution Tube Capacity	1.5 mL–1.7 mL
Centrifuge Compatibility	12,000 × <i>g</i>
Elution Volume	50 μL
DNA Recovery	Up to 95% (dependent on DNA fragment size)
DNA Fragment Size	40 bp to 10 kb

Contents and storage

All components of the PureLink[™] Quick Gel Extraction Kit are shipped at room temperature. Upon receipt, store all components at room temperature.

Note: For better long-term performance store Quick Gel Extraction Columns in Wash Tubes at 2°C to 8°C.

Component	Cat. no. K210012 (50 reactions)	Cat. no. K210025 (250 reactions)
Gel Solubilization Buffer (L3)	2 × 90 mL	10 × 90 mL
Wash Buffer (W1)	16 mL	5 × 16 mL
Elution Buffer (E5) (10 mM Tris-HCl, pH 8.5)	15 mL	5 × 15 mL
PureLink™ Quick Gel Extraction Columns in Wash Tubes	50 each	5 × 50 each
PureLink [™] Elution Tubes	50 each	5 × 50 each

Gel extraction and purification workflow

To purify DNA fragments from agarose gel using the PureLink[™] Quick Gel Extraction Kit , excise the desired band and dissolve the TAE or TBE agarose gel. Place the dissolved gel into the Quick Gel Extraction Column to bind the DNA to a silica membrane, then purify and elute the DNA fragment by centrifugation or vacuum protocol.



Figure 1 Centrifugation and Vacuum workflows for the PureLink™ Quick Gel Extraction Kit

Methods



Gel extraction protocol

The following protocol is used to purify DNA fragments from various percentage TAE and TBE agarose gels and from agarose gels with different melting points.

- The PureLink[™] Quick Gel Extraction Kit is suitable for purifying DNA fragments from 40 bp to 10 kb in size. You may purify larger DNA fragments from gels using these kits; however, this may result in lower DNA recovery.
- The PureLink[™] Quick Gel Extraction Kit is **not** designed to purify supercoiled plasmid DNA or genomic DNA from agarose gels. **Only linear DNA fragments can be purified from gels using these kits.**
- For best results, use one Quick Gel Extraction Column per 10 µg of DNA fragment loaded onto the gel, but up to 15 µg of DNA can be purified with each column. Use additional Quick Gel Extraction Columns if larger amounts of DNA are to be purified.

Before first use of the kit

Before beginning, prepare Wash Buffer (W1) with ethanol according to the following directions.

Kit K2100-25 (250 reactions) contains five 16-mL bottles of Wash Buffer (W1). Repeat the procedure for each bottle of Wash Buffer (W1).

Add ethanol to Wash Buffer (W1)

- 1. Add 64 mL of 96–100% ethanol to the bottle of Wash Buffer (W1).
- 2. Mark the checkbox on the Wash Buffer label to indicate that ethanol was added to the bottle.
- 3. Store the Wash Buffer with ethanol at room temperature.

Prepare the gel slice

General guidelines

- Ensure that the DNA fragment of interest is **completely** separated from other DNA fragments on the agarose gel.
- To minimize DNA degradation, always wear gloves and use a clean razor blade to cut the gel slice.
- Maintain a sterile working environment when handling DNA to avoid any contamination from DNases.
- Ensure that no DNase is introduced into the sterile solutions supplied with the kit.
- Ensure that all equipment that comes in contact with DNA is sterile, including pipette tips and tubes.



CAUTION!

- The Gel Solubilization Buffer (L3) contains guanidine isothiocyanate (an irritant). This chemical is harmful when in contact with the skin, or when it is inhaled or ingested.
- **Do not** add bleach or acidic solutions directly to solutions or sample preparation waste that contains guanidine isothiocyanate, as reactive compounds and toxic gases are formed.

Always wear a laboratory coat, gloves, and safety glasses when handling this chemical. Dispose of the buffer and chemicals in appropriate waste containers.

Required materials not supplied

- Agarose gel containing the DNA fragment
- Weighing paper or weigh trays
- Scale (sensitive to 0.001 g)
- Water bath or heat block set at 50°C
- 1.5-mL or 5 mL-polypropylene microcentrifuge tubes
- Clean, sharp razor blade
- (Optional:) 100% isopropanol



Excise the DNA band

After completion of agarose gel electrophoresis:

- 1. Excise the area of the gel with the desired DNA band using a clean, sharp razor blade. Minimize the amount of agarose surrounding the DNA band.
- **2.** Weigh the gel slice containing the DNA using a scale sensitive to 0.001 g, then place up to 400 mg of the excised gel slice into a polypropylene microcentrifuge tube.

Item	≤2% agarose gels	>2% agarose gels
Microcentrifuge tube	1.5-mL tube	5-mL

Note: If the gel slice with the desired DNA exceeds 400 mg, cut the gel into smaller slices so that no one piece exceeds 400 mg. Place each additional gel slice created into a separate microcentrifuge tube.

3. Proceed to "Dissolve the gel".

Dissolve the gel

1. Add Gel Solubilization Buffer (L3). to the excised gel slice containing the DNA fragment ("Excise the DNA band" on page 10).

Component	≤2% agarose gels	>2% agarose gels	
	3 volumes buffer:1 volume of gel	6 volumes buffer:1 volume of gel	
Gel Solubilization Buffer (L3)	1.2 mL	2.4 mL	

Note: For example, add 1.2 mL or 2.4 mL of Gel Solubilization Buffer for a 400 mg gel slice.

2. Incubate the tube(s) containing the excised gel slice at 50°C for at least 10 minutes in a water bath or heat block.

Invert the tube by hand every 3 minutes to mix and ensure gel dissolution.

Note: High concentration gels (>2% agarose) or large gel slices can take longer than 10 minutes to dissolve.

- 3. Incubate the tube for an additional 5 minutes after the gel slice appears to have dissolved.
- 4. (*Optional:*) For optimal DNA yields, add 1 gel volume of isopropanol to the dissolved gel slice. Mix well.
- 5. Proceed to "Purify DNA by centrifuge" on page 11, or "Purify DNA by vacuum" on page 12.

Purify DNA by centrifuge

This procedure is designed for purifying DNA fragments in approximately 30 minutes using a centrifuge.

Required materials not supplied

You will need the following items:

- 96–100% ethanol
- Microcentrifuge capable of >10,000 × g
- DNase-free pipettes and tips

Bind DNA

1. Pipet the dissolved gel piece containing the DNA fragment of interest (see "Dissolve the gel") onto the center of a Quick Gel Extraction Column inside a Wash Tube.

Note: Do not load >400 mg dissolved agarose per Quick Gel Extraction Column.

2. Centrifuge at $12,000 \times g$ for 1 minute. Discard the flow-through and replace the Quick Gel Extraction Column into the Wash Tube.

Wash DNA

- 1. Add 500 µL Wash Buffer (W1), containing ethanol (see page 8) to the Quick Gel Extraction Column.
- **2.** Centrifuge at $12,000 \times g$ for 1 minute. Discard the flow-through and replace the column into the Wash Tube.
- **3.** Centrifuge the column again at maximum speed for 1–2 minutes to remove any residual Wash Buffer and ethanol. Discard the Wash Tube and place the Quick Gel Extraction Column into an Elution Tube.

Elute DNA

- 1. Add 50 µL Elution Buffer (E5) to the center of the Quick Gel Extraction Column.
- 2. Incubate the column for 1 minute at room temperature.
- **3.** Centrifuge the Column at $12,000 \times g$ for 1 minute. Discard the Quick Gel Extraction Column. *The Elution Tube contains the purified DNA.*
- 4. Store the purified DNA (see "Store the purified DNA" on page 13), or proceed to downstream application of choice.

Purify DNA by vacuum

This procedure is designed for purifying DNA fragments in approximately 30 minutes using a vacuum manifold. **Perform all vacuum operations at room temperature.**

Required materials not supplied

You will need the following items:

- 96-100% ethanol
- Vacuum manifold (with vacuum source capable of producing pressure of 15–20 in. Hg or –500 to –700 mbar)
- DNase-free pipettes and tips

Bind DNA

- 1. Assemble the vacuum manifold according to the manufacturer's instructions.
- 2. Attach a Quick Gel Extraction Column to the vacuum manifold.
- **3.** Pipet the dissolved gel piece containing the DNA fragment of interest (see "Dissolve the gel") onto the center of the silica membrane of the Quick Gel Extraction Column. Apply the vacuum until all of the liquid passes through the column, then switch off the vacuum source.

Note: Do not load more than 400 mg dissolved agarose per column.

Wash DNA

- 1. Add 500–700 µL Wash Buffer (W1) containing ethanol (see page 8) to the center of the column.
- **2.** Apply the vacuum until all of the liquid passes through the column, then switch off the vacuum. Remove the Quick Gel Extraction Column from the vacuum and place it into a Wash Tube.
- **3.** Centrifuge the column with the Wash Tube at maximum speed for 1–2 minutes to remove any residual Wash Buffer and ethanol. Discard the flow-through and the Wash Tube. Place the Quick Gel Extraction Column into a Elution Tube.

Elute DNA

- 1. Add 50 µL Elution Buffer (E5) to the center of the Quick Gel Extraction Column.
- 2. Incubate the column for 1 minute at room temperature.
- **3.** Centrifuge the column at $12,000 \times g$ for 1 minute. Discard the Quick Gel Extraction Column. *The Elution Tube contains the purified DNA.*
- 4. Store the purified DNA (see "Store the purified DNA" on page 13), or proceed to downstream application of choice.

Store the purified DNA

Store the purified DNA at 4°C for immediate use, or prepare aliquots of the DNA and store at –20°C for long-term storage. Avoid repeated freeze thaw cycles of the DNA.

Estimate DNA yield and quality

After DNA purification, determine the quantity and quality of the purified DNA.

Determine DNA yield with Qubit[™] DNA Assay Kits

Measure DNA concentration using Qubit™ DNA Assay Kits or UV absorbance at 260 nm.

The Qubit[™] DNA Assay Kits (see "Additional products" on page 15 for ordering information) provide a rapid, sensitive, and specific method for measuring dsDNA concentration with minimal interference from RNA, protein, ssDNA (primers), or other common contaminants that affect UV absorbance.

The kit contains a state-of-the-art quantitation reagent, pre-diluted standards for standard curve, and a pre-made buffer. For optimal results, perform the quantitation using a Qubit[™] 4 Fluorometer (see "Additional products" on page 15). You can also use standard fluorescent microplate readers. Follow manufacturer's recommendations to perform the assay.

Estimate DNA quality

Typically, DNA isolated using the PureLink[™] Quick Gel Extraction Kit has an A₂₆₀/A₂₈₀ >1.8 when samples are diluted in Tris-HCl pH 7.5–8.5 indicating that the DNA is substantially free of contaminants that would otherwise affect UV absorbance.

To confirm the integrity and the size of the purified DNA fragment, perform agarose gel electrophoresis.

Expected results

DNA fragments of various sizes were purified using PureLink[™] Quick Gel Extraction Kit as described in the manual. The concentration of purified DNA was measured using Qubit[™] DNA Assay Kits (see "Additional products" on page 15 for ordering information). The DNA yields are listed in the table.

DNA Size	Amount Loaded	% DNA Yield ^[1]	
400 bp	0.5 µg	>85%	
740 bp	1 µg	>85%	
2.9 kb	1 µg	>85%	

^[1] Note: The DNA yield varies with the fragment size, amount of DNA loaded on the gel, gel slice size, elution volume, and incubation time.



Troubleshooting

Observation	Cause	Solution	
Low DNA yield	Incorrect ratio of gel to Gel Solubilization Buffer	Ensure that the correct volume of Gel Solubilization Buffer (L3) is added for every 1 volume of gel used, based on the agarose gel percentage (see "Dissolve the gel" on page 10).	
	Incomplete	Verify that the temperature of water bath or heat block is at 50°C.	
	solubilization of gel piece	Cut large gel slices into several pieces to accelerate the gel dissolution.	
		Mix gel slice in the buffer every 3 minutes during the dissolution step.	
	DNA fragment is too large	Increase the incubation time for elution to >10 minutes.	
	DNA is supercoiled	This kit is not designed to purify supercoiled plasmid DNA from agarose gels.	
Low A _{260/230} ratio	Guanidine carryover from the Gel Solubilization Buffer	 Do not get any buffer solution in the cap area of the tube. Add a second wash step with Wash Buffer (W1): After your first wash with Wash Buffer, followed by centrifugation: a. Add another 500–700 µL Wash Buffer, containing ethanol. b. Centrifuge at 12,000 × g. Discard the flow-through and return the column into the Wash Tube. c. Centrifuge at maximum speed for 2–3 minutes to remove residual Wash Buffer and ethanol. 	
Downstream enzymatic reactions are inhibited	Residual ethanol in the purified DNA	Traces of ethanol from the Wash Buffer can inhibit downstream enzymatic reactions. To remove Wash Buffer, discard Wash Buffer flow through from the Wash Tube. Place the column into the Wash Tube and centrifuge the column at $>12,000 \times g$ for 2–3 minutes to completely dry the column.	



Related products

Additional products

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

Product	Quantity	Catalog no.
Qubit™ dsDNA Assay Kit, High Sensitivity	500 assays	Q32854
Qubit™ dsDNA Assay Kit, Broad-Range	500 assays	Q32853
Qubit™ 4 Fluorometer	1 each	Q33238

E-Gel[™] agarose gels and DNA ladders

E-Gel[™] Agarose Gels are bufferless, pre-cast agarose gels designed for fast, convenient electrophoresis of DNA samples. E-Gel[™] agarose gels are available in different agarose percentages and well formats for your convenience.

A large variety of DNA ladders are available from Thermo Fisher Scientific for sizing DNA. For more details on these products, visit **thermofisher.com**.







WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, and so on). To obtain SDSs, visit thermofisher.com/support.

Chemical safety



WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below. Consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with sufficient ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if needed) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.



WARNING! HAZARDOUS WASTE (from instruments). Waste produced by the instrument is potentially hazardous. Follow the guidelines noted in the preceding General Chemical Handling warning.



WARNING! 4L Reagent and Waste Bottle Safety. Four-liter reagent and waste bottles can crack and leak. Each 4-liter bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position.

Biological hazard safety

WARNING! Potential Biohazard. Depending on the samples used on this instrument, the surface may be considered a biohazard. Use appropriate decontamination methods when working with biohazards.



WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Conduct all work in properly equipped facilities with the appropriate safety equipment (for example, physical containment devices). Safety equipment can also include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 6th Edition, HHS Publication No. (CDC) 300859, Revised June 2020 cdc.gov/labs/bmbl
- Laboratory biosafety manual, fourth edition. Geneva: World Health Organization; 2020 (Laboratory biosafety manual, fourth edition and associated monographs)
 who.int/publications/i/item/9789240011311



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 - Certificates of Analysis
 - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Limited product warranty

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