

Agarose Gel Extraction Kit

DNA Extraction by silica-gel membrane adsorption

DNA Cleanup

	Cat.-No.	Amount
	PP-202XS	10 preparations
	PP-202S	50 preparations
	PP-202L	250 preparations

For *in vitro* use only
Quality guaranteed for 12 months
Store at room temperature

Description

Agarose Gel Extraction Kit is designed for high-yield recovery of DNA from agarose gel with simultaneous removal of primer-dimers, primers, nucleotides, proteins, salt, agarose, ethidium bromide, and other impurities. The preparation is based on a silica-membrane technology for binding DNA in high-salt and elution in low-salt buffer. The kit provides a simple and efficient way to purify DNA in a size range between 100 bp and 10 kbp. It requires no organic extractions or precipitation and guarantees high and stable recovery rates.

Kit contents

Extraction Buffer
Activation Buffer
Washing Buffer (before use, add 96-99% Ethanol as indicated on the bottle)
Elution Buffer
Spin Columns and 2 ml Collection Tubes

To be provided by you

96-99% Ethanol
Isopropanol (optional)
1.5 ml microtubes

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Preparation procedure

The agarose gel is dissolved in the chaotropic Extraction Buffer followed by a simple binding, washing, and eluting procedure. Before start, add 96-99% Ethanol as indicated on the bottle to the Washing Buffer. The additional use of Isopropanol (not included in the kit) enhances yield and is recommended for fragments smaller than 200 bp or larger than 5 kbp. The optional primary washing step minimizes the salt content of the purification product but may significantly reduce the yield of DNA fragments <200 bp.

1. Excision of the Gel

- Cut the area of gel containing the DNA fragment.
- Transfer the excised gel to a clean 1.5 ml microtube.

2. Sample Preparation

- Add 3 volumes of *Extraction Buffer* to 1 volume of the sliced gel. For example, add 300 µl *Extraction Buffer* to each 100 mg (approx. 100 µl) gel. For gels containing >2.5% agarose, add 6 volumes of *Extraction Buffer* per gel volume.
- Incubate at 60°C for 10 min with occasional mixing to ensure gel dissolution.
- For DNA fragment sizes smaller than 200 bp or larger than 5 kbp and to enhance yield add 1 volume *Isopropanol* per gel volume to the dissolved gel and mix well.

3. Column Activation

- Place a *Spin Column* into a 2 ml collection tube
- Add 100 µl of *Activation Buffer* into the *Spin Column*
- Centrifuge at 10,000 g for 30 sec in a micro-centrifuge.

4. Column Loading

- Apply the sample mixture from step 2 into the *activated Spin Column*.
- Centrifuge at 10,000 g for 30 sec in a micro-centrifuge.
- Discard the flow-through.

5. Optional Primary Washing (only for DNA >200 bp)

- Place the DNA loaded *Spin Colum* into the used 2 ml tube.
- Apply 500 µl of *Washing Buffer* to the *Spin Column*.

- Centrifuge at 10,000 g for 1 min and discard the flow-through.

6. Column Washing

- Add 500 µl of *Washing Buffer* to the *Spin Column*.
- Centrifuge at 10,000 g for 2 min and discard the flow-through.

7. Elution

- Place the *Spin Column* into a clean 1.5 ml microtube (not provided in the kit).
- Add 30-50 µl *Elution Buffer* or dd-water to the center of the column membrane.
- Incubate for 1 min at room temperature.
- Centrifuge at 10,000 g for 1 min to elute DNA.