Manual

IFTA AG
Certified QMS according to
DIN EN ISO 9001:2000-12
Reg. No. IC 03214 034

Jena Bioscience

PCR Purification Kit

DNA Purification by silica-gel membrane adsorption

DNA Cleanup

	Cat.-No.	Amount
	PP-201XS	10 preparations
	PP-201S	50 preparations
	PP-201L	250 preparations

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

Description

PCR Purification Kit is designed for the work-up of PCR reactions (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 linear or circular DNA and is optimized for working with DNA amounts of up to 20 µg. It requires no organic extractions or precipitation and guarantees high and stable recovery rates.

Kit contents

Binding Buffer

Activation Buffer

Washing Buffer (before use, add 96-99% Ethanol as indicated on the bottle)

Elution Buffer

Spin Columns

2 ml Collection Tubes

To be provided by you

96-99% Ethanol

Isopropanol (for high yield sample preparation)

1.5 ml microtubes

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Preparation procedure
The DNA purification follows a simple binding, washing, and eluting procedure. Before start, add 96-99% Ethanol to the Washing Buffer as indicated on the bottle:

Cat.-No.	Amount	Ethanol to be added	Final volume
PP-201XS	10 preps	12 ml	15 ml
PP-201S	50 preps	60 ml	75 ml
PP-201L	250 preps	300 ml	375 ml

The additional use of Isopropanol is recommended for fragments smaller than 200 bp or larger than 5 kbp.
The optional secondary washing step minimizes the salt content of the purification product but may significantly reduce the yield of DNA fragments <200 bp.

1a Standard Sample Preparation
For DNA fragment sizes in the range of 200 bp to 5 kbp:

- Add 5 volumes of *Binding Buffer* to 1 volume of DNA sample and mix well. For example, if the volume of your DNA sample is 50 µl, add 250 µl *Binding Buffer*.

1b High Yield Sample Preparation
For DNA fragment sizes smaller than 200 bp or larger than 5 kbp:

- Add 3 volumes *Binding Buffer* and 2 volumes of Isopropanol to the PCR sample. For example, if the volume of your DNA sample is 50 µl, add 150 µl Binding Buffer and 100 µl *Isopropanol*.

2 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.

3 Column Loading

- Apply the sample mixture from step 1 into the *activated Spin Column*.
- Centrifuge at 10,000 g for 30 sec in a micro-centrifuge.
- Discard the flow-through.

4 Column Washing

- Place the DNA loaded *Spin Colum* into the used 2 ml tube.
- Apply 750 µl of *Washing Buffer* to the *Spin Column*.
- Centrifuge at 10,000 g for 30 sec and discard the flow-through.

  Optional Secondary Washing: Recommended only for DNA >200 bp, if highly purified DNA (for DNA sequencing, transfection etc.) is required.
  - o Add 750 µl of *Washing Buffer* to the Spin Column.
  - o Centrifuge at 10,000 g for 30 sec and discard the flow-through.

- Centrifuge again for 2 min to remove residual *Washing Buffer*.

5 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 at room temperature for 1 min.
- Centrifuge at 10,000 g for 1 min to elute DNA.