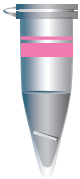
SMARTPURE Protocol

Gel kit
SK-GEPU-100

Sample Preparation



1. Excise **DNA fragment** out of the gel with a clean scalpel.
➜ Minimize the size of the gel slice by removing extra agarose.
2. Weigh the gel slice and transfer it to a clean tube.
3. Add 3 volumes of **Extraction Buffer** to 1 volume of gel slice.
➜ e.g.300 µL of Extraction Buffer to 100 mg of gel. The gel slice should be ≤400 mg per assay.
➜ The Yellow color indicates a pH <7.8
4. Incubate (5-10 min) at 50 °C until the gel melts and vortex the tube every 2-3 min during the incubation.
➜ If the color of the mixture becomes purple add 10 µl of 3 M sodium acetate (pH5.0) and mix. The color will return yellow.
Optional: if DNA Fragment <500 bp or > 4 Kb, add 1 volume of isopropanol to 1 volume of gel and mix well.

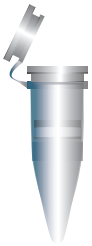
Binding

5. Place a Smart Pure column into a collection tube (2mL)
6. Apply **Sample** on the SmartPure column.
7. Centrifuge for 1 min at 6 000 × g.
8. Discard the flow-through.
➜ If the sample volume is higher than 750 µL, reload the remaining volume on the same SmartPure column and spin again.
9. Add 500 µL of **Extraction Buffer** on the SmartPure column
10. Centrifuge for 30-60 sec at 12 000 × g.
11. Discard the flow-through.

Washing

12. Add 750 µL of **Wash Buffer** to the SmartPure column
13. Centrifuge for 30-60 sec at 12 000 × g.
14. Discard the flow-through.
➜ If the DNA will be used for salt sensitive applications, let the spin column stand for 2-5 min after the addition of the Wash Buffer before centrifuging.
15. Centrifuge for 1 min at 12 000 × g to remove residual liquid.

Elution



16. Transfer the SmartPure column to a sterile 1.5ml microcentrifuge tube.
17. Add 30-100 µL of **Elution Buffer**, H_2O or TE Buffer to the SmartPure column and let it stand for 1 min at room temperature.
➜ The volume of elution buffer can be adjusted according to needs (min 20 µL).

Purified DNA

18. Centrifuge for 1 min at 12 000 × g to elute the DNA in the microcentrifuge tube.
➜ The purified DNA fragment can be used directly or stored at -20°C for long term storage.

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The SmartPure Gel Kit ensures an easy, fast and effective extraction and purification of DNA fragments from up to 3% standard agarose gel in TAE or TBE buffer.

First use

- Add to the SmartPure Wash Buffer 40 mL of ethanol and mix well.

Kit components (100 preps)

Component	Amount
SmartPure Extraction Buffer	170 mL
SmartPure Wash Buffer	2x10 mL
SmartPure Elution Buffer	20 mL
SmartPure Column	100

Material not supplied

- Sterile 1.5 mL microcentrifuge tubes
- 10/100/1000 µL tips
- Microcentrifuge (14 000 x g)
- Vortex mixer
- Absolute ethanol
- Isopropanol

Technical Information

Column Volume	750 µL
Sample Volume	up to 400 mg gel slice
DNA size range	from 60 bp to 23 kb
Elution Recovery	≥ 99%
Working time	16 min/2 samples
Agarose Gel	
Gel Type	Standard or high/low melting agarose
Electrophoresis buffer	TAE or TBE buffer
Incubation temperature	50°C for low-melting agarose 55°C for standard agarose.

Shipping & Storage conditions

The SmartPure Gel Kit is shipped at room temperature.

The SmartPure Gel Kit is stable for 18 months if stored in a dry environment at room temperature (15°C-25°C).

Troubleshooting

No recovery

Please check whether the ethanol had been added to the Wash Buffer bottle.

Low recovery

1] The extraction buffer is acidic. The pH may increase during the gel melting process and could lead to inefficient DNA binding. Please add 0.1 volume 3M sodium acetate (pH5.0).

2] A non fresh electrophoresis buffer may lead to a low recovery

3] Incubate the Elution Buffer at 30~60°C to increase the yields.

Absorbance problem

Please use the Elution Buffer to adjust zero value and dilute the sample.

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