

**Product Code:** 10088

Research Use Only

Store at Room Temp.

**Size:** 100 preparations

## Description

AddPrep Fragment Purification Kit offer simple, rapid and cost-effective method from PCR product, enzyme reaction mixture, and agarose gel in TAE or TBE buffer system. This kit is designed for the preparation of up 50~90% recovery yield from original reaction mixture. Purified DNA with mini kit is immediately used in ligation, sequencing and other downstream applications. Phenol extraction and ethanol precipitation are not required and high-quality purified DNA is eluted in a small volume of Elution Buffer.

## Kit Components

Solution & Material	Size	Solution & Material	Size
Spin column	100 ea	Washing	15 ml (Add Ethanol 60 ml)
Binding	60 ml	Elution	20 ml

## Storage and Stability

AddPrep Fragment Purification Kit is stable for 1.5 years when stored in a constant temperature 15 ~ 35°C.

## Before You Begin

1. Add ethanol to Optional Solution and Washing Solution before use.
2. Check Binding Solution for salt precipitation and salt precipitant can be dissolved by warming at 50°C.

## Extraction Protocol from PCR product and enzyme reaction mixture

1. **Add 5 volumes of Binding Solution to 1 volume of the sample and mix well by vortex.**  
Example of the PCR reaction product is 50 µl, add 250 µl of Binding Buffer.
2. **Centrifuge the tube briefly, 8,000rpm~13,000rpm, 10sec. at room temperature.**
3. **Transfer the mixture to a spin column.**
4. **Centrifuge for 30 sec ~ 1 min at 13,000. Discard the flow-through and put the spin column into the collection tube.**
5. **Add 700µl Washing Solution into spin column and centrifuge for 30sec at 13,000 rpm.**  
Discard the flow-through and put the spin column into the collection tube.
6. **Centrifuge for an additional 1 ~ 2 min at 13,000 rpm to remove residual Washing Buffer in spin column and glass fiber filter.**  
Residual ethanol of Washing Solution may inhibitor subsequent enzymatic reaction and other downstream applications.
7. **Transfer the spin column to new 1.5 ml micro-centrifuge tube:** The 1.5 ml micro-centrifuge tube is not provided.
8. **Add 50 µl of Elution Solution to the center of the glass fiber filter in the column, let stand for 1 min at room temperature and centrifuge for 1 min at 13,000 rpm.**  
For larger fragment(>5kb), use pre-warmed (more than 50°C) Elution Buffer for good elution.

## Extraction Protocol from agarose gel

1. **Cut out the interesting DNA fragment with a sharp scalpel or razor blade:** Minimize gel volume by cutting gel slice as small as possible.
2. **Weigh the gel slice in a micro-centrifuge tube. Add 3 volumes of Binding Solution to 1 volume of gel:**  
If the weight of agarose gel slice is 100 mg, add 300 µl of Binding Solution and if the concentration of agarose gel is more than 1.5%, add 6 volumes of Binding Solution.
3. **Vortex the mixture and incubate at 55°C until the agarose gel is completely melted (5~10min):**  
To help the dissolving of agarose gel, vortex the tube every 2~3 min during the incubation. Centrifuge the tube briefly at room temperature.
4. **(Optional) Add 1 gel volume of isopropanol to the sample and vortex to mix:**  
Do not centrifuge after mixing well. This step increases the yield of DNA fragment <200bp or >5kb.
5. **Transfer the mixture to a Spin column.**
6. **Centrifuge for 1 min at 13,000 rpm. Discard the flow-through and re-inserting the spin column to the collection tube.**
7. **(Optional) Add 500 µl of Binding Solution to the column and centrifuge for 30 sec at 13,000 rpm. Discard the flow-through and re-inserting the spin column to the collection tube:** This step is complete removal of agarose for very sensitive applications.
8. **Add 700µl Washing Buffer into spin column and centrifuge for 30sec at 13,000 rpm.**  
Discard the flow-through and put the spin column into the collection tube.
9. **Centrifuge for an additional 1 ~ 2 min at 13,000 rpm to remove residual Washing Buffer in spin column and glass fiber filter.**  
Residual ethanol of Washing Buffer may inhibitor subsequent enzymatic reaction and other downstream applications.
10. **Transfer the spin column to new 1.5 ml micro-centrifuge tube:** The 1.5 ml micro-centrifuge tube is not provided.
11. **Add 50 µl of Elution Buffer to the center of the glass fiber filter in the column, let stand for 1 min at room temperature and centrifuge for 1 min at 13,000 rpm.** For larger fragment(>5kb), use pre-warmed (more than 50°C) Elution Buffer for good elution.