ISOSPIN PCR Product

Manual (Ver. 03)

Code No. 315-08001

NIPPON GENE CO., LTD.

I Description

The ISOSPIN PCR Product enables a rapid and effective purification of DNA fragments from amplification reactions.

II Kit components

Component	(100 preps)	Note
ISB Buffer	100 ml x 1	
ISW Buffer	100 ml x 1	(includes ethanol)*
ISE Buffer	10 ml x 1	10 mM Tris-HCl (pH 8.5)
Spin Column (a Spin Column and Collection Tube set)	50 sets x 2	

* Keep the buffer bottle tightly closed after use.

III Storage conditions

All the kit components can be stored at room temperature (15-25°C).

IV Precautions

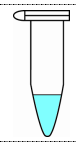
- The kit is intended research use only.

V Protocol

Material not supplied

- Micropipette
- Pipette tips
- 1.5 mL microcentrifuge tubes
- Microcentrifuge

Protocol



1. Add 1 volume of **Sample** (PCR reaction) in a 1.5 mL microcentrifuge tube. Add 5 volumes of **ISB Buffer**, and mix well by inverting. Spin down lightly.
   Example) Add 250 µl of ISB Buffer to 50 µl of the Sample.

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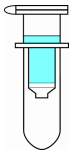

2. Prepare a **Spin Column** (a Spin Column and Collection Tube set). Apply the **Mixture** to the Spin Column.
3. Centrifuge for 1 min at 12,000 x g at room temperature.
4. Discard the flow-through. Place the Spin Column back into the same Collection Tube.
   Note) If there are the Mixture left over, apply the remaining Mixture to the Spin Column, and repeat 3 and 4.

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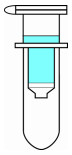

5. Apply 750 µl of **ISW Buffer** to the Spin Column.
6. Centrifuge for 1 min at 12,000 x g at room temperature.
7. Discard the flow-through. Place the Spin Column back into the same Collection Tube.

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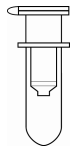

8. Centrifuge for 1 min at 12,000 x g at room temperature to remove residual liquid.

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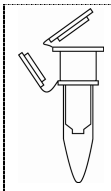
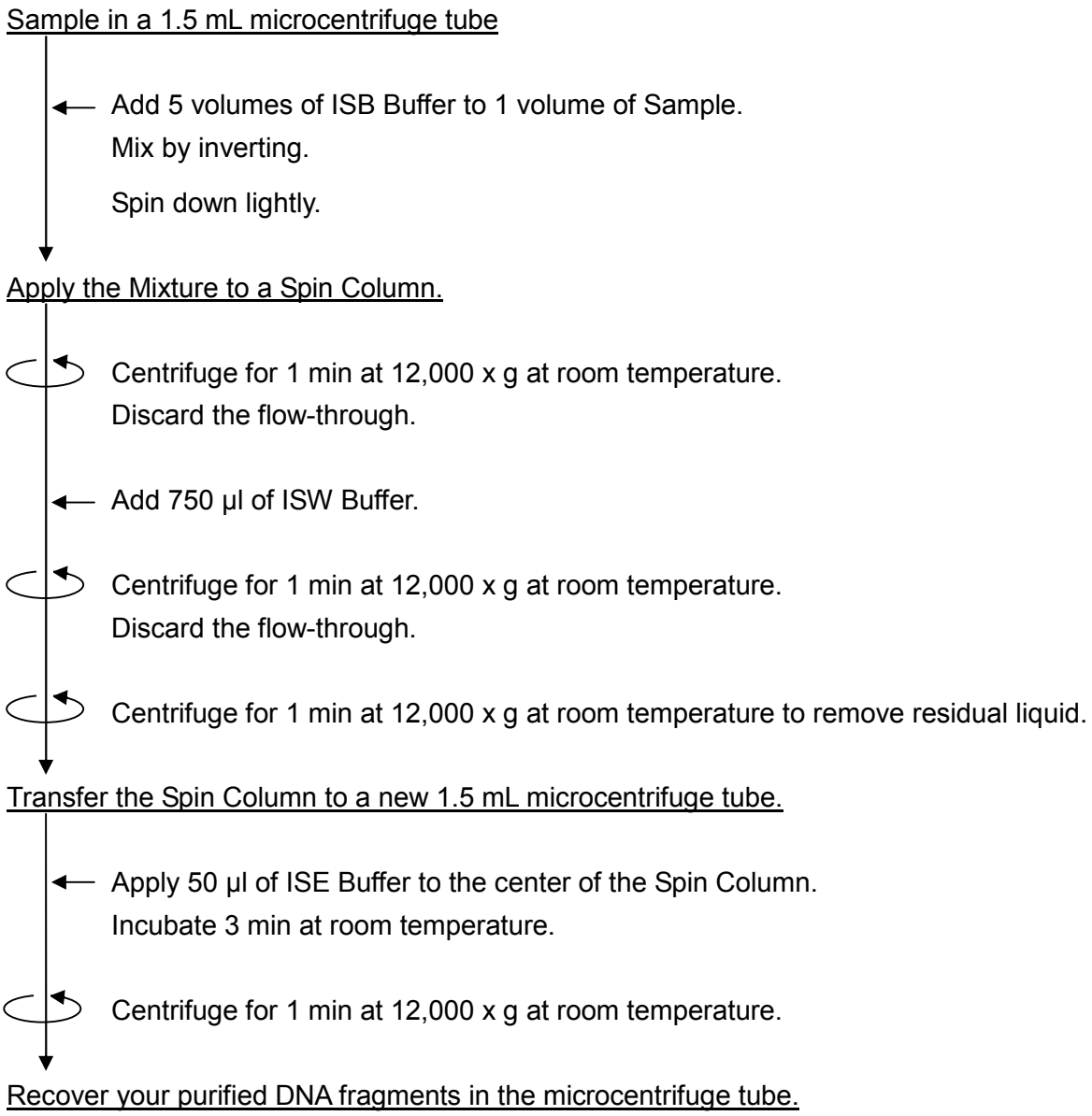


9. Transfer the Spin Column to a new 1.5 mL microcentrifuge tube.
10. Add 50 µl of **ISE Buffer** to the center of the Spin Column. Incubate 3 min at room temperature.
    Note) Nuclease-free water or TE (pH8.0) can also be used to elute the DNA in place of ISE Buffer (10 mM Tris-HCl, pH 8.5).
11. Centrifuge for 1 min at 12,000 x g at room temperature.
12. Recover your purified DNA fragments in the microcentrifuge tube.
    Note) The DNA fragments can be used directly or stored at -20°C for long term storage.

Simplified Protocol



VI Troubleshooting

Low DNA Yield

Ensure the ISE Buffer is applied to the center of the Spin Column so that elution is efficient. Larger elution volumes can increase yield of DNA at the cost of dilution of the sample.

Low DNA Performance

Please repeat wash step (Step 5, 6 and 7). 80% Ethanol can also be used to wash the DNA in place of ISW Buffer.

If ethanol has been carried-over, spin for 5 min, instead of 1 min in Step 8.

Sheared DNA

Mix well by inverting or flicking the tube. Do not vortex.

VII Data

Technical Information

Binding Capacity:	20 μg
Minimum Elution Volume:	10 μl
Typical DNA Recovery:	60-95% (100 bp to 20 kbp)
Column Volume:	900 μl
Removable Primer Size:	≤ 40 mer

The information in the descriptions of the products may be changed without prior notification.

NIPPON GENE CO., LTD.

If you have any questions, please contact us by web form.

http://www.nippongene.com/

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