

[Product Name] MagPure A4 XP

[Product specifications] 5ml, 50ml, 500ml

[Intended Use]

MagPure A4 XP utilize magen's solid-phase paramagnetic bead tehcnology for high-throughput purification of PCR amplicons. AmPure utilizes an optimized buffer to selectively bind PCR amplicons 100bp and larger to paramagnetic beads. Excess primes, nucleotides, salts and enzymes can be removed using a simple washing procedure. The resulting purified PCR product is essentially free of contamintants.

[Principle]

This product is based on the purification method of high binding magnetic particles. PCR amplicons mix with MagPure A4 XP, 100bp and larger DNA binds to magnetic beads. Excess primes, nucleotides, salts and enzymes can be removed using a simple washing procedure and finally DNA was eluted by Elution Buffer or Water.

[Main Composition]

Cat.No.	BP-2	BP-5	BP-50	BP-500
MagPure A4 XP	2 ml	5 ml	50 ml	500 ml

【Storage conditions and Validity】

MagPure A4 XP should be stored at $2-8^{\circ}$ C upon arrival and is stable up to 18 months under the condition. However, short-term storage (up to 4 weeks) at room temperature (15-25°C) does not affect its performance. — Shake the reagent well before use. It should appear homogenous and consistent in color

DO NOT FREEZE

【Preparation before Use】

- Elution Buffer: Reagent grade water, 0.1 mM EDTA (pH 8.0), or 0.5 mM EDTA (pH 8.0). The optimal elution buffer will vary depending on dye chemistry and reaction conditions.
- magnetic plate
- For 96 well format: Fresh 85% Ethanol made with Non-denatured Ethanol.

[Protocol]

- Determine whether or not a plate transfer is necessary.
 If the PCR reaction volume multiplied by 2.8 exceeds the volume of the PCR plate, a transfer to a 300 µL round bottom plate or a 1.2 mL deep-well plate is required.
- Shake the MagPure A4 XP bottle to resuspend any magnetic particles that may have settled.
 Then add MagPure A4 XP according to the sample reaction volume shown in Table.

Sample Reaction Volume (µL)	MagPure A4 XP (µL)		
1 OµL	18µL		
20µL	36µL		
50 _µ L	80µL		
100µL	180µL		

- Mix reagent and sample thoroughly by pipette mixing 10 times. Let the mixed samples incubate
 for 5 minutes at room temperature for maximum recovery.
 - This step binds DNA fragments 100 bp and larger to the magnetic beads. Pipette mixing is preferable to vortexing as it tends to be more reproducible. The color of the mixture should appear homogenous after mixing.
- 4. Place the reaction plate onto an Magnet Plate for 5 minutes to separate beads from the solution.

 IMPORTANT Wait for the solution to clear before proceeding to the next step.

- 5. Aspirate the cleared solution from the reaction plate and discard. Leave 5 μL of supernatant behind, otherwise beads are drawn out with the supernatant.
 - This step must be performed while the reaction plate is situated on the Magnet Plate. Do not disturb magnetic beads.
- Dispense 200µL of 70% ethanol to each well of the reaction plate and incubate for 30 seconds at room temperature. Aspirate out the ethanol and discard.

The beads are not drawn out easily when in alcohol, so it is not necessary to leave any supernatant behind. If the total volume of sample plus reagent exceeds 200 μ L, then use a wash volume of at least the volume of sample plus reagent.

- 7. Dispense 200µL of 70% ethanol to each well of the reaction plate and incubate for 30 seconds at room temperature. Aspirate out the ethanol and discard.
- 8. Let the reaction air-dry for 10 minutes at room temperature.

For fragments 10 kb and larger, do not over dry the bead ring (bead ring appears cracked if over dried) as this will significantly decrease elution efficiency.

9. Remove the reaction plate from the magnet plate, and then add 40 µL of elution buffer to each well of the reaction plate and pipette mix 10 times. Incubate for 2 minutes.

The liquid level will be high enough to contact the magnetic beads at a $40\mu L$ elution volume. A greater volume of elution buffer can be used, but using less than $40\,\mu L$ will require extra mixing (to ensure the liquid comes into contact with the beads), and may not be sufficient to elute the entire PCR product.

Place the reaction plate onto an Magnet Plate for 1 minute to separate beads from the solution.
 Transfer the eluate to a new plate.

Bead carryover into the final plate is usually not a cause for concern. The samples can be stored in the freezer with beads and the beads are inert in downstream enzymatic reactions. If bead carryover must be limited for any reason, $2~\mu L - 5~\mu L$ of eluate can be left behind in the original plate. In addition, a second transfer away from the beads is optional. To do so, place the final plate containing beads and eluate onto the magnet for 1 minute to separate the beads. Transfer the eluate into another clean plate.

This step should also be performed before placing the plate on the magnetic plate. If you are vortexing, use a medium speed and make sure the suspension is completely homogeneous before continuing. The volume of 85% ethanol needed has been calculated for common sequencing reaction volumes, as shown in Table.

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