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## Auto-Mag® PCR-Pure

Cat # AMD-S002

Version 2.0

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### Protocol for PCR clean-up (96-well Plate Format)

The following protocol exemplifies for removal of salts, primers, primer-dimers, dNTPs, from PCR reactions and selectively recovers amplicons greater than 100 bp.

#### Materials and Equipment to be supplied by User

- 100 % ethanol.
- Elution Buffer (diH<sub>2</sub>O, 10mM Tris-HCl pH 8.0 or TE buffer).
- For 96 well format: 96 well cycling plate.
- Appropriate magnetic separation device compatible with 96-well PCR plate.
- Well calibrated pipettor and Disposable pipette tips.

#### Before Starting

- Prepare fresh 80% ethanol (Prepare from absolute ethanol. Do not use denatured alcohol).
- Bring the Auto-Mag® PCR-Pure to room temperature for at least 30 min before use.
- Suspension Auto-Mag® PCR-Pure by vortexing.

#### Procedure

1. Completely float the Auto-Mag® PCR-Pure reagent until it appears homogeneous in color.
2. Add the appropriate volume of Auto-Mag® PCR-Pure reagent into the reaction plate containing 10-50ul PCR sample according to the instructions in the table below.

Table 2: Some common PCR reaction volumes and suggested Auto-Mag® PCR-Pure volumes

PCR Sample Volume (ul)	Auto-Mag® PCR-Pure Volume (ul)*
10	18
20	36
25	45
50	90

\* Formula used to calculate the volume of Auto-Mag® PCR-Pure reagent needed for PCR reaction: Auto-Mag® PCR-Pure reagent volume per reaction = 1.8 X PCR reaction volume.

3. Mix thoroughly the Auto-Mag® PCR-Pure reagent and PCR sample by pipetting up and down 10 times. Incubate the mixture for 10 minutes at room temperature.



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4. Place the sample plate on the 96 well magnetic separation device and allow the magnetic beads to settle on the magnet for 5 minutes or until the supernatant turns completely clear. Carefully remove and discard the supernatant by pipetting.

*Note: Do not disturb the attracted magnetic beads while aspirating the supernatant.*

5. Keep the sample plate on the magnet and add 200µl of 80% ethanol to each well and incubate for 30 seconds at room temperature. Carefully remove and discard the supernatant by pipetting.

*Note: Do not disturb the attracted magnetic beads while aspirating the supernatant. Significant bead loss will result in reduced yield.*

6. Repeat steps 5 for second 80% ethanol wash.

7. Keep the sample plate on the magnetic separation device and air dry the magnetic beads by incubating sample tube at room temperature for 5 minutes.

*Note: It is critical to completely remove all traces of alcohol but take caution in not over drying the beads as this will reduce the yield.*

8. Remove the sample plate from the magnetic separation device. Add 20-50µl of elution buffer (reagent grade water, Tris-HCl pH 8.0 or TE buffer) to each well and mix by pipetting up and down 10 times. Incubate for 5 minutes at room temperature.

9. Place the sample plate back on the magnetic separation device and wait 2 minutes or until the magnetic beads clear from solution.

10. Transfer the eluate (cleared supernatant) to a new plate and keep at -20°C for long term storage, or for subsequent applications.