omega BIO-TEK

innovations in nucleic acid isolation

Product Manual

Mag-Bind® Total Pure NGS

M1378-00	5 mL
M1378-01	50 mL
M1378-02	500 mL

Manual Date: November 2018
Revision Number: v2.1

For Research Use Only

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Mag-Bind® Total Pure NGS

Table of Contents

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Introduction and Principle

Omega Bio-tek's Mag-Bind® Total Pure NGS Kit allows rapid and reliable isolation of PCR products with high recovery rates. The system combines Omega Bio-tek's proprietary chemistries with the reversible nucleic acid-binding properties of magnetic beads to selectively bind DNA fragments 100 bp and larger and eliminate excess nucleotides, primers, and small, non-targeted amplification products, such as primer dimers. This kit is designed for both manual and fully automated purification of PCR samples. Purified PCR fragments can be used for microarrays, automated fluorescent DNA sequencing, restriction enzyme digestion, and other applications.

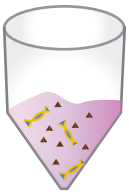
The Mag-Bind® Total Pure NGS magnetic particles technology provides a better solution for nucleic acid purification compared to centrifugation and vacuum-based technologies. The product can be easily scaled up while providing very user-friendly handling procedures. If using Mag-Bind® Total Pure NGS for the first time, please read this booklet to become familiar with the procedures. DNA products are first mixed and bind to Mag-Bind® Total Pure NGS particles. With two rapid wash steps, trace contaminants such as nucleotides, primers and small, non-targeted amplification products are removed. Pure DNA is eluted in Elution Buffer or water. Purified DNA can be directly used in downstream applications without the need for further purification. The volume of Mag-Bind® Total Pure NGS can be varied to selectivly bind difference size fragments. Mag-Bind® Total Pure NGS can be used with next generation sequencing library preparations protocols for reaction clean-ups at amounts specified by the library construction manufacturer.

New In this Edition

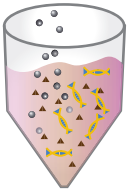
November 2018:

- Omega Bio-tek's logo has changed and minor edits were made to manual layout.

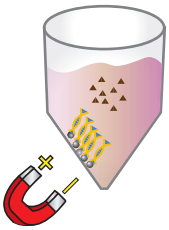
Illustrated Protocol



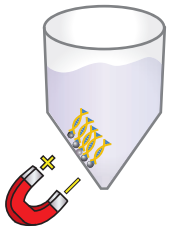
Determine the Reaction size



Add Mag-Bind® Total Pure NGS and Mix

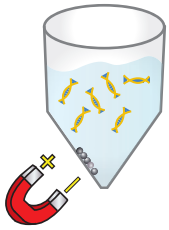


Magnetize and Remove Supernatant



Wash Twice with 70% Ethanol

Dry



Elute DNA

Kit Contents

Product Number	M1378-00	M1378-01	M1378-02
Mag-Bind® Total Pure NGS	5 mL	50 mL	500 mL
User Manual	✓	✓	✓

Storage and Stability

Mag-Bind® Total Pure NGS is guaranteed for at least 12 months from the date of purchase when stored at 2-8°C.

Mag-Bind® Total Pure NGS Protocol for 96-well Plates

Materials and Equipment to be Supplied by User:

- 96-well PCR plate containing PCR samples with capacity to hold sample and magnetic bead volumes desired
- Magnetic separation device (Recommend AlpAqua Cat# 001322). For elution volumes <30 µL AlpAqua Magnum FLX (Cat#A000400) is recommended.
- 96-well microplate or PCR Plate for elution
- Vortexer
- Multichannel pipettor
- Multichannel disposable reservoirs
- Sealing film
- 70% ethanol
- Elution Buffer (Cat#PDR048 or 10 mM Tris pH 8.0),TE Buffer, or nuclease-free water

Before Starting:

- Bring the Mag-Bind® Total Pure NGS to room temperature before use.

1. Read the manufacturer's instruction manual for the magnetic separation device, if provided.

2. Place the 96-well PCR plate on the bench and measure the volume of the PCR reaction. Determine the volume of Mag-Bind® Total Pure NGS that will be added to the reaction. If the reaction volume will exceed 200 µL transfer to a microtiter plate for processing.

   Note: PCR reactions >20 µL will need to be transferred to a processing plate.

3. Shake or vortex the Mag-Bind® Total Pure NGS to resuspend any particles that may have settled. Allow Mag-Bind® Total Pure NGS to come to room temperature before use.

4. Add the desired volume of Mag-Bind® Total Pure NGS to each well based upon desired fragment size to recover. Adding more Mag-Bind® Total Pure NGS binds smaller fragments while using less will exclude smaller sizes. Volumes to add to the sample is determined by the next generation sequencing library instruction manual.

   Example: 1.2X ratio required: 50 µL sample x 1.2 = add 60 µL Mag-Bind® Total Pure NGS

5. Pipet up and down 5-10 times or vortex for 30 seconds.

6. Let sit at room temperature for 5 minutes.

7. Place the plate on a magnetic separation device to magnetize the Mag-Bind® Total Pure NGS. Let sit at room temperature until the Mag-Bind® Total Pure NGS is completely cleared from solution.

8. Aspirate and discard the cleared supernatant. Do not disturb the Mag-Bind® Total Pure NGS.

9. With the plate remaining on the magnet, add 200 µL 70% ethanol to each well.

10. Let sit at room temperature for 1 minute. It is not necessary to resuspend the Mag-Bind® Total Pure NGS.

11. Aspirate and discard the cleared supernatant. Do not disturb the Mag-Bind® Total Pure NGS.

12. Repeat Steps 9-11 for a second 70% ethanol wash step.

13. Leave the plate on the magnetic separation device for 5-15 minutes to air dry the Mag-Bind® Total Pure NGS. Remove any residual liquid with a pipettor.

    Note: It is important to dry the Mag-Bind® Total Pure NGS before elution. Residual ethanol may interfere with downstream applications.

    Optional: Incubating the plate at 37°C can speed up evaporation.

14. Remove the plate from magnetic separation device.

15. Add 30-40 µL Elution Buffer (not provided) to each well.

16. Pipet up and down 20 times or vortex for 30 seconds.

17. Let sit at room temperature for 5 minutes.

18. Place the plate on a magnetic separation device to magnetize the Mag-Bind® Total Pure NGS. Let sit at room temperature until the Mag-Bind® Total Pure NGS is completely cleared from solution.

19. Transfer the cleared supernatant containing purified DNA to a new 96-well microplate and seal with non-permeable sealing film.

20. Store the plate at 2-8°C if storage is only for a few days. For long-term storage, samples should be kept at -20°C.

Mag-Bind® Total Pure NGS Protocol for 384-well Plates

Materials and Equipment to be Supplied by User:

- 384-well PCR plate containing PCR samples
- Magnetic separation device for 384-well PCR plates
- Skirted 384-well PCR plate with capacity to hold sample and magnetic bead volumes desired
- Vortexer
- Multichannel pipettor
- Multichannel disposable reservoirs
- Sealing film
- 70% ethanol
- Elution Buffer (Cat#PDR048 or 10 mM Tris pH 8.0),TE Buffer, or nuclease-free water

Before Starting:

- Bring the Mag-Bind® Total Pure NGS to room temperature before use.

1. Read the manufacturer's instruction manual for the magnetic separation device, if provided.

2. Place the 384-well PCR plate on the bench and measure the volume of the PCR reaction. Transfer the sample to a skirted 384-well PCR plate.

3. Shake the Mag-Bind® Total Pure NGS to resuspend any Mag-Bind® Total Pure NGS particles that may have settled. Allow Mag-Bind® Total Pure NGS to come to room temperature before use.

4. Add the desired volume of Mag-Bind® Total Pure NGS to each well based upon desired fragment size to recover. Adding more Mag-Bind® Total Pure NGS binds smaller fragments while using less will exclude smaller sizes. Volumes to add to the sample is determined by the next generation sequencing library instruction manual.

   Example: 1.2X ratio required: 50 µL sample x 1.2 = add 60 µL Mag-Bind® Total Pure NGS

5. Pipet up and down 5-10 times or vortex for 30 seconds.

6. Let sit at room temperature for 5 minutes.

7. Place the plate on a magnetic separation device to magnetize the Mag-Bind® Total Pure NGS. Let sit at room temperature until the Mag-Bind® Total Pure NGS is completely cleared from solution.

8. Aspirate and discard the cleared supernatant. Do not disturb the Mag-Bind® Total Pure NGS.

9. Add 30 µL 70% ethanol to each well.

10. Let sit at room temperature for 1 minute. It is not necessary to resuspend the Mag-Bind® Total Pure NGS.

11. Aspirate and discard the cleared supernatant. Do not disturb the Mag-Bind® Total Pure NGS.

12. Repeat Steps 9-11 for a second 70% ethanol wash step.

13. Leave the plate on the magnetic separation device for 5 minutes to air dry the Mag-Bind® Total Pure NGS. Remove any residual liquid with a pipettor.

    Note: It is important to dry the Mag-Bind® Total Pure NGS before elution. Residual ethanol may interfere with downstream applications.

    Optional: Incubating the plate at 37°C can speed up evaporation.

14. Remove the plate from magnetic separation device.

15. Add 30 µL Elution Buffer (not provided) to each well.

16. Pipet up and down 20 times or vortex for 30 seconds.

17. Let sit at room temperature for 2-3 minutes.

18. Place the plate on a magnetic separation device to magnetize the Mag-Bind® Total Pure NGS. Let sit at room temperature until the Mag-Bind® Total Pure NGS is completely cleared from solution.

19. Transfer the cleared supernatant containing purified DNA to a new 384-well microplate and seal with non-permeable sealing film.

20. Store the plate at 2-8°C if storage is only for a few days. For long-term storage, samples should be kept at -20°C.

Troubleshooting Guide

Please use this guide to troubleshoot any problems that may arise. For further assistance, please contact the technical support staff, toll free, at **1-800-832-8896**.

Possible Problems and Suggestions

Problem	Cause	Solution
Low yield	Low PCR product yield	Increase the number of amplification cycles for PCR
	Smaller PCR product size	Small PCR fragments normally give lower yield.
	Ethanol residue	During the drying step, remove any liquid from bottom of the well.
	Particle loss during the procedure	Increase magnetization time. Aspirate slowly.
	DNA remains bound to beads	Increase elution volume.
	Incomplete resuspension of the beads during elution	Vortex or pipet up and down to fully resuspend the beads.
Problem	Cause	Solution
Primer carryover	Insufficient wash of the particles	Wash the beads one more time with 70% ethanol.
Problem		Solution
Non-specific amplification products were not removed	The size of the non-specific amplification products are larger than 100 bp	Non-specific amplification products larger than 100 bp are not efficiently removed from PCR products.
Problem	Cause	Solution
Problems in downstream applications	Salt carryover	70% ethanol must be stored at room temperature.
	Ethanol carryover	Ensure the beads are completely dried before elution.

Ordering Information

The following components are available for purchase separately.
(Call Toll Free at 1-800-832-8896)

Product	Part Number
Mag-Bind® Total Pure NGS (50 mL)	M1378-01
Mag-Bind® Total Pure NGS (500 mL)	M1378-02
Elution Buffer (100 mL)	PDR048

For more purification solutions, visit **www.omegabiotek.com**

AVAILABLE FORMATS


Spin Columns

96-Well Silica Plates


Mag Beads

SAMPLE TYPES


Blood / Plasma

Plasmid


Cultured Cells


Plant & Soil


NGS Clean Up


Tissue


FFPE


Fecal Matter

omega BIO-TEK

innovations in nucleic acid isolation

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