AbraMag® DNA Size Selection Magnetic Beads are intended for research and *in vitro* use only. This product was not tested or certified for diagnostic use.

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AbraMag® DNA Size Selection Magnetic Beads

Product No. 544100 (5 mL), **544103** (60 mL), **544106** (450 mL)

1. General Description

AbraMag® DNA Size Selection Magnetic Beads are designed to purify DNA fragments of a desired size out of a sheared DNA or similar sample for use in downstream applications such as sequencing. Fragments are reversibly bound to paramagnetic particles while excess primers, salts, enzymes, and free nucleotides left in the reaction are washed away. The beads are designed for use with a magnetic separator rack.

2. Safety Instructions

Always use appropriate protective equipment (including but not limited to gloves, lab coats, and safety glasses) when working with nucleic acids. Refer to Safety Data Sheet for further information.

3. Storage and Stability

Upon delivery, store at 4°C. **Do not freeze the magnetic beads solution**. Do not use after the printed expiration date.

4. Principle

The **AbraMag® DNA Size Selection Magnetic Beads** process uses a simple, efficient, magnetic bead-based procedure for DNA fragment purification from a mixed reaction, as illustrated below in **Figure 1**:

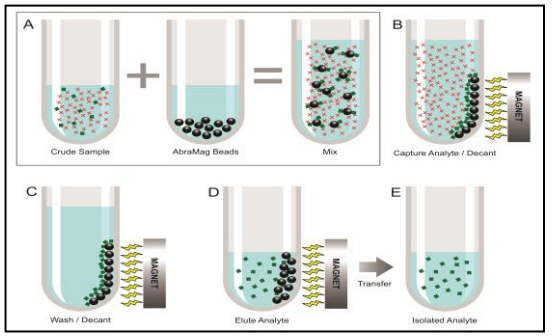

Figure 1. Schematic of the **AbraMag® DNA Size Selection Magnetic Beads** process.

4.A. **Mixing:** The mixed sample is added to washed AbraMag® magnetic beads.

4.B. **Binding:** DNA fragments of a desired size bind to the beads in the presence of the optimized buffer. A magnet is used to secure the beads.

4.C. **Washing:** Primers and/or other unwanted reagentsare washed away in a series of two wash steps.

4.D. **Elution:** Fragments are then eluted and transferred to a new tube.

4.E. **Downstream Applications:** Pure, high-quality isolated DNA fragments may then be used for downstream procedures such as cloning and sequencing.

5. Limitations and Precautions

Samples perform best when in molecular biology-grade water or Tris/TE solution prior to size selection. If a simple PCR clean-up (> 100 bp) is desired rather than size selection, use **AbraMag® PCR Clean-Up Magnetic Beads** (PN 555030 (5 mL), 555035 (60 mL), 555040 (450 mL)).

6. Working Instructions

6.A. **Materials Provided**

1. AbraMag® DNA Size Selection Magnetic Beads Solution

6.B. **Additional Materials and Equipment Required** (not included with the product)

1. Sheared DNA or other sample
2. Laboratory-grade water
3. Freshly prepared 70% Ethanol
4. Molecular-grade TE buffer pH 8.0 (10 mM Tris, 1 mM EDTA pH 8.0)
5. Disposable gloves and other protective equipment
6. Micro-pipettes with disposable plastic filter barrier tips
7. 1.5 mL sterile, nuclease-free microcentrifuge tubes
8. 4°C refrigerator
9. Magnetic microcentrifuge tube separator, Solo (PN 472270), Multi-6 (PN 472260), Microtiter Plate Side-Pull (PN 472235) or Bottom-Pull (PN 472236), or similar
10. Vortexer

6.C. **Selecting Bead Ratio for Desired Size Selection**

The size cutoff, favoring retention of DNA fragments from 50 to 400+ bp, can be controlled by altering the ratio of AbraMag® DNA Size Selection Magnetic Beads to the starting sample volume. **Figure 2** below shows a gel electrophoresis analysis of size selection according to increasing ratios. Check **Table 1** for example ratios.

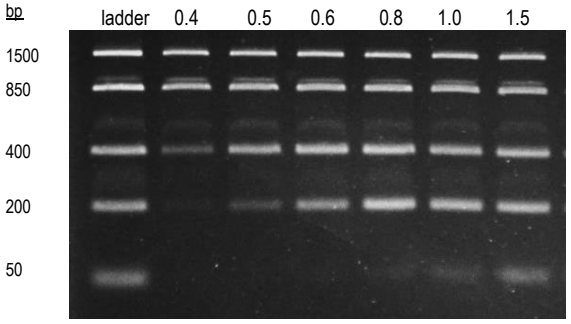

Figure 2. 2% agarose gel electrophoresis of DNA ladder showing effect of bead solution to sample ratios on fragment size exclusion. 100V for 25 minutes.

Ratio	Size Cutoff (bp)	Amount of bead solution (µL)
0.4	400 +	20
0.6	200 +	30
0.8	100 +	40
1.5	50 +	75

Table 1. Example bead ratios and resulting size selection using a starting sample of 50 µL.

6.D. **Procedure**

1. Transfer the sample to be purified to a 1.5 mL microcentrifuge tube. Dilute the sample to at least 50 µL using laboratory-grade water.
   Note: To use a larger volume of starting sample, simply multiply it by the desired ratio (Section 6.C).
   Ex: 100 µL sample x 1.5 = use 150 µL of bead solution in Section 6.D.2.
   Note: A small amount of starting product can be saved for gel analysis. See Section 7.B.
2. Vortex the **AbraMag® DNA Size Selection Magnetic Beads** with ~1 second pulses. Ensure that the solution is completely resuspended. Check Section 6.C to determine the amount of bead solution to use according to the desired ratio and add that amount of beads to the sample. Pipette up and down to mix well. It is recommended to try a range of ratios closest to the desired size selection in order to optimize the fragment size selection.
3. Incubate the sample/bead mixture for 5 minutes at room temperature.
4. Place the sample on the magnetic separator until the solution is clear (~1 minute). Leaving the tube on the separator, aspirate and discard the supernatant without disturbing the beads that have gathered at the magnet.
5. Leaving the tube on the separator, gently add 200 µL of freshly prepared 70% ethanol, without dislodging the beads from the side of the tube. Let sit for 30 seconds. Aspirate and discard the supernatant.
6. Repeat Step 5 for a second wash. Carefully remove all pipettable ethanol.
7. Remove the tube from the magnetic separator and leave at room temperature for ~3 minutes with the cap open to completely evaporate any residual ethanol.
8. Add 40 µL TE buffer pH 8.0 or laboratory-grade water to the sample. Pipette up and down to mix well. Incubate at room temperature for 2-3 minutes. Return the tube to the magnetic separator for 1 minute. Leaving the tube on the separator, transfer the eluate to a new microcentrifuge tube using a pipette. ***The eluate contains the size selected DNA fragments.***

7. Analyzing Results

7.A. **Product Recovery**

Product recovery can be determined by using a fluorometer with intercalating dye, gel electrophoresis analysis, and/or spectrophotometer.

7.B. **Size Selection Confirmation**

Gel electrophoresis can be used to evaluate efficacy of the size selection, and to check optimization of the ratio. Using an agarose or PAGE gel, run the eluted sample recovered in Section 6.D.8 compared to a molecular weight ladder to ensure that the desired fragment sizes have been selected.