



Corporate Headquarters
400 Valley Road
Warrington, PA 18976
1-800-523-2575
FAX 1-800-343-3291
Email: info@polysciences.com
www.polysciences.com

Europe - Germany
Polysciences Europe GmbH
Handelsstr. 3
D-69214 Eppelheim, Germany
(49) 6221-765767
FAX (49) 6221-764620
Email: info@polysciences.de

TECHNICAL DATA SHEET 530

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BioMag[®] Nuclease-Free Streptavidin

Description

BioMag Nuclease-Free Streptavidin is a suspension of BioMag particles approximately 1µm in size which are attached to streptavidin. The suspension is supplied in phosphate buffered saline (pH 7.4) containing 0.1% BSA. Azide has been added as a stabilizer. Shake vigorously before use. Magnetically separate the BioMag particles, aspirate the supernatant and resuspend in an appropriate buffer.

Storage and Stability

Freezing, drying or centrifuging BioMag results in extensive aggregation and loss of binding activity. DO NOT FREEZE OR DRY THIS PRODUCT.

BioMag Concentration: 1 mg/ml

Binding Capacity

1mg of BioMag Streptavidin will bind:

- > 1500 pmoles of free biotin
- > 1000 pmoles of a 20-mer biotinylated oligonucleotide
- > 200 pmoles of a 100-mer biotinylated oligonucleotide
- > 70 pmoles of a 300 bp 5'biotinylated double stranded DNA
- > 25 pmoles of a 1Kbp 5'biotinylated double stranded DNA

Researchers are advised to optimize the use of BioMag in any application as procedures designed by other manufacturers may not be ideal.

For research use only, not intended for use in humans or in vitro diagnostics use.

Should any of our materials fail to perform to our specifications, we will be pleased to provide replacements or return the purchase price. We solicit your inquiries concerning all needs for life sciences work. The information given in this bulletin is to the best of our knowledge accurate, but no warranty is expressed or implied. It is the user's responsibility to determine the suitability for his own use of the products described herein, and since conditions of use are beyond our control, we disclaim all liability with respect to the use of any material supplied by us. Nothing contained herein shall be construed as a recommendation to use any product or to practice any process in violation of any law or any government regulation.

mRNA PURIFICATION PROCEDURE USING BIOMAG NUCLEASE-FREE STREPTAVIDIN

The following procedure is an example of a BioMag Nuclease-Free Streptavidin application for the isolation of 1-2µg of mRNA from approximately 75-100µg of total RNA. The total isolation time is less than 30 minutes.

1. Dispense 200µl of BioMag Streptavidin into a nuclease-free microcentrifuge tube. Using a magnetic separation unit (Cat.#84111S or similar rare earth magnetic separator), pull the magnetic particles to the side of the microcentrifuge tube for 30 seconds. Remove and discard the supernatant. Resuspend the BioMag Streptavidin in 100µl of **Binding Buffer (20 mM Tris, 0.5 M NaCl, pH 8.0)**.
2. Incubate 2.5µl (2.5µg) of 5'-Biotinylated Oligo (dT) (or an appropriate amount of biotinylated molecule) with the 100µl of BioMag Streptavidin from Step 1. for 15 minutes at room temperature.
3. Magnetically separate for 30 seconds and discard the supernatant. Wash the Oligo (dT) bound particles from Step 2. with 100µl of Binding Buffer 2 times, leaving the magnetic particles as a wet cake.
4. Bring up the total RNA sample in DEPC-treated water to a total volume of 90µl.
5. Incubate the RNA sample at 55°C for 5 minutes to disrupt secondary structures.
6. Add 10µl of 5M NaCl to achieve a final concentration of 0.5 M NaCl.
7. Add the total RNA to the washed magnetic particles from Step 3. Mix gently and hybridize at room temperature for 3 minutes.
8. Magnetically separate and wash the particles with 100µl of **Wash Buffer (7 mM Tris, 0.17M NaCl, pH 8.0)** 2 times.
9. Elute the bound mRNA with 25-50µl of DEPC-treated water at 55°C for 2 minutes.

10. Magnetically separate and transfer the supernatant to a nuclease-free microcentrifuge tube.

11. Repeat elution of mRNA with 25-50µl of DEPC-treated water at 55°C for another 2 minutes in order to completely elute the bound mRNA from the particles. Magnetically separate and transfer the supernatant to the tube containing the first elution of mRNA from Step 10.

Ordering Information:

Cat. #	Description	Size
8MB4804B	BioMag® Nuclease-Free Streptavidin Molecular Biology Grade	10ml
8-MB4804C	BioMag® Nuclease-Free Streptavidin Molecular Biology Grade	25ml
8-MB4804E	BioMag® Nuclease-Free Streptavidin Molecular Biology Grade	100ml

To Order:

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Order online anytime at www.polysciences.com

References:

1. Hornes E., K.S. Jakobsen, O.S. Gabrielsen, L.S. Korsnes, E.B. Jansen and M. Espelund. 1991. Purification of mRNA and DNA binding proteins using magnetic beads. In: Kemshead J.T., ed. Magnetic separation techniques applied to cellular and molecular biology. Somerset, Wordsmiths' conference publications. 197-205.
2. Morrisey, David V., Massimo Lombardo, John K. Eldredge, Kevin R. Kearney, E. Patrick Groody and Mark K. Collins. 1989. Nucleic Acid Hybridization Assays Employing dA-Tailed Capture Probes. Anal. Biochem. 181:345-359.