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ProductInformation

Streptavidin Iron Oxide Particles
from *Streptomyces avidinii*

Product Number **S 2415**
Storage Temperature 2-8 C

Product Description

Synonym: BioMag® Streptavidin

Concentration: 1 mg/ml
Particles per ml: 5 x 10^8
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Particle size: 0.5-1.5 µm

The product is a suspension of iron oxide particles approximately 1 µm in size, which are attached to streptavidin. The particles are suspended in 10 mM phosphate buffered saline with 1% bovine serum albumin.

The binding capacity of 1 mg is approximately 1500 pmoles of free biotin, >1000 pmoles of 20-mer biotinylated oligonucleotides, >200 pmoles of a 100-mer biotinylated oligo, >70 pmoles of a 300 base pair 5'-biotinylated double stranded DNA, and >25 pmoles of 1 kilobase 5'-biotinylated double stranded DNA.

The product has been used to purify a complex transcription factor, yeast transcription factor IIIC (TFIIIC). Biotinylated DNA containing the TFIIIC recognition sequence attached to the magnetic beads binds the protein specifically in the presence of the competitor DNA. Nearly homogenous THIIIC was obtained with three cycles of absorption and was shown to be fully active in transcription and DNA binding assays.[1]

Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

Freezing, drying, or centrifuging this product results in extensive aggregation and loss of binding activity. Do not freeze or dry this product.

Procedure

Shake vigorously before use. Researchers are advised to optimize the use of this product in any application, because procedures for related products from other manufacturers may not be ideal.

The following protocol can be used to isolate 1-2 µg of messenger RNA from 75 - 100 µg of total RNA using this product. The total isolation time is less than 30 minutes.

1. Dispense 200 l of S 2415 into a nuclease free microcentrifuge tube. Using a magnetic separation unit (Product No. M 1167) 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 in 100 µl of binding buffer (20 mM Tris, pH 8.0, 0.5 M NaCl).
2. Incubate 2.5 µl (2.5 µg) of 5' biotinylated oligo(dT) (or an appropriate amount of biotinylated molecule) with 100 µl of the suspension for 15 minutes at room temperature.
3. Magnetically separate for 30 seconds and discard the supernatant. Wash the oligo(dT) bound particles with 100 µl of binding buffer twice, leaving the magnetic particles as a wet cake.
4. Bring up the total RNA sample with DEPC treated water to a total volume of 90 µl.
5. Incubate the RNA sample at 55 °C for 5 minutes to disrupt secondary structures.
6. Add 10 µl of 5 M NaCl to the RNA sample to achieve a final concentration of 0.5 M NaCl.
7. Add the total RNA sample to the washed magnetic particles from step 3. Mix gently and hybridize at room temperature for three minutes.
8. Magnetically separate and wash the particles with 100 µl of wash buffer (7 mM Tris, pH 8.0, 0.17 M NaCl) twice.

9. Elute the bound mRNA with 25 - 50 μl of DEPC treated water at 55 °C for two minutes.
10. Magnetically separate and transfer the supernatant to a nuclease free microcentrifuge tube.
11. Repeat the elution of mRNA with 25 - 50 μl of DEPC treated water at 55 °C for another two 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.

Additional procedures on a related product have also been published.[2]

References

1. Hornes, E., et al., in Magnetic Separation Techniques Applied to Cellular and Molecular Biology, Proceedings of the John Ugelstad Conference, Kemshead, J. T., ed., Wordsmiths' Conference Publications (Somerset, U.K.: 1991).
2. Morrissey, D., et al., Nucleic acid hybridization assays employing dA-tailed capture probes. I. Multiple capture methods. Anal. Biochem., **181(2)**, 345-359 (1989).

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