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BioMag[®] Oligo (dT)20, Nuclease-free Catalog Number: 8MB4803

DESCRIPTION

BioMag[®] Oligo dT(20) is a nuclease-free suspension of BioMag[®] particles approximately 1.5µm in size, which are covalently bound with Oligo dT(20). The suspension is supplied in a buffer (pH 8.0) containing 20 mM Tris, 0.5 M sodium chloride, and 0.1% sodium azide. Shake vigorously before use. Magnetically separate the BioMag[®] particles, aspirate the supernatant, and resuspend in an appropriate buffer for your application.

CHARACTERISTICS

Mean Diameter: ~1.5µm Particle Concentration: 5mg/ml Binding Capacity: 100µL of BioMag® Oligo dT(20) is sufficient to isolate ~1-2µg of polyadenylated RNA from

~100µg of total RNA

MATERIAL

Material Supplied

BioMag[®] Oligo dT(20): 2ml

Material Required

- Binding Buffer: 20mM Tris and 0.5M NaCl at pH 8.0
- Wash Buffer: 7mM Tris and 0.17M NaCl at pH 8.0
- DEPC-treated water
- Nuclease-free microcentrifuge tubes
- Magnetic separator

PROCEDURE

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

The following is a procedure for the isolation of 1-2µg of polyadenylated RNA from approximately 100µg of total RNA. The 2mL of BioMag[®] Oligo dT(20) supplied is sufficient for 20 isolations of 1-2µg of mRNA. (More or less mRNA can be isolated by modifying the procedure.) The total isolation time is approximately 15 minutes.

- Dispense 100µL of BioMag[®] Oligo dT(20) into a nuclease-free microcentrifuge tube. Using a magnetic separation unit, pull the magnetic particles to the side of the microcentrifuge tube for 30 seconds. Remove and discard the supernatant. Wash the BioMag[®] Oligo dT(20) once with 200µL of the Binding Buffer. Magnetically separate, discard the supernatant, and resuspend in 100µL of the Binding Buffer.
- 2. Bring up the total RNA sample in DEPC-treated water to a total volume of 90µL.

- 3. Incubate the RNA sample at 55°C for 5 minutes to disrupt secondary structures.
- 4. Add 10µL of 5M NaCl to achieve a final concentration of 0.5M NaCl.
- 5. Add the total RNA to the BioMag[®] Oligo dT(20) from Step 1. Mix gently and hybridize at room temperature for 3 minutes.
- 6. Magnetically separate and wash the particles 2 times with 100μL of the Wash Buffer.
- Elute the bound polyadenylated RNA with 25-50µL of DEPC-treated water at 55°C for 2 minutes. Greater than 90% of polyadenylated RNA is eluted in this step.
- 8. Magnetically separate and transfer the supernatant to a nucleasefree microcentrifuge tube.
- Repeat elution of polyadenylated RNA 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 7.

REFERENCES

- Hornes, E., L. Korsnes. 1990. Magnetic DNA hybridization properties of oligonucleotide probes attached to superparamagnetic beads and their use in the isolation of poly(A) mRNA from eukaryotic cells. *Genet Anal Tech Appl*, 7(6):145-150.
- Morrissey, D.V., M. Lombardo, J.K. Eldredge, K.R. Kearney, E.P. Groody, M.L. Collins. 1989. Nucleic acid hybridization assays employing dA-tailed capture probes. Multiple capture methods. *Anal Biochem*, 181(2):345-359.

STORAGE AND SAFETY

Storage Store at 4°C. Freezing, drying, or centrifuging BioMag[®] may result in irreversible aggregation and loss of binding activity. To ensure stability, BioMag[®] Oligo dT(20) must be stored in the buffer in which it is supplied.

Safety This particle suspension contains sodium azide. Sodium azide may react with lead and copper plumbing to form explosive metal azides. Upon disposal of material, flush with a large volume of water to prevent azide accumulation. Please consult the Safety Data Sheet for more information.

These products are for research use only and are not intended for use in humans or for *in vitro* diagnostic use.

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ORDERING INFORMATION

Cat. #	Description	Sizes
8MB4803-2	BioMag [®] Oligo (dT)20, Nuclease-free	2ml

TO ORDER

In The U.S. Call:	1(800) 523-2575 • (215) 343-6484
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