# Product Information



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**Monoclonal Anti-Maltose Binding Protein**
**Clone MBP-17**
produced in mouse, purified immunoglobulin

Catalog Number **M1321**

## Product Description

Monoclonal Anti-Maltose Binding Protein (MBP) (mouse IgG1 isotype) is derived from the hybridoma MBP-17 produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a purified recombinant MBP fusion protein. The isotype is determined using a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Catalog Number ISO2.

Monoclonal Anti-Maltose Binding Protein recognizes native, as well as denatured-reduced, forms of purified MBP or MBP fusion proteins by immunoblotting,[1] dot blot, luminometric immunoassay,[2] and ELISA.

Recombinant DNA technology enables the attachment of genes of interest to specific sequences or genes that can provide 'affinity handles' (tags) designed to enable the selective identification and purification of the protein of interest.[3-5] These sequences of tails or tags are genetically engineered away from the protein active site, by insertion at the N- or C-terminus. It has been reported that the addition of a maltose binding protein (MBP) tag creates a stable fusion product that does not appear to interfere with the bioactivity of the protein or with the biodistribution of the MBP tagged product.[6, 7] The expression of polypeptides in-frame with maltose binding protein (MBP) allows for their easy purification from bacterial extracts under mild conditions, which employ a single affinity chromatographic step on amylose resin.[6] This system and others based on the expression of fusion proteins utilize a specific protease cleaving site to facilitate correct cleavage of the fusion protein.[5] Thus, the MBP system incorporates a factor Xa cleavage site at the carboxy terminus of the MBP sequence,[7] and cleavage by factor Xa separates MBP from its partner protein. Many recombinant proteins[6-8] have been engineered with MBP tags to facilitate the detection, isolation and purification of the proteins. Monoclonal antibody reacting specifically with MBP may be useful in various immunotechniques to identify the expression of an MBP fusion protein in bacteria or in cells and tissues transfected with a MBP fusion protein expressing vectors.

## Reagent

Supplied as a solution in 0.01M PBS, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody concentration: ~ 2 mg/mL

## Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

## Storage/Stability

For extended storage, freeze at -20 °C in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discard if not used within 12 hours.

## Product Profile

Immunoblotting: a working concentration of 0.05-0.1 μg/mL is recommended using purified recombinant MBP.

**Note**: In order to obtain best results in different techniques and preparations we recommend determining optimal working concentration by titration test.

## Procedure

Immunoblotting
All incubation steps should be performed at room temperature.

1. Separate MBP-tagged proteins from sample lysates using a standard SDS-PAGE protocol.
   Load 2.5-20 μg of total lysate protein per lane. The amount of lysate to be loaded per lane depends on the level of protein expression and may vary between experiments.
2. Transfer proteins from the gel to a nitrocellulose membrane.

3. Block the membrane using a solution of 5% non-fat dry milk in phosphate buffered saline (Dulbecco's Phosphate Buffered Saline, Catalog Number D8537) for at least 60 minutes.
4. Wash the membrane three times for 5 minutes each in PBS containing 0.05% TWEEN 20®, Catalog Number P3563.
5. Incubate the membrane with Anti-Maltose Binding Protein using an optimized concentration in PBS containing 1% bovine serum albumin (BSA, Catalog Number A9647) for two hours.
6. Wash the membrane three times for 5 minutes each in PBS containing 0.05% TWEEN 20.
7. Incubate the membrane for 60 minutes with Anti-Mouse IgG-Peroxidase, e.g. Catalog Number A9917, A3682, or A2304, or with Anti-Mouse IgG-Alkaline Phosphatase, e.g. Catalog Number A1293, A2179 or A1682, as the secondary antibody at the recommended concentration in PBS containing 0.05% TWEEN 20. Adjust the antibody concentration to maximize detection sensitivity and to minimize background.
8. Wash the membrane three times for 5 minutes each in PBS containing 0.05% TWEEN 20.
9. Treat the membrane with either a peroxidase or an alkaline-phosphatase substrate, as appropriate.

**References**


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