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ProductInformation

MONOCLONAL ANTI-BACTERIAL ALKALINE PHOSPHATASE (BAP, PhoA)
CLONE BAP-77
Mouse Ascites Fluid

Product Number **B 6804**

Product Description

Monoclonal Anti-Bacterial Alkaline Phosphatase (BAP, PhoA) (mouse IgG1 isotype) is derived from the BAP-77 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a BALB/c mouse immunized with a BAP tagged fusion protein. The isotype is determined using Sigma ImmunoType™ Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2).

Monoclonal Anti-Bacterial Alkaline Phosphatase (BAP, PhoA) recognizes BAP (approx. 50 kDa) by immunoblotting, dot blot and ELISA. The antibody reacts with fusion protein expressed by prokaryotes expression vectors. It does not react with alkaline phosphatase derived from human placenta, bovine liver, calf intestinal mucosa or from shrimp. The antibody does not identify BAP in whole extract of wild type *E. coli* by immunoblotting.

Recombinant DNA technology enables the insertion of genes of interest to specific sequences or genes, which can provide 'affinity handles' (tags) for the selective identification and purification of the protein of interest.[1-3] The addition of BAP tag to a given gene, creates a stable fusion product that does not appear to interfere with the bioactivity of the protein, or with the biodistribution of the BAP tagged product. BAP (also known as PhoA, EC 3.1.3.1) is an approx. 50 kDa protein, derived from *E. coli*. Alkaline phosphatase appears to require export to the periplasm to show enzymatic activity.[4] Fusions of the secreted alkaline phosphatase to an integral cytoplasmic membrane protein of *E. coli* shows different activities depending on where with the membrane protein the alkaline phosphatase is fused.[5] Fusions to positions in or near the periplasmic (extracellular) domain lead to high alkaline phosphatase activity, whereas those to positions in the cytoplasmic domain give low activity. Consequently, analysis of alkaline phosphatase fusions to membrane proteins of unknown structure are generally useful in determining their membrane topologies.[4] Expression of enzymatically active protein fusions in *E. coli* could facilitate the analysis of proteins and even replace some reagents frequently used in immunology such as chemically-produced antibody-enzyme conugates.[6,7] Monoclonal antibody reacting specifically with BAP may be useful in various immunotechniques, to identify the expression of a BAP fusion protein *in situ or* by immunoblotting, in bacteria, bacterial lysates or in cells and tissues transfected with BAP fusion protein expressing vectors. Expression system that exploits the BAP signal sequence to translocate a target protein to the periplasm, may be used to evaluate how changes in the composition and sequence of amino acids near the BAP-target protein junction influence the periplasmic accumulation of the target recombinant protein.[8]

Reagents

Monoclonal Anti-Bacterial Alkaline Phosphatase (BAP, PhoA) is supplied as ascites fluid containing 15 mM sodium azide.

Precautions and Disclaimer

Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution.

Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. 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 discarded if not used within 12 hours.

Product Profile

A minimum working dilution of 1:20,000 is determined by immunoblotting, using a purified *E. coli* BAP preparation.

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

References

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