

Product Information

C-TERMINAL FLAG-BAP[™] CONTROL PROTEIN

Product No. **P 7457**

Store at 0 to -20 °C

Product Description

C-Terminal FLAG-BAP control protein is a 466 amino acid C-terminal FLAG[®] fusion protein of *E. coli* bacterial alkaline phosphatase (BAP) with a calculated molecular weight of 49.1 kD.

The C-terminal FLAG-BAP control protein migrates as a 45-55 kD band by SDS-PAGE depending on electrophoresis conditions.

C-Terminal FLAG-BAP control protein has been found to be useful for assurance of the functional integrity of anti-FLAG M2 monoclonal antibody in immunological procedures such as Western blotting, ELISA, immunoprecipitation, fluorescence microscopy, light microscopy and FACS. It is also useful as a functional control in immunoaffinity chromatography with the anti-FLAG M2 affinity gel (Product No. A 1205).

The C-terminal FLAG-BAP control protein is supplied in 10 mM Tris, 120 mM NaCl, 0.05 mM ZnCl₂ in 50% glycerol, pH 8.0.

Reagents Required but Not Provided

- Tris buffered saline (TBS), 0.05 M Tris, 0.15 M NaCl, pH 7.4
- Non-fat dry milk
- Anti-FLAG M2 monoclonal antibody (Product No. F 3165)
- Anti-mouse IgG peroxidase conjugate
- Luminol (5-amino-2,3-dihydro-1,4-phthalazine-dione, Product No. A 4685) or other peroxidase substrate

Preparation Instructions

Dilute the anti-FLAG M2 antibody solution to 10 µg/ml in TBS. Adjust the antibody concentration to maximize detection sensitivity and to minimize background.

Procedure

Procedure for Western Blot

1. Transfer the C-terminal FLAG-BAP control protein to a nitrocellulose membrane.
2. Block the membrane using a solution of 5% non-fat dry milk in TBS at 37 °C for 1 hour.
3. Wash the membrane twice for 1-2 minutes each in TBS at room temperature.
4. Incubate the membrane with anti-FLAG M2 antibody as the primary antibody at room temperature for 30 minutes.
5. Wash the membrane three times for 1-2 minutes each in TBS at room temperature.
6. Incubate the membrane with anti-mouse IgG peroxidase conjugate as the secondary antibody at the manufacturer's recommended concentration in TBS. Incubate at room temperature for 30 minutes. Adjust the antibody concentration to maximize detection sensitivity and to minimize background.
7. Wash the membrane three times for 15 minutes each in TBS at room temperature.
8. Treat the membrane with luminol or other peroxidase substrate.

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