Technical Bulletin

Anti-HA-FITC antibody, Mouse monoclonal

Clone HA-7, purified from hybridoma cell culture

B9183

Product Description

Recombinant DNA technology enables the attachment of genes of interest to specific sequences or genes that can provide "affinity handles" or tags. These tags are designed to enable the selective identification and purification of the protein of interest.[1-5] These sequences or tags are genetically engineered away from the protein active site, by insertion at the N- or C-terminus.

Human influenza hemagglutinin (HA) is a surface glycoprotein required for the infectivity of the virus.[6] The short sequence derived from the HA molecule, has been used as a tag, known as the HA-Tag. Many recombinant proteins have been engineered to express the HA tag, which does not appear to interfere with the bioactivity or the biodistribution of the recombinant protein. This tag facilitates the detection, isolation and purification of the proteins.[4,5]

Anti-HA-FITC antibody, Mouse monoclonal, is derived from the HA-7 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice, immunized with a synthetic peptide corresponding to a fragment (YPYDVPDYA) of human influenza virus hemagglutinin (HA), conjugated to KLH. The antibody is isolated from ascites fluid and conjugated to fluorescein isothiocyanate (FITC), isomer I.

Anti-HA-FITC antibody, Mouse monoclonal recognizes the HA tag expressed on the N- or C-terminus on HA tagged proteins. The antibody reacts specifically with HA tagged proteins by immunocytochemistry. This staining is specifically inhibited by the immunizing peptide HA peptide (Cat. No. I2149).

Reagent

This product is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA and 15 mM sodium azide as a preservative.

Specific Antibody concentration: ~1.0 mg/mL (exact value on Certificate of Analysis for particular lot)

F/P Molar Ratio: 3-8 (exact value on Certificate of Analysis for particular lot)

Storage/Stability

- **Store the product protected from light.**
- For continuous use, store at 2-8 °C for up to one month.
- For extended storage, freeze 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 discarded if not used within 12 hours.

Precautions and Disclaimer

Because of the sodium azide content, a Safety Data Sheet for this product has been sent to the attention of the safety officer of your institution. Consult the Safety Data Sheet for information regarding hazardous and safe handling practices.

Product Profile

Immunocytochemistry

1-5 µg/mL of the antibody detects HA-tagged proteins in mammalian cells.

Note: To obtain best results in various techniques and preparations, it is recommended to determine optimal working dilutions by titration.

Procedure for Direct Immunofluorescent Staining of Cultured Cells

1. Grow transfected cultured cells that express the HA-tagged protein of choice on sterile coverslips at 37 °C.
2. Wash the cells briefly in PBS.
3. Fix the cells for 10 minutes with 3% paraformaldehyde. Immediately permeabilize with 0.5% Triton® X-100.
4. Wash the specimens twice in PBS (5 minutes each wash).
5. Incubate the specimens cell-side-up with Anti-HA-FITC in PBS at room temperature for 1 hour.
6. Wash three times in PBS (5 minutes each wash).
7. Add one drop of aqueous mounting medium on the coverslip and invert carefully on a glass slide. Avoid air bubbles.
8. Examine using a fluorescence microscope with appropriate filters.

References

1. Jarvik, W., and Telmer, C.A., *Annu. Rev. Genet.*, **32**, 601-618 (1998).
2. Olins, P.O., and Lee, S.C., *Curr. Opin. Biotechnol.*, **4(5)**, 520-525 (1993).
3. Uhlen, M., and Moks, T., *Methods Enzymol.*, **185**, 129-143 (1990).
4. Kolodziej, P.A., and Young, R.A., *Methods Enzymol.*, **194**, 508-519 (1991).
5. Woychik, N.A., and Young, R.A., *Trends Biochem. Sci.*, **15(9)**, 347-351 (1990).
6. Wilson, I.A. *et al.*, *Cell*, **37(3)**, 767-778 (1984).

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