Technical Bulletin

Anti-Biotin–FITC Antibody, Mouse Monoclonal

Clone BN-34, purified from hybridoma cell culture

F4024

Product Description

Monoclonal Anti-Biotin (mouse IgG1 isotype) is derived from the hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. Biotinylated KLH was used as the immunogen. The isotype is determined using ImmunoType™ Kit (Cat. No. ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Cat. No. ISO-2).

Biotin is an essential vitamin required by cells in living organisms or in cultures. The discovery of the high binding affinity of biotin to egg white or bacteria-derived avidin has been exploited in the design of immunoassays and immunohistologic staining techniques to serve as a basis for identifying antigen-antibody interactions. The most popular applications involve localization of the antigen with a primary antibody, addition of a biotinylated antibody to bind to primary antibody, and finally application avidin-FITC.

While standard assay methods using the avidin-biotin-fluorochrome complex will suffice for most studies, there are occasions when enhanced sensitivity is needed to detect minute amounts of antigen or localize low densities of antigens in histologic sections. Sensitivity of avidin-biotin immunoassays is enhanced by selectively enlarging the avidin-biotin-fluorochrome by adding FITC conjugated antibody to biotin to the avidin-biotin-FITC complex, thereby increasing the signal from substrate conversion. In addition, such an antibody can be used in many other applications where biotin can be introduced as target label.

For instance, it can be used as a method of choice in detection of DNA and mRNA in routine paraffin sections by sensitive, non-isotopic in-situ hybridization. It has also been used successfully for detection of micro-injected biotin-haptenized cytoskeletal proteins that enables the direct examination of the pattern of incorporation and turnover of cytoskeletal proteins in living cells.

Reagents

The immunoglobulin fraction of the ascites fluid is conjugated to Fluorescein Isothiocyanate (FITC) and then further purified to remove unconjugated FITC. The conjugate is provided as a liquid in 0.01 M phosphate buffer, pH 8.0, with 1% BSA and 15 mM sodium azide as a preservative.

Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C 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.

Product Profile

FITC Conjugated Monoclonal Anti-Biotin recognizes free biotin or when it is conjugated to various immunoglobulins using various immunochemical assays such as immunocytochemistry, immunohistochemistry, and flow cytometry.

A working dilution of 1:80 to 1:160 was determined by indirect immunohistology using biotinylated monoclonal or polyclonal antibodies.

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