

**MONOCLONAL ANTI-BIOTIN CLONE BN-34**

Mouse Ascites Fluid

Product No. **B7653**

Lot 107H4836

Monoclonal Anti-Biotin (mouse IgG1 isotype) is derived from the BN-34 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with biotinylated KLH. The isotype is determined using Sigma ImmunoType™ Kit (Sigma Stock No. ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Sigma Stock No. ISO-2). The product is provided as ascites fluid with 0.1% sodium azide (see MSDS)\* as a preservative.

**Specificity**

Monoclonal Anti-Biotin recognizes the free biotin molecule and biotin conjugated to various immunoglobulins in ELISA and immunohistochemical techniques. Specificity was verified by using biotinylated goat antibodies reactive against human and rabbit antigens coated on microtiter plates.

**Description**

Biotin is an essential vitamin required by cells in living organisms or in culture. The high binding affinity to egg white or bacteria-derived avidin has been exploited in the design of immunoassays and immunohistologic staining techniques.<sup>1</sup> The most popular procedure involves localization of the antigen with a primary antibody, addition of a biotinylated antibody to bind to the primary antibody, application of a complex of avidin and biotinylated enzyme (usually horseradish peroxidase) and finally, reaction with a chromogenic substrate.<sup>2,3</sup> While standard assay methods using the avidin-biotin-enzyme complex will suffice for most studies, there are occasions when enhanced sensitivity is needed to detect smaller amounts of antigen or localize low density antigens in histologic sections. Conventional immunoassay methods are improved by the use of Monoclonal Anti-Biotin<sup>4</sup>, which enhances the sensitivity of avidin-biotin immunoassays by bridging a second layer of avidin-biotin-enzyme complex.<sup>3</sup> This antibody can be used in many other applications where biotin can be introduced as a target label. For instance, it has been used in detection of low copy human papilloma virus DNA and mRNA in routine paraffin sections of

cervix by sensitive non-isotopic in-situ hybridization.<sup>5</sup> It has also been used successfully for the detection of microinjected biotin-haptenized cytoskeletal proteins to examine directly the pattern of incorporation and turnover of cytoskeletal proteins in living cells.<sup>6</sup>

**Uses**

Monoclonal Anti-Biotin may be used in a wide range of applications including blotting (Western, Southern and Dot Blots), immunocytochemistry, in-situ nucleic acids hybridization, ELISA, fluorescent activated cell-sorting (FACS) and electron microscopy.

**Titer:** 1;2,000

The antibody titer was determined by direct ELISA using biotinylated goat anti-rabbit IgG coated on polystyrene microtiter plates (10 µg/ml coat).

In order to obtain best results in different techniques and preparations, it is recommended that each individual user determine their optimum working dilution by titration assay.

**Storage**

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.

\* 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.

**References**

1. Wilchek, M., and Bayer, E., Immunol. Today, **5**, 39 (1984).

2. Guerin-Reverchon, I., et al., J. Immunol. Meth., **123**, 167 (1989).
3. Allan, G., et al., J. Virol. Meth., **24**, 181 (1989).
4. Dakshinamurti, K., et al., Biochem.J., **237**, 477 (1986).
5. Burns, J., et al., J. Clin. Pathol., **40**, 858 (1987).
6. Okabe, S. and Hirokawa, N., PNAS (USA), **86**, 4127 (1989).

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