

Labeled and Unlabeled Anti-Biotin

A11242 anti-biotin, mouse IgG₁, monoclonal 2F5

A31801 anti-biotin, mouse IgG₁, monoclonal 2F5, Alexa Fluor® 488 conjugate

A31800 anti-biotin, mouse IgG₁, monoclonal 2F5, Alexa Fluor® 594 conjugate

Quick Facts

Storage upon receipt:

- $\leq -20^{\circ}\text{C}$ (A11242); $\leq 6^{\circ}\text{C}$ (A31800, A31801)
- Dessicate (A11242)
- Protect from light (A31801, A31800)
- Avoid freeze-thaw cycles

Abs/Em: 495/519 nm (A31801);
590/617 nm (A31800)

Introduction

The high affinity of avidin for biotin was first exploited in histochemical applications in the mid-1970s.^{1,2} The use of avidin–biotin techniques has since become standard for diverse detection schemes,³ although limitations of this method have also been recognized.⁴ As an alternative to avidin reagents, monoclonal antibodies to biotin have been developed.^{3,5,6} Molecular Probes now offers labeled (A31801, A31800) and unlabeled (A11242) versions of a high-affinity mouse monoclonal antibody (MAb) to biotin. Anti-biotin MAb 2F5 (isotype IgG_{1,k}) can potentially be used to detect biotinylated molecules in immunohistochemistry, *in situ* hybridization, ELISA and Western blot applications. The Alexa Fluor® 488 and Alexa Fluor 594 dye conjugates, which have fluorescence excitation and emission maxima similar to those of fluorescein and Texas Red® dyes, respectively, are especially useful for indirect immunofluorescence, as they exhibit remarkably bright, photostable and pH-insensitive fluorescence.

Contents

Product A11242 is supplied lyophilized in unit sizes of 100 μg . Products A31800 and A31801 are supplied in a unit size of 100 μL as a 1 mg/mL solution in phosphate-buffered saline (PBS), pH 7.2, plus 5 mM sodium azide.

Storage and Handling

Upon receipt, store product A11243 desiccated at $\leq -20^{\circ}\text{C}$. Store products A31800 and A31801 at $\leq 6^{\circ}\text{C}$, and, if frozen, avoid freeze-thaw cycles. Protect the Alexa Fluor dye conjugates from light.

To prepare stock solutions of the lyophilized product (A11242), reconstitute the solid material in PBS, containing 1% bovine serum albumin (BSA) and 2 mM sodium azide. For example, to make a 500 $\mu\text{g}/\text{mL}$ stock solution, add 200 μL of buffer directly to the vial. Reconstituted antibody solutions can be stored at $2\text{--}6^{\circ}\text{C}$ for up to two weeks. For longer storage, divide the solutions into aliquots and freeze at $\leq -20^{\circ}\text{C}$. When properly stored, these products are stable for at least six months. **AVOID REPEATED FREEZING AND THAWING. PROTECT FLUORESCENT CONJUGATES FROM LIGHT.**

Properties

The purity and yield of each MAb preparation has been assessed by SDS polyacrylamide gel electrophoresis and immunofluorescent staining of fibroblasts in culture. In addition, these antibodies have been tested in a cytological experiment to ensure low nonspecific staining. At the time of preparation, the fluorescent anti-biotin conjugates are certified to be free of unconjugated dye.

Approximate absorption and fluorescence emission maxima for the Alexa Fluor 488 dye conjugate are 495 nm and 519 nm, respectively; for the Alexa Fluor 594 dye conjugate the absorption and emission maxima are 590 nm and 617 nm, respectively.

Application

Because protocols vary with the application, the appropriate dilution of anti-biotin should be determined empirically. For initial experiments, we recommend concentrations of 1–10 $\mu\text{g}/\text{mL}$.

In the case of the fluorescent conjugates, it is a good practice to centrifuge the antibody solution briefly in a microcentrifuge before use; only the supernatant should then be added to the experiment. This step will eliminate any protein aggregates that may have formed during storage, thereby reducing nonspecific background staining.

References

1. Proc Natl Acad Sci USA 71, 3537 (1974); 2. Biochim Biophys Acta 264, 165 (1972); 3. Methods Enzymol 184, 111 (1990); 4. Methods Biochem Anal 26, 1 (1980); 5. Methods Enzymol 279, 451 (1997); 6. J Immunol Methods 165, 177 (1993).

Product List *Current prices may be obtained from our Web site or from our Customer Service Department.*

Cat #	Product Name	Unit Size
A11242	anti-biotin, mouse IgG ₁ , monoclonal 2F5.....	100 µg
A31801	anti-biotin, mouse IgG ₁ , monoclonal 2F5, Alexa Fluor® 488 conjugate *1 mg/mL*	100 µL
A31800	anti-biotin, mouse IgG ₁ , monoclonal 2F5, Alexa Fluor® 594 conjugate *1 mg/mL*	100 µL

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

Please visit our Web site — www.probes.com — for the most up-to-date information.

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