

Product No. B 2766
Lot 057H8818

Anti-Mouse IgA (α -chain specific)
Biotin Conjugate

Antibody Developed in Goat
Affinity Isolated Antigen Specific Antibody

Antiserum is developed in goat using purified mouse IgA as the immunogen. Affinity isolated antigen specific antibody is obtained from goat anti-mouse IgA antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, that do not specifically bind to the α -chain of mouse IgA. Goat anti-mouse IgA is conjugated to N-Hydroxysuccinimidobiotin (Sigma Product No. H 1759) according to the method of Bayer, et al.¹ The conjugate is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA and 15 mM sodium azide (see MSDS)* as a preservative.

Specificity

Specificity of the biotin conjugated anti-mouse IgA antibodies is determined by Enzyme Linked Immunosorbent Assay (ELISA). The conjugate is specific for mouse IgA when tested against purified mouse IgA, IgG and IgM myeloma proteins.

Identity and Purity

Identity and purity of the antibody is established by immunoelectrophoresis, prior to conjugation. Electrophoresis of the antibody preparation followed by diffusion versus anti-goat IgG and anti-goat whole serum results in single arcs of precipitation.

Antibody Content

The product is provided with a specific antibody content of 0.4 mg/ml (prior to the addition of BSA).

Working Dilution: 1:61,000

Working dilution is defined as the dilution of conjugate that give a change in absorbance of 1.0 at 492 nm after 30 minutes of substrate conversion at 25°C (Voller, et al., and Guedson et al.)^{2,3}. Microtiter plates are coated with purified mouse IgA at a concentration of 200 ng/ml in 0.05 M carbonatebicarbonate buffer pH 9.6 (Carbonate-Bicarbonate Buffer capsules are available as Sigma Product No. C 3041). Following incubation with the biotinylated antibody, a solution of Avidin-Horseradish Peroxidase (Sigma Product No. A 3151, diluted in 0.01 M phosphate buffered saline, pH 7.4, containing 0.05% Tween 20 and 0.5% BSA) is added.

Substrate: 0.04% *o*-Phenylenediamine Dihydrochloride* (OPD, Sigma Product No. P 8412), and 0.012% Hydrogen Peroxide* (H₂O₂, Sigma Product No. H 1009) in phosphate-citrate buffer, pH 5.0 [25.7 ml 0.2 M dibasic sodium phosphate (Sigma Product No. S 0876), 24.3 ml 0.1 M citric acid (Sigma Product No. C 7129) and 50 ml deionized water].

*Add immediately before use.

In order to obtain best results it is recommended that each individual user determine their optimum working dilution by titration assay.

Storage

For continuous use, store at 2-8°C up to one month. For extended storage, the solution may be frozen 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.

References

1. Bayer, E.A., et al., Methods in Enzymology, **62**, 308 (1979).
2. Voller, A., et al., Bulletin WHO, **53**, 55 (1976).
3. Guedson, J.L., et al., J. Histochem. and Cytochem., **27**, 1131 (1979).

*Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.