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Product Information

Anti-Mouse IgG (whole molecule)—Alkaline Phosphatase produced in goat, affinity isolated antibody

Catalog Number A3562

Product Description

Antiserum is produced in goat using purified mouse IgG as the immunogen. Affinity isolated antibody is obtained from anti-mouse IgG antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to mouse IgG. Anti-Mouse IgG is conjugated to alkaline phosphatase by protein cross-linking with 0.2% glutaraldehyde.¹

Specificity of the antiserum is determined by immunoelectrophoresis (IEP) and Ouchterlony Double Diffusion (ODD) assays, prior to conjugation. By IEP, the antiserum reacts specifically with normal mouse serum and mouse IgG. By ODD, the antiserum reacts with mouse IgG1, IgG2a, IgG2b, IgG3, IgA, and IgM.

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

Reagent

Supplied as a solution in 0.05 M Tris buffer, pH 8.0, containing 1% BSA, 1 mM MgCl₂, 50% glycerol, and 15 mM sodium azide as a preservative.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage and Stability

Store at 2-8 °C.

Product Profile

<u>Dot blot</u>: a minimum titer of 1:30,000 is determined. Diluted conjugate detects ≤20 ng mouse lgG bound to nitrocellulose.

Substrate: 5-Bromo-4-chloro-3-indolyl Phosphate/Nitroblue Tetrazolium (BCIP/NBT), SIGMA*FAST*[™]Tablets, Catalog Number B5655.

<u>Direct ELISA</u>: a minimum titer of 1:30,000 is determined Titer is defined as the dilution of conjugate sufficient to give a change in absorbance of 1.0 at 405 nm after 30 minutes of substrate conversion at 25 °C.² Microtiter plates are coated with purified mouse IgG at

Microtiter plates are coated with purified mouse IgG at a concentration of 5 μ g/mL in 0.05 M carbonate-bicarbonate buffer, pH 9.6.

Carbonate/Bicarbonate Buffer capsules are available as Cat. No. C3041.

Substrate: *p*-Nitrophenyl phosphate (pNPP), Catalog Number N2765, 1.0 mg/mL in 10% diethanolamine buffer, pH 9.8, containing 0.5 mM MgCl₂.

Indirect Immunohistology: a minimum titer of 1:50 is determined by an indirect assay using formalin-fixed, paraffin-embedded sections of human tonsil and Monoclonal Anti-Human IgG, Catalog Number I6760, as the primary antibody.

Substrate: Fast Red TR/AS-MX Napthol Phosphate³ (SIGMA*FAST* Tablets Catalog Nos. F4523 or F4648.

 $\frac{Immunoblotting}{Immunoblotting}: a minimum titer of 1:30,000 is determined. Mouse IgG was detected directly using 10 <math display="inline">\mu g$ protein under reducing conditions on an SDS-PAGE gradient (4-20%) gel. The protein was transferred to nitrocellulose, blocked with 5% BSA in 0.05 M Tris and then incubated with the conjugate.

Substrate: 5-Bromo-4-chloro-3-indolyl Phosphate/Nitroblue Tetrazolium (BCIP/NBT), SIGMA*FAST* Tablets, Catalog Number B5655.

Note: Working dilutions should be determined by titration assays. Due to differences in assay systems, these titers may not reflect the user's actual working dilution.

References

1. Avrameas, V., Immunochemistry, 6, 43, (1969).

- 2. Voller, A., et al., Bull. World Health Org., **53**, 55 (1976).
- 3. Pluzek, K. and Ramlau, R., Alkaline Phosphatase Labeled Reagents, in CRC Handbook of Immunoblotting of Proteins, Bjerrum, O., and Heegaard, N., (Eds.), CRC Press Inc., Boca Raton, FL, 1, 177 (1988).

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