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Product Information

Anti-Horse IgG (whole molecule)–Alkaline Phosphatase
produced in rabbit, affinity isolated antibody

Catalog Number **A6063**

Product Description

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

Specificity of the antiserum is determined by immunoelectrophoresis, prior to conjugation, versus normal horse serum and horse IgG.

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

Reagent

Provided as a solution in 0.05 M Tris buffer, pH 8.0, containing 1% BSA, 1 mM $MgCl_2$, 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

Store at 2-8 °C.

Product Profile

Direct ELISA: Minimum 1:30,000
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 horse IgG at a concentration of 5 µg/ml in 0.05 M carbonate-bicarbonate buffer, pH 9.6
Carbonate/Bicarbonate Buffer capsules are available as Catalog Number C3041.

Substrate: *p*-Nitrophenyl Phosphate (pNPP), Catalog Number N2765, 1.0 mg/ml in 10% diethanolamine buffer, pH 9.8, containing 0.5 mM $MgCl_2$.

Western Blotting: Minimum 1:30,000
Horse IgG (10 µg) was run under reducing conditions on an SDS-PAGE gradient (4-20%) gel. The protein was transferred to nitrocellulose, blocked with 0.5% BSA in 0.05 M Tris and then incubated with 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., *Bulletin WHO*, **53**, 55 (1976).
3. Pluzek, K.J. and R. Ramlau, Alkaline Phosphatase Labeled Reagents, in CRC Handbook of Immunoblotting of Proteins, Vol. 1: Technical Descriptions, O.J. Bjerrum and N.H.H. Heegaard, Eds., (CRC Press Inc., Boca Raton, FL), p. 177, 1988.

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