

Product Information

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

Catalog Number **A3687**

Product Description

Antiserum is produced in goat using purified rabbit IgG as the immunogen. Affinity isolated antibody is obtained from goat antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulin, which do not specifically bind to rabbit IgG. Anti-Rabbit 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 rabbit serum and rabbit IgG.

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

Supplies 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/Stability

Store at 2-8 °C.

Product Profile

Dot Blot: minimum dilution 1:30,000

Diluted conjugate detects up to 20 ng rabbit IgG bound to nitrocellulose.

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

Direct ELISA: minimum titer 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 rabbit 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 MgCl₂.

Immunohistology: minimum dilution 1:50

Determined by an indirect assay using formalin- fixed, paraffin-embedded sections of human tonsil and Anti-Human IgG, Catalog Number I2011, as the primary antibody.

Substrate: Fast Red TR/AS-MX Naphthol Phosphate³ SIGMAFAST Tablets Catalog Nos. F4523 or F4648.

Western Blotting: minimum dilution 1:30,000

Rabbit IgG was detected directly using 10 µg 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), SIGMAFAST 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. and Ramlau, R., 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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