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Product Information

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

Catalog Number **A8438**

Product Description
Anti-Rat IgG (whole molecule) is produced in goat using purified rat IgG as the immunogen. Affinity isolated antibody is obtained from anti-rat IgG antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to rat IgG. Anti-Rat IgG is conjugated to alkaline phosphatase by protein cross-linking with 0.2% glutaraldehyde.[1]

Specificity of Anti-Rat IgG (whole molecule)–Alkaline Phosphatase is determined by ELISA.

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.

Reagent
Supplied 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
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
Protein concentration : 0.5-8.0 mg conjugate/mL

Dot Blot: minimum dilution 1:30,000
Diluted conjugate detects < 20 ng rat IgG bound to nitrocellulose.
Substrate: 5-Bromo-4-chloro-3-indolyl Phosphate/ Nitroblue Tetrazolium (BCIP/NBT), SIGMA*FAST*™ 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 rat IgG at a concentration of 5 µg/mL in 0.05M 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 $MgCl_2$.

Direct Immunohistochemistry: minimum 1:50
Titer was determined using frozen mouse spleen sections.

Substrate: Fast Red TR/AS-MX Napthol Phosphate[3] SIGMA*FAST* Tablets, Catalog Nos.F4523 or F4648.

Immunoblotting: minimum 1:30,000
Rat IgG was detected directly using 10 µg protein per lane. Reducing conditions on a SDS-PAGE gradient (4-20%) gel were used. The protein was transferred to nitrocellulose, blocked with 0.5% BSA in 0.05 M Tris, transferred and then incubated with the conjugate.

Substrate: BCIP/NBT, SIGMA*FAST* Tablets.

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 Hlth. Organ*., **53**, 55 (1976).

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