

## Product Information

### Anti-Guinea Pig IgG (whole molecule)- Alkaline Phosphatase

produced in goat, affinity isolated antibody

Catalog Number **A5062**

#### Product Description

Anti-Guinea Pig IgG (whole molecule) is produced in goat using purified guinea pig IgG as the immunogen. Affinity isolated antibody is obtained from anti-guinea pig IgG antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to guinea pig IgG. Anti-Guinea Pig IgG is conjugated to alkaline phosphatase by protein cross linking with 0.2% glutaraldehyde.<sup>1</sup>

Specificity of the antiserum is determined by immunoelectrophoresis, prior to conjugation, versus normal guinea pig serum and guinea pig 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

Provided as a solution in 0.05 M Tris buffer, pH 8.0, containing 1 mM MgCl<sub>2</sub>, 50% glycerol, and 15 mM sodium azide.

#### 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 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.<sup>2</sup> Microtiter plates are coated with purified guinea pig 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 Cat. No. C3041.

Substrate: *p*-Nitrophenyl Phosphate (pNPP), Cat. No. N2765, 1.0 mg/mL in 10% diethanolamine buffer, pH 9.8, containing 0.5 mM MgCl<sub>2</sub>.

#### Dot Blot

Diluted conjugate detects a minimum of 0.5 ng guinea pig IgG bound to nitrocellulose.

Substrate: 5-Bromo-4-chloro-3-indolyl Phosphate/Nitroblue Tetrazolium (BCIP/NBT), SIGMAFAST™ Tablets, Cat. No. B5655.

#### Immunohistology

The minimum working antibody dilution of 1:50 was determined by an indirect assay using formalin-fixed, paraffin-embedded sections of human skin and Anti-Keratin, Cat. No. K4252, as the primary antibody.

Substrate: Fast Red TR/AS-MX naphthol phosphate<sup>3</sup> SIGMAFAST Tablets Cat. Nos. F4523 or F4648.

#### Western Blotting

Minimum working antibody dilution: 1:30,000 Guinea Pig 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 incubated with the conjugate.

Substrate: 5-Bromo-4-chloro-3-indolyl Phosphate/Nitroblue Tetrazolium (BCIP/NBT, SIGMAFAST Tablets, Cat. No. 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 Organ.*, **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**, p. 177, 1988.

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