

## Product Information

**Anti-Rabbit IgG (whole molecule),  
F(ab')<sub>2</sub> fragment-Alkaline Phosphatase**  
produced in goat, affinity isolated antibody

Catalog Number **A3937**

### Product Description

Anti-Rabbit IgG is produced in goat using purified rabbit IgG as the immunogen. The F(ab')<sub>2</sub> fragment of the antibody is obtained from pepsin digested antiserum by immunospecific methods of purification. Affinity isolation removes essentially all goat serum proteins, including immunoglobulins which do not specifically bind to rabbit IgG. Anti-Rabbit IgG is conjugated to alkaline phosphatase, by protein cross linking with glutaraldehyde.<sup>1</sup>

Specificity of the antiserum is determined by immunoelectrophoresis (IEP) versus normal rabbit serum and rabbit IgG, prior to conjugation.

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. The antibody preparation is found to consist only of the F(ab')<sub>2</sub> fragment of goat IgG as determined by SDS-PAGE.

No contamination with goat IgG whole molecule is observed.

### Reagent

Supplied as a solution in 0.05 M Tris buffer, pH 8.0, containing 1% BSA, 1 mM MgCl<sub>2</sub>, 50% glycerol, with 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

**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.<sup>2</sup>

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 MgCl<sub>2</sub>.

**Dot Blot:** 1:30,000

Diluted conjugate detects less than 20 ng rabbit IgG bound to nitrocellulose.

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

**Immunohistochemistry:** Minimum 1:50

Determined by an indirect assay using formalin-fixed, paraffin-embedded sections of human tonsil and Anti-Human IgG, Catalog Number I9764, as the primary antibody.

Substrate: Fast Red TR/AS-MX Naphthol Phosphate<sup>3</sup> SIGMAFAST™ Tablets, Catalog Nos. F4523 or F4648.

**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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