

## Product Information

### **Monoclonal Anti-Rabbit IgG ( $\gamma$ -chain specific)- Alkaline Phosphatase, clone RG-96**

produced in mouse, purified immunoglobulin

Catalog Number **A2556**

#### **Product Description**

Monoclonal Anti-Rabbit IgG (mouse IgG1 isotype) is derived from the hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. Purified rabbit IgG was used as the immunogen. The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Catalog Number ISO2. The immunoglobulin fraction of the ascites fluid containing anti-rabbit immunoglobulins is conjugated to alkaline phosphatase by protein cross-linking with 0.2% glutaraldehyde.

Monoclonal Anti-Rabbit IgG ( $\gamma$ -chain specific) recognizes an epitope located on the  $\gamma$  (heavy)-chain of rabbit IgG. In immunoblotting, the antibody recognizes both native and denatured forms of rabbit IgG. In ELISA, the antibody is specific for rabbit IgG, and shows no cross-reactivity with rabbit IgA and IgM or human IgG, IgA, and IgM. No cross-reaction is observed with IgG from the following species: bovine, cat, chicken, dog, goat, guinea pig, horse, pig, rat, or sheep.

Rabbit antibodies against many analytes are in wide use as primary antibodies in various assay techniques, both in research and clinical applications. Secondary antibodies often lack species specificity for the primary rabbit immunoglobulins. In many instances, such antibodies also recognize non-related immunoglobulins that appear in the preparation being tested resulting in increased levels of background staining and false positives. To resolve this, extensive adsorbing steps must be incorporated into the manufacturing process. Alkaline Phosphatase-Monoclonal Anti-Rabbit Immunoglobulins, which do not recognize human or many other species immunoglobulins, can serve as an essential tool, especially when used as a secondary reagent in immunohistochemistry.

#### **Reagent**

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**

ELISA (direct): Minimum titer 1:50,000

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>1</sup>

Microtiter plates are coated with purified rabbit IgG at a concentration of 5  $\mu$ g/mL in 0.05 M carbonate-bicarbonate buffer, pH 9.6. Carbonate-Bicarbonate Buffer, capsules, Cat. No. C3041.

Substrate: 4-Nitrophenyl phosphate (pNPP) disodium salt, Cat. No. N2765, 1.0 mg/ml in 10% diethanolamine buffer, pH 9.8, containing 0.5 mM  $MgCl_2$ .

#### Immunoblotting

Working dilution of 1:200,000-1:400,000 is determined using an immunoblot assay detecting Actin in total cell extract of HeLa cells (5-10  $\mu$ g per well)

#### Immunohistochemistry

A minimum antibody dilution of 1:100 was determined in an indirect assay using formalin-fixed, paraffin-embedded human tonsil and Anti-Human IgG, Cat. No. I8635, as the primary antibody.

**Note:** Working dilutions should be determined by titration assay. Due to differences in assay systems, these titers may not reflect the user's actual working dilution.

## Reference

1. Voller, A., et al., Bulletin WHO, **53**, 55 (1976).

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