

3050 Spruce Street, St. Louis, MO 63103 USA Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757 email: techservice@sial.com sigma-aldrich.com

# **Product Information**

# Anti-Human IgM (μ-chain specific) Peroxidase Conjugate

F(ab')<sub>2</sub> Fragment of Affinity Isolated Antigen Specific Antibody Antibody Developed in Goat

Product No. A4290

## **Product Descripton**

Anti-Human IgM is developed in goat using purified Human IgM as the immungen. The  $F(ab')_2$  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 the  $\infty$ -chain of human IgM. Goat anti-Human IgM is conjugated to Sigma Horseradish Peroxidase, Type VI (Product No. P 8375) by a modification of the periodate method of Wilson and Nakane.  $^1$ 

Specificity of the peroxidase conjugated anti-human IgM antibodies for the ∞-chain of human IgM is determined by ELISA. The conjugate is specific for human IgM when tested against purified human IgA, IgG, IgM, Bence Jones Kappa and Bence Jones Lambda myeloma proteins.

Identity and purity of the antibody is established by immunoelectrophoresis (IEP) versus anti-goat IgG and anti-goat whole serum. Following incubation, a single arc of precipitation is observed against the anti-goat IgG and the anti-goat whole serum. The antibody preparation is found to consist of only the F(ab')<sub>2</sub> fragment of goat IgG as determined by SDS-polyacrylamide gel electrophoresis (PAGE). No contamination with goat IgG whole molecule is observed.

#### Reagents

The conjugate is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA with preservative.

## **Product Profile**

Titer: Minimum 1:10,000 (Direct ELISA)
We are now reporting lot specific information as a titer
by direct ELISA rather than as a working dilution. Titer

is defined as the dilution of conjugate sufficient to give a change in absorbance of 1.0 at 450 nm after 30 minutes of substrate conversion at 25 °C (Voller, *et al.*<sup>2</sup>). Microtiter plates are coated with purified human IgM at a concentration of 5 ∞g/mL in 0.05 M carbonate/bicarb-onate buffer, pH 9.6 carbonate/bicarbonate buffer capsules are available as Product No. C 3041). Substrate: *o*-Phenylenediamine dihydrochloride (OPD, Product No. P 8287), 0.4 mg/ml in 0.05 M phosphate-citrate buffer, pH 5.0 containing 0.03% sodium perborate (phosphate-citrate buffer capsules with sodium perborate are available as Product No. P 4922).

Working dilution should be determined by titration assay. Due to product improvement and changes in the assay procedure, we now list a lot specific titer by direct ELISA for this product. Due to differences in assay systems, this titer may not reflect the user's actual working dilution.

## Storage

For continuous use, store at 2-8 °C. For extended storage, the solution may be frozen in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

### References

- Wilson, M.B. and P.K. Nakane, Immunofluorescence and Related Staining Techniques (Elsevier/ North- Holland Biomedical Press, Amsterdam), p. 215 (1978).
- 2. Voller, A., et al., Bulletin WHO, 53, 55 (1976).

Pcs, AD, SM 08/20-1