

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone (800) 325-5832 (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

Anti-Human Polyvalent Immunoglobulins (IgA, IgG, IgM)
FITC Conjugate
Antibody developed in Rabbit
IgG Fraction of Antiserum

Product Number F 4637

Product Description

Anti-Human Polyvalent Immunoglobulins to human IgA, IgG, and IgM is developed in rabbit using purified human IgA, IgG, and IgM as the immunogens. Whole antisera is fractionated and then further purified by ion exchange chromatography to provide the IgG fraction of antiserum. This fraction is essentially free of other rabbit serum proteins. The IgG fraction is then conjugated to crystalline fluorescein isothiocyanate (FITC) in an alkaline reaction and then further purified to remove free FITC.

Specificity for each immunoglobulin is determined by immunoelectrophoresis (IEP) 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), prior to conjugation. Electrophoresis of the antibody preparation followed by diffusion against anti-rabbit whole serum result in single arcs of precipitation in the gamma region.

Reagents

The conjugate is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Precautions and Disclaimer

Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for a maximum of one month. 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.

Product Profile

- The minimum dilution of 1:128 was determined by direct immunofluorescent labeling of human peripheral blood lymphocytes.
- In an A.N.A (anti-nuclear antibody) assay, a minimum dilution of 1:128 was used on acetone-fixed rat liver cells with A.N.A positive serum as the primary antibody.
- A dilution of 1:80 was determined by direct immunofluorescent labeling of formalin-fixed, paraffin-embedded human tonsils.

In order to obtain best results, it is recommended that each individual user determine optimum working dilutions for their system by titration assay.

Protein Concentration: 10-20 mg/ml by absorbance at 280 nm ($E_{280}^{1\%}$ = 14.0).

F/P Molar Ratio: 2.5 to 6.5

The F/P molar ratio is determined spectrophotometrically as follows:

$$F/P = \frac{A_{495} \times 1.4}{A_{280} - (0.36 \times A_{495}) \times 0.2} \times 0.41$$

Where:

0.2 = The extinction coefficient of bound FITC at a concentration of 1 μg/ml at pH 7.2.

0.36 = The fluorochrome absorbance correction factor (non-protein absorbance).

0.41 = The factor for conversion of fluorochrome to protein ratios from weight to molar ratios.

In an agar diffusion assay, the conjugate produces a precipitation arc at a minimum dilution of 1:16 against a dilution of normal human serum.

JWM/KMR 09/02