Product No. A-5175
Lot 056H4813

Anti-Human Lambda Light Chain (Bound & Free) Peroxidase Conjugate

Antibody developed in Goat
Affinity Isolated Antigen Specific Antibody

Antiserum is developed in goat using purified human lambda light chains as the immunogen. Affinity isolated antigen specific antibody is obtained from goat anti-human lambda antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to lambda light chains (bound and free). Goat anti-human lambda affinity isolated antibody is conjugated to horseradish peroxidase by protein cross linking with 0.2% glutaraldehyde. The conjugate is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 0.01% thimerosal as a preservative.

Specificity

Specificity of the Peroxidase Conjugated Anti-Human Lambda is determined by Enzyme Linked Immunosorbent Assay (ELISA). The conjugate is specific for human lambda light chain (bound and free) when tested against human IgA, IgG, IgM, Bence Jones Kappa and Lambda myeloma proteins.

Identity and Purity

Identity and purity of the antibody is established by immunoelectrophoresis (IEP), 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.

Enzyme Activity: 445 purpurogallin units/ml

Enzyme activity is determined using 5% Pyrogallol (Sigma Product No. P-0381) in deionized water, pH 6.0, at 20°C. One purpurogallin unit will form 1 mg of purpurogallin from pyrogallol in 20 seconds at pH 6.0, 20°C.

Titers

1. 1:35,000 (Direct ELISA)

We are now reporting lot specific information as a titer by direct ELISA rather than a working dilution (see below). 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.).[1] Microtiter plates are coated with purified human lambda light chain (bound) at a concentration of 5 µg/ml in 0.05 M carbonate-bicarbonate buffer, pH 9.6 (Carbonate-Bicarbonate Buffer Capsules are available as Sigma Product No. C-3041).

Substrate: *o*-Phenylenediamine Dihydrochloride (OPD, Sigma 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 Sigma Product No. P-4922).

2. Dot Blot

a. A dilution of 1:1,000 was determined by direct assay using 40 ng human IgG per dot.
b. A dilution of 1:4,000 was determined by direct assay using 40 ng Bence Jones lambda per dot.

3. Immunohistology

A dilution of 1:50 was determined in a direct assay using paraffin-embedded, formalin-fixed human tonsils.

Working Dilution

Working dilution should be determined by titration assay. 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

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