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Total Human IgG ELISA Assay



Catalog No. E4-007

*For Research Use Only*

Intended Use:

To quantitate total human Immunoglobulin G (IgG)

Principle of Procedure:

Solid phase capture sandwich ELISA assay using a microwell format.

Shelf Life:

The expiration date for the package and each component is stated on the label(s). Store all components at 2° - 8° degrees C with the exception of the standard, which should be stored at -20°C.

Specimen and Standard Dilutions:

Dilute each serum or plasma specimen to be tested initially 1:100 in phosphate buffered saline (PBS), then dilute 1:100 in the IgG specimen diluent provided. The final dilution dilution factor will be 1:10,000.

Prepare serial two fold dilutions of the human IgG standard: Neat (N), 1:2, 1:4, 1:8 etc. with the specimen diluent provided. Use the specimen diluent alone as the blank control well.

Materials Supplied:

Anti-Human IgG coated microwell strips 12x8 with plastic frame  
 HRP conjugated goat anti-human IgG -12mL  
 IgG standard (pre-diluted)- 1 ml  
 TMB/peroxide substrate color developer -12mL  
 IgG specimen diluent -1 X 60mL  
 Sulfuric acid termination reagent (0.5N) -12mL  
 15 X Wash buffer concentrate - 60mL

Limitations of the Procedure:

No single assay should be used as the only basis for arriving at a diagnostic conclusion. For research use only.

Dynamic Range:

0.156 mg/mL to 10.0 mg/mL.

Reproducibility:

C.V. 6%-10% depending upon the region of the standard curve.

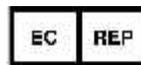
Assay Procedure:

\* Caution: All human fluids should be treated as infectious agents that could carry HIV.

\*Allow each reagent to reach room temperature before use

1. Add 100uL of diluted specimen or standard to each microwell
2. Incubate at room temperature for 45 minutes
3. Decant and wash each microwell four times with wash buffer (dilute buffer 1:15 with reagent grade water)
4. Add 100uL of HRP conjugated goat anti-human IgG to each well
5. Incubate at room temperature for 45 minutes
6. Decant and wash as in step 3
7. Add 100uL of TMB/peroxide substrate and incubate at room temperature for 15 minutes
8. Terminate the reaction with 100uL of 0.5N sulfuric acid
9. Zero the microwell reader at 450nm using the specimen diluent zero control well
10. Determine the optical density (O.D.) of the remaining wells
11. Construct a standard curve using the O.D. values obtained for each of the standards
12. Interpolate the unknowns from the standard curve

\*Interpolated concentrations greater than 10 mg/ml should be sub diluted 1:4 and re-assayed then corrected mathematically



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