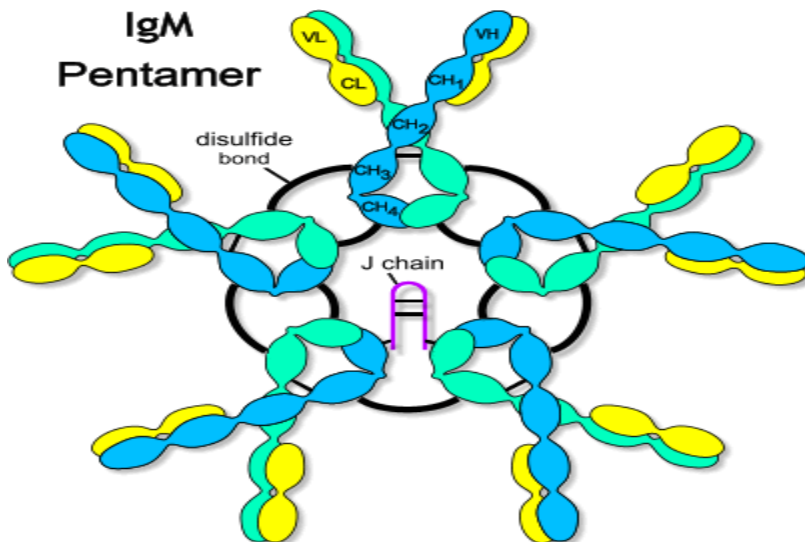


# Total Human IgM Assay:



## Intended Use:

To quantitate total human Immunoglobulin M (IgM)

## 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 components at 2°-8°C, except for standard, which should be stored at -20°C.

## Patient and Standard Dilutions:

Dilute each serum or plasma specimen to be tested initially 1:1000 in phosphate buffered saline (PBS), e.g. 10ul of specimen into 10 ml of PBS, then subdilute 1:10 with the IgM specimen diluent provided for a final dilution of 1:10,000. Prepare serial two fold dilutions of the human IgM standard: Neat, 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 IgM coated microwell strips 12x8 with plastic frame  
HRP conjugated goat anti-human IgM -12mL  
IgM standard (pre-diluted 1: 10,000)  
TMB/peroxide substrate color developer -12mL  
IgM specimen diluent (Specimen Diluent Green II) -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:

0.031 $\mu$ g/mL - 2.0 $\mu$ g/mL.

### Reproducibility:

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

### Assay Procedure:

\*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 60 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 IgM to each well
5. Incubate at room temperature for 60 minutes
6. Decant and wash as in step 3
7. Add 100uL of TMB/peroxide substrate and incubate at room temperature for 30 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

**Note: Constructed 4 Parameter Curve displayed.**

### Typical Standard Curve:

