Reagents and supplies for Zika IgM ELISA

Coating buffer:
Carbonate/bicarbonate buffer pH 9.6
1.59g Na₂CO₃ + 2.93g NaHCO₃ diluted in 1L water

Wash buffer:
Phosphate buffered saline (PBS), 0.05% Tween 20, pH 7.2
PBS is available in powdered form from multiple commercial sources

Blocking buffer:
PBS/5% milk/ 0.5% Tween 20

Coating antibody:
Goat anti-human IgM
Kirkegaard and Perry Laboratories cat# 01-10-03

Positive control:
6B6C-1 huIgM chimera (CDC) or Zika IgM-positive serum

Negative control:
Known virus-negative serum

Viral antigen:
Sucrose-acetone extracted suckling mouse brain viral antigens, gamma-irradiated tissue culture antigen, non-infectious, previously titrated, or COS-1 antigen (non-infectious) (CDC)

Normal antigen:
Sucrose-acetone extracted suckling mouse brain antigen, tissue culture antigen, or normal COS-1 antigen (CDC)

Detecting antibody conjugate:
Horseradish peroxidase conjugated monoclonal antibody 6B6C-1 (commercially available from Hennessy Research and InBios. Note: the Hennessy detecting antibody conjugate cannot be used with COS-1 antigens)*.

Substrate:
3,3',5,5' tetramethylbenzidine base (Enhanced K-Blue TMB substrate), Neogen Corp. Cat# 308175

Stop solution:
1 N H₂SO₄ (50 µl/well)

Plates:
Immulon 2 HB flat-bottomed 96 well plates
Dynatech Technologies cat# 3455

Microplate washer
Microplate reader with 450 nm filter

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Refrigerator
Incubator (37C)
Single and multi-channel pipettors
Reagent reservoirs
Ziploc bags, paper towels

*For updated information regarding conjugates, please refer to LRN guidance.