

Product Information

VZV IgM ELISA

Catalog Number **SE120137**
Storage Temperature 2–8 °C

TECHNICAL BULLETIN

Product Description

Varicella zoster virus causes chickenpox, a highly contagious disease, acquired by touching blisters or respiratory secretions, or through the air. A person is usually infectious 1–2 days before the rash to 4–5 days after the start of the rash, or until the blisters have formed crusts. Symptoms start about 2–3 weeks after exposure and include fever, tiredness, and an itchy rash with small blisters that dry up and form scabs in 2–4 days. More severe but rare problems or complications that could occur are pneumonia (especially in adults), skin infection, blood infection, and encephalitis. Approximately 90% of chickenpox cases are in children 1–14 years of age and 90% of people have had chickenpox by their early 20's. The reactivated form (herpes zoster, shingles) of VZV infection generally occurs in older adults whose immunity has waned, in infants or children exposed to VZV in the perinatal period, or in the immunocompromised. VZV infection during pregnancy infrequently leads to maternal pneumonia. Chickenpox can occur during pregnancy in women seropositive for VZV, especially when seropositive at low titer, with low-avidity, largely IgG3 antibodies. Maternal VZV infection during pregnancy (especially between 13–20 weeks gestation) can be associated with outcomes ranging from skin scarring or limb hypoplasia to multi system involvement and death.

Because VZV and herpes simplex virus (HSV) can cross-react, viral culture can be used to detect and differentiate HSV from VZV, but PCR testing may prove the most valuable for diagnosing and differentiating active infection. IgG antibodies can be detected 9 days after the onset of rash in varicella, 10 days in zoster; immunoreactivity peaks at an average 66 and 27 days, respectively. The IgM response to varicella is detected at 6–7 days post-onset and peaks at an average 14 days; IgM response to zoster is detectable at 8–10 days and peaks at 18–19 days.

The VZV IgM ELISA Kit is intended for the detection of IgM antibody to VZV in human serum or plasma. Diluted serum (serum diluent contains sorbent to remove Rheumatoid Factor and human IgG interference) is added to wells coated with purified antigen. IgM specific antibody, if present, binds to the antigen. All unbound materials are washed away and the enzyme conjugate is added to bind to the antibody-antigen complex, if present. Excess enzyme conjugate is washed off and substrate is added. The plate is incubated to allow the oxidation of the substrate by the enzyme. The intensity of the color generated is proportional to the amount of IgM specific antibody in the sample.

Components

Materials Provided	96 Tests
Microwell coated with VZV antigen	12 x 8 x 1
Sample Diluent: 1 bottle (ready to use)	22 mL
Calibrator: 1 vial (ready to use)	1 mL
Positive Control: 1 vial (ready to use)	1 mL
Negative Control: 1 vial (ready to use)	1 mL
Enzyme conjugate: 1 bottle (ready to use)	12 mL
TMB Substrate: 1 bottle (ready to use)	12 mL
Stop Solution: 1 bottle (ready to use)	12 mL
Wash concentrate 20x: 1 bottle	25 mL

Reagents and Equipment Required but Not Provided.

1. Distilled or deionized water
2. Precision pipettes, Disposable pipette tips,
3. ELISA reader capable of reading absorbance at 450 nm
4. Absorbent paper or paper towel
5. Graph paper

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation InstructionsSample Preparation

1. Collect blood specimens and separate the serum.
2. Specimens may be refrigerated at 2–8 °C for up to seven days or frozen for up to six months. Avoid repetitive freezing and thawing.

20x Wash Buffer Concentrate

Prepare 1x Wash buffer by adding the contents of the bottle (25 mL, 20x) to 475 mL of distilled or deionized water. Store at room temperature (18–26 °C).

Storage/Stability

Store the kit at 2–8 °C

Procedure

Notes: The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.

Optimal results will be obtained by strict adherence to the test protocol. Precise pipetting as well as following the exact time and temperature requirements is essential.

The test run may be considered valid provided the following criteria are met:

1. If the O.D. of the Calibrator is >0.250 .
2. The Ab index for Negative control should be <0.9 .
3. The Ab index for Positive control should be >1.2 .

Bring all specimens and kit reagents to room temperature (18–26 °C) and gently mix.

1. Place the desired number of coated strips into the holder.
2. Negative control, positive control, and calibrator are ready to use. Prepare 21-fold dilution of test samples, by adding 10 μL of the sample to 200 μL of Sample Diluent. Mix well.
3. Dispense 100 μL of diluted sera, calibrator, and controls into the appropriate wells. For the reagent blank, dispense 100 μL of Sample Diluent in 1A well position. Tap the holder to remove air bubbles from the liquid and mix well. Incubate for 20 minutes at room temperature.
4. Remove liquid from all wells. Wash wells three times with 300 μL of 1x wash buffer. Blot on absorbent paper or paper towel.
5. Dispense 100 μL of enzyme conjugate to each well and incubate for 20 minutes at room temperature.
6. Remove enzyme conjugate from all wells. Wash wells three times with 300 μL of 1x wash buffer. Blot on absorbent paper or paper towel
7. Dispense 100 μL of TMB substrate and incubate for 10 minutes at room temperature.
8. Add 100 μL of stop solution.
9. Read O.D. at 450 nm using ELISA reader within 15 minutes. A dual wavelength is recommended with reference filter of 600–650 nm.

Results

Calculations

1. Check Calibrator Factor (CF) value on the calibrator bottle. This value might vary from lot to lot. Make sure the value is checked on every kit.
2. Calculate the cut-off value: Calibrator OD x Calibrator Factor (CF).
3. Calculate the Ab (Antibody) Index of each determination by dividing the O.D. value of each sample by cut-off value.

Example of typical results:

Calibrator mean OD = 0.8

Calibrator Factor (CF) = 0.5

Cut-off Value = $0.8 \times 0.5 = 0.400$

Positive control O.D. = 1.2

Ab Index = $1.2/0.4 = 3$

Patient sample O.D. = 1.6

Ab Index = $1.6/0.4 = 4.0$

Interpretation

The following is intended as a guide to interpretation of VZV IgM antibody index (Ab Index) test results; each laboratory is encouraged to establish its own criteria for test interpretation based on sample populations encountered.

<0.9 – No detectable antibody to VZV IgM by ELISA

0.9–1.1 – Borderline positive. Follow-up testing is recommend if clinically indicated.

>1.1 – Detectable antibody to VZV IgM by ELISA

Notes: To enhance sensitivity and specificity of this IgM test, the provided sample diluent has been formulated to block IgG and Rheumatoid Factor (RF) interferences. Turbidity could be seen after diluting serum with sample diluent. This turbidity is due to the blocking of serum IgG and has shown interference with test results. It can be removed by centrifugation.

In specimens with high RF and high autoimmune antibodies, the possibility of eliminating the interferences cannot be ruled out entirely.

Lipemic or hemolyzed samples may cause erroneous results.

References

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