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Product Information

EBV-VCA IgA ELISA

Catalog Number **SE120046**
Storage Temperature 2–8 °C

TECHNICAL BULLETIN

Product Description
Epstein-Barr virus (EBV) is a herpes virus known to cause infectious mononucleosis (IM). EBV infection may demonstrate a wide spectrum of clinical symptoms. The majority of primary EBV infections are transmitted via saliva, occur during childhood, and are sub-clinical. In the U.S., 50% of the population demonstrate EBV antibodies before the age of 5 years; 80% by adulthood. Transfusion-associated EBV infections have also been reported. Epstein-Barr virus has also been associated in the pathogenisis of two human cancers, Burkitt's lymphoma and nasopharyngeal carcinoma. Burkitt's lymphoma is primarily observed in Sub-Sahara Africa, especially in African children, and in New Guinea. Nasopharyngeal carcinoma is observed in Asia, most notably in Southern China.

The EBV-VCA IgA ELISA Kit is intended for the detection of IgA antibody to EBV-VCA in human serum or plasma.

The diluted serum is added to wells coated with purified antigen. IgA 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 IgA specific antibody in the sample.

Components

Materials Provided	96 Tests
Microwell coated with EBV-VCA antigen	12 × 8 × 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 20×: 1 bottle	25 ml

Reagents and Equipment Required but Not Provided.
- Distilled or deionized water
- Precision pipettes
- Disposable pipette tips
- ELISA reader capable of reading absorbance at 450 nm
- Absorbent paper or paper towel
- 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 Instructions
Sample 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 of serum sample.

20× Wash Buffer Concentrate
Prepare 1× Wash buffer by adding the contents of the bottle (25 mL, 20×) 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.

It is recommended that standards, controls, and serum samples be run in duplicate

Optimal results will be obtained by strict adherence to this protocol. Accurate and precise pipetting, as well as following the exact time and temperature requirements prescribed are essential. Any deviation from this may yield invalid data.

The test run may be considered valid provided the following criteria are met:
1. The O.D. of the Calibrator should be >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 3 times with 300 μl of 1× 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 3 times with 300 μl of 1× 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 × 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 × 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

Notes: The test results obtained using this kit serve only as an aid to diagnosis and should be interpreted in relation to the patient’s history, physical findings, and other diagnostic procedures.

Lipemic or hemolyzed samples may cause erroneous results.

Interpretation

The following is intended as a guide to interpretation of this EBV-VCA IgA 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 EBV-VCA IgA by ELISA
0.9–1.1 – Borderline positive. Follow-up testing is recommended if clinically indicated.
>1.1 – Detectable antibody to EBV-VCA IgA by ELISA

References

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