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Cat#: EV010G (96 Tests)
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# **EBV-VCA IgG ELISA**

Catalog No.: EV010G (96 Tests)

# **INTENDED USE**

The Calbiotech, Inc. (CBI) EBV-VCA IgG ELISA Kit is intended for the detection of IgG antibody to EBV-VCA in human serum or plasma.

# SUMMARY AND EXPLANATION

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 majorities 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.

# PRINCIPLE OF THE TEST

Diluted patient serum is added to wells coated with purified antigen. IgG 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 IgG specific antibody in the sample.

	MATERIALS PROVIDED	96 Tests
1.	Microwells coated with EBV-VCA antigen	12x8x1
2.	Sample Diluent: 1 bottle (ready to use)	22 ml
3.	Calibrator: 1 Vial (ready to use)	1ml
4.	Positive Control: 1 vial (ready to use)	1ml
5.	Negative Control: 1 vial (ready to use)	1ml
6.	Enzyme conjugate: 1 bottle (ready to use)	12ml
7.	TMB Substrate: 1 bottle (ready to use)	12ml
8.	Stop Solution: 1 bottle (ready to use)	12ml
9.	Wash concentrate 20X: 1 bottle	25ml

#### MATERIALS NOT PROVIDED

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

# STORAGE AND STABILITY

- Store the kit at 2-8° C.
- Keep microwells sealed in a dry bag with desiccants.
- 3. The reagents are stable until expiration of the kit.
- Do not expose test reagents to heat, sun or strong light.

#### WARNINGS AND PRECAUTIONS

- 1. For Research Use Only. Not for use in diagnostic procedures.
- 2. For laboratory use.
- 3. Potential biohazardous materials:
  - The calibrator and controls contain human source components which have been tested and found non-reactive for hepatitis B surface antigen as well as HIV antibody with FDA licensed reagents. However, there is no test method that can offer complete assurance that HIV, Hepatitis B virus or other infectious agents are absent. These reagents should be handled at the Biosafety Level 2, as recommended in the Centers for Disease Control/National Institutes of Health manual, "Biosafety in Microbiological and Biomedical Laboratories" 1984.
- 4. 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.
- 5. Do not pipette by mouth. Do not smoke, eat, or drink in the areas in which specimens or kit reagents are handled.
- The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.
- 7. Control sera and sample diluent contain preserved with sodium azide. Sodium azide may react with lead and copper plumbing to form explosive metal azide. On disposal, flush with a large volume of water.

#### SPECIMEN COLLECTION AND HANDLING

- Collect blood specimens and separate the serum.
- 2. Typically, 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.

# SPECIMEN COLLECTION AND HANDLING

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#### REAGENT PREPARATION

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 (20-25°C).

#### **ASSAY PROCEDURE**

Bring all specimens and kit reagents to room temperature (20-25°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 1:21 dilution of test samples, by adding 10 µl of the sample to 200 µl of sample diluent. Mix well.
- 3. Dispense 100  $\mu$ l of diluted sera, calibrator and controls into the appropriate wells. For the reagent blank, dispense 100 $\mu$ l 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  $\mu$ l of 1X wash buffer. Blot on absorbance paper or paper towel.
- 5. Dispense 100  $\mu$ l 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  $\mu$ l of 1X wash buffer. Blot on absorbance paper or paper towel.
- 7. Dispense 100  $\mu$ l 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 min. A dual wavelength is recommended with reference filter of 600-650 nm.

#### **CALCULATION OF RESULTS**

- 1. Check Calibrator Factor (CF) value on the calibrator bottle. This value might vary from lot to lot. Make sure you check the value 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.

# LIMITATIONS OF THE TEST

1. Lipemic or hemolyzed samples may cause erroneous results.