ErbaLisa® EBV-VCA IgG

Catalog No. IME00093 (96 Tests)

INTENDED USE

The ERBAlisa 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
Microwells coated with EBV-VCA antigen	12x8x1
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
20X Wash Concentrate: 1 Bottle	25 ml

MATERIALS NOT PROVIDED

1. Distilled or deionized water
2. precision pipettes
3. Disposable pipette tips
4. Micortiter well reader capable of reading absorbance at 450nm
5. Absorbance paper or paper towel
6. Graph paper

STORAGE AND STABILITY

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

WARNINGS AND PRECAUTIONS

1. 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, as 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
2. This test kit is designed for IVD Use.
3. Do not pipette by mouth. Do not smoke, eat, or drink in the areas in which specimens or kit reagents are handled.
4. The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.
5. It is recommended that standards, control and serum samples be run in duplicate.
6. 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.

SPECIMEN COLLECTION HANDLING

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

REAGENTS PREPARATION

1. All reagents should be brought to room temperature (20-25°C) before use.

2. 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

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 µl of diluted sera, calibrator and controls into the appropriate wells. For the reagent blank, dispense 100µl 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 absorbance 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 absorbance 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 to stop reaction.
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.

Example of Typical Results

Calibrator mean OD = 0.8 Calibrator Factor (CF) = 0.5
Cut-off Value = 0.8 x 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

QUALITY CONTROL

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

1. The O.D. of the Calibrator should be greater than 0.250.
2. The Ab index for Negative control should be less than 0.9.
3. The Ab Index for Positive control should fall within the range specified on the COA/label.

INTERPRETATION

The following is intended as a guide to interpretation of test results; each laboratory is encouraged to establish its own criteria for test interpretation based on sample populations encountered.

Antibody Index Interpretation

<0.9 No detectable Antibody to EBV-VCA IgG by ELISA.
0.9-1.1 Borderline positive. Follow-up testing is recommended if clinically indicated.
>1.1 Detectable antibody to EBV-VCA IgG by ELISA.

LIMITATIONS OF THE TEST

1. 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.
2. Lipemic or hemolyzed samples may cause erroneous results.

PERFORMANCE CHARACTERISTICS

1. **Sensitivity and Specificity:** 398 patient sera were tested by this EBV-VCA IgG ELISA and a reference ELISA method. 346 sera were positive and 36 were negative by both methods (96% agreement). The results are summarized below:

		EBV-VCA IgG ELISA		
		+	–	Total
Reference ELISA Kit	+	346	7	353
	–	9	36	45
	Total	355	43	398

2. **Precision:**

Intra-assay study:

Serum	No. of replicates	Mean	Standard Deviation	CV %
1	16	2.29	0.080	3.49
2	16	1.78	0.042	2.36
3	16	0.22	0.007	3.20

Inter-assay study

Serum	No. of replicates	Mean	Standard Deviation	CV %
1	10	1.52	0.104	6.80
2	10	1.16	0.053	4.57
3	10	0.24	0.021	8.75

REFERENCES

1. Gray JJ. Avidity of EBV VCA-specific IgG antibodies: distinction between recent primary infection, past infection and reactivation. J Virol Methods 1995;52(1-2):95-104.
2. Liu MT; Yeh CY. Prognostic value of anti-Epstein-Barr virus antibodies in nasopharyngeal carcinoma (NPC). Radiat Med 1998;16(2):113-7.
3. Hadar T; Margalith M; Sagiv E; Sarov B; Sarov I. The significance of serum IgM IgA and IgG antibodies specific for Epstein-Barr virus as determined by immunoperoxidase assay in the rapid diagnosis of infectious mononucleosis. Isr J Med Sci 1995;31(5):280-3.
4. Levine PH; Stemmermann G; Lennette ET; Hildesheim A; Shibata D; Nomura A. Elevated antibody titers to Epstein-Barr virus prior to the diagnosis of Epstein-Barr-virus-associated gastric adenocarcinoma. Int J Cancer 1995;60(5):642-4.
5. Debyser Z; Reynders M; Goubau P; Desmyter J. Comparative evaluation of three ELISA techniques and an indirect immunofluorescence assay for the serological diagnosis of Epstein-Barr virus infection. Clin Diagn Virol 1997;8(1):71-81.

2017-9-6

Calbiotech Inc.,1935 Cordell Ct., El Cajon, CA 92020 Tel (619) 660-6162, Fax (619) 660-6970,www.calbiotech.com

CE IVD 2° 8° EC REP

CEpartner4U, Esdoornlaan 13, 3951 DB Maarn, The Netherlands **Tel** +31 (0)6.516.536.26