CE

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IVD	See external label	2°C-8°C	Σ=96 tests	REF Cat # 1401Z

Herpes Simplex 1 IgG (HSV 1 IgG)

Cat # 1401Z

Test	HSV 1 IgG ELISA
Method	ELISA: Enzyme Linked Immunosorbent Assay
Principle	Indirect: Antibody Capture
Detection Range	Qualitative Positive; Negative control & Cut off
Sample	5ul Serum
Specificity	100%
Sensitivity	100%
Total Time	~ 90 min
Shelf Life	12 -18 Months

** Laboratory results can never be the only base of a medical report. The patient history and further tests have to be taken into account*

NAME AND INTENDED USE

The DIAGNOSTIC AUTOMATION ELISA, HSV 1 IgG is intended for use in evaluating a patient's serologic status to herpes simples virus (HSV) infection, or for evaluating paired sera for the presence of a significant increase in herpes specific IgG.

SUMMARY AND EXPLANATION OF THE TEST

Herpes Simplex Virus is a common pathogen and its primary infection is usually asymptomatic. There are two immunologically distinct types of HSV: Type 1 and Type 2. HSV 1 is generally associated with oral infection and lesions above the waist, and HSV 2 is associated with genital infections and lesions below the waist. Clinical cases primarily are 1) eczema herpeticum with eczematous skin changes with numerous lesions, 2) Gingivo-stomatitis and 3) Herpes sepsis, almost only found in newly born of premature infants. DIAGNOSTIC AUTOMATION ELISA HSV IgG is an accurate serologic method to detect HSV specific antibody in serum sample.

PRINCIPLE OF THE TEST

Purified HSV antigen is coated on the surface of microwells. Diluted patient serum is added to wells, and the HSV 1 IgG specific antibody, if present, binds to the antigen. All unbound materials are washed away. After adding enzyme conjugate, it binds to the antibody-antigen complex. Excess enzyme conjugate is washed off and TMB Chromogenic substrate is added. The enzyme conjugate catalytic reaction is stopped at a specific time. The intensity of the color generated is proportional to the amount of IgG specific antibody in the sample. The results are read by a microwell reader compared in a parallel manner with calibrator and controls.

MATERIALS PROVIDED

1. Microwell Strips: Purified HSV1 antigen coated wells	(12 X 8 wells)
2. Sample Diluent: Blue Color Solution	1 vial (22 ml)
3. Calibrator: Factor value (f) stated on label. Red Cap	1 vial (150µl)
4. Negative Control: Range Stated on label. Natural Cap	1 vial (150µl)
5. Positive Control: Range stated on label. Green Cap	1 vial (150µl)
6. Washing Concentrate 10X: White Cap	1 bottle (100 ml)
7. Enzyme Conjugate: Red Color solution	1 vial (12 ml)
8. TMB Chromogenic Substrate: Amber Bottle	1 vial (12ml)
9. Stop Solution	1 vial (12 ml)

STORAGE AND STABILITY

1. Store the kit at 2 - 8 ^{O}C.
2. Keep microwells sealed in pouch with desiccants. We recommend you use up all wells within weeks after initial opening of the pouch.
3. The reagents are stable until expiration of the kit.
4. Do not expose test reagents to heat, sun or strong light during storage or usage.

WARNINGS AND PRECAUTIONS

1. Potential biohazardous materials:
   The calibrator and controls contain human source components which have been tested and found nonreactive 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 in vitro diagnostic 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 a integral unit. The components of different lots should not be mixed.
5. This product contains components 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

1. Collect blood specimens and separate the serum.
2. Specimens may be refrigerated at 2 - 8 ^{o}C for up to seven days or frozen for up to six months. Avoid repetitive freezing and thawing of serum sample.

PREPARATION FOR ASSAY

1. Prepare 1x washing buffer.
   Prepare washing buffer by adding distilled or deionized water to 10x wash concentrate to a final volume of 1 liter.
2. Bring all specimens and kit reagents to room temperature (20-25 ^{o}C) and gently mix.

ASSAY PROCEDURE

1. Place the desired number of coated strips into the holder.
2. Prepare 1:40 dilutions by adding 5µl of the samples, negative calibrator, positive calibrator, and Cut-off calibrator to 200µl of sample diluent. Mix well.
3. Dispense 100 µl of diluted sera, and calibrators into the appropriate wells. For the reagent blank, dispense 100 µl sample diluent in 1A well position. Tap the holder to remove air bubbles from the liquid and mix well. Incubate for 30 minutes at room temperature.
4. Remove liquid from all wells. Repeat washing three times with washing buffer.
5. Dispense 100 µl of enzyme conjugate to each well and incubate for 30 minutes at room temperature.
6. Remove enzyme conjugate from all wells. Repeat washing three times with washing buffer.
7. Dispense 100 µl of TMB Chromogenic Substrate to each well and incubate for 30 minutes at room temperature.
8. Add 100 µl of 2 N HCl to stop reaction.
   Make sure there are no air bubbles in each well before reading
9. Read O.D. at 450 nm with a microwell reader.

CALCULATION OF RESULTS

1. To obtain the Cut off OD value: Multiply the OD of Calibrator by Factor (f) printed on label of calibrator.
2. Calculate the IgG Index of each determination by dividing the OD values of each sample by obtained OD value of cut off.

For example:

If Factor (f) Value on label = 0.4
This factor (f) is a variable value. It could be 0.35 or 0.5 etc. printed on the label of calibrator.

Obtained Calibrator O.D = 1.100
Cut off O.D. = 1.100 X 0.4 = 0.44 (by definition IgG Index = 1)

Patient sample O.D. = 0.580

IgG Index = 0.580 / 0.44 = 1.32 (Positive Result)

Patient Sample O.D. = 0.320
IgG Index = 0.320 / 0.44 = 0.73 (Negative Result)

QUALITY CONTROL

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

1. The O.D. value of the reagent blank against air from a microwell reader should be less than 0.250.
2. If the O.D. value of the Calibrator is lower than 0.250, the test is not valid and must be repeated.

INTERPRETATION

Negative: HSV 1 IgG Index of 0.90 or less are seronegative for IgG antibody to HSV 1.
Equivocal: HSV 1 IgG between 91~99 is equivocal. Sample should be retested.
Positive: HSV 1 IgG 1.0 or greater are seropositive.

PERFORMANCE CHARACTERISTICS

Specificity and Sensitivity:
A total of 66 patient samples were used to evaluate specificity and sensitivity of the test. The DIAGNOSTIC AUTOMATION ELISA HSV 1 IgG test results were compared to a commercial ELISA kit results.

		Reference ELISA			
		N	E	P	Total
DIAGNOSTIC	N	20 (D)	0	0 (B)	20
AUTOMATION	E	0	0	1	1
ELISA	P	0 (C)	0	45 (A)	45
	Total	20	0	46	66

Sensitivity = **A / (A+B) = 45 / (45+0) =100%**

Specificity =**D / (C+D) = 20 / (0+20) =100%**

Accuracy = **(A+D) / (A+B+C+D)**

= 45+20 / (45+0+0+20) =65 / 65 = 100%

Precision:
The precision of the assay was evaluated by testing three different sera of eight replicates over a period of one week.

The intra-assay and inter-assay C.V. are summarized below:

	Negative	Low positive	Positive
Intra-assay	8..9%	7..5%	6..8%
Inter-assay	10..3%	8..3%	7..5%

LIMITATION OF THE PROCEDURE

1. As with other serological assays, the results of these assays should be used in conjunction with information available from clinical evaluation and other diagnostic procedures.
2. Samples obtained too early during primary infection may not contain detectable antibody.
3. A single serum sample should not be used to aid in the diagnosis of recent infection. Paired samples should be collected and tested simultaneously to look for seroconversion.

REFERENCES

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3. Coleman, R.M., L. Pereira, P.D. Bailey, D. Dondero, C. Wickliffe, and A.J. Nahmias. Determination of herpes simplex virus type-specific antibodies by enzyme-linked immunosorbent assay. J. Clin. Microbiol. 18(1983) 287.

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