

Measles (Rubeola) IgG ELISA Kit

(Catalog # E4662-100, 96 assays; Store at 2-8°C)

10/18

I. Introduction:

Measles is an acute, highly contagious viral disease. Although measles is usually considered a childhood disease, it can be contracted at any age. Measles is spread by direct contact with nasal or throat secretions of infected people or, less frequently, by airborne transmission. Measles symptoms generally appear in two stages. In the first stage, the individual may have a runny nose, cough and a slight fever. The second stage begins on the third to seventh day and consists of high fever and red blotchy rash lasting four to seven days. The rash usually begins on the face and then spreads over the entire body. Symptoms usually appear in 10-12 days, although they may occur between 8-13 days after exposure. The presence of IgG antibody to measles virus is indicative of previous exposure or vaccination. In individuals with acute measles, a significant increase in measles IgG antibody level is indicative of recent infection. IgM antibodies to measles virus are often detectable with onset of the rash and typically persist for 4 weeks. At least 80% of patients will be positive for measles IgM at 6 days and 100% at 16 days after onset of symptoms.

II. Application:

Detection of IgG antibody to Measles

III. Sample Type:

Human serum or plasma

IV. Kit Contents:

Components	E4662-100	Part No.
Microplate	12 strips x 8 wells	E4662-100-1
Sample Diluent	22 ml	E4662-100-2
Calibrator	1 ml	E4662-100-3
Positive Control	1 ml	E4662-100-4
Negative Control	1 ml	E4662-100-5
Enzyme conjugate	12 ml	E4662-100-6
TMB Substrate	12 ml	E4662-100-7
Stop Solution	12 ml	E4662-100-8
Wash Buffer (20X)	25 ml	E4662-100-9

V. User Supplied Reagents and Equipment:

- Microplate reader capable of measuring absorbance at 450 nm.
- Absorbent paper.
- Adjustable pipettes and pipette tips.

VI. Storage Conditions and Reagent Preparation:

Store kit at 2-8°C. Keep microwells sealed in a dry bag with desiccants. Spin tubes briefly to bring down all components to the bottom of tubes. Reagents are stable until the expiration of the kit. Do not expose reagent to heat, sun, or strong light.

- **Wash Buffer:** 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).

VII. Warning & Precautions:

- 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."
- 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.
- 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.
- 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.

VIII. Sample Preparation and Storage:

Collect blood specimens & separate the serum immediately. Specimens may be stored refrigerated at (2-8°C) for 7 days. Store frozen at (-20°C) for up to six month. Avoid multiple freeze-thaw cycles. Prior to assay, frozen sera should be completely thawed and mixed well.

IX. Assay Protocol:

Prior to assay, bring all reagents to room temperature. Gently mix all reagents before use.

1. Place the desired no. of coated strips into the holder. Replace any unused microwell strips back into the aluminum bag, seal and store at 2-8°C.
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 100 µl 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 μ l of 1X wash buffer. Blot on absorbance paper or paper towel.
7. Dispense 100 μ l of TMB substrate and incubate for 10 minutes at room temperature.
8. Add 100 μ l 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.

X. Calculation

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. Calculate the cut-off value: Calibrator OD x Calibrator Factor (CF). 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 = $1.6 / 0.4 = 4.0$

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 IgG antibody 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 IgG antibody by ELISA.
- 0.9 - 1.1 Borderline positive. Follow-up testing is recommend if clinically indicated.
- > 1.1 Detectable antibody to IgG antibody 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 patients history, physical findings and other diagnostic procedures.
2. Lipemic or hemolyzed samples may cause erroneous results.

Sensitivity and Specificity

327 human sera were tested by this Measles IgG ELISA and a reference ELISA method. 284 sera were positive and 33 were negative by both methods (97% agreement). The results are summarized below:

		Measles IgG ELISA		
		+	-	Total
Reference ELISA kit	+	284	4	288
	-	6	33	39
Total		290	37	327

XI. RELATED PRODUCTS:

- Measles (Rubeola) IgM ELISA Kit (E4663)
- Influenza Neuraminidase Inhibitor Susceptibility Assay Kit (K524)
- Cardiolipin IgG, IgA, IgM ELISA Kit (E4658)
- Cardiolipin IgA ELISA Kit (E4659)
- Cardiolipin IgG ELISA Kit (E4660)

FOR RESEARCH USE ONLY! Not to be used on humans.