



DIAGNOSTIC AUTOMATION, INC.

23961 Craftsman Road, Suite D/E/F,
Calabasas, CA 91302

Tel: (818) 591-3030 Fax: (818) 591-8383

onestep@rapidtest.com

technicalsupport@rapidtest.com

www.rapidtest.com

IVD



See external label



2°C-8°C



Σ=96 tests

REF

Cat # 9070-11

Rubella IgM

CHEMILUMINESCENCE

Cat # 9070-11

SUMMARY OF ASSAY PROCEDURE

Step	(20-25°C Room temp.)	Volume	Incubation time
1	Sample dilution 1:40 =5 µl / 200 µl		
2	Diluted samples, controls & calibrator	100 µl	30 minutes
3	Washing buffer (3 times)	350 µl	
4	Enzyme conjugate	100 µl	30 minutes
5	Washing buffer (3 times)	350 µl	
6	Substrate A and Substrate B mixture	100µl	5 minutes
7	Read with Luminometer in 5~30 minutes		

NAME AND INTENDED USE

Rubella IgM Chemiluminescence ELISA is intended for use in the detection of IgM antibody to the rubella virus.

SUMMARY AND EXPLANATION OF THE TEST

Rubella is a herpes virus. Generally rubella is considered a mild adolescence disease. However a maternal infection could be transmitted through the placenta to the fetus, causing congenital rubella. Congenital rubella may result in chronic cardiac disease, growth retardation, hepatosplenomegaly, malformations and other severe anomalies. Children born asymptomatic may develop these abnormalities later in life.

To reduce risk of such severe complications, accurate serological methods must be performed to determine the serologic status of childbearing aged women.

PRINCIPLE OF THE TEST

Purified rubella antigen is coated on the surface of microwells. Diluted patient serum is added to wells, and the rubella IgM 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 substrate A & substrate B mixture is added. The light generated (RLU) is proportional to the amount of IgM specific antibody in the sample. The results are read by a microwell luminometer compared in a parallel manner with calibrator and controls.

MATERIALS PROVIDED

1. Microwell Strips: purified *Rubella* antigen coated wells. (12 x 8 wells)
2. Absorbent Solution: Black Cap. 1 vial (22 ml)
3. Washing Concentrate 10x: White Cap. 1 bottle (100 ml)
4. Enzyme Conjugate: Red Color Solution. 1 vial (12 ml)
5. Cut-off Calibrator: Yellow Cap. *Rubella* M Index = 1.0 (150 µl/vial)
6. Negative Control: Range stated on label. Natural Cap. (150 µl/vial)
7. Positive Control: Range stated on label. Red Cap. (150 µl/vial)
8. Substrate A: H₂O₂ in buffer. Natural bottle. 1 vial (7 ml)
9. Substrate B: Luminol in buffer. Amber bottle. 1 vial (7 ml)

STORAGE AND STABILITY

1. Store the kit at 2 - 8 °C.

2. Always keep microwells tightly sealed in pouch with desiccants. We recommend you use up all wells within 4 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. Do not pipette by mouth. Do not smoke, eat, or drink in the areas in which specimens or kit reagents are handled.
3. The components in this kit are intended for use as a integral unit. The components of different lots should not be mixed.
4. 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 °C for up to seven days or frozen for up to six months. Avoid repetitive freezing and thawing of serum sample.
3. If rubella is suspected clinically, a blood specimen should be taken within three days after onset of a rash and a second specimen taken at least two weeks later. Test both serums for antibody simultaneously.

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 °C) and gently mix.

ASSAY PROCEDURE

1. Prepare 1:40 dilutions by adding 5 µl of the samples, negative control, positive control, and calibrators to 200 µl of absorbent solution. Mix well.
2. Place the desired number of coated strips into the holder.
3. Dispense 100 µl of diluted sera, calibrators, and controls into the appropriate wells. 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 and 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. Mix equal volume of Substrate A & Substrate B, then dispense 100 µl of this mixture to each well.
8. Read RLU with a microwell luminometer within 5~30 minutes.

CALCULATION OF RESULTS

Determination of Index values

1. Calculate the mean of duplicate RLU values (B).
2. Calculate the Rubella M Index of each determination by dividing the mean values of each sample (B) by Cut-off calibrator mean value (C).

For example:

Sample	Well No	RLU (A)	Mean RLU (B)	INDEX B/C
Cut-off Calibrator	A1	546741	541280 (C)	1
	B1	535819		
Positive Control	C1	1554000	1595850	2.9
	D1	1637700		
Negative Control	E1	41839	43827	0.08
	F1	45815		
Patient Sample 1	G1	194694	188895	0.35
	H1	183096		
Patient Sample 2	A2	1105962	1115238	2.1
	B2	1124513		

QUALITY CONTROL

1. In order for the assay results to be considered valid the controls should be within the ranges indicated on the labels.
1. The RLU values vary with the different luminometer used.
2. Each laboratory should assay controls at levels in low, normal and elevated ranges for monitoring assay performance. Quality control trends should be maintained to monitor batch to batch consistency.

INTERPRETATION

Rubella IgM Index	Interpretation
< 0.90	Negative for IgM to Rubella
0.91 ~ 0.99	Equivocal, sample should be retested
1 ~ 2	Low positive
2 ~ 2.5	Moderate positive
> 2.5	High positive

LIMITATIONS OF THE PROCEDURE

1. To prevent false negative and false positive IgM test results caused by the presence of specific IgG and rheumatoid factor (RF) in some specimens, reagents provided in this kit has been formulated to resolve these interferences. However, specimens with extremely high RF and high autoimmune antibodies, the possibility of these interferences cannot be ruled out entirely.
2. 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.

PERFORMANCE CHARACTERISTICS

Specificity and Sensitivity:

A total of 42 patient samples were used to evaluate specificity and sensitivity of the test. Rubella IgM test results were compared to a commercial ELISA kit results:

		Reference ELISA			
		N	E	P	Total
Rubella IgM Chemiluminescence ELISA	N	26 (D)	0	0 (B)	26
	E	0	0	0	0
	P	0 (C)	0	16 (A)	16
	Total	26	0	16	42

$$\text{Sensitivity} = A / (A+B) = 16 / 16 = 100\%$$

$$\text{Specificity} = D / (C+D) = 28 / 28 = 100\%$$

$$\text{Accuracy} = (A+D) / (A+B+C+D) = 42 / 42 = 100\%$$

Expected Values:

49 random samples were determined with Rubella IgM Chemiluminescence ELISA. The test results were computed as IgM Index using a chosen reference serum (cut off) as IgM index 1. None were found to be positive (0%), and 49 were found to be negative (100%). Others reported 0% positivity from a study of 182 sera. A third set of 48 random samples, the positivity was found 8.30%.

Precision:

The precision of the assay was evaluated by testing three different sera eight replicates on 3 days. The intra-assay and inter-assay C.V. are summarized below:

	Negative	Low positive	Positive
Intra-assay	10.7%	9.4%	8.6%
Inter-assay	12.8%	10.4%	8.9%

REFERENCES

1. Gravell, M., P.H. Dorcett, O. Gutenson, and A.C. Ley. Detection of antibody to rubella virus by enzyme-linked immunosorbent assay. J. Infect. Dis. 136:S300, 1977.
2. Hermann, K.L. Rubella virus. Manual of Clinical Microbiology, 3rd Edition. Lennette, Balows, Hausler, Truant (ed). Chapt. 86:862, 1980.
3. Katz, S.L. Rubella (German measles). Zinssmer Microbiology, 18th Edition. Jolik, Willett, Amos (ed). Chapt. 75:1067, 1985.

Date Adopted	Reference No.
2007-10-19	DA-Rubella IgM-2009



DIAGNOSTIC AUTOMATION, INC.

23961 Craftsman Road, Suite D/E/F, Calabasas, CA 91302

Tel: (818) 591-3030 Fax: (818) 591-8383

ISO 13485-2003



Revision Date: 02-18-2009