

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY**

A. 510(k) Number:

K092587

B. Purpose for Submission:

New device

C. Measurand:

IgM antibodies specific to Rubella and Cytomegalovirus (CMV).

D. Type of Test:

Multiplex flow immunoassay (multiplexed fluoromagnetic bead assay)

E. Applicant:

Bio-Rad Laboratories Inc.

F. Proprietary and Established Names:

BioPlex™ 2200 Rubella and CMV IgM Kit
BioPlex™ 2200 Rubella and CMV IgM Calibrator Set
BioPlex™ 2200 Rubella and CMV IgM Control Set

G. Regulatory Information:

Product code	Classification	Regulation section	Panel
LFX: Enzyme Linked Immunoabsorbent Assay, Rubella	Class II	866.3510; Rubella Virus Serological Reagents	Microbiology
JIX: Calibrator, multi- analyte mixture	Class II	862.1150 - Calibrator	Clinical Chemistry
JJX: Single (specified) analyte controls (assayed and unassayed)	Class I	862.1660 - Quality Control Material (assayed and unassayed)	Clinical Chemistry

Note: The BioPlex™ 2200 Rubella and CMV IgM Kit is a multiplex immunoassay for the detection of IgM antibodies to Rubella and CMV. This device is classified as Class II as described above and no new product code is assigned for this device. The first product code assigned for the device is listed under the regulation section for Rubella reagents.

The following is the regulation section and product code that is applicable to the other analyte detected by the device subject of this submission.

866.3175 - Cytomegalovirus serological reagents. Class II (Microbiology). Antibody IgM, IF, cytomegalovirus virus (LKQ) and Enzyme Linked Immunoabsorbent Assay, Cytomegalovirus (LFZ)

H. Intended Use:

1. Intended use(s):

The BioPlex™ 2200 Rubella and CMV IgM kit

The BioPlex™ 2200 Rubella and CMV IgM kit is a multiplex flow immunoassay intended for the qualitative detection of IgM antibodies to Rubella and Cytomegalovirus (CMV) in human serum and potassium EDTA or sodium heparin plasma.

The BioPlex 2200 Rubella and CMV IgM kit is intended for use with the Bio-Rad BioPlex 2200 System.

This kit is intended as an aid in the diagnosis of a current or recent Rubella and/or CMV infection, in individuals suspected of having one of the respective disease states including women of child bearing age.

This assay is not FDA cleared or approved for use in testing (screening) blood or plasma donors.

Performance characteristics for the Rubella and CMV IgM assays have not been evaluated in immunosuppressed or organ transplant individuals. Performance characteristics of this kit have not been established for use in neonatal screening or for use at point of care facilities.

The BioPlex 2200 Rubella and CMV IgM Calibrator Set

The BioPlex® 2200 Rubella and CMV IgM Calibrator Set is intended for the calibration of the BioPlex 2200® Rubella and CMV IgM Reagent Pack.

The BioPlex 2200 Rubella and CMV IgG Control Set

The BioPlex® 2200 Rubella and CMV IgM Control Set is intended for use as an assayed quality control to monitor the overall performance of the BioPlex 2200 instrument and BioPlex 2200® Rubella and CMV IgM Reagent Pack in the clinical laboratory. The performance of the BioPlex 2200® Rubella and CMV IgM Control Set has not been established with any other Rubella and CMV IgM assays.

2. Indication(s) for use:

Same as Intended Use

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

The Bio-Rad BioPlex 2200 System

I. Device Description:

The BioPlex 2200 Rubella and CMV IgM kit is a fully automated multiplexed micro particle bead based immunoassay for the qualitative detection of IgM antibodies to Rubella, and CMV in human serum and EDTA or heparinized plasma using the Luminex flow cytometry technology. It consists of a reagent kit and the Bioplex 2200 Instrument.

The BioPlex 2200 Rubella and CMV IgM Reagent Pack consists of:

1. A bead set containing dyed beads coated with lysates of Rubella, and CMV, an Internal Standard bead (ISB), a Serum Verification bead (SVB), and a Reagent Blank bead (RBB) in buffer with Glycerol and protein stabilizers (bovine), ProClin™ 300 (0.3%) and sodium azide (< 0.1%) as preservatives.
2. Phycoerythrin-conjugated donkey polyclonal anti-human IgM antibody and phycoerythrin –conjugated murine monoclonal anti-human FXIII antibody, in buffer with stabilizers (bovine and equine), ProClin® 300 (0.3%), sodium benzoate (0.1%) and sodium azide (<0.1%) as preservatives.
3. Sample diluent; containing goat anti-human IgG antibody and protein stabilizers (bovine and equine) in buffer, ProClin® 300 (0.3%) and sodium azide (<0.1%) as preservatives.

The device requires other materials which are provided independently from Bio-Rad and these include the BioPlex 2200 Instrument and Software System, the Bioplex 2200 sheath fluid and wash solution.

The BioPlex 2200 Rubella and CMV IgM Calibrator Set consists of three distinct serum based calibrators in three vials. They are made from defibrinated plasma with added known concentrations of Rubella and CMV antibodies from human disease state plasma. The calibrators are used for the qualitative calibration of the device. All calibrators contain ProClin® 300 (0.3%), sodium benzoate (0.1%) and sodium azide (<0.1%) as preservatives.

The BioPlex 2200 Rubella CMV IgM Control Set includes a negative control and a one positive control for Rubella and another for CMV, intended to be used as assayed quality control material with the BioPlex Rubella and CMV IgM Test. All antibodies

are derived from human disease state plasma. All controls contain Amikacin (0.003%), Cycloheximide (C15H23NO4) (0.009%), Amphotericin B (0.002%), Cefotaxime Sodium (0.002%), Ciprofloxacin (0.005%), ProClin[®] 300 (<0.3%), Sodium benzoate (<0.1%) and sodium azide (<0.1%).

J. Substantial Equivalence Information:

1. Predicate device name(s):

There is no single predicate device for the panel as a whole; the following is a list of the predicate devices for each of the analytes in the panel. These predicate devices are used as comparators to assess the performance of the device and determine the substantial equivalence. The final classification of the new multiplexed device follows that of the Rubella predicate.

ADVIA Centaur[®] Rubella IgM
 bioMérieux VIDAS[®] CMV IgM

2. Predicate 510(k) number(s):

The 510(k) numbers for the Rubella and CMV predicate devices are K010668 and K933549 respectively.

3. Comparison with predicates:

1. Comparison with- ADVIA Centaur[®] Rubella IgM (K010668)

Similarities		
Item	Device	Predicates
Intended Use	Intended for the qualitative detection of IgM antibodies to Rubella , and Cytomegalovirus (CMV)	Intended for the qualitative detection of IgM antibodies to the rubella virus in serum or plasma (EDTA, heparin)
Indications	As an aid in the diagnosis of a current or recent Rubella and/or CMV infection, in individuals suspected of having one of the respective disease states including women of child bearing age.	As an aid in the presumptive diagnosis of current or recent infection with rubella .
Measurand	Rubella and CMV IgM	Rubella IgM
Detection	Qualitative detection	same
Matrices	Serum, EDTA or Heparinized Plasma	same
Controls	Negative Control and Positive Controls specific for rubella and CMV IgM	Negative Control and Positive Control specific for rubella IgM

Differences		
Item	Device	Predicate
Calibrators	Multiple Calibrators	Single calibrator
Analytes detected	Multiple Analytes	Single Analyte
Technology	Multiplexed flow immunoassay	Sandwich immunoassay using direct, chemiluminometric technology
Signal Detection	Fluorescence	Chemiluminescence
Solid phase	Antigen-coated paramagnetic microbead reagent. Microbeads are infused with red and infrared fluorescent dyes for bead classification.	anti-human IgM FC monoclonal antibody is covalently coupled to paramagnetic particles

2. Comparison with- bioMérieux VIDAS[®] CMV IgM - K933549

Similarities		
Item	Device	Predicates
Intended Use	Intended for the qualitative detection of IgM antibodies to Rubella, and Cytomegalovirus (CMV)	Intended for the qualitative detection of anti-CMV IgM antibodies in human serum.
Indications	As an aid in the diagnosis of a current or recent Rubella and/or CMV infection, in individuals suspected of having one of the respective disease states including women of child bearing age.	As an aid in the diagnosis of cytomegalovirus infection.
Measurand	Rubella and CMV IgM	CMV IgM
Detection	Qualitative detection	same
Controls	Negative Control and Positive Controls specific for rubella and CMV IgM	Negative Control and Positive Controls specific for CMV IgM
Signal Detection	Fluorescence	same

Differences		
Item	Device	Predicate
Matrices	Serum, EDTA or Heparinized Plasma	Serum
Analytes detected	Multiple Analytes	Single Analyte
Calibrators	Multiple calibrators	Single Calibrator
Technology	Multiplexed flow immunoassay, using Antigen-coated paramagnetic microbead reagent.	Two-step enzyme immunoassay sandwich method with fluorescent detection (ELFA)
Solid Phase	Antigen-coated paramagnetic	Antigen-coated solid phase

Differences		
Item	Device	Predicate
	microbead reagent. Microbeads are infused with red and infrared fluorescent dyes for bead classification.	receptacles

K. Standard/Guidance Document referenced (if applicable):

1. CLSI *In- vitro* guideline; User Demonstration of Performance for Precision and Accuracy; Approved Guideline (EP15-A).
2. CLSI *In- vitro* guideline; Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline-Second Edition (EP05-A2).
3. CLSI *In- vitro* guideline; Interference Testing in Clinical Chemistry; Approved Guideline - Second Edition (EP07-A2).

The following FDA guidance documents were also referenced in this submission:

1. Guidance on Informed Consent for In Vitro Diagnostic Device Studies Using Leftover Human Specimens that are Not Individually Identifiable - Guidance for Sponsors, Institutional Review Boards, Clinical Investigators and FDA Staff
<http://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm078384.htm>
2. Guidance for Industry and FDA Staff - Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests
<http://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm071148.htm>

L. Test Principle:

The BioPlex™ 2200 Rubella and CMV IgM kit uses multiplex flow immunoassay, a methodology that greatly resembles traditional EIA, but permits simultaneous detection and identification of many antibodies in a single tube.

The kit employs a panel of two different populations of dyed beads coated with cell lysates bearing Rubella or CMV antigens to identify the presence of IgM antibodies to Rubella, and CMV antigens as follows.

The BioPlex 2200 System combines an aliquot of patient sample, sample diluent, and bead reagent into a reaction vessel; the mixture is incubated at 37°C. After a wash cycle, anti-human IgM antibody, conjugated to phycoerythrin (PE), is added to the dyed beads, and this mixture is incubated at 37°C. The excess conjugate is removed in another wash cycle, and the beads are re-suspended in wash buffer. The bead mixture then passes through the detector. The identity of the dyed beads is determined by the fluorescence of the dyes, and the amount of antibody captured by the antigen is determined by the

fluorescence of the attached PE. Raw data are reported as relative fluorescence intensity (RFI). Additionally, the ISB beads, SVB beads and a RBB beads are present in each reaction mixture to verify detector response, the addition of serum or plasma to the reaction vessel and the absence of significant non-specific binding in serum or plasma.

The BioPlex 2200 Rubella and CMV IgM Calibrator Set calibrates the instrument using a set of three distinct serum based calibrators. Calibrators are used in a test system to establish points of reference that are used in the determination of qualitative numeric measurement of IgM antibodies to Rubella, and CMV in human serum and EDTA or heparinized plasma. The Calibrator Set should be loaded and assayed at minimum in duplicate every 30 days or with each new Re-agent Pack lot.

The BioPlex 2200 Rubella and CMV IgM Control Set includes a negative control as well as a CMV IgM positive control and a Rubella IgM positive control. The BioPlex Rubella and CMV IgM Positive Controls are manufactured to give positive results, with values above the cut-off for each specific analyte. The BioPlex Rubella and CMV IgM Negative Control are manufactured to give negative results, with values below the cut-off for each specific analyte. The recommended frequency for performing quality control is once every 24-hour testing period. Performing quality control is also necessary after each new assay calibration and certain service procedures. A BioPlex 2200 Rubella and CMV IgM Control Lot Data disk is available to load the necessary value assignment data into the instrument. The result for each of the tested antibodies is expressed as an antibody index (AI). For Rubella and CMV, results of ≤ 0.8 AI are negative, 0.9 and 1.0 AI are equivocal and results of ≥ 1.1 AI are reported as positive.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Separate internal and external reproducibility studies were conducted to evaluate the reproducibility of the BioPlex 2200 Rubella and CMV IgM kit on the BioPlex 2200 instrument.

The internal precision study was conducted in-house by Bio-Rad Laboratories in accordance with CLSI EP5-A2, Evaluation of Precision Performance of Quantitative Measurement Methods.

For the internal reproducibility study, three (3) panels made from serum and plasma (EDTA and heparinized) were assayed two (2) times in two (2) separate daily runs over 20 days (n=80).

The data were analyzed for within-run, between-run, between-day, and total precision and the standard deviation (SD) and percent coefficient of variation (% CV) were calculated.

The within-run precision for rubella IgM samples ranged from 4.3% to 5.5% in a serum matrix, 6.3% to 6.9% for EDTA and 4.8% to 9.5% for heparin plasma matrices. The within-run precision for CMV IgM samples ranged from 5.3% to 9.2% in a serum matrix, 4.9% to 11.8% for EDTA and 5.2% to 12.8% for heparin plasma matrices.

The external reproducibility study was conducted in accordance with CLSI EP15-A2 during the external clinical evaluation at each of three clinical study sites. Three lots of reagent packs, three lots of the BioPlex 2200 Rubella and CMV IgM Calibrator Set and three lots of BioPlex 2200 Rubella and CMV IgM Control Set were evaluated. The sample panels made from serum and plasma (EDTA and heparinized) at varying levels of analytes, were tested in quadruplicate over five (5) days (4 replicates x 1 run x 5 days x 3 testing sites = 60 replicates per panel member).

The data were analyzed for within-run, between-run, between-day, between-site, and total precision and the standard deviation (SD) and percent coefficient of variation (% CV) were calculated.

The within run precision for positive samples greater than or equal to the cut-off (≥ 1.1 AI) in all sample matrices ranged from 2.7% to 13.3% for Rubella IgM and 2.8% to 13.5% for CMV IgM. The total precision for positive samples greater than or equal to the cut-off (≥ 1.1 AI) ranged from 2.3% to 13.5% for Rubella IgM and 3.0% to 14.2% for CMV IgM.

The mean Antibody Index (AI), standard deviation (SD), and percent coefficient of variation (%CV) for each panel member were calculated. The data for the serum panel is represented below:

BioPlex 2200 Rubella IgM Serum

Rubella IgM Panel Member	Mean (AI)	Site 1		Site 2		Site 3	
		Within-Run	Total	Within-Run	Total	Within-Run	Total
		%CV	%CV	%CV	%CV	%CV	%CV
Negative 1	0.2	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
Negative 2	0.2	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
High Negative 1	0.3	14.2%	14.9%	9.5%	9.5%	0.0%	0.0%

Rubella IgM Panel Member	Mean (AI)	Site 1		Site 2		Site 3	
		Within-Run	Total	Within-Run	Total	Within-Run	Total
		%CV	%CV	%CV	%CV	%CV	%CV
High Negative 2	0.5	0.0%	0.0%	0.0%	0.0%	4.4%	4.4%
Low Positive 1	1.0	7.2%	7.7%	4.9%	5.5%	6.4%	6.7%
Low Positive 2	1.5	4.3%	5.3%	4.2%	4.2%	6.6%	6.6%
High Positive 1	1.8	5.5%	5.5%	2.8%	2.8%	4.3%	5.1%
High Positive 2	2.0	6.3%	6.3%	2.7%	2.7%	3.7%	3.9%
Positive Control	1.4	3.4%	3.4%	6.4%	6.7%	4.2%	4.4%

BioPlex 2200 CMV IgM Serum

CMV IgM Panel Member	Mean (AI)	Site 1		Site 2		Site 3	
		Within-Run	Total	Within-Run	Total	Within-Run	Total
		%CV	%CV	%CV	%CV	%CV	%CV
Negative 1	0.2	0.0%	0.0%	15.1%	17.8%	10.9%	10.9%
Negative 2	0.3	14.8%	14.8%	9.5%	9.5%	7.3%	7.3%
High Negative 1	0.8	0.0%	0.0%	5.0%	5.3%	7.1%	7.1%
High Negative 2	0.7	11.1%	11.5%	8.3%	8.3%	6.7%	7.4%
Low Positive 1	1.5	7.2%	7.2%	6.4%	6.4%	9.9%	9.9%
Low Positive 2	1.4	12.7%	12.7%	5.6%	5.6%	4.8%	5.5%
High Positive 1	2.1	5.6%	5.9%	2.8%	3.0%	6.6%	6.6%
High Positive 2	2.6	6.4%	8.4%	3.4%	3.5%	4.0%	4.0%
Positive Control	1.8	4.6%	4.6%	7.2%	7.2%	4.5%	6.0%

b. *Linearity/assay reportable range:*

Not applicable as this assay is a qualitative assay.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

The traceability, stability and expected values for the controls and calibrators were evaluated per the manufacturer's protocols and met the acceptance criteria for all product claims.

BioPlex 2200 ToRC IgM Calibrator Set: Calibrator assignment is established from replicate analyses using a master set of calibrators and a specific lot of BioPlex 2200 ToRC IgM Reagent Pack on multiple BioPlex 2200 instruments.

BioPlex 2200 ToRC IgM Control Set: The mean values listed in the package insert for the BioPlex 2200 ToRC IgM Control Set are derived from replicate analyses. It is recommended that individual laboratories establish their own limits for each parameter and use those provided only as guides.

Stability Studies: Stability studies have been performed to determine the open vial and shelf life stability for the BioPlex 2200 ToRC IgM Control and Calibrator Sets. Stability studies were conducted for Rubella, and CMV IgM and Factor XIIIb. The measurement of Factor XIIIb is used to calibrate the serum verification assay, which ensures that serum or plasma is present in the reaction vessels.

d. *Detection limit:*

Not applicable as this assay is a qualitative assay.

e. *Analytical specificity:*

Cross-Reactivity

A cross-reactivity study was performed to determine if samples from various disease states and other potentially cross-reacting agents interfere with test results when tested with the BioPlex 2200 Rubella and CMV IgM kit.

Samples known to be positive for each of the potential cross-reactants, as determined by FDA cleared devices, were evaluated with the BioPlex 2200 Rubella and CMV IgM assay. All samples were pre-tested by the predicate devices and only those that tested negative by the predicate devices were further evaluated by the BioPlex 2200 Rubella and CMV IgM kit. Table G summarizes the number of samples scored negative by the BioPlex 2200 Rubella and CMV IgM assay within each of the cross-reactant panels. No significant cross-reactivity was observed except for potential cross-reactivity

of Myeloma IgM samples tested with the Rubella and CMV IgM assays, and EBV VCA IgM, Parvovirus B19 IgM and dsDNA samples tested with the CMV IgM assay.

Potential Cross-Reactant	BioPlex 2200 Rubella and CMV IgM Kit Results			
	N	Rubella IgM	N	CMV IgM
ANA Screen	10		10	9
CMV IgM	10	10	0	N/A
dsDNA (SLE clinical)	10	10	10	9 ^b
EBV VCA IgM	10	9	8	6
hCG (pregnancy)	10	10	10	10 ^a
HIV IgG	10	10	10	10
HSV-1 or 2 IgM	9	8	10	10 ^b
Hypergammaglobulinemia IgM	10	9	10	10
Influenza (compliment fixation)	10	10	10	10
Measles IgM	10	10	10	10
Mumps IgM	10	10	10	9
Myeloma IgM	9	7	7	5
Parvovirus B19 IgM	10	10 ^a	9	4
Rheumatoid Factor (Total)	9	9	10	9 ^a
Rubella IgM	0	N/A	10	10
<i>T. gondii</i> IgM	10	10	10	10
VZV IgM	10	10	10	10

a. One BioPlex 2200 equivocal result.

b. Two BioPlex 2200 equivocal results.

Note: The highlighted areas indicate that the potential cross reactivity with either Rubella or CMV IgM could not be ruled out.

Interfering Substances

Testing for interfering substances was conducted according to CLSI Protocol EP7-A2 (Vol. 25, No. 27). Samples were prepared by blending a pool of negative human serum with samples positive for Rubella or CMV IgM to achieve values of 1.7 to 2.3 AI for Rubella and 1.7 to 2.3 AI for CMV. Interferent or solvent (negative control) was added exogenously at levels indicated in Table H below. Test and control samples were evaluated in replicates of ten. Changes in results ranged from -13.6 to 10.0%. No significant interference was observed in any of the substances tested.

Substance	Concentration
Hemoglobin	≤ 500 mg/dL
Bilirubin (unconjugated)	≤ 20 mg/dL
Bilirubin (conjugated)	≤ 30 mg/dL
Cholesterol	≤ 500 mg/dL
Red Blood Cells	≤ 0.4% (v/v)
Gamma Globulin	≤ 6 g/dL
Triglycerides	≤ 3300 mg/dL
Beta Carotene	≤ 0.6 mg/dL
Protein (total)	≤ 12 g/dL
Ascorbic Acid	≤ 3 mg/dL
Lithium Heparin	≤ 8000 units/dL
Sodium Heparin	≤ 8000 units/dL
EDTA	≤ 800 mg/dL
Sodium Citrate	≤ 1000 mg/dL

f. Assay cut-off:

The BioPlex 2200 Rubella and CMV IgM assays report results as an antibody index, (AI). AI values are calculated by using linear regression analysis of a two point assay calibration curve. The cut-off value and assignment of the calibrators are determined by performing concordance and Receiver Operating Characteristic (ROC) analysis, using predicate results as the standard. Analyze-it software is used for the ROC analysis. This software does not employ an equivocal range for the on-test condition. Therefore, predicate equivocal results are not included in the calculations for establishing cut-off. After establishment of the cut-off, an equivocal range is set bracketing the cut-off value. Based on the results, calibrator values were adjusted such that the cut-off values equal to 1.1 AI for Rubella and CMV IgM assays. A total of 586 and 593 patient samples were evaluated to confirm rubella and CMV IgM cut-off values, respectively. ROC Analysis was performed for each analyte using this population of samples.

Results obtained using a cut-off of 1.0 AI in the BioPlex Rubella and CMV IgM assay ROC analyses demonstrated >98% negative agreement and >98% positive

agreement for CMV and >95% negative agreement and >84% positive agreement for Rubella. The BioPlex 2200 Rubella and CMV IgM assays met the concordance specifications for both analytes.

2. Comparison studies:

a. *Method comparison with predicate device:*

Comparative Testing: Prospective

Performance of the Rubella and CMV IgM kit was evaluated against corresponding commercially available Rubella and CMV IgM immunoassays. Three clinical sites tested 700 prospective samples submitted for : Rubella IgM (300 - U.S.), and CMV IgM testing (400; 300 U.S. + 100 Europe). Of the 300 samples submitted for Rubella IgM testing, 71 females were pregnant women. Results are shown in the Table below.

Equivocal results obtained on the predicate devices were re-tested on two other commercially available EIA and the consensus result of 2 out of the 3 tests was used in the analysis.

Test Ordered			BioPlex 2200 Rubella and CMV IgM kit						
			Pos (+)	Eqv	Neg (-)	Total	Pos (+) % Agreement	Neg (-) % Agreement	
Commercially Available Immunoassay	Rubella IgM	Test Ordered	Pos (+)	2	0	1	3	66.7% (2/3) 95% CI 20.8 – 93.9%	95.6% (282/295) 95% CI 92.6 – 97.4%
		Eqv	0	2	0	2			
		Neg (-)	10	3	282	295			
		Total	12	5	283	300			
	Pregnant Women*	Pos (+)	0	0	0	0	N/A	96.8% (70/71) 95%CI 92.8% - 99.8%	
		Eqv	0	0	0	0			
		Neg (-)	1	0	70	71			
		Total	1	0	70	71			
	CMV IgM	Test Ordered	Pos (+)	7 ^a	0	3 ^a	10	53.8% (7/13) 95%CI 29.1 – 76.8%	97.7% (377/386) 95%CI 95.6 – 98.8%
		Eqv	1	1	3	5			
		Neg (-)	5 ^b	3	377 ^a	385			
		Total	13	4	383	400			

- a. One sample that was equivocal by the predicate device was adjudicated by two out of three FDA cleared devices.
- b. Two samples that were equivocal by the predicate device were adjudicated by two out of three FDA cleared devices.

* The pregnant women population is a subset of the test ordered population.

Comparative Testing: Retrospective

Performance of the Rubella and CMV IgM kit was evaluated against corresponding commercially available Rubella and CMV IgM immunoassays. Three clinical sites tested 107 Rubella (45 females with 89% in the 15 - 45 age group, 49 males and 13 with unknown gender) and 229 CMV (144 females with 84% in the 15 - 45 age group and 85 males) presumptive IgM positive samples. The pregnancy status of the preselected samples was not available. Presumed positive banked samples for Rubella and CMV IgM were further selected by the respective predicate device used for the comparative analysis. Results are shown in the Table below.

Presumptive Positive for Rubella or CMV IgM			BioPlex Rubella and CMV IgM kit				
			Pos(+)	Eqv	Neg(-)	Total	Pos(+) % Agreement
Commercially Available Immunoassay	Rubella IgM	Pos(+)	103	1	3	107	96.3%
		Eqv	0	0	0	0	(103/107)
		Neg(-)	0	0	0	0	95% CI
		Total	103	1	3	107	90.3 – 98.5%
	CMV IgM	Pos(+)	209 ^a	3	17 ^b	229	91.3%
		Eqv	0	0	0	0	(209/229)
		Neg(-)	0	0	0	0	95% CI
		Total	209	3	17	229	86.9 – 94.3%

a. One sample was that was equivocal by the predicate device was adjudicated by two out of three FDA cleared devices.

b. Two samples that were equivocal by the predicate device were adjudicated by two out of three FDA cleared devices.

b. Matrix comparison:

Matched serum and plasma (potassium EDTA and sodium heparin) samples drawn from 20 individual donors were acquired from commercial sources. All

samples were evaluated in replicates of 10. Mean plasma values were compared to matched mean serum AI values. Scatter plots comparing the performance of serum samples against potassium EDTA and sodium heparin plasma samples along with the corresponding slopes of regression and correlation coefficient (r) are shown in Figures 1-4. All assays pass the slope specification of 1.0 ± 0.2 , intercept specification of ± 0.2 , and correlation coefficient (r) of ≥ 0.98 .

Figure 1. Rubella IgM : Serum vs. EDTA Plasma (N=20)

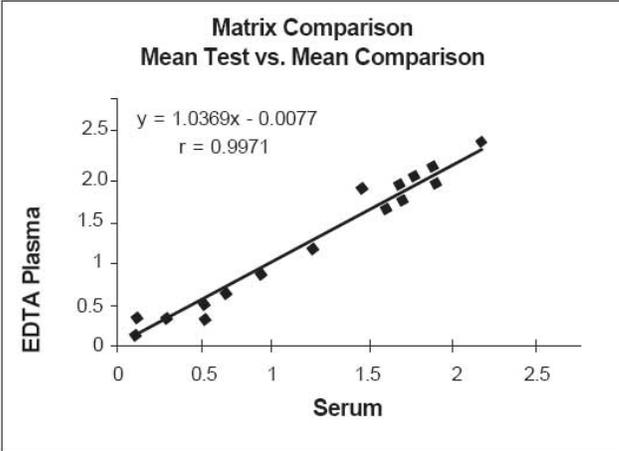


Figure 2. Rubella IgM : Serum vs. Heparin Plasma (N=20)

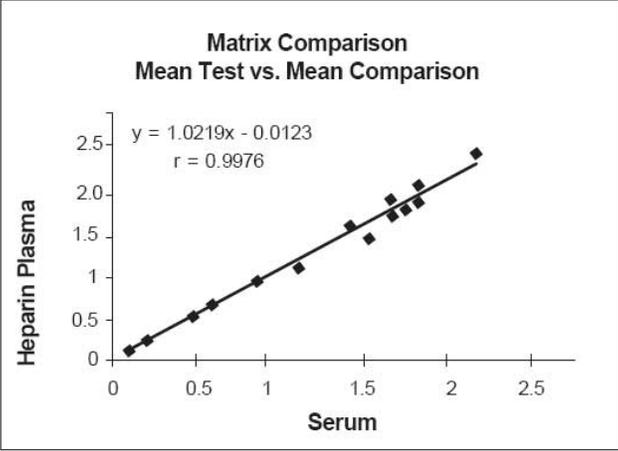


Figure 3. CMV IgM : Serum vs. EDTA Plasma (N=20)

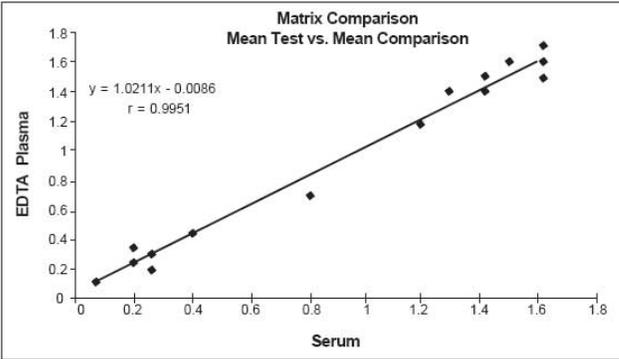
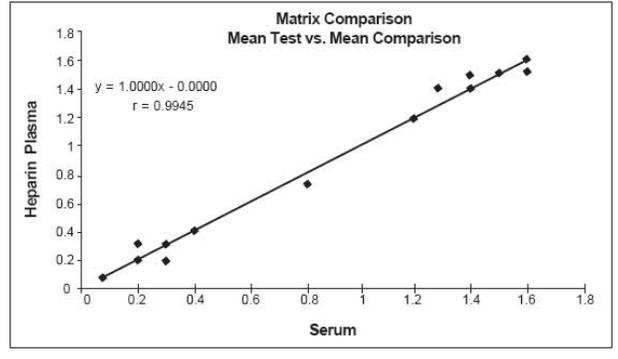


Figure 4. CMV IgM : Serum vs. Heparin Plasma (N=20)



Clinical studies:

a. *Clinical Sensitivity:*

Not Applicable

b. *Clinical specificity:*

Not Applicable

c. Other clinical supportive data (when a. and b. are not applicable):

IgM Detection

Rubella and CMV IgM-positive samples were selected and supplemented with matched specific IgG. The sample pools were split and further supplemented with dithiothreitol (DTT) which inactivates IgM activity. The samples were assayed neat and diluted into assay range in replicates of two. IgM was measured using Rubella and CMV IgM kit. The results are shown in below.

BioPlex 2200 Rubella and CMV IgM kit - Rubella IgM Specificity

Sample	Rubella IgM (AI) Before Treatment	DTT Treatment AI (% recovery)
Sample1	3.6	0.2 (5.6%)
Sample2	3.3	0.2 (6.1%)
Sample3	3	0.2 (6.7%)
Sample4	2.7	0.2 (7.4%)
Sample5	2.6	0.2 (7.7%)
Sample6	2.5	0.2 (8%)
Sample7	2.3	0.2 (8.7%)
Sample8	2.2	0.2 (9.1%)
Sample9	2.2	0.2 (9.1%)
Sample10	2.2	0.2 (9.1%)

BioPlex 2200 Rubella and CMV IgM kit - CMV IgM Specificity

Sample	CMV IgM (AI)* Before Treatment	DTT Treatment AI (% recovery)
Sample1	460	30.2 (6.6%)*
Sample2	341.5	26.4 (7.7%)*
Sample3	109	0.4 (0.4%)
Sample4	107	0.7 (0.7%)
Sample5	85.5	0.7 (0.8%)
Sample6	37.1	0.6 (1.6%)
Sample7	35.5	0.6 (1.7%)
Sample8	28.1	0.3 (1.1%)
Sample9	23.8	0.3 (1.3%)
Sample10	22.5	0.7 (3.1%)

*Values derived from dilution

Seroconversion Testing

Rubella IgM

Three Liquichek™ Rubella IgM seroconversion panels obtained from Bio-Rad Laboratories were tested with the BioPlex 2200 Rubella and CMV IgM kit. The results shown in below were compared to a commercial method.

Panel RP001	Rubella IgM	
Day	Commercial Method Index	BioPlex 2200 Rubella and CMV IgM kit AI
0	0.20 (Neg)	0.3 (Neg)
2	0.10 (Neg)	0.2 (Neg)
7	0.15 (Neg)	0.2 (Neg)
9	0.00 (Neg)	0.2 (Neg)
14	0.29 (Neg)	0.3 (Neg)
17	0.91 (Eq)	1.1 (Pos)
21	9.57 (Pos)	>4.0 (Pos)
24	7.95 (Pos)	>4.0 (Pos)
28	1.87 (Pos)	1.9 (Pos)
31	4.92 (Pos)	3.7 (Pos)
35	7.31 (Pos)	3.1 (Pos)
38	3.54 (Pos)	1.8 (Pos)
42	2.43 (Pos)	1.7 (Pos)
45	1.83 (Pos)	1.3 (Pos)
50	1.95 (Pos)	1.2 (Pos)

The BioPlex 2200 Rubella and CMV IgM test detected Rubella IgM four days earlier demonstrating greater sensitivity.

Panel RP011	Rubella IgM	
Day	Commercial Method Index	BioPlex 2200 Rubella and CMV IgM kit AI
0	0.21 (Neg)	0.2 (Neg)
3	0.00 (Neg)	0.2 (Neg)
9	0.02 (Neg)	0.2 (Neg)
12	0.00 (Neg)	0.2 (Neg)
16	1.64 (Pos)	1.1 (Pos)
19	5.94 (Pos)	>4.0 (Pos)
24	8.95 (Pos)	>4.0 (Pos)
27	7.69 (Pos)	>4.0 (Pos)
31	4.50 (Pos)	3.7 (Pos)
36	2.70 (Pos)	2.4 (Pos)
39	1.78 (Pos)	1.8 (Pos)
43	1.12 (Pos)	1.2 (Pos)
46	0.77 (Neg)	0.8 (Neg)
50	0.43 (Neg)	0.8 (Neg)
53	0.32 (Neg)	0.6 (Neg)
57	0.32 (Neg)	0.6 (Neg)

60	0.13 (Neg)	0.4 (Neg)
64	0.37 (Neg)	0.5 (Neg)
67	0.00 (Neg)	0.4 (Neg)
71	0.00 (Neg)	0.5 (Neg)

The BioPlex 2200 Rubella and CMV IgM test showed comparable performance to a commercial method.

Panel RP014	Rubella IgM	
Day	Commercial Method Index	BioPlex 2200 Rubella and CMV IgM kit AI
0	0.00 (Neg)	<0.2 (Neg)
5	0.00 (Neg)	<0.2 (Neg)
7	0.00 (Neg)	<0.2 (Neg)
12	0.00 (Neg)	<0.2 (Neg)
14	0.23 (Neg)	0.7 (Neg)
19	2.44 (Pos)	2.3 (Pos)
21	2.55 (Pos)	2.4 (Pos)
26	2.58 (Pos)	2.2 (Pos)
28	2.15 (Pos)	1.8 (Pos)
33	1.5 (Pos)	1.6 (Pos)
35	1.5 (Pos)	1.2 (Pos)
40	1.00 (Pos)	1.0 (Eq)
42	0.76 (Neg)	0.8 (Neg)

The BioPlex 2200 Rubella and CMV IgM test showed comparable performance to a commercial method, except for the day 40 sample which scored equivocal in the BioPlex 2200 Rubella and CMV IgM test and positive (at the cutoff) in the comparison test.

CMV IgM

Bio-Rad Laboratories Liquichek™ CMV IgM Seroconversion panel was assayed with BioPlex 2200 Rubella and CMV IgM kits and a commercial method. The results are shown in below. The BioPlex 2200 Rubella and CMV IgM test was able to detect CMV IgM in all the samples compared to a commercial test which scored equivocal at 59 days.

Panel RP003	CMV IgM	
Day	Commercial Methos Index	BioPlex 2200 Rubella and CMV IgM kit AI
1	1.16 (Pos)	>4.0 (Pos)
4	1.62 (Pos)	>4.0 (Pos)
8	2.44 (Pos)	>4.0 (Pos)
51	1.16 (Pos)	3.4 (Pos)
55	1.04 (Pos)	3.0 (Pos)

59	0.84 (Eq)	2.1 (Pos)
65	0.85 (Eq)	2.3 (Pos)
67	0.75 (Eq)	2.0 (Pos)
72	0.68 (Neg)	1.7 (Pos)
74	0.62 (Neg)	1.5 (Pos)
79	0.64 (Neg)	1.8 (Pos)
84	0.54 (Neg)	1.7 (Pos)
88	0.72 (Eq)	2.1 (Pos)
95	0.59 (Neg)	1.7 (Pos)
99	0.65 (Neg)	1.6 (Pos)

4. Clinical cut-off:

See section (1f).

5. Expected values/Reference range:

The observed prevalence for the Rubella and CMV IgM using the BioPlex 2200 Rubella and CMV IgM assay was determined using samples submitted for Rubella (300, US) or CMV IgM (400; 300 US +100 Europe) testing. The results are presented in the tables below. The predictive values of the test are dependent on the prevalence. As rubella incidence decreases, the predicative positive value of rubella IgM results decreases.

Note: Each laboratory should establish frequency distributions for their specific patient populations.

Expected values using the BioPlex 2200 Rubella and CMV IgM kit in test ordered population

Age	Gender	Rubella IgM (US)		CMV IgM (US)		CMV IgM (EU)	
		Pos/Total	% Prevalence	Pos/Total	% Prevalence	Pos/Total	% Prevalence
0 - 10	F	0/13	0.0%	0/4	0.0%	0/0	N/A
	M	0/11	0.0%	0/6	0.0%	0/2	0.0%
11 - 20	F	0/44	0.0%	1/19	5.3%	0/4	0.0%
	M	0/12	0.0%	0/20	0.0%	0/1	0.0%
21 - 30	F	4/80	5.0%	1/41	2.4%	1/36	2.8%
	M	0/8	0.0%	0/10	0.0%	0/2	0.0%
31 - 40	F	4/55	7.3%	1/34	2.9%	3/42	7.1%
	M	0/8	0.0%	3/17	17.6%	0/1	0.0%

41 - 50	F	3/26	11.5%	1/35	2.9%	0/6	0.0%
	M	1/12	8.3%	0/19	0.0%	0/2	0.0%
51 - 60	F	0/19	0.0%	1/23	4.3%	0/1	0.0%
	M	0/9	0.0%	1/33	0.0%	0/1	0.0%
61 - 70	F	0/1	0.0%	0/16	0.0%	0/1	0.0%
	M	0/1	0.0%	0/14	0.0%	0/0	N/A
71 +	F	0/0	N/A	0/2	0.0%	0/0	N/A
	M	0/1	0.0%	0/7	0.0%	0/1	0.0%
Total		12/300*	4.0%**	9/300	3.0%	4/100	4.0%

*One of pregnant women (N=71) was Rubella IgM positive.

** Since 2001 the incidence of Rubella in the US has been less than 10/1,000,000 population.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.