510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k062211

B. Purpose for Submission:

This is an original bundled application for a new multiplexed flow immunoassay for the qualitative detection of IgG antibodies of 3 separate analytes; Epstein-Barr Virus Nuclear Antigen (EBV NA-1), Viral Capsid Antigen (EBV VCA), and Early Antigen diffuse (EBV EA-D) in human serum to aid in the diagnosis of infectious mononucleosis.

C. Measurand:

Epstein-Barr Virus Nuclear Antigen (EBV NA-1), Viral Capsid Antigen (EBV VCA), and Early Antigen diffuse (EBV EA-D)

D. Type of Test:

Multiplexed flow immunoassay

E. Applicant:

BIO-RAD LABORATORIES, INC.

F. Proprietary and Established Names:
BioPlex 2200 EBV IgG Panel on the BioPlex 2200 Multi-Analyte Detection System
BioPlex 2200 EBV IgG Control Set
BioPlex 2200 EBV IgG Calibrator Set

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
LSE	I	866.3235 - EPSTEIN-BARR VIRUS SEROLOGICAL REAGENTS	Microbiology (83)

H. Intended Use:

  1. <u>Intended use(s):</u>
The BioPlex™ 2200 EBV IgG kit is a multiplex flow immunoassay intended for the qualitative detection of IgG antibodies to three (3) separate EBV antigens; Epstein-Barr Virus Nuclear Antigen-1 (EBV NA-1), Viral Capsid Antigen (EBV VCA), and Early Antigen diffuse (EBV EA-D) in human serum. The test system can be used in conjunction with the BioPlex 2200 EBV IgM kit as an aid in the laboratory diagnosis of infectious mononucleosis (IM).

The EBV IgG kit is intended for use with the Bio-Rad BioPlex 2200 System.

Assay performance characteristics have not been established for immunocompromised or immunosuppressed patients, cord blood, neonatal specimens, or infants. Assay performance characteristics have not been established for the diagnosis of nasopharyngeal carcinoma, Burkitt's lymphoma, and other EBV-associated lymphomas.

  2. <u>Indication(s) for use:</u>

See above

  3. <u>Special conditions for use statement(s):</u>

Prescription use only

  4. <u>Special instrument requirements:</u>

The BioPlex 2200 EBV IgG kit is intended for use with the BioPlex 2200 System instrument and software.

I. Device Description:

   The EBV IgG kit is a multiplex flow immunoassay for the qualitative detection of IgG antibodies to three (3) separate EBV antigens; Epstein-Barr Virus Nuclear Antigen-1 (EBV NA-1), Viral Capsid Antigen (EBV VCA), and Early Antigen diffuse (EBV EA-D) in human serum. See test principle below for more details.

J. Substantial Equivalence Information:

Predicate Device 1: EBNA IGG EIA TEST SYSTEM by Immuno Probe, Inc.

510k number: K951549

Component	Similarities	
	Device	Predicate
Measurand	EBNA-1 IgG	EBNA-1 IgG
Matrices	Serum	Serum
Intended Use	Aid in diagnosis of infectious mononucleosis	Aid in diagnosis of infectious mononucleosis
	Differences	
	Device	Predicate
Technology	Multiplexed flow immunoassay	Traditional ELISA
Intended Use	Qualitative detection	Semi-quantitative detection

Predicate Device 2: EBV VCA IGG EIA TEST SYSTEM by Immuno Probe, Inc.

510k number: K980912

Component	Similarities	
	Device	Predicate
Measurand	EBV VCA IgG	EBV VCA IgG
Matrices	Serum	Serum
Intended Use	Qualitative detection of EBV VCA IgG to aid in diagnosis of infectious mononucleosis	Qualitative detection of EBV VCA IgG to aid in diagnosis of infectious mononucleosis
	Differences	
	Device	Predicate
Technology	Multiplexed flow immunoassay	Traditional ELISA

Predicate Device 3: EBV EA-D IGG EIA TEST SYSTEM by Immuno Probe, Inc.

510k number: K973123

Component	Similarities	
	Device	Predicate
Measurand	EBV EA-D IgG	EBV EA-D IgG
Matrices	Serum	Serum
Intended Use	Qualitative detection of EBV EA-D IgG to aid in diagnosis of infectious mononucleosis	Qualitative detection of EBV EA-D IgG to aid in diagnosis of infectious mononucleosis
	Differences	
	Device	Predicate

Technology	Multiplexed flow immunoassay	Traditional ELISA

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

STANDARDS
Title and Reference Number
Interference Testing in Clinical Chemistry; Approved Guideline (EP 7-A)
Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline-Second Edition (EP5-A2)

Other Standards			
GUIDANCE			
Document Title	**Office**	**Division**	**Web Page**
Review Criteria for In Vitro Diagnostic Devices for Detection of IGM Antibodies to Viral Agents	**OIVD**	**DIHD**	http://www.fda.gov/cdrh/ode/527.pdf
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	**CBER/ OIVD**		http://www.fda.gov/cdrh/oivd/guidance/1588.html

L. Test Principle:

The EBV IgG 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. Three (3) different populations of beads are coated with *E. coli* derived recombinant proteins, EBV NA-1 (28kD and 45kD), EBV VCA p18 (40kD), and EBV EA-D (28kD) associated with infectious mononucleosis. 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, antihuman IgG 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 is calculated in relative fluorescence intensity (RFI).

Three additional dyed beads, an Internal Standard Bead (ISB), a Serum Verification Bead (SVB) and a Reagent Blank Bead (RBB) are present in each reaction mixture to verify detector response, the addition of serum to the reaction vessel and the absence of significant non-specific binding in serum. Refer to the BioPlex 2200 System Operation Manual for more information. The instrument is calibrated using a set of seven (7) distinct calibrator vials, supplied separately by Bio-Rad Laboratories. A combination of four (4) vials representing four (4) different antibody concentrations are used for calibration. The result for each of these antibodies is expressed as an antibody index (AI).

M. Performance Characteristics (if/when applicable):

1. <u>Analytical performance:</u>

   a. *Precision/Reproducibility:*

   Precision Studies:
   A precision panel, consisting of six (6) panel members was prepared by Bio-Rad Laboratories. Two (2) of the six (6) panel members had high levels of the antibodies contained in the BioPlex 2200 EBV IgG kit (EBV NA-1 IgG, EBV VCA IgG, and EBV EA-D IgG) and two (2) of the six (6) panel members had antibody levels near the cutoff, both prepared from positive patient samples. Two (2) of the six (6) panel members were negative (one high negative and one low negative) for both of the analytes. Precision testing was performed at Bio-Rad Laboratories on one lot of the EBV IgG kit, one lot of the EBV IgG Calibrator Set and one lot of the EBV IgG Control Set. Each of the six (6) panel members was tested in duplicate (x2) on two (2) runs per day for ten (10) days using one (1) lot of EBV IgG kit, one (1) lot of EBV IgG Calibrator Set and one (1) lot of EBV IgG Control Set (2 times x 2 runs x 10 days = 40 replicates per panel member). The data were analyzed for intra-assay and inter-assay precision according to the principles described in the Clinical Laboratory Standards Institute guidance EP5-A2, revised November 2004 and ISO/TR 22971:2205. The standard deviation (SD) and percent coefficient of variation (%CV) were calculated. Results can be found in Tables S - U.

Table S. Precision Results; BioPlex 2200 EBV NA-1 IgG

EBV NA-1 IgG Panel Members	Sample N*	AI Mean	Within-Run		Between-Day		Between-Run		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	42	4.6	0.2	4.2%	0.3	6.3%	0.3	5.9%	0.4	9.6%
High Positive 2	42	4.6	0.2	4.0%	0.2	5.4%	0.4	8.6%	0.5	10.9%
Low Positive 1	42	1.9	0.1	5.5%	0.0	0.0%	0.2	11.9%	0.2	13.1%
Low Positive 2	42	2.2	0.1	5.0%	0.0	0.0%	0.2	10.4%	0.3	11.5%
High Negative	43	0.7	0.1	7.1%	0.0	3.8%	0.1	12.1%	0.1	14.5%
Low Negative	44	0.0	0.0	0.0%	0.0	0.0%	0.0	0.0%	0.0	0.0%

*Additional samples were run.

Table T. Precision Results; BioPlex 2200 EBV VCA IgG

EBV VCA IgG Panel Members	Sample N*	AI Mean	Within-Run		Between-Day		Between-Run		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	42	3.5	0.1	3.9%	0.1	3.9%	0.2	6.8%	0.3	8.7%
High Positive 2	42	3.4	0.2	5.0%	0.1	3.7%	0.3	9.1%	0.4	11.0%
Low Positive 1	42	1.6	0.1	8.4%	0.0	2.6%	0.1	8.1%	0.2	11.9%
Low Positive 2	42	1.3	0.1	7.4%	0.1	4.1%	0.1	10.2%	0.2	13.3%
High Negative	43	0.6	0.1	10.8%	0.0	0.0%	0.1	11.4%	0.1	15.7%
Low Negative	44	0.2	0.0	16.3%	0.0	6.8%	0.0	5.1%	0.0	18.4%

*Additional samples were run.

Table U. Precision Results; BioPlex 2200 EBV EA-D IgG

EBV EA-D IgG Panel Members	Sample N*	AI Mean	Within-Run		Between-Day		Between-Run		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	42	4.3	0.2	4.6%	0.0	0.0%	0.3	6.9%	0.4	8.3%
High Positive 2	42	4.2	0.3	6.0%	0.3	6.3%	0.3	7.2%	0.5	11.2%
Low Positive 1	42	2.3	0.2	10.3%	0.1	4.5%	0.2	9.8%	0.3	14.9%
Low Positive 2	42	2.3	0.2	7.0%	0.2	6.8%	0.2	9.1%	0.3	13.4%
High Negative	43	0.7	0.1	13.4%	0.0	0.0%	0.1	12.9%	0.1	18.6%
Low Negative	44	0.2	0.1	29.8%	0.0	5.3%	0.0	0.0%	0.1	30.3%

*Additional samples were run.

Reproducibility Studies:
A reproducibility panel, consisting of nine (9) panel members was prepared by Bio-Rad Laboratories. Two (2) of the nine (9) panel members had high levels of EBV NA-1 and EBV VCA; two (2) of the nine (9) panel members had high levels

of EBV EA-D; two (2) of the nine (9) panel members had antibody levels near the cutoff for EBV NA-1 and EBV VCA; two (2) of the nine (9) panel members had antibody levels near the cutoff for EBV EA-D. All were prepared from positive patient samples. One (1) of the nine (9) panel members was negative for all three (3) analytes contained in the BioPlex 2200 EBV IgG kit. In addition, three (3) lots of the EBV IgG Control Set [1 positive control (antibody positive) and a negative control (antibody negative)] were also tested. Reproducibility testing was performed at each of three (3) US testing facilities on a total of three (3) lots of the EBV IgG kit, three (3) lots of the EBV IgG Calibrator Set and three (3) lots of the EBV IgG Control Set. Each testing facility evaluated reproducibility using one (1) kit lot of EBV IgG with matched calibrators and controls. The panels were provided to each of the testing sites. Each of the nine (9) panel members and positive and negative controls was tested in quadruple (x4) on each day for three (3) days at each of three (3) US testing facilities using one (1) lot of EBV IgG reagent pack, one (1) lot of EBV IgG Calibrator Set and one (1) lot of EBV IgG Control Set (4 times x 3 days x 3 sites = 36 replicates per panel member and controls). The data were analyzed for intra-assay and inter-assay reproducibility according to the principles described in the Clinical Laboratory Standards Institute guidance EP5-A2, revised November 2004 and ISO/TR 22971:2205. The standard deviation (SD) and percent coefficient of variation (%CV) were calculated. Positive results can be found in Tables P-R.

Table P. Reproducibility; BioPlex 2200 EBV NA-1 IgG

EBV NA-1 IgG Panel Members	Sample N	Grand Mean AI	Within-Run		Between-Day		Between-Run		Between-Site*		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	36	4.2	0.1	3.2%	0.0	0.0%	0.1	2.9%	0.3	7.4%	0.4	8.5%
High Positive 2	36	4.3	0.1	3.4%	0.1	2.5%	0.1	2.8%	0.3	7.5%	0.4	9.0%
Low Positive 1	36	1.5	0.1	4.8%	0.0	0.0%	0.1	5.7%	0.2	11.6%	0.2	13.8%
Low Positive 2	36	2.1	0.1	3.2%	0.0	1.1%	0.1	3.7%	0.2	9.4%	0.2	10.7%
Positive Control	36	2.9	0.1	1.9%	0.0	1.1%	0.1	2.4%	0.5	17.2%	0.5	17.5%

*Between site variance includes between lot variance.

Table Q. Reproducibility; BioPlex 2200 EBV VCA IgG

EBV VCA IgG Panel Members	Sample N	Grand Mean AI	Within-Run		Between-Day		Between-Run		Between-Site*		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	36	3.3	0.1	3.5%	0.0	0.0%	0.1	3.3%	0.4	12.7%	0.4	13.5%
High Positive 2	36	3.2	0.1	3.4%	0.0	0.0%	0.1	3.7%	0.2	7.3%	0.3	8.9%
Low Positive 1	36	1.5	0.1	5.0%	0.0	0.0%	0.1	5.5%	0.2	16.2%	0.3	17.8%
Low Positive 2	36	1.3	0.1	5.8%	0.0	0.0%	0.1	5.0%	0.1	7.6%	0.1	10.8%
Positive Control	36	2.3	0.1	2.8%	0.0	0.0%	0.1	2.7%	0.1	5.6%	0.2	6.9%

*Between site variance includes between lot variance.

Table R. Reproducibility; BioPlex 2200 EBV EA-D IgG

EBV EA-D IgG Panel Members	Sample N	Grand Mean AI	Within-Run		Between-Day		Between-Run		Between-Site*		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
High Positive 1	36	4.1	0.2	4.4%	0.0	0.8%	0.0	0.0%	0.4	8.7%	0.4	9.8%
High Positive 2	36	4.0	0.1	3.7%	0.0	0.7%	0.1	3.5%	0.2	6.2%	0.3	8.1%
Low Positive 1	36	2.3	0.2	8.8%	0.1	2.8%	0.0	0.0%	0.1	5.7%	0.2	10.9%
Low Positive 2	36	2.2	0.1	4.6%	0.0	0.0%	0.1	3.5%	0.1	4.2%	0.2	7.2%
Positive Control	36	3.0	0.1	3.1%	0.0	0.0%	0.1	2.5%	0.6	18.8%	0.6	19.2%

*Between site variance includes between lot variance.

b. *Linearity/assay reportable range:*

Not Applicable.

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

Not Applicable.

d. *Detection limit:*

Not applicable.

e. Analytical specificity:

A cross-reactivity study was performed to determine if samples from various disease states and other potentially interfering factors interfere with test results when tested with the BioPlex 2200 EBV IgG kit. A panel of ten (10) specimens* positive for each cross reactant were evaluated for possible cross reactivity with the BioPlex 2200 EBV IgG kit for each of the three EBV IgG antibody assays. Due to the high prevalence of EBV IgG antibodies in the normal population, the test specimens were also evaluated on corresponding commercially available microplate EIAs. Most of the samples evaluated were high positive for each disease state. The majority of all samples that elicited a positive result were also confirmed positive by the corresponding commercially available microplate EIA, indicating reactivity to EBV IgG antibodies rather than cross reactivity with a potentially interfering factor. Results can be found in Table S.

Due to limited availability of samples, only four E. coli specimens were evaluated.

Table S. Cross-Reactivity

Cross Reactives	N	Method	BioPlex 2200 EBV IgG		
			EBV NA-1 IgG	EBV VCA IgG	EBV EA-D IgG
ANA	10	BioPlex 2200	9	10	7
		EIA	9	10	7
		Discrepants	0	0	0
Rheumatoid Factor	10	BioPlex 2200	10	10	1
		EIA	10	10	1*
		Discrepants	0	0	0
Toxo IgG	10	BioPlex 2200	9	9	2
		EIA	9	9*	2*
		Discrepants	0	0	0
Rubella IgG	10	BioPlex 2200	10	10	2
		EIA	10	10	1*
		Discrepants	0	0	1
CMV IgG	10	BioPlex 2200	10	10	2
		EIA	10	10	2**
		Discrepants	0	0	1
VZV IgG	10	BioPlex 2200	8	8	1
		EIA	9	8	1
		Discrepants	1	0	0
HSV-1 IgG	10	BioPlex 2200	10	10	2
		EIA	10	10	3
		Discrepants	0	0	1
HSV-2 IgG	10	BioPlex 2200	10	10	3
		EIA	10	10	4
		Discrepants	0	0	1
HIV	10	BioPlex 2200	10	9	1
		EIA	10	10	2**
		Discrepants	0	1	1
E. coli	4	BioPlex 2200	4	4	0
		EIA	4	4	0*
		Discrepants	0	0	0
Pregnant women	10	BioPlex 2200	9	9	3
		EIA	9	10	3*
		Discrepants	0	1	0

*One Equivocal Sample; **Two Equivocal Samples

Interfering Substances:

Testing for interfering substances was conducted according to CLSI Protocol EP7-A (Vol. 22, No. 27). No significant interference was observed in any of the substances tested. The following substances, listed in Table T, were tested (N=10) at maximum levels on one reagent lot.

Table T. Interfering Substances

Substance	Concentration
Hemoglobin	≤ 500 mg/dL
Bilirubin (unconjugated)	≤ 20 mg/dL
Bilirubin (conjugated)	≤ 20 mg/dL
Triglycerides	≤ 3000 mg/dL
Protein (total)	≤ 12 g/dL
Cholesterol	≤ 500 mg/dL
Red Blood Cells	≤ 0.4% Concentrate
Gamma-globulin	≤ 2.5 g/dL
Ascorbic Acid	≤ 3 mg/dL

f. Assay cut-off:

A final cut-off of 1 AI was established for all BioPlex 2200 EBV IgG kit assays based on an evaluation of 808 serum samples with the BioPlex 2200 EBV IgG kit assays and corresponding commercially available microplate EIA tests. ROC analysis was performed for each BioPlex 2200 EBV IgG assay using this population of samples. Samples that were equivocal on the commercially available microplate EIA tests were excluded from the analysis. This analysis was used to optimize sensitivity and specificity for the BioPlex 2200 EBV IgG kit assays. A final cut-off of 1 AI was established for all BioPlex 2200 EBV IgG kit assays based on the data collected.

2. Comparison studies:

a. Method comparison with predicate device:
Performance of the BioPlex 2200 EBV IgG kit was tested against corresponding commercially available microplate EIAs. A total of 621 banked serum samples from patients for which an EBV test were ordered were tested at 3 U.S. clinical testing sites. The BioPlex 2200 EBV IgG kit was run in conjunction with the BioPlex 2200 EBV IgM kit to allow for a complete antibody response profile. The characterization by antibody response was not compared with clinical data regarding presence, absence or status of disease. Two (2) samples were excluded due to RBB analysis error messages during BioPlex 2200 EBV IgM testing. One (1) sample was excluded due to RBB

analysis error messages during BioPlex 2200 EBV IgG testing. Using Table A as a guideline, results were analyzed by BioPlex 2200 EBV IgG analytes and corresponding EBV IgG reference assays according to serological characterization based on reference assay results. For the purpose of percent agreement calculations, BioPlex 2200 EBV IgG equivocal results were assigned to the opposite clinical interpretation than that of the corresponding reference assay result. Likewise, the reference IgG assay equivocal results were assigned to the opposite clinical interpretation than that of the corresponding BioPlex 2200 EBV IgG result. Results from all sites are shown and summarized in Tables H-M.

Table H. BioPlex 2200 EBV NA-1 IgG vs. EIA: Comparison by Serological Pattern Characterization

EBV Serological Status	Reference EBV NA-1 IgG Interpretation									Total
	Positive			Equivocal			Negative			
	BioPlex 2200 EBV NA-1 IgG			BioPlex 2200 EBV NA-1 IgG			BioPlex 2200 EBV NA-1 IgG			
	Pos	Ind	Neg	Pos	Ind	Neg	Pos	Ind	Neg	
	N	N	N	N	N	N	N	N	N	N
Primary Acute	0	0	0	0	0	0	0	0	31	31
Late Acute	104	0	4	0	0	0	0	0	2	110
Recovering	0	0	0	0	0	0	0	0	4	4
Previous Infection	285	1	4	0	0	0	4	0	11	305
Susceptible	0	0	0	0	0	1	1	0	125	127
Inconclusive	20	0	9	0	0	0	0	0	12	41
Overall	409	1	17	0	0	1	5	0	185	618

Table I. BioPlex 2200 EBV NA-1 IgG vs. EIA: Percent Agreement & Confidence Intervals by Serological Pattern Characterization

EBV Serological Status	Positive Agreement		95% CI	Negative Agreement		95% CI
Primary Acute	(0/0)	N/A*	N/A*	(31/31)	100%	89.0 -100%
Late Acute	(104/108)	96.3%	90.9 - 98.6%	(2/2)	100%	34.2 - 100%
Recovering	(0/0)	N/A*	N/A*	(4/4)	100%	51.0 -100%
Previous Infection	(285/290)	98.3%	96.0 - 99.3%	(11/15)	73.3%	48.0 - 98.1%
Susceptible	(0/1)	0.0%	N/A*	(125/126)	99.2%	95.6 - 99.9%
Inconclusive	(20/29)	69.0%	50.8 - 82.7%	(12/12)	100%	75.8 - 100%
Overall	(409/428)	95.6%	93.2 - 97.1%	(185/190)	97.4%	94.0 - 98.9%

*In cases where agreement resulted in a numerator of zero (0), 95% confidence interval could not be calculated; in cases where agreement resulted in (0/0) samples, percent agreement and 95% confidence interval could not be calculated.

Table J. BioPlex 2200 EBV VCA IgG vs. EIA: Comparison by Serological Pattern Characterization

EBV Serological Status	Reference EBV VCA IgG Interpretation									Total
	Positive			Equivocal			Negative			
	BioPlex 2200 EBV VCA IgG			BioPlex 2200 EBV VCA IgG			BioPlex 2200 EBV VCA IgG			
	Pos	Ind	Neg	Pos	Ind	Neg	Pos	Ind	Neg	
	N	N	N	N	N	N	N	N	N	N
Primary Acute	4	0	3	0	0	1	0	0	23	31
Late Acute	106	0	4	0	0	0	0	0	0	110
Recovering	4	0	0	0	0	0	0	0	0	4
Previous Infection	296	1	8	0	0	0	0	0	0	305
Susceptible	0	0	0	0	0	0	0	0	127	127
Inconclusive	16	0	0	0	0	0	1	0	24	41
Overall	426	1	15	0	0	1	1	0	174	618

Table K. BioPlex 2200 EBV VCA IgG vs. EIA: Percent Agreement & Confidence Intervals by Serological Pattern Characterization

EBV Serological Status	Positive Agreement		95% CI	Negative Agreement		95% CI
Primary Acute	(4/8)	50.0%	21.5 - 78.5%	(23/23)	100%	85.7 - 100%
Late Acute	(106/110)	96.4%	91.0 - 98.6%	(0/0)	N/A*	NA*
Recovering	(4/4)	100%	51.0 - 100%	(0/0)	N/A*	NA*
Previous Infection	(296/305)	97.0%	94.5 - 98.4%	(0/0)	N/A*	NA*
Susceptible	(0/0)	N/A*	NA*	(127/127)	100%	97.1 - 100%
Inconclusive	(16/16)	100%	80.6 - 100%	(24/25)	96.0%	80.5 - 99.3%
Overall	(426/443)	96.2%	93.9 - 97.6%	(174/175)	99.4%	96.8 - 99.9%

*In cases where agreement resulted in (0/0) samples, percent agreement and 95% confidence interval could not be calculated.

Table L. BioPlex 2200 EBV EA-D IgG vs. EIA: Comparison by Serological Pattern Characterization

EBV Serological Status	Reference EBV EA-D IgG Interpretation									Total
	Positive			Equivocal			Negative			
	BioPlex 2200 EBV EA-D IgG			BioPlex 2200 EBV EA-D IgG			BioPlex 2200 EBV EA-D IgG			
	Pos	Ind	Neg	Pos	Ind	Neg	Pos	Ind	Neg	
	N	N	N	N	N	N	N	N	N	N
Primary Acute	18	1	2	0	0	0	2	1	7	31
Late Acute	72	1	3	4	0	0	6	3	21	110
Recovering	4	0	0	0	0	0	0	0	0	4
Previous Infection	0	0	0	5	3	3	23	22	249	305
Susceptible	0	0	0	0	1	1	0	9	116	127
Inconclusive	10	2	1	0	0	0	2	2	24	41
Overall	104	4	6	9	4	4	33	37	417	618

Table M. BioPlex 2200 EBV EA-D IgG vs. EIA: Percent Agreement & Confidence Intervals by Serological Pattern Characterization

EBV Serological Status	Positive Agreement		95% CI	Negative Agreement		95% CI
Primary Acute	(18/21)	85.7%	65.4 - 95.0%	(7/10)	70.0%	39.7 - 89.2%
Late Acute	(72/76)	94.7%	87.2 - 97.9%	(21/34)	61.8%	45.0 - 76.1%
Recovering	(4/4)	100%	51.0 - 100%	(0/0)	N/A*	N/A*
Previous Infection	(0/3)	0.0%	N/A*	(249/299)	83.3%	78.6 - 87.1%
Susceptible	(0/1)	0.0%	N/A*	(116/125)	92.8%	86.9 - 96.2%
Inconclusive	(10/13)	76.9%	49.7 - 91.8%	(24/28)	85.7%	68.5 - 94.3%
Overall	(104/118)	88.1%	81.1 - 92.8%	(417/496)	84.1%	80.6 - 87.0%

*In cases where agreement resulted in a numerator of zero (0), 95% confidence interval could not be calculated; in cases where agreement resulted in (0/0) samples, percent agreement and 95% confidence interval could not be calculated.

Comparison of Characterization EBV Serological Status:
Using Table A as a guideline, samples characterized into serological status associated with EBV disease, using the commercially available microplate EIA and agglutination tests, were compared with characterizations using BioPlex 2200 EBV IgG and IgM kits. The EBV IgG kit was run in conjunction with the EBV IgM kit to allow for a complete antibody response profile. The characterization by antibody response was not compared with clinical data regarding presence, absence or status of disease. Results from 618 serum samples tested at 3 U.S. clinical testing sites are shown in Table N.

Table N. Comparison of EBV Serological Status

EBV Serological status		Primary Acute	Late Acute	Recovering	Previous Infection	Susceptible	Inconclusive	Total	% Serological Agreement	95% Confidence Interval
		BioPlex 2200 EBV IgG & IgM Profile								
Commercially Available Assays	Primary Acute	30	0	0	0	0	1	31	96.8%	83.8 - 99.4%
	Late Acute	5	90	1	13	0	1	110	81.8%	73.6 - 87.9%
	Recovering	1	0	3	0	0	0	4	75.0%	30.0 - 95.4%
	Previous Infection	0	31	2	263	4	5	305	86.2%	81.9 - 89.7%
	Susceptible	4	0	0	0	122	1	127	96.1%	91.1 - 98.3%
	Inconclusive	6	10	0	7	11	7	41	17.1%	8.5 - 31.3%
	Overall	46	131	6	283	137	15	618	83.3%	80.2 - 86.1%

Note: Calculations are performed for unshaded areas only.

   b. Matrix comparison:

   Not Applicable.

3. Clinical studies:

   Not Applicable

4. Clinical cut-off**:**

   Not Applicable

5. Expected values/Reference range:
   Expected values for the EBV IgG kit are presented by age and gender in the following tables for serum samples from unselected hospitalized pediatric and adult patients (N=303) and patients for which an EBV test was ordered (N=620). A total of 621 serum samples from patients for which an EBV test was ordered were tested. One (1) sample from the patients for which an EBV test was ordered population was excluded due to RBB analysis error messages during BioPlex 2200 EBV IgG testing. For all analytes, results of ≤0.8 AI are negative, 0.9 and 1.0 AI are indeterminate, and ≥1.1 AI are reported as positive.

Table B. Hospitalized Patient Samples: EBV NA-1 IgG

| Age | Gender | BioPlex 2200 EBV NA-1 IgG | | | | | | Total |
| | | Positive | | Indeterminate | | Negative | | |
		N	%	N	%	N	%	N
< 5 years of age	F	11	41%	0	0%	16	59%	27
	M	6	30%	0	0%	14	70%	20
5-12 years of age	F	13	59%	0	0%	9	41%	22
	M	15	44%	0	0%	19	56%	34
13-20 years of age	F	28	80%	0	0%	7	20%	35
	M	10	67%	0	0%	5	33%	15
21-30 years of age	F	5	83%	0	0%	1	17%	6
	M	1	50%	0	0%	1	50%	2
31-40 years of age	F	10	100%	0	0%	0	0%	10
	M	11	100%	0	0%	0	0%	11
41-50 years of age	F	13	100%	0	0%	0	0%	13
	M	7	100%	0	0%	0	0%	7
51-60 years of age	F	22	96%	0	0%	1	4%	23
	M	18	95%	0	0%	1	5%	19
61-70 years of age	F	11	100%	0	0%	0	0%	11
	M	12	100%	0	0%	0	0%	12
71-80 years of age	F	11	100%	0	0%	0	0%	11
	M	6	100%	0	0%	0	0%	6
81-90 years of age	F	11	100%	0	0%	0	0%	11
	M	5	83%	0	0%	1	17%	6
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	2	100%	0	0%	0	0%	2
Total		228	75%	0	0%	75	25%	303

Table C. Hospitalized Patient Samples: EBV VCA IgG

| Age | Gender | BioPlex 2200 EBV VCA IgG | | | | | | Total |
| | | Positive | | Indeterminate | | Negative | | |
		N	%	N	%	N	%	N
< 5 years of age	F	11	41%	0	0%	16	59%	27
	M	5	25%	0	0%	15	75%	20
5-12 years of age	F	14	64%	0	0%	8	36%	22
	M	15	44%	0	0%	19	56%	34
13-20 years of age	F	29	83%	0	0%	6	17%	35
	M	9	60%	0	0%	6	40%	15
21-30 years of age	F	6	100%	0	0%	0	0%	6
	M	1	50%	0	0%	1	50%	2
31-40 years of age	F	10	100%	0	0%	0	0%	10
	M	11	100%	0	0%	0	0%	11
41-50 years of age	F	13	100%	0	0%	0	0%	13
	M	7	100%	0	0%	0	0%	7
51-60 years of age	F	22	96%	0	0%	1	4%	23
	M	18	95%	0	0%	1	5%	19
61-70 years of age	F	10	91%	1	9%	0	0%	11
	M	11	92%	1	8%	0	0%	12
71-80 years of age	F	10	91%	1	9%	0	0%	11
	M	6	100%	0	0%	0	0%	6
81-90 years of age	F	11	100%	0	0%	0	0%	11
	M	5	83%	0	0%	1	17%	6
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	2	100%	0	0%	0	0%	2
Total		226	75%	3	1%	74	24%	303

Table D. Hospitalized Patient Samples: EBV EA-D IgG

| Age | Gender | BioPlex 2200 EA-D IgG | | | | | | Total |
| | | Positive | | Indeterminate | | Negative | | |
		N	%	N	%	N	%	N
< 5 years of age	F	2	7%	0	0%	25	93%	27
	M	1	5%	1	5%	18	90%	20
5-12 years of age	F	4	18%	0	0%	18	82%	22
	M	5	15%	0	0%	29	85%	34
13-20 years of age	F	9	26%	2	6%	24	69%	35
	M	3	20%	3	20%	9	60%	15
21-30 years of age	F	2	33%	0	0%	4	67%	6
	M	1	50%	1	50%	0	0%	2
31-40 years of age	F	4	40%	2	20%	4	40%	10
	M	6	55%	0	0%	5	45%	11
41-50 years of age	F	5	38%	0	0%	8	62%	13
	M	2	29%	0	0%	5	71%	7
51-60 years of age	F	8	35%	5	22%	10	43%	23
	M	10	53%	1	5%	8	42%	19
61-70 years of age	F	3	27%	0	0%	8	73%	11
	M	5	42%	1	8%	6	50%	12
71-80 years of age	F	4	36%	2	18%	5	45%	11
	M	1	17%	0	0%	5	83%	6
81-90 years of age	F	7	64%	1	9%	3	27%	11
	M	5	83%	0	0%	1	17%	6
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	1	50%	0	0%	1	50%	2
Total		88	29%	19	6%	196	65%	303

Table E. Samples from Patients for which an EBV Test was Ordered: EBV NA-1 IgG

Age	Gender	BioPlex 2200 EBV NA-1 IgG						Total
		Positive		Indeterminate		Negative		
		N	%	N	%	N	%	N
< 5 years of age	F	6	20%	0	0%	24	80%	30
	M	9	26%	0	0%	25	74%	34
5-12 years of age	F	22	35%	0	0%	40	65%	62
	M	24	39%	0	0%	38	61%	62
13-20 years of age	F	46	59%	1	1%	31	40%	78
	M	19	49%	0	0%	20	51%	39
21-30 years of age	F	42	91%	0	0%	4	9%	46
	M	25	76%	0	0%	8	24%	33
31-40 years of age	F	50	96%	0	0%	2	4%	52
	M	22	92%	0	0%	2	8%	24
41-50 years of age	F	33	100%	0	0%	0	0%	33
	M	30	97%	0	0%	1	3%	31
51-60 years of age	F	26	96%	0	0%	1	4%	27
	M	21	81%	0	0%	5	19%	26
61-70 years of age	F	11	85%	0	0%	2	15%	13
	M	19	90%	0	0%	2	10%	21
71-80 years of age	F	2	100%	0	0%	0	0%	2
	M	3	100%	0	0%	0	0%	3
81-90 years of age	F	2	100%	0	0%	0	0%	2
	M	2	100%	0	0%	0	0%	2
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	0	0%	0	0%	0	0%	0
Total		414	67%	1	0%	205	33%	620

Table F. Samples from Patients for which an EBV Test was Ordered: EBV VCA IgG

Age	Gender	BioPlex 2200 EBV VCA IgG						Total
		Positive		Indeterminate		Negative		
		N	%	N	%	N	%	N
< 5 years of age	F	6	20%	0	0%	24	80%	30
	M	11	32%	0	0%	23	68%	34
5-12 years of age	F	21	34%	0	0%	41	66%	62
	M	28	45%	0	0%	34	55%	62
13-20 years of age	F	47	60%	0	0%	31	40%	78
	M	20	51%	0	0%	19	49%	39
21-30 years of age	F	43	93%	1	2%	2	4%	46
	M	26	79%	0	0%	7	21%	33
31-40 years of age	F	52	100%	0	0%	0	0%	52
	M	22	92%	0	0%	2	8%	24
41-50 years of age	F	32	97%	0	0%	1	3%	33
	M	31	100%	0	0%	0	0%	31
51-60 years of age	F	27	100%	0	0%	0	0%	27
	M	24	92%	0	0%	2	8%	26
61-70 years of age	F	12	92%	0	0%	1	8%	13
	M	18	86%	0	0%	3	14%	21
71-80 years of age	F	2	100%	0	0%	0	0%	2
	M	3	100%	0	0%	0	0%	3
81-90 years of age	F	2	100%	0	0%	0	0%	2
	M	1	50%	0	0%	1	50%	2
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	0	0%	0	0%	0	0%	0
Total		428	69%	1	0%	191	31%	620

Table G. Samples from Patients for which an EBV Test was Ordered: EBV EA-D IgG

Age	Gender	BioPlex 2200 EA-D IgG						Total
		Positive		Indeterminate		Negative		
		N	%	N	%	N	%	N
< 5 years of age	F	3	10%	3	10%	24	80%	30
	M	6	18%	2	6%	26	76%	34
5-12 years of age	F	7	11%	3	5%	52	84%	62
	M	6	10%	4	6%	52	84%	62
13-20 years of age	F	16	21%	8	10%	54	69%	78
	M	12	31%	2	5%	25	64%	39
21-30 years of age	F	16	35%	2	4%	28	61%	46
	M	9	27%	3	9%	21	64%	33
31-40 years of age	F	15	29%	5	10%	32	62%	52
	M	7	29%	1	4%	16	67%	24
41-50 years of age	F	10	30%	2	6%	21	64%	33
	M	4	13%	1	3%	26	84%	31
51-60 years of age	F	13	48%	3	11%	11	41%	27
	M	10	38%	1	4%	15	58%	26
61-70 years of age	F	6	46%	1	8%	6	46%	13
	M	5	24%	3	14%	13	62%	21
71-80 years of age	F	0	0%	1	50%	1	50%	2
	M	1	33%	0	0%	2	67%	3
81-90 years of age	F	0	0%	0	0%	2	100%	2
	M	1	50%	0	0%	1	50%	2
91-100 years of age	F	0	0%	0	0%	0	0%	0
	M	0	0%	0	0%	0	0%	0
Total		147	24%	45	7%	428	69%	620

N. Proposed Labeling:

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

O. Conclusion:

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

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