

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

**A. 510(k) Number:**

K073382

**B. Purpose for Submission:**

New device.

**C. Measurand:**

Epstein - Barr virus (EBV) specific IgG antibodies to the EBV nuclear antigen (EBV NA-1), viral capsid antigen (EBV VCA), and early antigen-diffuse (EBV EA-D)

**D. Type of Test:**

Multiplexed micro particle immunoassay based on Luminex xMAP technology

**E. Applicant:**

Focus Diagnostics, Inc.

**F. Proprietary and Established Names:**

Plexus™ EBV IgG Multi-Analyte Diagnostics (MP0500G)

**G. Regulatory Information:**

1. Regulation section:

21 CFR 866.3235 – Epstein Barr Virus Serological Reagents

2. Classification:

Class I, non exempt

3. Product code:

LSE

4. Panel:

Microbiology (83)

## **H. Intended Use:**

### 1. Intended use(s):

Focus Diagnostics' Plexus™ EBV IgG Multi-Analyte Diagnostics test kit is intended for qualitatively detecting the presence or absence of human IgG class antibodies to viral capsid antigen (VCA), early antigen-diffuse (EA-D), and nuclear antigen-1 (EBNA-1) of Epstein-Barr virus in human sera. The test is indicated as an aid in the diagnosis of EBV infection and EBV-associated infectious mononucleosis.

The performance of this assay has not been established for use in the diagnosis of nasopharyngeal carcinoma and Burkitt's lymphoma, for testing of immunocompromised patients, for use by a point of care facility or for use with automated equipment. This assay has not been evaluated for donor screening.

### 2. Indication(s) for use:

Same as Intended Use

### 3. Special conditions for use statement(s):

For prescription use only

### 4. Special instrument requirements:

Instrument: The Luminex xMAP® System.

Software: Plexus™ Multi Analyte Diagnostic Software for Luminex xMAP instrument with Luminex IS 2.3 software (SW.MP0001)

## **I. Device Description:**

The Focus Diagnostics Plexus™ EBV IgG Multi-Analyte Diagnostic uses an antigen bead suspension that contains three distinct EBV antigen bead types (viral capsid antigen (VCA) , early antigen- D (EA-D) and nucleic antigen-1 (EBNA-1)) and one process control bead type that fluoresce at different wavelengths and/or intensities.

## **J. Substantial Equivalence Information:**

### 1. Predicate device name(s):

Predicate devices: Diamedix EBNA-1 IgG ELISA (K946353), Diamedix EA-D IgG ELISA (K884829), Diamedix VCA IgG (K884591).

Consensus comparator: For VCA IgG the performance was compared to a consensus based algorithm based on a 2 out of 3 rule,

consisting of the predicate device Diamedix VCA IgG ELISA (K884951) and 2 other devices; Athena Multi-Lyte EBV IgG Test System (K042118) and Focus EBV-VCA Antibody (IgG) IFA (K884591)

2. Predicate K number(s):

See J1 above

3. Comparison with predicate:

VCA IgG:

Component	Similarities			
	Device	K	K	K
Measurand	EBV VCA IgG	EBV VCA IgG	EBV VCA IgG	EBV VCA IgG
Matrix	Serum	Serum	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	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			
Technology	Multiplexed flow immunoassay	Traditional ELISA	Multiplex bead immunoassay	Immunofluorescence Antibody (IFA) test
Antigen	EBV-VCA: VCA gp 125, affinity purified antigen	EBV-VCA: Recombinant 47 kDa fusion half of p18	EBV VCA gp25	purified protein

EBNA-1 IgG:

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
Antigen	Recombinant EBNA-1, truncated, 35 kDa	27 kDa purified native protein

EA-D IgG:

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
Antigen	EBV-EA: Recombinant EA-D	EBV-EA: Recombinant EA-D 28 kDa

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

Guidance for Industry and FDA Staff, Format for Traditional and Abbreviated 510(k), 08/12/2005, (<http://www.fda.gov/cdrh/ode/guidance/1567.pdf>)

Off-The-Shelf Software Use in Medical Devices, 09/9/1999, (<http://www.fda.gov/cdrh/ode/guidance/585.pdf>)

Cyber security for Networked Medical Devices Containing Off-The-Shelf (OTS) Software, 01/14/2005, (<http://www.fda.gov/cdrh/comp/guidance/1553.pdf>)

Guidance for the Content of Premarket Submissions for Software Contained in Medical Devices, 05/11/2005, (<http://www.fda.gov/cdrh/ode/guidance/337.pdf>)

General Principles of Software Validation, 01/11/2002, (<http://www.fda.gov/cdrh/comp/guidance/938.pdf>)

**L. Test Principle:**

The Focus Diagnostics Plexus™ EBV IgG Multi-Analyte Diagnostic is a three step procedure.

1. Patient sera are diluted and incubated with multiplexed bead suspension consisting of a mixture of distinguishable sets of polystyrene beads. Conjugated to the primary set of beads are the following EBV antigens; EA-D, EBNA- 1 and VCA. The bead mix also contains one background bead set designed to check for non-specific reactivity in the patient sample. If EBV antibodies are present, then the

antibodies bind to the corresponding antigen beads. The beads are rinsed to remove non-reactive serum proteins.

2. Phycoerythrin (PE)-conjugated goat anti human IgG, (Conjugate) is added, and the conjugate binds to the bound EBV antibody. The beads are rinsed to remove non-reactive conjugate.

3. Fluorescence from each distinct EBV antigen bead type is measured and compared against a Cutoff calibrator. The fluorescence is read in a plate reader, similar to an ELISA reader. Beads are first read with an orange laser to identify the bead type (e.g., EB-D, VCA, EBNA-1, or background), and then read with a green laser to the measure amount of fluorescence from the PE contained in the conjugate.

**M. Performance Characteristics (if/when applicable):**

1. Analytical performance:

a. *Precision/Reproducibility:*

The inter/intra-assay reproducibility and the inter-laboratory reproducibility testing were performed at three laboratories. Each of the three laboratories tested twelve samples in triplicate on five different days. The results of the study are summarized in the table below:

Plexus VCA IgG						Plexus EBNA IgG						Plexus EA IgG					
ID	Intra-assay & Inter-assay %CV			Inter-Lab		ID	Intra-assay & Inter-assay %CV			Inter-Lab		ID	Intra-assay & Inter-assay %CV			Inter-Lab	
	Mean Index	Intra-assay	Inter-assay	Mean Index	% CV		Mean Index	Intra-assay	Inter-assay	Mean Index	% CV		Mean Index	Intra-assay	Inter-assay	Mean Index	% CV
5	5.38	2.1%	8.1%	5.38	7.4%	12	6.66	2.1%	8.7%	6.66	8.9%	12	4.42	3.9%	7.5%	4.42	2.0%
19	5.12	2.9%	10.0%	5.12	10.1%	15	4.41	4.1%	6.9%	4.42	5.0%	4	2.69	3.6%	19.9%	2.69	7.0%
20	5.02	2.4%	8.4%	5.03	8.1%	8	3.66	2.5%	6.7%	3.66	5.8%	16	1.88	4.7%	7.5%	1.88	4.7%
6	4.21	3.2%	6.7%	4.20	3.7%	4	2.68	5.6%	18.3%	2.67	7.8%	15	1.39	5.6%	15.6%	1.39	15.7%
4	3.37	4.3%	16.6%	3.37	2.5%	18	2.05	3.8%	9.0%	2.05	8.6%	19	0.99	6.0%	12.0%	0.99	7.9%
16	2.21	4.1%	28.4%	2.22	27.0%	5	1.84	3.7%	8.0%	1.84	4.8%	8	0.91	3.7%	8.6%	0.91	2.1%
18	2.11	4.0%	6.0%	2.11	3.7%	6	1.47	5.1%	11.8%	1.46	11.7%	5	0.76	4.7%	12.6%	0.76	5.6%
2	2.01	3.9%	7.9%	2.01	4.4%	2	1.00	4.0%	13.4%	1.00	13.6%	20	0.63	5.3%	8.5%	0.63	2.6%
11	1.16	6.3%	13.3%	1.16	11.9%	3	0.81	6.1%	16.0%	0.81	11.9%	18	0.32	5.3%	9.6%	0.32	6.6%
8	1.11	4.7%	77.0%	1.12	31.1%	16	0.77	6.1%	59.1%	0.77	50.9%	6	0.29	6.1%	33.8%	0.29	4.7%
15	0.38	8.7%	19.6%	0.38	13.4%	20	0.65	5.4%	11.1%	0.65	8.9%	2	0.21	5.2%	61.9%	0.21	12.4%
1	0.17	18.2%	69.2%	0.17	59.3%	9	0.15	9.8%	304.1%	0.15	134%	3	0.21	8.2%	64.6%	0.21	10.1%
9	0.11	15.2%	117.9%	0.11	72.5%	19	0.11	7.8%	23.8%	0.11	19.8%	11	0.18	6.6%	106.1%	0.18	27.0%
3	1.71	5.4%	11.4%	1.71	5.8%	1	0.07	14.0%	41.1%	0.07	34.7%	1	0.15	9.1%	96.5%	0.15	16.9%
12	4.69	2.6%	7.7%	4.69	5.4%	11	0.04	11.2%	32.9%	0.04	23.1%	9	0.10	8.0%	40.1%	0.10	28.1%

The inter-lot reproducibility was evaluated with fifteen (15) samples in triplicates on three (3) lots of Plexus EBV kit. The results of the study are summarized in the table below:

Plexus VCA IgG			Plexus EBNA IgG			Plexus EA-D IgG		
ID	Mean	%CV	ID	Mean	%CV	ID	Mean	%CV
5	6.00	3.2%	12	7.35	3.6%	12	4.51	5.3%
19	5.88	2.3%	15	4.76	4.6%	4	2.86	3.3%
20	5.57	2.4%	8	4.01	4.5%	16	1.84	2.0%
12	5.08	3.7%	4	2.62	4.8%	15	1.36	9.0%
6	4.66	3.3%	18	2.39	6.7%	19	1.00	4.6%
4	3.61	1.3%	5	1.66	3.5%	8	0.85	7.2%
18	2.08	3.7%	6	1.44	5.8%	5	0.68	4.8%
2	2.01	2.4%	2	0.93	5.0%	20	0.59	2.3%
16	1.77	4.8%	3	0.74	2.3%	18	0.30	8.6%
3	1.72	4.0%	20	0.58	3.5%	6	0.29	4.2%
11	1.00	3.8%	16	0.36	3.2%	2	0.21	8.3%
8	0.75	2.8%	19	0.10	7.7%	3	0.20	5.8%
15	0.31	7.8%	1	0.06	5.7%	11	0.18	14.4%
1	0.11	7.7%	11	0.04	8.1%	1	0.15	10.8%
9	0.02	0.0%	9	0.02	30.0%	9	0.11	6.4%

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:*

The sponsor evaluated the potential cross-reactivity of the assay as follows: samples positive for similar disease states and potentially cross-reactive factors as determined by an FDA cleared device were tested with the Plexus EBV kit for each of the three (VCA, EA-D and EBNA-1) IgG analytes. Additionally 41 samples negative for EBV by an FDA cleared device and positive for some of possible cross reacting agents were included. The panel consisted of (ANA= 28, CMV= 31, HSV-1= 29, HSV-2=13, HSV-6= 4, Rubella Virus=1, Mumps = 1, Rubella Virus=39, *Toxoplasma gondii* =19, and VZA= 35). Because of the high prevalence of EBV IgG antibodies in the normal population, the test samples were also evaluated on commercially available ELISA. The majority of all samples that did elicit a positive result were also confirmed positive by the corresponding commercially available

ELISA, indicating reactivity to EBV IgG antibodies rather than cross reactivity with a potentially interfering factor. ANA showed possible cross reactivity with the EBNA-1 analyte, and HSV2 showed possible cross reactivity with the VCA analyte.

Potential cross reactivity with *E. coli* and *Pichia pastoris* which are the recombinant vector for the EAD and EBNA-1 antigens used in the assay was not assessed, due to difficulties in obtaining the appropriate samples.

Cross-Reactivity											
Cross Reactives	N	Method	EBV VCA IgG			EBV EBNA IgG			EBV EAD IgG		
			Positive	Equivocal	Negative	Positive	Equivocal	Negative	Positive	Equivocal	Negative
ANA	28	Plexus	27	0	1	28	0	0	8	0	20
		ELISA	28	0	0	28	0	0	5	5	18
		Discrepant	1			0			6 <sup>5</sup>		
Cytomegalovirus (CMV)	31	Plexus	24	1	6	24	0	7	4	0	27
		ELISA	26	0	5	25	0	6	3	1	27
		Discrepant	2 <sup>1</sup>			1			2 <sup>1</sup>		
HSV-1	29	Plexus	27	1	1	26	0	2	3	1	25
		ELISA	27	0	2	28	1	1	3	0	26
		Discrepant	2 <sup>1</sup>			2 <sup>1</sup>			1 <sup>1</sup>		
HSV-2	13	Plexus	13	0	0	12	0	1	2	3	8
		ELISA	13	0	0	13	0	0	2	1	10
		Discrepant	0			1			4 <sup>4</sup>		
HHV-6	4	Plexus	0	0	4	0	0	4	0	0	4
		ELISA	0	0	4	0	0	4	0	0	4
		Discrepant	0			0			0		
Measles (Rubeola)	1	Plexus	0	0	1	0	0	1	0	0	1
		ELISA	0	0	1	0	0	1	0	0	1
		Discrepant	0			0			0		
Mumps	1	Plexus	0	0	1	0	0	1	0	0	1
		ELISA	0	0	1	0	0	1	0	0	1
		Discrepant	0			0			0		
Rubella Virus	39	Plexus	26	0	13	25	1	13	4	1	34
		ELISA	26	1	12	27	0	12	3	2	34
		Discrepant	8 <sup>1</sup>			2 <sup>1</sup>			1 <sup>1</sup>		
<i>Toxoplasma gondii</i>	19	Plexus	17	0	2	18	0	1	1	4	14
		ELISA	19	0	0	18	0	1	1	2	16
		Discrepant	2			0			4 <sup>4</sup>		
Varicella-zoster (VZV)	35	Plexus	26	0	9	22	0	13	4	1	30
		ELISA	24	0	11	22	0	13	4	2	29
		Discrepant	4			0			4 <sup>3</sup>		

<sup>1</sup>One Equivocal Sample; <sup>2</sup>Two Equivocal Samples; <sup>3</sup>Three Equivocal Samples; <sup>4</sup>Four Equivocal Samples; <sup>5</sup>Five Equivocal Samples

The sponsor assessed the test's performance with potentially interfering substances by spiking four samples with two levels of four different potentially interfering substances. Four samples, two positive and two negative for EBV IgG antibodies by Plexus EBV IgG were used in the study. Baseline levels for triglycerides, albumin, bilirubin, and hemoglobin were established for each sample. The remaining serum was spiked with purchased interfering substances at levels that exceeded the expected human range. The spiked samples were tested again in the assay to determine if the elevated levels of interfering substances affected the assay. It was thus concluded that no interference was observed for any of the interfering substances in either the positive or negative sample.

*f. Assay cut-off:*

Establishment of the cutoff values for the EBV IgG and IgM Plexus was performed using 585 patient serum samples submitted for EBV testing. These samples were first tested on the predicate devices (Diamedix ELISA for VCA IgG, VCA IgM, EA, EBNA, and the Accutest for infectious mononucleosis). Each sample was classified as positive, negative or equivocal for each of these assays. The serum samples were then run on the EBV IgG and IgM Plexus assay. Comparisons were made for each analyte with its respective predicate test (excluding equivocal samples on the predicated device) on a Receiver Operating Characteristics (ROC) analysis. Based on the ROC analysis graphs a cutoff value was obtained.

<b>Plexus™ EBV IgG Multi-Analyte Diagnostics (MP0500G)</b>					
Antigen	Positive With predicate Device	% Positive Agreement (sensitivity)	Negative With predicate Device	% Negative Agreement (specificity)	Cutoff score
EBNA	359	96.1% 345/359	219	100% 219/219	0.301
EA	130	82.3% 107/130	409	94.6% 387/409	0.874
VCA IgG	417	95.7% 399/417	150	68% 102/150	0.600

2. Comparison studies:

*a. Method comparison with reference method:*

Performance of the Plexus EBNA-1 IgG and Plexus EA-D IgG analytes was compared to the Diamedix EBNA-1 and EA-D ELISA tests. The performance of the Plexus EBV VCA IgG analyte was compared to a consensus comparator consisting of the predicate device the Diamedix VCA IgG ELISA and 2 other devices; Athena Multi-Lyte EBV IgG Test System and Focus

EBV-VCA Antibody (IgG) IFA. For each sample, a consensus based algorithm (2/3) was used to determine the predicate result for comparison with the Plexus VCA IgG result. The studies were conducted at three United States testing sites: a hospital laboratory located in Northeast (n = 350), a pediatric hospital laboratory located in the Mid-West (n=249), and Focus (n=124) with serum samples in which EBV tests were ordered. The sera were sequentially submitted to the laboratory, archived, and masked. Samples were collected at three sites and include both prospective (n = 723) and retrospective (n = 150) specimens all tested by the Mid-West investigator. Retrospective samples were pre-selected based on EBV VCA IgM positive results from a FDA cleared device. Results are summarized per analyte, broken by the serological classification.

Serological status was determined by the use of the Diamedix ELISA assays for the EBV analytes EBNA-1 IgG, VCA IgG, EA-D IgG and VCA IgM and a commercially available heterophile rapid test for the heterophile antibody, and following a generally accepted classification chart.

**EBV VCA IgG vs. Consensus Predicate: Comparison by Serological Status (Prospective Population Samples N = 723)**

EBV VCA IgG Results							
Serostatus by Predicates		Consensus Predicate		Plexus			% Agreement
			n	Positive	Equivocal	Negative	
Acute	Primary Acute	Positive	57	57	0	0	100%(57/57), 95% CI:93.7-100%
		Negative	1	0	0	1	33.3%(1/3, 95% CI:6.1-79.2%
		No consensus <sup>1</sup>	2	2	0	0	NA
	Late Acute	Positive	72	70	1	1	97.2%(70/72), 95% CI:90.4-99.2%
		Negative	0	0	0	0	NA
		No consensus	0	0	0	0	NA
Recovering	Positive	1	1	0	0	100%(1/1), 95% CI:20.7-100%	
	Negative	0	0	0	0	NA	
	No consensus	0	0	0	0	NA	
Previous Infection	Positive	292	282	1	9	96.6%(282/292), 95% CI:93.8-98.1%	
	Negative	6	1	0	5	83.3%(5/6), 95% CI:43.6-97%	
	No consensus	0	0	0	0	NA	

EBV VCA IgG Results						
Serostatus by Predicates	Consensus Predicate		Plexus			% Agreement
		n	Positive	Equivocal	Negative	
No Infection	Positive	9	3	0	6	30.0%(3/10), 95% CI:10.8-60.3%
	Negative	217	13	1	203	93.5%(203/217), 95% CI:89.5-96.1%
	No consensus <sup>1</sup>	1	0	0	1	NA
Indeterminate	Positive	50	49	0	1	98%(49/50), 95% CI:89.5-99.6%
	Negative	15	0	0	15	100%(15/15), 95% CI:79.6-100%
	No consensus	0	0	0	0	NA

<sup>1</sup> No consensus results: the combination of three predicates could not yield a conclusive result for these samples – a 2/3 majority could not be obtained.

### EBV EBNA-1 vs. Predicate: Comparison by Serological Status (Prospective Population Samples N = 723)

EBV EBNA-1 IgG Results							
Serological Status by Predicates	Predicate ELISA		Plexus			% Agreement	
		n	Positive	Equivocal	Negative		
Acute	Primary Acute	Positive	0	0	0	0	NA
		Equivocal	0	0	0	0	NA
		Negative	60	0	0	60	100%(60/60), 95% CI:94-100%
	Late Acute	Positive	69	65	0	4	94.2%(65/69), 95% CI:86-97.7%
		Equivocal	0	0	0	0	NA
		Negative	3	0	0	3	100%(3/3), 95% CI:43.8-100%
Recovering	Positive	0	0	0	0	NA	
	Equivocal	0	0	0	0	NA	
	Negative	1	0	0	1	100%(1/1), 95% CI:20.7-100%	
Previous Infection	Positive	284	266	2	16	93.7%(266/284), 95% CI:90.2-96%	
	Equivocal	0	0	0	0	NA	
	Negative	14	1	0	13	92.9%(13/14), 95% CI:68.5-98.7%	

EBV EBNA-1 IgG Results						
Serological Status by Predicates	Predicate ELISA		Plexus			% Agreement
		n	Positive	Equivocal	Negative	
No Infection	Positive	0	0	0	0	NA
	Equivocal	0	0	0	0	NA
	Negative	227	1	0	226	99.6%(226/227), 95% CI:97.5-99.9%
Indeterminate	Positive	36	29	0	7	76.3%(29/38), 95% CI:60.8-87%
	Equivocal	2	0	0	2	0%(0/2), 95% CI:0-65.8%
	Negative	27	0	0	27	100%(27/27), 95% CI:87.5-100%

**EBV EA-D vs. Predicate: Comparison by Serological Status (Prospective Population Samples N = 723)**

EA-D IgG Results							
Serological Status by Predicates	Predicate ELISA		Plexus			% Agreement	
		n	Positive	Equivocal	Negative		
Acute	Primary Acute	Positive	43	40	0	3	93%(40/43), 95% CI:81.4-97.6%
		Equivocal	0	0	0	0	NA
		Negative	17	4	0	13	76.5%(13/17), 95% CI:52.7-90.4%
	Late Acute	Positive	51	41	1	9	80.4%(41/51), 95% CI:67.5-89%
		Equivocal	0	0	0	0	NA
		Negative	21	2	2	17	81%(17/21), 95% CI:60-92.3%
Recovering	Positive	1	0	0	1	0%(0/1), 95% CI:0-79.3%	
	Equivocal	0	0	0	0	NA	
	Negative	0	0	0	0	NA	
Previous Infection	Positive	0	0	0	0	NA	
	Equivocal	0	0	0	0	NA	
	Negative	298	11	1	286	96%(286/298), 95% CI:93.1-97.7%	

EA-D IgG Results							
		Predicate ELISA		Plexus			
Serological Status by Predicates			n	Positive	Equivocal	Negative	% Agreement
No Infection	Positive		0	0	0	0	NA
	Equivocal		0	0	0	0	NA
	Negative		227	1	3	223	98.2%(223/227), 95% CI:95.6-99.3%
Indeterminate	Positive		12	7	0	5	26.9%(7/26), 95% CI:13.7-46.1%
	Equivocal		30	14	2	14	6.3%(2/32), 95% CI:1.7-20.1%
	Negative		23	0	0	23	62.2%(23/37), 95% CI:46.1-75.9%

**EBV VCA IgG vs. Consensus Predicate: Comparison by Serological Status (Retrospective Presumed Acute Population Samples N = 150)**

EBV VCA IgG Results							
		Consensus Predicate		Plexus			
Serostatus by Predicates			n	Positive	Equivocal	Negative	% Agreement
Acute	Primary Acute	Positive	106	99	1	6	93.4%(99/106), 95% CI:87-96.8%
		Negative	0	0	0	0	NA
		No consensus	0	0	0	0	NA
	Late Acute	Positive	8	8	0	0	100%(8/8), 95% CI:67.6-100%
		Negative	0	0	0	0	NA
		No consensus	0	0	0	0	NA
No Infection	Positive	1	1	0	0	100%(1/1), 95% CI:20.7-100%	
	Negative	1	1	0	0	0%(0/1), 95% CI:0-79.3%	
	No consensus	0	0	0	0	NA	
Indeterminate	Positive	33	31	0	2	93.9%(31/33), 95% CI:80.4-98.3%	
	Negative	0	0	0	0	NA	
	No consensus <sup>1</sup>	1	1	0	0	NA	

<sup>1</sup> No consensus results: the combination of three predicates could not yield a conclusive result for these samples – a 2/3 majority could not be obtained.

**EBV EBNA-1 vs. Predicate: Comparison by Serological Status (Retrospective Presumed Acute Population Samples N = 150)**

EBV EBNA-1 IgG Results							
Serological Status by Predicates		Predicate ELISA		Plexus			% Agreement
			n	Positive	Equivocal	Negative	
Acute	Primary Acute	Positive	0	0	0	0	NA
		Equivocal	0	0	0	0	NA
		Negative	106	1	0	105	99.1%(105/106), 95% CI:94.8-99.8%
	Late Acute	Positive	4	0	0	4	0%(0/4), 95% CI:0-49%
		Equivocal	0	0	0	0	NA
		Negative	4	0	0	4	100%(4/4), 95% CI:51-100%
No Infection	Positive	0	0	0	0	NA	
	Equivocal	0	0	0	0	NA	
	Negative	2	0	0	2	100%(2/2), 95% CI:34.2-100%	
Indeterminate	Positive	4	0	0	4	0%(0/8), 95% CI:0-32.4%	
	Equivocal	4	0	0	4	0%(0/4), 95% CI:0-49.0%	
	Negative	26	0	0	26	100%(26/26), 95% CI:87.1-100%	

**EBV EA-D vs. Predicate: Comparison by Serological Status (Retrospective Presumed Acute Population Samples N = 150)**

EBV EA-D IgG Results							
Serological Status by Predicates		Predicate ELISA		Plexus			% Agreement
			n	Positive	Equivocal	Negative	
Acute	Primary Acute	Positive	61	57	1	3	93.4%(57/61), 95% CI:84.3-97.4%
		Equivocal	0	0	0	0	NA
		Negative	45	7	3	35	77.8%(35/45), 95% CI:63.7-87.5%
	Late Acute	Positive	3	3	0	0	100%(3/3), 95% CI:43.8-100%
		Equivocal	0	0	0	0	NA
		Negative	5	1	0	4	80%(4/5), 95% CI:37.6-96.4%

EBV EA-D IgG Results						
Serological Status by Predicates	Predicate ELISA		Plexus			% Agreement
		n	Positive	Equivocal	Negative	
No Infection	Positive	0	0	0	0	NA
	Equivocal	0	0	0	0	NA
	Negative	2	0	0	2	100%(2/2), 95% CI:34.2-100%
Indeterminate	Positive	10	8	0	2	66.7%(8/12), 95% CI:39.1-86.2%
	Equivocal	14	12	0	2	0%(0/14), 95% CI:0-21.5%
	Negative	10	0	1	9	40.9%(9/22), 95% CI:23.3-61.3%

b. *Matrix comparison:*

Not Applicable

3. Clinical studies:

a. *Clinical Sensitivity:*

Not Applicable

b. *Clinical specificity:*

Not Applicable

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

Not Applicable

4. Clinical cut-off:

See 1 f

5. Expected values/Reference range:

Expected values for the EBV IgG kit are presented by age and gender in the following tables for (723) serum samples from patients for which EBV tests were ordered.

For all analytes, index values of <0.90 are negative,  $\geq 0.90$  to  $\leq 1.10$  are equivocal and > 1.10 are positives.

**Expected Values Plexus VCA IgG:**

Age	Gender	Positive		Equivocal		Negative		Total
		n	%	n	%	n	%	
<5	F	8	53.3	0	0.0	7	46.7	15
<5	M	17	47.2	0	0.0	19	52.8	36
5-12	F	43	43.0	1	1.0	56	56.0	100
5-12	M	46	48.4	0	0.0	49	51.6	95
13-20	F	118	70.7	0	0.0	49	29.3	167
13-20	M	82	63.1	1	0.8	47	36.2	130
21-30	F	37	97.4	0	0.0	1	2.6	38
21-30	M	15	83.3	0	0.0	3	16.7	18
31-40	F	14	87.5	0	0.0	2	12.5	16
31-40	M	13	92.9	0	0.0	1	7.1	14
41-50	F	19	100	0	0.0	0	0.0	19
41-50	M	12	92.3	0	0.0	1	7.7	13
51-60	F	15	93.8	0	0.0	1	6.3	16
51-60	M	8	72.7	0	0.0	3	27.3	11
61-70	F	9	100	0	0.0	0	0.0	9
61-70	M	8	80.0	0	0.0	2	20.0	10
>70	F	7	100	0	0.0	0	0.0	7
>70	M	7	77.8	1	11.1	1	11.1	9
Total		478	66.1	3	0.4	242	33.5	723

**Expected Values Plexus EBNA-1 IgG:**

Age	Gender	Positive		Equivocal		Negative		Total
		n	%	n	%	n	%	
<5	F	3	20.0	0	0.0	12	80.0	15
<5	M	12	33.3	0	0.0	24	66.7	36
5-12	F	28	28.0	0	0.0	72	72.0	100
5-12	M	36	37.9	1	1.1	58	61.1	95
13-20	F	85	50.9	0	0.0	82	49.1	167
13-20	M	52	40.0	0	0.0	78	60.0	130
21-30	F	27	71.1	0	0.0	11	28.9	38
21-30	M	10	55.6	0	0.0	8	44.4	18
31-40	F	15	93.8	0	0.0	1	6.3	16

Age	Gender	Positive		Equivocal		Negative		Total
		n	%	n	%	n	%	
31-40	M	11	78.6	0	0.0	3	21.4	14
41-50	F	17	89.5	0	0.0	2	10.5	19
41-50	M	10	76.9	0	0.0	3	23.1	13
51-60	F	16	100.0	0	0.0	0	0.0	16
51-60	M	10	90.9	0	0.0	1	9.1	11
61-70	F	9	100.0	0	0.0	0	0.0	9
61-70	M	8	80.0	0	0.0	2	20.0	10
>70	F	6	85.7	0	0.0	1	14.3	7
>70	M	7	77.8	1	11.1	1	11.1	9
Total		362	50.1	2	0.3	359	49.7	723

#### Expected Values Plexus EA-D IgG:

Age	Gender	Positive		Equivocal		Negative		Total
		n	%	n	%	n	%	
<5	F	1	6.7	1	6.7	13	86.7	15
<5	M	4	11.1	0	0.0	32	88.9	36
5-12	F	11	11.0	0	0.0	89	89.0	100
5-12	M	9	9.5	1	1.1	85	89.5	95
13-20	F	27	16.2	2	1.2	138	82.6	167
13-20	M	25	19.2	2	1.5	103	79.2	130
21-30	F	9	23.7	0	0.0	29	76.3	38
21-30	M	5	27.8	0	0.0	13	72.2	18
31-40	F	5	31.3	0	0.0	11	68.8	16
31-40	M	2	14.3	0	0.0	12	85.7	14
41-50	F	5	26.3	1	5.3	13	68.4	19
41-50	M	1	7.7	1	7.7	11	84.6	13
51-60	F	3	18.8	0	0.0	13	81.3	16
51-60	M	3	27.3	1	9.1	7	63.6	11
61-70	F	4	44.4	0	0.0	5	55.6	9
61-70	M	3	30.0	0	0.0	7	70.0	10
>70	F	3	42.9	0	0.0	4	57.1	7
>70	M	0	0.0	0	0.0	9	100.0	9
Total		120	16.6%	9	1.2%	594	82.2%	723

**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.