510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT

I Background Information:

A 510(k) Number

K243485

B Applicant

Abbott Molecular Inc.

C Proprietary and Established Names

Alinity m CMV

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
PAB	Class II	21 CFR 866.3180 - Quantitative Cytomegalovirus Nucleic Acid Tests For Transplant Patient Management	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain clearance for the modification of a previously approved device, Alinity m CMV assay, which allows the use of an alternative DNA polymerase.

B Measurand:

Cytomegalovirus (CMV) DNA.

C Type of Test:

Quantitative in vitro nucleic acid amplification test.

III Intended Use/Indications for Use:

A Intended Use(s):

The Alinity m CMV assay is an in vitro polymerase chain reaction (PCR) assay for use with the automated Alinity m System to quantitate cytomegalovirus (CMV) DNA in human EDTA plasma. The Alinity m CMV assay is intended for use as an aid in the management of Hematopoietic Stem Cell Transplant and Solid Organ Transplant patients who are undergoing anti-cytomegalovirus therapy. The Alinity m CMV assay can be used to assess virological response to anti-cytomegalovirus therapy.

The results from the Alinity m CMV test must be interpreted within the context of all relevant clinical and laboratory findings. The Alinity m CMV test is not intedned as a screening test for the presence of CMV DNA in blood or blood products.

B Indication(s) for Use:

See Intended Use above.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

For use with the Alinity m System

IV Device/System Characteristics:

A Device Description:

The Alinity m CMV assay is an in vitro polymerase chain reaction (PCR) assay for use with the automated Alinity m System to quantitate cytomegalovirus (CMV) DNA in human EDTA plasma. The Alinity m CMV assay is intended for use as an aid in the management of Hematopoietic Stem Cell Transplant (HSCT) and Solid Organ Transplant (SOT) patients who are undergoing anti-cytomegalovirus therapy. The Alinity m CMV assay can be used to assess virological response to anti-cytomegalovirus therapy.

The Alinity m CMV assay requires 3 separate assay specific kits:

- Alinity m CMV AMP Kit (List No. 09N46-095) consisting of 2 types of multi-well assay trays. The amplification trays (AMP TRAY 1) contain lyophilized, unit-dose PCR amplification/detection reagents and lyophilized, unit-dose internal control (IC) with proteinase K in separate wells, and the activation trays (ACT TRAY 2) contain liquid unit-dose activation reagent. The intended storage condition for the Alinity m CMV AMP Kit is 2°C to 8°C.
- Alinity m CMV CTRL Kit (List No. 09N46-085) consisting of negative controls, low-positive controls and high-positive controls, each supplied as liquid in single-use tubes. The intended storage condition for the Alinity m CMV CTRL Kit is – 25°C to –15°C.

- Alinity m CMV CAL Kit (List No. 09N46-075) consisting of 2 calibrator levels, each supplied as liquid in single-use tubes. The intended storage condition for the Alinity m CMV CAL Kit is – 25°C to – 15°C.

B Principle of Operation:

The Alinity m CMV assay utilizes real-time polymerase chain reaction (PCR) to amplify and detect CMV genomic DNA sequences that have been extracted from human plasma specimens. The steps of the Alinity m CMV assay consist of sample preparation, PCR assembly, amplification/detection, and result calculation and reporting. All steps of the Alinity m CMV assay procedure are executed automatically by the Alinity m System. Manual dilutions may be performed for low-volume specimens to meet the minimum volume requirement.

The Alinity m System is designed to be a random access analyzer that can perform the Alinity m CMV assay in parallel with other Alinity m assays on the same instrument.

CMV DNA from human plasma is extracted using the Alinity m Sample Prep Kit 2, Proteinase K, Alinity m Lysis Solution, and Alinity m Diluent Solution. The Alinity m System employs magnetic microparticle technology to facilitate nucleic acid capture, wash, and elution. The resulting purified DNA is then combined with liquid unit-dose Alinity m CMV activation reagent and lyophilized unit-dose Alinity m CMV amplification/detection reagents and transferred into a reaction vessel. Alinity m Vapor Barrier Solution is then added to the reaction vessel which is then transferred to an amplification/detection unit for PCR amplification, and real-time fluorescence detection of CMV.

At the beginning of the Alinity m CMV sample preparation process, a lyophilized unit-dose internal control (IC) on the AMP Tray is rehydrated by the Alinity m System and delivered into each sample preparation reaction. The IC is then processed through the entire sample preparation and real-time PCR procedure along with the specimens, calibrators, and controls to demonstrate proper sample processing and validity. The Alinity m CMV amplification/detection reagents consist of enzymes, primers, probes, and activation reagents that enable polymerization, and detection.

A CMV calibration curve is required for determination of CMV DNA concentration. Two levels of calibrators are processed through sample preparation and PCR to generate the calibration curve. The concentration of CMV DNA in specimens and controls is then calculated from the stored calibration curve.

Assay controls are tested at or above an established minimum frequency to help ensure that instrument and reagent performance remains satisfactory. During each control event, a negative control, a low positive control, and a high positive control are processed through sample preparation and PCR procedures that are identical to those used for specimens.

Calculation

Quantitative viral load results are reported for patient specimens with CMV viral concentrations within the assay's quantitation range. The concentration of CMV DNA in a specimen is calculated from the calibration curve by the system software. The Alinity m System reports the results in International Units as IU/mL or Log [IU/mL].

Interpretation of results

Result	Interpretation	Interpretation Additional Information
Not Detected	CMV DNA not detected	
<LLoQ	CMV DNA detected but not quantified	CMV DNA concentration is below the Lower Limit of Quantitation (LLoQ) of the assay
LLoQ to ≤ULoQ	CMV DNA detected and quantified	CMV DNA concentration is within the linear range of the assay (≥LLoQ to ≤ULoQ)
>ULoQ	CMV DNA detected	CMV DNA concentration is above the Upper Limit of Quantification (ULoQ) of the assay.

C Instrument Description Information:

1. Instrument Name:

   Alinity m System

2. Specimen Identification:

   Cytomegalovirus DNA in Human EDTA plasma

3. Specimen Sampling and Handling:

   Plasma specimens may be tested for viral load determination. For the Alinity m CMV assay, only use collection tubes as described: K_2 EDTA, K_3 EDTA, Plasma Preparation Tube (PPT)

4. Calibration:

   Alinity m CMV CAL Kit consisting of 2 calibrator levels, each supplied as liquid in single-use tubes.

5. Quality Control:

   Alinity m CMV CTRL Kit consisting of negative controls, low-positive controls, and high-positive controls, each supplied as liquid in single-use tubes.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Alinity m CMV

B Predicate 510(k) Number(s):
P210022

C Comparison with Predicate(s):

Device & Predicate Device(s):	P210022	K243485
Device Trade Name	Alinity m CMV	Same
Regulation No./Product Code	21 CFR 814 / PAB	21 CFR 866.3180 / PAB
Device Class	Class III	Class II
General Device Characteristic Similarities		
Intended Use/Indications For Use	The Alinity m CMV assay is an in vitro polymerase chain reaction (PCR) assay for use with the automated Alinity m System quantitate cytomegalovirus (CMV) DNA in human EDTA plasma. The Alinity m CMV assay is intended for use as an aid in the management of Hematopoietic Stem Cell Transplant and Solid Organ Transplant patients who are undergoing anti-cytomegalovirus therapy. The Alinity m CMV assay can be used to assess virological response to anti-cytomegalovirus therapy. The results from the Alinity m CMV test must be interpreted within the context of all relevant clinical and laboratory findings. The Alinity m CMV test is not intended as a screening test for the presence of CMV DNA in blood or blood products.	Same
Assay Type	Quantitative	Same
Assay Targets	Conserved regions of CMV genome (UL34 and UL80.5)	Same
Specimen Types	EDTA Plasma	Same
Specimen Collection and Transport	K_2 EDTA, K_3 EDTA, Plasma Preparation Tube (PPT) Plasma may be stored in primary tubes (with or without gel) and secondary tubes	Same
Principles of the Procedure	See above for description of principles of procedure	Same

Instrument and System Components	Alinity m System: Fully integrated and automated diagnostics analyzer which utilizes real-time PCR technology	Same
Sample Preparation Instrument Components	Automated liquid handling and robotic manipulation platform	Same
Assay Controls	• Negative Control • Low Positive Control • High Positive Control • Internal Control (IC)	Same
Results Reporting	• Not Detected • Detected < LLoQ • Detected > ULoQ • Concentration from LLoQ to ≤ ULoQ, reported in IU/mL or Log IU/mL For low volume specimens tested with the Specimen Dilution Procedure, quantitative results generated on the Alinity m System represent the CMV DNA concentration in the specimen prior to dilution.	Same
Reagents and Storage Conditions	• Alinity m CMV AMP Kit: 2°C to 8°C • Alinity m CMV CTRL Kit: –25°C to –15°C • Alinity m CMV CAL Kit: –25°C to –15°C	Same
General Device Characteristic Differences		
Amplification Enzymes	Original DNA Polymerase is the enzyme used for DNA amplification.	Original DNA Polymerase or alternative DNA Polymerase is the enzyme used for DNA amplification.

VI Standards/Guidance Documents Referenced:

Special Control (89 FR 77448), labeling requirements under 21 CFR 809.10(b), the information for special controls pertaining to cytomegalovirus (CMV) quantitative nucleic acid test intended for transplant patient management.

CLSI EP05-A3: Clinical Laboratory Standards Institute. *Evaluation of the Precision of Quantitative Measurement Procedures; Approved Guideline* – Third Edition. CLSI Guideline EP05-A3, CLSI: Wayne, PA, 2014.

CLSI EP35: Clinical Laboratory Standards Institute. *Assessment of Equivalence or Suitability of Specimen Types for Medical Laboratory Measurement Procedures; Approved Guideline* – First Edition. CLSI Guideline EP35. CLSI: Wayne, PA: 2019.

CLSI EP09c: Clinical and Laboratory Standards Institute. *Measurement Procedure Comparison and Bias Estimation Using Patient Sample* – Third Edition. CLSI Guideline EP09c. Wayne, PA: CLSI; 2018.

CLSI EP06: Clinical Laboratory Standards Institute. *Evaluation of Linearity of Quantitative Measurement Procedures; Approved Guideline* – Second Edition. CLSI Guideline EP06, CLSI: Wayne, PA, 2020.

CLSI EP17-A2: Clinical Laboratory Standards Institute. *Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline* – Second Edition. CLSI Guideline EP17-A2, CLSI: Wayne, PA, 2012.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Precision:

Precision of Alinity m CMV assay with the alternative DNA polymerase enzyme was determined by testing 8 panel members with CMV concentrations that span the targeted quantitation range of the assay (30 to 10^8 IU/mL [1.48 to 8.00 Log IU/mL]). Panel members with multiple targeted concentration levels were prepared with clinical sample (1.48 to 2.7 Log IU/mL), cultured virus (4.0 to 5.0 IU/mL) or plasmid DNA (7.0 to 8.0 IU/mL). The quantitation of the panel members was traceable to international standard material for Human Cytomegalovirus (CMV) DNA (code 09/162). Each panel member was tested in 3 replicates, twice each day for 15 days on 1 Alinity m System operated by 3 operators using 3 lots of CMV AMP kit, for a total of 270 replicates per panel member. The study design and analysis were derived from recommendations in CLSI Guideline EP05-A3 3rd Edition.

The precision study results are shown in **Table 1**.

Table 1. Precision Analysis (All lots)

Panel Member	N	Mean Conc. (Log IU/mL)	Within-Run		Between-Run		Between-Day		Within-Laboratory[a]		Between-Lot		Total[b]	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
01	270	1.52	0.26	17.2	0.06	3.7	0.00	0.0	0.27	17.6	0.02	1.3	0.27	17.6
02	270	1.70	0.16	9.6	0.02	0.9	0.03	1.5	0.17	9.7	0.01	0.4	0.17	9.8
03	270	2.13	0.21	9.8	0.05	2.4	0.00	0.0	0.22	10.1	0.00	0.2	0.22	10.1
04	270	2.83	0.05	1.8	0.02	0.9	0.00	0.0	0.06	2.0	0.00	0.1	0.06	2.0
05	270	4.04	0.04	1.0	0.03	0.7	0.00	0.0	0.05	1.3	0.02	0.4	0.05	1.3
06	270	5.03	0.03	0.7	0.03	0.5	0.00	0.0	0.04	0.8	0.01	0.3	0.04	0.9
07	270	7.19	0.04	0.5	0.02	0.3	0.00	0.0	0.04	0.6	0.02	0.3	0.05	0.7
08	270	8.15	0.05	0.6	0.00	0.0	0.01	0.1	0.05	0.6	0.03	0.3	0.05	0.7

[a] Within-Laboratory includes Within-Run, Between-Run, and Between-Day Components.

[b] Total includes Within-Run, Between-Run, Between-Day, and Between-Lot Components.

Reproducibility:
Reproducibility of Alinity m CMV assay with the alternative DNA polymerase enzyme was determined by testing 9-member reproducibility panel. Eight positive panel members with multiple targeted concentration levels were prepared with clinical sample (1.0 to 2.0 Log IU/mL), cultured virus (3.0 to 5.0 Log IU/mL), and or plasmid DNA (> 5.0 Log IU/mL). The quantitation of the panel members was traceable to international standard material for CMV DNA (code 09/162).

Each panel member was tested using 1 reagent lot, 1 lot of calibrators and controls, 3 external sites with 1 instrument per site, 4 replicates per panel per run, 5 non-consecutive days, 2 runs per day. The study design contained a total of 120 replicates across all runs for each panel member tested. The study design and analysis were derived from recommendations in CLSI Guideline EP05-A3 3rd Edition.

The reproducibility results are summarized in **Table 2**.

Table 2. Reproducibility for Positive Panel Members (Log IU/mL)

Panel Member	N	Mean Conc. (Log IU/mL)	Within-Run		Between-Run		Between-Day		Within-Laboratory[a]		Between-Site		Total[b]	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	117	7.87	0.05	0.7	0.02	0.3	0.00	0.0	0.06	0.7	0.09	1.1	0.11	1.4
2	117	6.85	0.05	0.7	0.01	0.2	0.01	0.2	0.05	0.7	0.07	1.0	0.08	1.2
3	119	5.11	0.05	1.0	0.00	0.0	0.01	0.3	0.05	1.1	0.03	0.5	0.06	1.2
4	119	4.03	0.04	1.0	0.02	0.4	0.00	0.1	0.04	1.1	0.04	1.0	0.06	1.5
5	117	3.09	0.06	1.9	0.02	0.7	0.01	0.2	0.06	2.1	0.05	1.7	0.08	2.7
6	118	1.87	0.16	8.5	0.03	1.7	0.00	0.0	0.16	8.7	0.09	4.9	0.19	10.0
7	116	1.44	0.25	17.4	0.11	7.5	0.00	0.0	0.27	19.0	0.13	8.7	0.30	20.9
8	111	1.20	0.32	26.6	0.10	8.2	0.00	0.0	0.33	27.9	0.17	14.4	0.38	31.4

[a] Within-Laboratory includes Within-Run, Between-Run, and Between-Day Variance Components.
[b] Total (Overall) includes Within-Run, Between-Run, Between-Day, and Between-Site Variance Components.

For the negative panel member the overall negative percent agreement with the expected result was 100.0% (119/119) **Table 3**. All three external sites had 100.0% negative percent agreement with the expected result.

Table 3. Reproducibility for Negative Panel Member

Panel Member	Site	Total N	Positive	Negative	Negative Rate (%)	95% CI
9	all	119	0	119	100%	(96.9-100%)

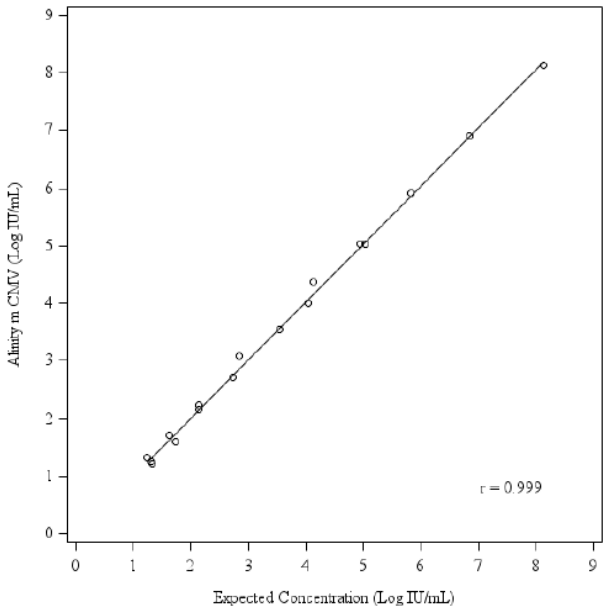
2. Linearity:

Linearity was evaluated by testing 17 panel members that spanned the intended linear range of the assay (30 to 100,000,000 IU/mL), including a panel member below the expected Lower Limit of Quantification (LLoQ) at 15 IU/mL (1.18 Log IU/mL), and a Panel member exceeding the expected Upper Limit of Quantification (ULoQ) at 200,000,000 IU/mL (8.30 Log IU/mL). Panel members with multiple concentration levels were prepared with clinical sample (1.18 to 3.4 Log IU/mL), cultured virus (1.3 to 5.0 Log IU/mL) and plasmid DNA

(1.4 to 8.3 Log IU/mL). The quantitation of the panel members was traceable to international standard material for CMV DNA (code 09/162). The study was conducted using 1 lot of Alinity m CMV on 1 instrument with a minimum of 27 replicates for each panel member. Linearity study design and evaluation was based on the recommendations in CLSI EP06 2nd Edition.

Alinity m CMV was linear across the quantitation range from 30 to 100,000,000 IU/mL (1.48 to 8.0 Log IU/mL). Representative results for Alinity m CMV linearity performance are shown in **Figure 1**.

Figure 1. Alinity m CMV Linearity



3. Analytical Specificity/Interference:

   Refer to P210022.

4. Assay Reportable Range:

   Refer to P210022. The analytical measuring interval of the Alinity m CMV for plasma is from the lower limit of quantitation (LLoQ) of 30 IU/mL (1.48 Log IU/mL) to the upper limit of quantitation (ULoQ) of 100,000,000 IU/mL (8.0 Log IU/mL).

5. Traceability and Stability:

   Traceability to international standard material:
   Refer to P210022.

   Reagent Stability:
   The data submitted support the following storage conditions for the Alinity m CMV assay with alternative DNA polymerase.

Table 4. Alinity m CMV AMP kit stability

Stability study	Expiration
Reagent On-board	30 days
Reagent Shelf-life	15 months

6. Detection Limit:

The Limit of Detection (LoD) was assessed by testing dilutions of the international standard material for CMV DNA (code 09/162) prepared in CMV-negative human plasma. The three concentration panel members (20.0 IU/mL, 30.0 IU/mL, and 50.0 IU/mL) were selected based on expected LoD of the on-market Alinity m CMV. Testing for each CMV DNA concentration was performed with 3 lots of reagents across multiple days. LoD Study design and analysis follow CLSI EP17-A2 and demonstrate a detection rate of ≥ 95.0% for samples at the claimed LoD of 30 IU/mL (1.48 Log IU/mL) and above.

7. Lower Limit of Quantitation (LLoQ):

LLoQ is defined as the lowest concentration at which CMV DNA is reliably quantitated within an acceptable total error. The LLoQ was evaluated based on the recommendation in CLSI EP17-A2, section 7. For each panel member from the CMV LoD study, Total Analytical Error (TAE) and Total Error in difference between two measurements (TE) were calculated for each lot and for all lots.

The results are presented in **Table 5** and **Table 6**.

Table 5. Alinity m CMV TAE and TE (overall)

Panel Member	Target concentration (IU/mL)	N	Target Concentration (Log IU/mL)	Mean (Log IU/mL)	Bias (Log IU/mL)	SD	TAE	TE
1	20	139	1.30	1.30	-0.00	0.31	0.62	0.88
2	30	139	1.48	1.42	-0.06	0.27	0.59	0.75
3	50	144	1.70	1.74	0.04	0.17	0.38	0.48

Table 6. Alinity m CMV TAE and TE (by Lot)

Panel Member	Target Concentration (IU/mL)	Lot	N	Target Concentration (Log IU/mL)	Mean concentration (Log IU/mL)	Bias (Log IU/mL)	SD	TAE	TE
1	20	1	46	1.30	1.30	0.00	0.26	0.52	0.74
		2	48	1.30	1.26	-0.04	0.36	0.76	1.01
		3	45	1.30	1.34	0.04	0.30	0.64	0.86
2	30	1	45	1.48	1.36	-0.12	0.29	0.71	0.83
		2	47	1.48	1.41	-0.07	0.27	0.62	0.77
		3	47	1.48	1.49	0.01	0.21	0.43	0.60
3	50	1	48	1.70	1.76	0.06	0.15	0.37	0.43
		2	48	1.70	1.76	0.06	0.19	0.44	0.54
		3	48	1.70	1.69	-0.01	0.16	0.33	0.46

The panel member with the target concentration at the assay's claimed LoQ of 30 IU/mL has a TAE and TE less than or equal to 1.00 Log IU/mL.

8. Accuracy (Instrument):

   Accuracy was reviewed in P210022.

9. Carry-Over:

   Carry-over was reviewed in P210022/S008.

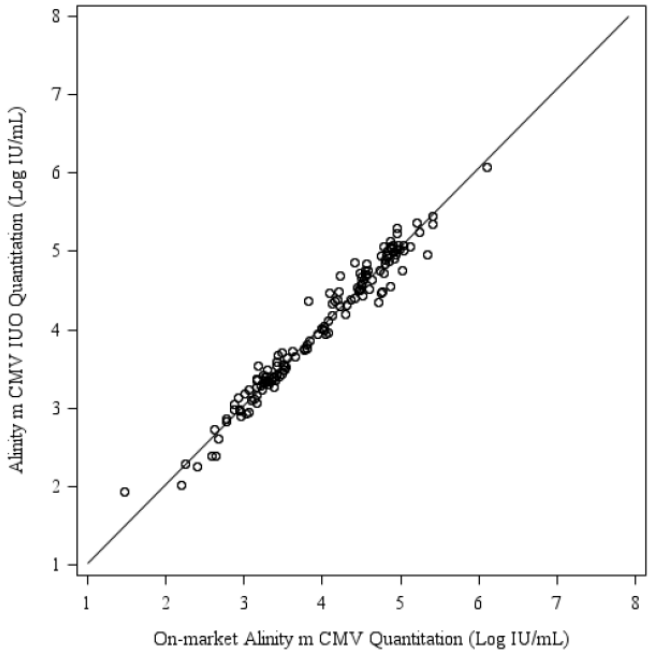
B Comparison Studies:

1. Method Comparison with Predicate Device:

   The Alinity m CMV clinical specimen method comparison testing was conducted at one testing site (internal testing site) using 3 reagent lots of the Alinity m CMV with alternative and 1 lot of the on-market Alinity m CMV. The specimens were obtained from hematopoietic stem cell transplant (HSCT) and solid organ transplant (SOT) recipients. Samples were selected such that CMV levels in the samples span the measuring range of the Alinity m CMV assay. To ensure coverage of the measuring range, both neat clinical specimens and samples made with unique individual high-titer clinical specimens spiked into unique individual negative specimens were included. All samples were tested with both the on-market Alinity m CMV assay with the original DNA polymerase and the Alinity m CMV assay with the alternative DNA polymerase.

   Figure 2 shows the Deming regression performed using the on-market Alinity m assay result as the independent variable and the respective Alinity m with alternative assay result as the dependent variable on 141 clinical samples. The Deming regression analysis results have correlation coefficient (r) of 0.983, a slope of 1.01 and intercept of 0.01, and the 95% confidence intervals (-0.13, 0.16) (**Figure 2**).

Figure 2. Deming Regression Plot – All data



Systematic Difference was calculated for selected viral load levels (2.70, 3.30, 4.00 Log IU/mL) along with the two-sided 95% confidence intervals (**Table 7**).

Table 7. Systematic Difference at Selected Viral Load Levels

Target Viral Load Levels	Systematic Difference	95% CI
2.70 Log IU/mL	0.04 Log IU/mL	(-0.01, 0.09)
3.30 Log IU/mL	0.05 Log IU/mL	(0.01, 0.08)
4.00 Log IU/mL	0.05 Log IU/mL	(0.03, 0.08)

The Alinity m CMV assay with alternative DNA polymerase has viral load values with a mean bias relative to the comparator assay of less than or equal to 0.25 Log IU/mL within the quantitation range.

2. Matrix Comparison:

N/A

C Clinical Studies:

1. Clinical Sensitivity:

N/A

2. Clinical Specificity:

N/A

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

N/A

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

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