510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number: K163569

B. Purpose for Submission: Adding a previously cleared assay on a new instrument platform

C. Measurand: IgM antibodies to CMV

D. Type of Test: Electrochemiluminescence immunoassay

E. Applicant: Roche Diagnostics, Inc.

F. Proprietary and Established Names: Elecsys CMV IgM

G. Regulatory Information:

1. Regulation section: 21 CFR 866.3175
2. Classification: Class II
3. Product code(s): LFZ – Cytomegalovirus serological reagents
   JJE – Discrete photometric chemistry analyzer for clinical use
4. Panel: Microbiology

H. Intended Use:

1. Intended use(s):

   Immunoassay for the in vitro qualitative detection of IgM antibodies to CMV in human serum, lithium‑heparin plasma, K2‑EDTA plasma, and K3‑EDTA plasma. The test is intended as an aid in the diagnosis of recent or current CMV infection in individuals for which a CMV IgM test was ordered, including pregnant women.

   Performance characteristics have not been evaluated in immunocompromised or immunosuppressed individuals. This test is not intended for use in neonatal screening or for use at point of care facilities. This test is not intended for use in screening blood and plasma donors.

   The electrochemiluminescence immunoassay "ECLIA" is intended for use on cobas e immunoassay analyzers.

2. Indication(s) for use:
   Same as intended use.

3. Special conditions for use statement(s):
   For prescription use

4. Special instrument requirements:
   cobas e 801 Immunoassay Analyzer

I. Device Description:

The Elecsys CMV IgM is a μ-capture immunoassay that uses streptavidin microparticles, biotinylated recombinant CMV-specific antigen labeled with a ruthenium complex, and electrochemiluminescence detection. The test system contains human serum-based calibrators intended for use with the system.

The toal duration of the assay is 18 minutes. The assay steps are as follows:

- During the first incubation, 6 μL of sample are automatically prediluted 1:20 with diluent (Diluent Universal). Biotinylated monoclonal anti-human IgM specific antibodies are also added.
- During the second incubation, CMV-specific recombinant antigen labeled with a ruthenium complex and streptavidin-coated microparticles are added. Anti-CMV IgM antibodies present in the sample react with the ruthenium-labeled CMV-specific recombinant antigen. The complex becomes bound to the solid phase via interaction of biotin with streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell II M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- The analyzer automatically calculates the cutoff based on the measurement of Calibrator 1 and Calibrator 2. The result of the samples is given either as reactive, borderline, or non-reactive as well as in the form of a cutoff index (signal of sample/cutoff)(COI) as shown in Table 1.

Table 1: Interpretation of Results

Numeric Result (COI)	Result Message	Interpretation/Further Steps
COI < 0.7	Non-reactive	CMV IgM-specific antibodies not detected.
0.7 ≤ COI > 1.0	Borderline	Re-test the sample. If the result is indeterminate (borderline), collect and test a second sample within the following 2 weeks.
COI ≥ 1.0	Reactive	CMV IgM-specific antibodies detected.

The magnitude of the measured result above the cutoff is not indicative of the total amount of antibody present in the sample

The following reagents are provided in the Elecsys CMV IgM assay kit:

1) The reagent rackpack (cobas e pack) consists of reagents M, R1, and R2, and is labeled as CMVIGM:
    a. M: Streptavidin-coated microparticles, 1 bottle, 14.1mL
    b. R1: Anti-h-IgM-Ab~biotin, 1 bottle, 19.7 mL
    c. R2: CMV-Ag~Ru(bpy), 1 bottle, 19.7 mL
        CMV-specific antigen labeled with ruthenium complex

2) CMVIGM Cal1: Negative calibrator 1, 1 bottle of 1.0 mL
    Human serum negative for anti-CMV IgM; preservative.

3) CMVIGM Cal2: Positive calibrator 2, 1 bottle of 1.0 mL
    Anti-CMV IgM (human serum) in HEPES buffer, bovine albumin, and preservative

The following materials are required but not provided:

1) PreciControl CMV IgM 8 x 1.0 mL each of PreciControl CMV IgM 1 and 2
2) Diluent Universal, 45.2 mL sample diluent
3) CalSet Vials, 2 x 56 empty snap-cap bottles
4) General laboratory equipment
5) Cobas e 801 Immunoassay Analyzer
6) Accessories for the cobas e 801 analyzer

The cobas e 801 Immunoassay Analyzer is a fully automated, software controlled analyzer system for in vitro determination of analytes in human body fluids. It is part of the cobas 8000 modular analyzer series. It uses electrochemiluminescent technology for signal generation and measurement. It is a modified version of the cobas e 601 analyzer module, part of the cobas 6000 modular analyzer.

J. Substantial Equivalence Information:

1. Predicate device name(s): Elecsys CMV IgM (on the cobas e 601)

2. Predicate 510(k) number(s): K142133

3. Comparison with predicate:

Similarities		
Item	Candidate Device: Elecsys CMV IgM (on the cobas e 801) K163569	Predicate Device: Elecsys CMV IgM (on the cobas e 601) K142133
Indications for Use	Immunoassay for the in vitro qualitative detection of IgM antibodies to CMV in human serum, lithium‑heparin plasma, K2‑EDTA plasma, and K3‑EDTA plasma. The test is intended as an aid in the diagnosis of recent or current CMV infection in individuals for which a CMV IgM test was ordered, including pregnant women. Performance characteristics have not been evaluated in immunocompromised or immunosuppressed individuals. This test is not intended for use in neonatal screening or for use at point of care facilities. This test is not intended for use in screening blood and plasma donors. The electrochemiluminescence immunoassay "ECLIA" is intended for use on cobas e immunoassay analyzers.	Immunoassay for the in vitro qualitative detection of IgM antibodies to CMV in human serum, lithium-heparin plasma, K2-EDTA plasma, and K3-EDTA plasma. The test is intended as an aid in the diagnosis of recent or current CMV infection in individuals for which a CMV IgM test was ordered, including pregnant women. Performance characteristics have not been evaluated in immunocompromised or immunosuppressed individuals. This test is not intended for use in neonatal screening or for use at point of care facilities. This assay is not intended for use in screening blood and plasma donors. The electrochemiluminescence immunoassay "ECLIA" is intended for use on Elecsys immunoassay analyzers.
Sample Types	Serum, serum with separating gel, Li-heparin, K_2EDTA, and K_3EDTA plasma	Same
Detection Technology	Electrochemiluminescence immunoassay (ECLIA)	Same
Assay Protocol	μ-Capture	Same
Antibody	Biotinylated monoclonal anti-h-IgM antibody (mouse) CMV-specific antigen (recombinant, E. coli) labeled	Same

Similarities		
Item	Candidate Device: Elecsys CMV IgM (on the cobas e 801) K163569	Predicate Device: Elecsys CMV IgM (on the cobas e 601) K142133
	with ruthenium complex Streptavidin-coated microparticles	
Calibrators	CMV IgM Cal 1 and CMV IgM Cal 2	Same
Controls	PreciControl CMV IgM 1 &2	Same

Differences		
Item	Candidate Device: Elecsys CMV IgM (on the cobas e 801) K163569	Predicate Device: Elecsys CMV IgM (on the cobas e 601) K142133
Throughput	300 tests/hour/module	170 tests/hour/module
Sample Volume	6 µL	10 µL
Sample Capacity Onboard	300	150
Instrument Platform	Part of the cobas 8000 modular analyzer series	Part of the cobas 6000 modular analyzer series
Onboard Storage Temperature	5-10°C	18-22°C

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

- CLSI EP5-A3: Evaluation of Precision Performance of Clinical Chemistry Devices; Approved Guideline – 3rd Edition
- CLSI EP15-A2: User Verification of Performance for Precision and Trueness; Approved Guideline - 2nd Edition
- CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline – 2nd Edition

L. Test Principle:

The Elecsys CMV IgM immunoassay is based on the µ-Capture test format. During the first incubation step, biotinylated monoclonal anti-h-IgM-specific antibodies binds specifically to IgM the diluted test specimen. CMV-specific recombinant antigen labeled with a ruthenium complex and streptavidin-coated microparticles are then added for the second incubation. Anti-CMV IgM antibodies present in the sample react with the ruthenium-labeled CMV-specific recombinant antigen. The complex becomes bound to the solid phase via interaction of biotin with streptavidin. The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell II M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   a. *Precision/Reproducibility:*
   Precision of the Elecsys CMV IgM assay was evaluated according to CLSI EP05-A3 on one cobas e 801 Immunoassay Analyzer with one reagent lot. Two replicates per run for each control [PreciControl (PC) CMV IgM, level 1 and 2] and five native human serum and plasma samples (HSP) from single donors were measured (except for plasma sample 5 which was pooled). Measurements were performed in 2 runs per day for 21 days. All results met the pre-defined acceptance criteria and are shown in Table 2:

Table 2: Precision Results

			Repeatability		Intermediate Precision	
Sample	**n**	**Mean [COI]**	**SD[c] [COI]**	**CV[d] [%]**	**SD [COI]**	**CV [%]**
HSP[a] 1*	84	0.202	0.002	1.2	0.006	2.8
HSP 2#	84	0.847	0.011	1.3	0.015	1.8
HSP 3#	84	1.09	0.018	1.6	0.020	1.8
HSP 4#	84	3.46	0.033	1.0	0.049	1.4
HSP 5^	84	1.28	0.013	1.0	0.023	1.8
PC[b] CMV IgM 1	84	0.225	0.002	0.9	0.006	2.6
PC CMV IgM 2	84	1.85	0.036	1.9	0.041	2.2

*serum sample, # plasma sample, ^plasma pool
[a]HSP=human specimen [b]PreciControl [c]Standard Deviation
[d]Coefficient of Variation

   b. *Linearity/assay reportable range:*
   Not Applicable.

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

   Reagent Stability: Two studies were conducted to evaluate the stability of the Elecsys CMV IgM assay reagents on the cobas e 801: onboard reagent stability and reagent real-time stability. These studies were reviewed and found to be acceptable. The reagents are stable when stored unopened at 2°C to 8°C for 12 months or onboard (5 to 10°C) for 16 weeks.

   Sample Stability: Three studies were conducted to evaluate sample stability: sample stability at 2-8°C, sample stability at 20-25°C, and sample stability at -20°C. These studies were reviewed and found to be acceptable. Specimens are stable for 4 weeks at 2°C to 8°C, 7 days at 20-25°C, or 3 months at -20°C (±5°C).

d. Detection limit:

Limit of Blank: The Limit of Blank (LoB) of the Elecsys CMV IgM assay on the cobas e 801 Immunoassay Analyzer was determined according to CLSI EP17-A2 as the 95th percentile of measurements of blank samples. Five blank serum samples were assayed with two replicates per run for six runs, using three reagent lots on one instrument, for a total of 60 blank replicates per reagent lot. The LoB was calculated to be 0.243 COI.

Limit of Detection: The Limit of Detection (LoD) of the Elecsys CMV IgM assay on the cobas e 801 Immunoassay Analyzer was determined according to CLSI EP17-A2 as the lowest amount of analyte in a sample that can be detected with 95% probability. Five samples were prepared by spiking human serum samples from single donors with low levels of analyte. The samples were tested with two replicates per sample per run, six runs, with three reagent lots, on one instrument for a total of 60 replicates per sample per reagent lot. The LoD was calculated to be 0.267 COI using the following equation: LoD=LoB+1.653 x SD.

e. Analytical specificity:

Endogenous Interferences: To evaluate the effect of elevated levels of biotin (10 to 100 ng/mL), intralipid (150 to 1500 mg/dL), hemoglobin (50 to 500 mg/dL), and bilirubin (2 to 20 mg/dL) on the Elecsys CMV IgM assay, four CMV IgM samples (low negative, low positive, positive, and high positive) were tested on the cobas e 801. The recovery (% COI) was calculated for each sample and compared to the reference unspiked sample.
Acceptance criteria: Negative/borderline samples (COI<1.0), deviation $\pm$ 0.2 COI; Positive samples (COI $\geq$ 1.0), deviation $\pm$ 20% (Recovery 80-120%).
The results met the acceptance criteria for all concentrations of endogenous interferent tested. Roche will receive the following claims for no interference observed at the following concentrations: biotin $\leqq$100 ng/mL, intralipid $\leqq$ 1500 mg/dL, hemoglobin $\leqq$ 500 mg/dL, and bilirubin $\leqq$ 20 mg/dL.

To evaluate the effect of rheumatoid factor (RF), native high RF samples were diluted 1:1 with RF negative-CMV IgM positive and RF negative-CMV negative samples. The results of the CMV positive sample diluted with a RF high sample is compared to a 1:1 dilution of the same CMV sample with RF negative samples with similar total IGM levels as the RF high sample. Seven samples with a CMV IgM COI from 0.203 to 5.77 were diluted 1:1 with 3 RF high/CMV-IgM negative samples. As a reference, the sample samples were diluted with RF negative/CMV-IgM negative samples. RF concentrations of 462 to 899 IU/mL were tested and met the acceptance criteria. Roche will receive a claim for no interference for RF $\leq$ 899 IU/mL.

Exogenous Interfering Substances (Drugs): The effect of quantitation of analyte in the presence of drugs was determined by comparing values obtained from native

human serum and plasma samples from single donors on the cobas e 801. The samples were spiked with 17 commonly used and 2 special pharmaceutical compounds and tested on the Elecsys 2010 Immunoassay Analyzer. Acetylcysteine and phenylbutazone were measured on the cobas e 411 Immunoassay Analyzer. Two samples negative and positive for CMV IgM were divided into aliquots and spiked with the potential interferent. The concentration of CMV IgM in the samples was approximately 0.2 COI and 1.9 (2.4 for acetylcysteine and phenylbutazone)COI. The CMV IgM concentration of the spiked aliquots was determined in 9-fold determination and compared to the CMV IgM concentration determined for the reference aliquot on the Elecsys 2010 and the cobas e 411 Immunoassay Analyzers, respectively. Acceptance criteria: Negative/borderline samples (COI<1.0), devation ±0.2COI; Positive samples (COI>1.0),deviation ±10%. Each compound was found to be non-interfering at the concentration listed in the Table 3.

Table 3:Drug Interences: Common Therapeutic Drugs

Potential Interfering Exogenous Substance	Concentration Tested [mg/mL]
Acetylcystein*	1660
Ampicillin-Na	1000
Ascorbic acid	300
Ca-Dobesilate	200
Cyclosporine	5
Heparin	5000 U/L
Levodopa	20
Methyldopa +1.5	20
Metronidazole	200
Phenylbutazone*	400
Doxycycline	50
Acetylsalicylic Acid	1000
Rifampicin	60
Acetaminophen	200
Ibuprofen	500
Theophyllin	100
Ganciclovir	800 [mg/L]
Valganciclovir	900 [mg/L]

*Data measured on the cobas e 411 Immunoassay Analyzer

f. Assay cut-off:

Please refer to the Decision Summary for K142133 for a summary of the assay cutoff (performed on the Elecsys instrument family member Elecsys 2010). Validation of the assay cut-off was performed by external clinical studies on the Elecsys 2010 (K142133) and a Method Comparison study on the cobas e 801.

The analyzer automatically calculates the cut-off based on the measurement of CMVIGM Cal1 and CMVIGM Cal2. The results are in the form of a cut-off index (signal sample/cut-off) as previously shown in Table 1.

g. *High-dose hook effect:*
Testing with the Elecsys CMV IgM assay on the cobas e 801 demonstrated no high-dose hook effect. Three naturally positive human serum and plasma samples from single donors with high CMV IgM concentrations were diluted in a series of 11 dilution steps. Each dilution was tested in singlicate. No high-dose hook effect was observed up to 18.7 COI.

h. *Verification of IgM Specificity:*
Please refer to the Decision Summary for K142133 (performed on the Elecsys instrument family member Elecsys 2010).

2. Comparison studies:

a. *Method comparison with predicate device:*
A method comparison study was performed to compare the Elecsys CMV IgM on the cobas e 801 and the Elecsys CMV IgM on the cobas e 601 analyzer (predicate). A total of 223 leftover deidentified native human plasma samples from single donors were measured in singlicate. Measured values ranged between 0.210 to 20.7 COI for the cobas e 601 and between 0.189 to 21.0 COI for the cobas e 801. The results are shown in Table 4.

Table 4: Method Comparison

		cobas e 601 (original)			**Total cobas e 801**
		Negative COI <0.7	**Borderline 0.7 ≥ COI > 1.0**	**Positive COI ≥ 1.0**	
cobas e 801 (new)	**Negative COI < 0.7**	142	1	0	143
	Borderline 0.7 ≥ COI > 1.0	0	6	0	6
	Positive COI ≥ 1.0	0	1	73	74
Total cobas e 601		142	8	73	223

The positive percent agreement rate was 100% (100/100), the negative percent agreement rate was 100% (73/73), and the percent agreement for Borderline results was 75% (6/8). None of the samples changed from positive to false negative or negative to false positive.

b. *Matrix comparison:*
The study was conducted to assess the effect of anticoagulants on quantitation of analyte by the Elecsys CMV IgM immunoassay on the cobas e 801. The following matrices were evaluated: Li-heparin plasma, K_2-EDTA plasma, K_3-EDTA plasma, and serum from separator tubes. Data from using the serum separator tubes was generated on the Elecsys instrument family member Elecsys 2010 (K142133) and the

data is summarized here. Samples were collected into matched serum and plasma collection tubes and assayed in duplicate. The study was conducted using negative (non-reactive) and positive (reactive) samples. The results are summarized in Table 5 and Table 6 and support the use of the following sample types : Li-heparin plasma, K_2-EDTA plasma, K_3-EDTA plasma, and serum separator tubes.

Table 5: Matrix Equivalency Negative Samples

Sample Matrix	Mean COI	Percent of samples showing differences in recovery relative to serum (COI) in non-reactive specimens		
		< 0.07 COI	0.07 – 0.1 COI	> 0.1 COI
Li-Heparin Plasma	0.238	100%	0	0
K_2-EDTA Plasma	0.237	100%	0	0
K_3-EDTA Plasma	0.232	100%	0	0
Serum Separator Tubes	0.265	100%	0	0

Table 6: Matrix Equivalency Positive Samples

Sample Matrix	Mean COI	Percent of samples showing differences in recovery relative to serum (COI) in reactive specimens		
		< 10 %	10 – 20 %	> 20 %
Li-Heparin Plasma	2.15	77 %	23 %	0 %
K_2-EDTA Plasma	2.27	100 %	0 %	0 %
K_3-EDTA Plasma	2.32	100 %	0 %	0 %
Serum Separator Tubes	2.27	90 %	10 %	0 %

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):
   Please refer to the Decision Summary for K142133 for a summary of clinical study results (performed on the Elecsys instrument family member Elecsys 2010).

4. Clinical cut-off:
   Not Applicable.

5. Expected values/Reference range:

   Not Applicable.

N. Instrument Name:

cobas e801 Immunoassay Analyzer

O. System Descriptions:

1. Modes of Operation:

   Does the applicant's device contain the ability to transmit data to a computer, webserver, or mobile device?

   Yes ____X____ or No _____

   Does the applicant's device transmit data to a computer, webserver, or mobile device using wireless transmission?

   Yes _____ or No ____X____

2. Software:

   FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

   Yes ____X____ or No _____

3. Specimen Identification:

   Barcode

4. Specimen Sampling and Handling:

   Input and transport of samples using universal sample racks, modular sample buffer input, core/transportation unit and STAT port.

5. Calibration:

   Calibration must be performed once per reagent lot using fresh reagent (i.e., not more than 24 hours since the reagent kit was registered on the analyzer. Renered calibration is recommended after 12 weeks when using the same reagent lot, after 28 days when using the same cobas e pack on the analyzer, and as required (e.g., quality control finding outside the defined limits).

6. Quality Control:

   Controls should be run individually at least once every 24 hours when the test is in use, once per cobas e pack, and following each calibration. Values should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside defined limits.

P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:

The software documentation was reviewed and found to be acceptable. The firm provided documentation to support that the device was designed, developed, and maintained under appropriate software lifecycle processes.

Q. Proposed Labeling:

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

R. Conclusion:

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