Capture-CMV®

Solid Phase System for the Detection of IgG and IgM Antibodies to Cytomegalovirus(CMV)

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Rx ONLY

325-17

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IMMUCOR

Intended Use:

Capture-CMV is an in vitro qualitative solid phase red cell adherence test system for the detection of antibodies (IgG plus IgM) to cytomegalovirus (CMV) in human serum or plasma. Capture-CMV is intended to be used in screening of blood and plasma donors or patients for serological evidence of previous infection by CMV.

Summary of the Test:

Cytomegalovirus (CMV) is a common human viral pathogen which belongs to the family of herpes viruses. The presence of CMV antibodies in an individual indicates prior infection by the virus. The possibility exists that viral reactivation can occur in such individuals. CMV infection is usually asymptomatic, and can persist as a latent or chronic infection.[1] Viral transmission may occur through transfusion of blood or transplantation of organs from seropositive donors.[2]

Immunocompromised patients, such as premature neonates, organ transplant patients, and oncology patients, are at greater risk of developing more severe manifestations of CMV infections which can be a major direct or indirect cause of mortality in such patients.[2] Congenitally infected newborns are especially prone to developing severe cytomegalic inclusion disease (CID).[3] The severe form of CID may be fatal or may cause permanent neurological sequelae, such as mental retardation, deafness, microcephaly, and motor dysfunction. A CMV mononucleosis-type syndrome can result from the transfusion of CMV-infected blood products or the transplantation of CMV-infected donor organs in a seronegative immunocompromised patient.[1] Low birth weight neonates are also at high risk to CMV mononucleosis through transfusion of CMV-infected blood products.

One method of preventing or reducing CMV infection in seronegative immunocompromised patients is to select CMV seronegative blood donors or living organ donors that have been tested by serological screening test for antibodies to CMV. Capture-CMV is a solid phase red cell adherence antibody detection system based on procedures of Plapp et al.[4] This procedure is a modification of the mixed agglutination tests for antigen and antibody detection of Coombs et al[5] and Hogman[6] employing anti-IgG and IgG-coated red cells as the indicator system. Capture assays for the detection of antibodies to red cells or platelets use anti-IgG-coated red cells as the indicator.[7] Capture-CMV uses anti-IgG plus anti-IgM-coated indicator red cells.

Principle of the Test:

The CMV antigen utilized in this test is obtained from the cytomegalovirus strain AD 169 grown in human foreskin (HF) fibroblast cells. The inactivated virus is coated onto microtitration wells. The wells are dried and supplied to users along with necessary reagents and controls.

The assay procedure is a two step solid phase red cell adherence test carried out in microtitration wells coated with inactivated CMV virus. Serum or plasma samples are added to the viral-coated wells. The samples are incubated for five minutes; during which antibodies specific for CMV proteins bind to immobilized viral proteins. Unbound immunoglobulins are washed from the wells and replaced with a suspension of anti-IgG- plus anti-IgM-coated indicator red cells. Centrifugation brings the indicator red cells in contact with antibodies bound to the immobilized viral proteins. In the case of a positive test, the migration of the indicator red cells to the bottom of the well is impeded as the anti-IgG and anti-IgM bridges are formed between the indicator red cells and the viral-bound antibodies. As a consequence, the indicator red cells adhere over the surface of the microtitration well. In contrast, in the absence of viral antigen-antibody interactions (ie, a negative test) the indicator red cells are not impeded during their migration, and pellet to the bottom of the well as a packed, well-defined cell button.

Key:
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Reagents:

Capture-CMV Microtitration Wells: Rigid U-bottom microtitration well coated with glycine-extracted and purified CMV antigen obtained from cytomegalovirus strain AD 169 grown in human foreskin (HF) fibroblast cells. The wells are enclosed in foil pouches to which a desiccant and moisture indicator have been added. Each microtitration well is ready to be used as supplied. Store the wells between 1-30°C. If the humidity indicator enclosed within each pouch shows the presence of moisture (by the humidity indicator turning from blue to pink), the wells should not be used. Unused microtitration wells, desiccant and humidity indicator should be immediately resealed within the foil pouch to prevent the uptake of moisture. Carefully reseal pouch to minimize leakage of moisture into pouch during storage. Microtitration wells resealed in a pouch should be used within two weeks (but not beyond the expiration date) provided the humidity indicator does not show the presence of moisture.

Microtitration wells removed from pouches should be used within one (1) hour.

Adjunct Reagents to Capture Test Wells: (Purchased separately)

Capture-CMV Indicator Red Cells: A suspension of human red blood cells coated with rabbit anti-human IgG plus goat anti-human IgM molecules. The red blood cells are suspended in a buffered solution to which chloramphenicol (0.25 mg/mL), neomycin sulfate (0.1 mg/mL) and gentamycin sulfate (0.05 mg/mL) have been added as preservatives. Store at 1-10°C.

Capture-CMV Positive Control Serum (weak): Human serum containing IgG antibodies to CMV viral proteins. Capture-CMV Positive Control Serum (weak) is manufactured to represent the reactivity obtained by weak CMV antibody positive donors. Weak CMV antibody positive donors have a titration endpoint of 1:2 or less. Weak CMV antibody positive reactions are found in less than 2% of the donor population. Sodium azide (0.1%) has been added as a preservative*. Store at 1-10°C. Slight turbidity may occur with age.

Capture-CMV Negative Control Serum: Human serum containing no antibodies to CMV. Sodium azide (0.1%) has been added as a preservative*. Store at 1-10°C. Slight turbidity may occur with age.

Capture LISS: a low ionic strength solution containing glycine, bromcresol purple dye and the preservative sodium azide (0.1%)*. Store at 1-10°C.

On receipt, Capture-CMV adjunct reagents should be stored at 1-10°C.

The in-date components (Capture-CMV test wells, Capture LISS, Capture-CMV Controls, Capture-CMV Indicator Red Cells) used to perform Capture-CMV assays can be used interchangeably with other components, irrespective of their lot numbers, providing the components are within their expiration dates.

Precautions:

For *in vitro* diagnostic use.

This reagent contains 0.1% sodium azide. Warning: H302 Harmful if swallowed. Sodium azide may react with lead and copper plumbing to form explosive compounds. If discarded into the sink, flush with a large volume of water to prevent azide build-up.

CAUTION: ALL BLOOD PRODUCTS SHOULD BE TREATED AS POTENTIALLY INFECTIOUS. SOURCE MATERIAL FROM WHICH THIS PRODUCT WAS DERIVED WAS FOUND NEGATIVE WHEN TESTED IN ACCORDANCE WITH CURRENT FDA REQUIRED TESTS. NO KNOWN TEST METHODS CAN OFFER ASSURANCE THAT PRODUCTS DERIVED FROM HUMAN BLOOD WILL NOT TRANSMIT INFECTIOUS AGENTS.

Bring all refrigerated Capture-CMV microtitration wells to room temperature (18-30°C) before testing.

The format for the expiration date is expressed as CCYY-MM-DD (year-month-day).

Do not use Capture-CMV microtitration wells beyond the expiration date. Leaking vials should not be used.

Specimens should be handled according to OSHA Bloodborne Pathogens Rules. Blood specimens and all materials coming into contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions. Wear disposable gloves while handling kit reagents or specimens and thoroughly wash hands afterwards. Never pipet by mouth and avoid contact with skin and mucous membranes. Avoid splashing or forming an aerosol.

Avoid microbial contamination of reagents or incorrect results may occur. Use aseptic technique. Store all assay components at their proper temperatures when not in use.

Handle and dispose of all specimens and materials used to perform the test as if they contained infectious agents.

Cross contamination of samples can cause falsely positive results. Change pipet tips between samples to eliminate contamination/carryover with the previous specimen.

Tests should be performed using properly calibrated equipment, including centrifuges and microplate washers. Use of improperly calibrated or maintained equipment may result in false-positive or false-negative results.

Do not allow plates to dry once assay has begun.

Do not reuse wells of a plate.

Incubation times or temperatures other than those specified may give erroneous results.

Store saline for washing in a clean container to avoid contamination.

Reusable glassware must be washed out and thoroughly rinsed free of all detergents.

The performance of Capture-CMV is not impacted by icteric, lipemeic, or hemolyzed specimens containing bilirubin up to 30 mg.dL, triglycerides up to 600 mg/dL and hemoglobulin, up to 2+ hemolysis or by specimens containing albumin up to 5.2 g/dL.

Specimen Collection and Preparation:

Draw a blood specimen using an acceptable phlebotomy technique.[8] Serum or plasma (EDTA, CPD, CP2D, CPDA-1, ACD) may be used in this assay. Testing should be performed as soon as possible to minimize the chance that falsely positive or falsely negative reactions will occur due to improper storage or contamination of the specimen. Should delays in testing occur, serum or EDTA, CPD, and ACD anticoagulated whole blood specimens should be stored at 1-10°C for up to one week. Specimens collected in CP2D and CPDA-1 anticoagulants and stored as whole blood specimens at 1-10°C may be tested up to 42 days postdonation. Testing of CPDA-1 and ACD anticoagulated specimens stored at 18-25°C for five days is acceptable. Alternatively, serum or plasma can be separated from red cells and stored frozen at –20°C in a nondefrosting refrigerator. Samples should not be repeatedly frozen and thawed. Weakly reactive antibodies may deteriorate and become undetectable in samples stored beyond five days at room temperature or in serum or anticoagulated samples stored at 1-10°C beyond the recommended storage time. Do not use specimens drawn into tubes containing neutral gel separators. False-positive results may occur with such samples.

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Procedure:

Materials Provided:

1. Capture-CMV Microtitration Wells in sealed pouches

Additional Reagents:

1. Capture-CMV Indicator Red Cells
2. Capture-CMV Positive Control Serum (weak)
3. Capture-CMV Negative Control Serum
4. Capture LISS

Additional materials required: (as applicable)

1. Automatic or manual microtitration plate washer, vacuum source, and trap; or hand-held multi-channel refilling syringe. Note: The Capture-CMV system has been designed so that automated microtitration plate washers may be incorporated in washing steps to reduce the risks associated with handling patient samples.
2. Centrifuge rotor and carriers capable of accommodating microtitration wells.*
3. Micropipettors and tips capable of delivering 50 µL and 100 µL, or disposable pasteur pipets capable of delivering a 50±5 µL drop. Alternatively, an automatic pipettor-dilutor may be used.
4. Glass or plastic test tubes.
5. Stop watch or interval timer.
6. Illuminated white translucent surface.
7. Phosphate-buffered (approximately 15mM) isotonic saline, pH 6.5-7.5
8. Galileo*
9. Galileo Neo*
10. NEO Iris*

*It is the user's responsibility to validate an accessory device for its intended use. Validation results should be maintained as part of the laboratory's records for review by regulatory agencies.

Automated Instrument Users: For testing with automated instrumentation, refer to instructions provided in the instrument operator manual.

Test Method:

Manual and Semi-automated:

1. Bring reagents to room temperature.
2. Remove Capture-CMV Microtitration Wells from their protective pouch. If the presence of moisture is shown by the humidity indicator enclosed within each pouch the wells should not be used.

NOTE: A batch (or run) is considered to be the largest number of wells used that will be incubated, washed and centrifuged as a unit. Minimum batch size is one strip of wells. The maximum batch size is twelve 1x8 strips fitted into one frame holder. Positive and negative controls should be included with each batch or run.

3. Add 2 drops (100 µL) of Capture LISS to all test wells.

NOTE: The purple color of the Capture LISS will change to a sky or turquoise blue in the presence of serum. The retention of the purple color may indicate that test serum/plasma has inadvertently been omitted from the well.

4. Add 1 drop (50±5 µL) of Capture- CMV Positive Control Serum (weak) to the first well.
5. Add 1 drop (50±5 µL) of Capture- CMV Negative Control Serum to the second well.
6. Add 1 drop (50±5 µL) of the first test serum/plasma to the third well.
7. Add 1 drop (50±5 µL) of the next test sample to the fourth well.
8. Proceed in this manner until all test sera or plasma samples have been added. A clean pipette or new pipette tip must be used for each test sample to prevent cross-contamination.

NOTE: Each centrifugation run of test samples must contain at least one set of control reagents (Weak Positive and Negative Controls) to ensure that the wells have been centrifuged and/or washed properly.
Samples should be dispensed within 20 minutes. Once samples are dispensed, steps 9 through 12 should be completed without interruption.

9. Incubate the Capture-CMV wells at 18-30°C for a minimum of 5 minutes, but no more than 30 minutes.
10. Decant or aspirate the serum-LISS mixture from the wells and wash wells using a manual or automated wash technique.
    a. Manual Washing Technique
        i. Decant fluid from the wells.
        ii. Fill the wells of the strip with saline dispensed from a multichannel dispenser or manifold designed for microplates. Alternatively, a saline wash bottle can be used to dispense the saline. Saline should not be added with excessive force since this may cause the antigen bound to the bottom of the well to disengage from the plate.

iii. Decant the wells thoroughly by manually inverting the strip wells over a sink or waste receptacle and with several rapid, sharp motions, decanting the saline from the wells.
iv. Wash the wells a minimum of six times with saline.

b. Semi-automated Washing Technique

For semi-automated washing, refer to instructions provided in the washer operator manual.

NOTE: The automated washing device must be adjusted such that approximately 4-8 uL of saline remains in each well after aspiration. Wells should not be aspirated until they are dry.

11. Resuspend Capture-CMV Indicator Red Cells by gently inverting the vial. Immediately add 1 drop (50±5 µL) of Capture-CMV Indicator Red Cells to each test well.
12. Centrifuge the wells at 450-600 x g for 1 minute. Allow the centrifuge rotor to come to a complete stop. Centrifuge wells again at 1000-1400 x g for an additional minute. (The g force is an approximation of the speed required to produced the required degree of adherence. The appropriate g force (or rpms) and centrifugation time must be determined individually for each centrifuge used.)

NOTE: Overcentrifugation of the tests, following addition of the Capture-CMV Indicator Red Cells, may result in falsely negative or indeterminate positive reactions due to the collapse of the adherent indicator layer. Failure to obtain fully adherent reactions with the Positive Control Serum (strong) and/or partially adherent reactions with the Positive Control Serum (weak) may indicate that the microtitration wells have been overcentrifuged. The acceleration or deceleration characteristics of the centrifuge in use may affect the type of reactions obtained at the end of the assay. Failure to apply the braking mechanism in units with long deceleration times may result in falsely negative reactions. Conversely, braking of centrifuges with short deceleration times may also cause erroneous test results. Acceleration and deceleration parameters must be determined for each centrifuge type. In most cases, acceleration times of 5-30 seconds will permit satisfactory results. Excessive vibration of test wells during deceleration will lead to weak false-positive results. In such cases, results with the negative control will not be valid. Do not recentrifuge test wells. Repeat tests using new test wells and centrifuge using an alternative centrifuge.

13. Place the microtitration wells on an illuminated surface and examine for adherence or the absence of Indicator Cell adherence. For test results to be considered valid, the following reactions must be obtained with the Capture-CMV Control Sera:

Positive Control (Weak) = adherence of Indicator Red Cells over part or all of the reaction surface.

Negative Control Serum = a button of Indicator Red Cells at the bottom of the test wells with no area of adherence.

14. Compare each antibody detection test result with those obtained with the positive and negative control sera. A test should be repeated if a doubtful reaction (irregular, nonconcentric adherence) is obtained or if the control sera do not perform properly.

Galileo, Galileo Neo and NEO Iris:

1. Bring reagents and blood samples to 18-30°C before testing.
2. Centrifuge the blood samples to separate the plasma/serum from the red blood cells/clot.
3. Remove the Capture-CMV microplate frame and the desired number of Capture-CMV strips from the pouch.
4. Remove reagent vial caps.
5. Add one stirball to each new vial of Capture-CMV Indicator Red Cells to be used. Gently agitate each vial to resuspend the red blood cells.
6. Load reagents, microplates, and blood samples onto the instrument following the procedures in the Operator Manual Chapter 6 – Instrument Testing Operation.
7. Assign the CMV assay to the blood samples, either manually or following the upload worklist procedure.
8. Start the CMV assay following the procedures in the instrument Operator Manual Chapter 6 – Instrument Testing Operation. The instrument automatically performs the CMV assay, and records and interprets the blood sample results.
9. At the completion of the CMV Assay, press the Results button on the main menu bar to access the blood sample results.

Stability of the reaction: (Manual and Semi-automated Methods)

Following centrifugation, tests can be read immediately. Since positive reactions are permanent, wells can be covered following centrifugation to prevent evaporation, stored at 1-10°C, and read or reread up to 2 days following testing.

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Quality Control:

The reactivity of the Capture-CMV assay is evaluated at each centrifugation run by inclusion of the negative and weak positive controls. If, in any test run, the Positive Control Serum does not produce positive results and/or the Negative Control Serum does not produce negative results, the test run is invalid and all the tests performed in the run must be repeated. Continued failure of the control sera to perform properly may indicate that either one or more of the test reagents has deteriorated, or that the tests are not being performed correctly.

Interpretation of Results:

Negative test: A button of the Capture-CMV Indicator Red Cells at the bottom of the test well with no area of adherence indicates the test sample has no detectable CMV antibody and the person has not yet been infected with CMV and is presumed to be susceptible to primary infection.

Positive test: Adherence of Capture-CMV Indicator Red Cells to part or all of the reaction surface indicates a person with previous or current infection and who is presumed to be at risk of transmitting CMV infection but who is not necessarily currently contagious.

Limitations:

1. Erroneous test results can occur from bacterial or chemical contamination of test materials, inadequate incubation periods, improper centrifugation, inadequate washing of test wells, or omission of test reagents or steps.
2. Serum or plasma specimens obtained from tubes containing neutral gel separators may produce falsely positive results. Tubes with gel separators are not designed for blood bank use.
3. Addition of Capture-CMV Indicator Red Cells in excess of amounts described in this insert may result in falsely negative or indeterminate test reactions.
4. The Capture-CMV assay is designed to detect IgG plus IgM antibodies. The assay is neither designed to detect antibodies of the IgA or IgE class nor differentiate between IgM and IgG antibodies to CMV.
5. Positive test results may not be valid in persons who have received blood transfusions or other blood products within the past several months.
   Samples taken early during infection before antibodies have developed may be negative, thus a negative result does not rule out infection.
6. Care should be taken in interpreting test results of neonatal samples. A positive test usually indicates the presence of antibodies passively transferred from mother to fetus. A negative test may be helpful in excluding possible infection, but a diagnosis of active CMV infection may require viral culture.[13, 15]
7. The presence of IgG or total complement-fixing antibody does not assure protection from disease.[11]
8. The titer of a single specimen should not be used to aid in the diagnosis of recent infection. Paired samples (acute and convalescent) should be collected and tested concurrently to look for seroconversion which is indicative of primary infection.[12] Samples obtained too early during primary infection may not contain detectable antibodies. If CMV infection is suspected, a second sample should be obtained 2-7 weeks later and tested in parallel with the first specimen to look for seroconversion which is indicative of primary infection.
9. Positive test results in symptomatic patients require careful interpretation since false-positive reactions or heterotypic IgM responses may occur with sera from patients with heterophile-positive mononucleosis[9,13] or varicella zoster infection.[14]
10. Heterotypic IgM antibody responses to CMV have been reported in as many as 30% of persons with infectious mononucleosis[10] and polyclonal stimulation of B lymphocytes by EBV seems the most likely mechanism. However, reactivation of latent CMV is also a possibility. The reverse, that is heterotypic IgM antibody responses to EBV in CMV infections, is less frequently seen, but has been reported.[16] Reactivation of latent EBV would seem to be a possible mechanism.[13]
11. Over centrifugation of the tests, following addition of the Capture-CMV Indicator Red Cells, may result in falsely negative or indeterminate positive reactions due to the collapse of the adherent indicator layer. Undercentrifugation will lead to falsely positive results.

Expected Values:

The incidence of CMV infection is dependent upon geographical, socioeconomic, and age factors. Serological studies indicate that the incidence of antibodies to CMV is between 15% and 70% in adult populations.[17]

Specific Performance Characteristics:

The performance of this product is dependent upon adhering to the insert's recommended methodology.

The expiration for Capture-CMV Indicator Red Cells is 60 days from the date of the manufacture which is the earliest date blood used in this product is withdrawn from any donor.

Performance in Manual and Semi-Automated Methods:
The performance of the Capture-CMV assay (manual and semi-sutomated methods) was evaluated at seven (7) separate test sites on 6,506 specimens. The specimens comprised a mixture of blood bank donors and hospital patients in which CMV antibody status was routinely requested.

Performance in Patient Populations:
The performance of the Capture-CMV assay was compared to commercially available latex agglutination test at two (2) separate test sites on 308 patient specimens. Specimens giving discrepant results between the Capture-CMV assay and the latex agglutination test were evaluated at an independent test site using commercially available enzyme immunoassay (EIA) for total antibody and an in-house direct immunofluorescence assay (IFA) for CMV IgM antibody. A consensus result of positive or negative antibodies to CMV was assigned to each discrepant specimen based on the EIA and IFA results. The results are summarized in the following table.

		Capture-CMV	
		Positive	Negative
Consensus Result	Positive	138	0
	Negative	1	169

Relative Sensitivity = 100% (138/138)

Relative Specificity = 99.4% (169/170)

The commercially available latex agglutination test exhibited a relative sensitivity of 100% (138/138) and a relative specificity of 99.4% (169/170) at these test sites.

Performance in Donor Populations:
The performance of the Capture-CMV assay was compared to a commercially available latex agglutination test at two (2) separate sites on 508 donor specimens and to a commercially available automated passive hemagglutination assay at three (3) separate test sites on 4,367 specimens. The donor populations tested consisted of serum and plasma (EDTA) samples. Specimens giving discrepant results between the Capture-CMV assay and the latex agglutination tests were evaluated at an independent test site using commercially available enzyme immunoassay (EIA) for total antibody and an in-house indirect immunofluorescence assay (IFA) for CMV IgM antibody. Similarly, specimens giving discrepant results between the Capture-CMV assay and passive hemagglutination tests were evaluated at an independent test site using a commercially available enzyme immunoassay (EIA) for total antibody to CMV. A consensus result of positive or negative for antibodies to CMV was assigned to each discrepant specimen based on the EIA and IFA results. The relative sensitivity and relative specificity of the Capture-CMV assay with the calculated 95% confidence intervals at each donor test site are summarized in the following table:

Test Site	Relative Sensitivity		Relative Specificity	
	Value	†95% CI	Value	95% CI
Site #2	100%	99.6-100	98.9%	96.7-100
Site #3	100%	99.3-100	99.5%	98.7-100
Site #5	100%	99.9-100	100%	99.9-100
Site #6	99.3%	98.7-99.9	99.5%	99.0-100
Site #7	99.2%	98.5-99.99	99.4%	99.1-99.7

†95% CI= 95% two-sided Confidence Interval

The latex agglutination test exhibited a relative sensitivity of 99.5% (211/212) and a relative specificity of 99.3% (294/296) at test sites #2 and #3. The automated passive hemagglutination assay demonstrated a relative sensitivity of 96.0% (1313/1368) and a relative specificity of 99.2% (2573/2593) at test sites #5-7.

Performance on Galileo and Galileo Neo:
The performance of Capture-CMV when tested on the Galileo with a donor and patient population of 3,537 samples resulted in 99.7% detection of reactive samples and 99.6% detection of non-reactive samples compared to 99.2% and 98.4%, respectively, with manual and semi-automated methods.
The performance of Capture-CMV when tested on the Galileo Neo with a donor and patient population of 630 samples resulted in 100% Positive Percent Agreement and 98.9% Negative Percent Agreement with the Galileo results.

Performance on NEO Iris:

Method comparison studies were performed at four clinical sites, three external sites and internally at Immucor, Inc. for donor specimens and at two external sites and internally at Immucor, inc. for patient specimens. Specimens were tested on NEO Iris and Galileo Neo. Test results were evaluated for agreement between analyzers. Specimens with discordant results were further tested with a commercially available particle agglutination assay for total antibody (IgG+IgM) to CMV.

Specimen testing by sites:

Sites	Donor Specimens			Patient Specimens		
	Total	Serum	Plasma	Total	Serum	Plasma
1	474	57	417	26	18	8
2	289	59	230	0	0	0
3	103	20	83	195	70	125
Immucor	382	70	312	250	212	38

CMV Initial Results Donor Samples N=1248		Galileo Neo	
		Positive	Negative
NEO Iris	Positive	612	8
	Negative	27	601
CMV Resolved Results		Galileo Neo / Anti-CMV PA*	
		Positive	Negative
NEO Iris	Positive	612	8
	Negative	1	627
Sensitivity 99.8% (99.1%, 95% 2-sided LCI)			
Specificity 98.7% (97.5%, 95% 2-sided LCI)			

LCI – Lower Confidence Interval

CMV Initial Results Donor Serum Samples N=206		Galileo Neo	
		Positive	Negative
NEO Iris	Positive	103	1
	Negative	5	97
CMV Resolved Results		Galileo Neo / Anti-CMV PA*	
		Positive	Negative
NEO Iris	Positive	103	1
	Negative	0	102
Sensitivity 100.0% (96.5%, 95% 2-sided LCI)			
Specificity 99.0% (94.7%, 95% 2-sided LCI)			

CMV Initial Results Donor Plasma Samples N=1042		Galileo Neo	
		Positive	Negative
NEO Iris	Positive	509	7
	Negative	22	504
CMV Resolved Results		Galileo Neo / Anti-CMV PA*	
		Positive	Negative
NEO Iris	Positive	509	7
	Negative	1	525
Sensitivity 99.8% (98.9%, 95% 2-sided LCI)			
Specificity 98.7% (97.3%, 95% 2-sided LCI)			

CMV Initial Results	Galileo Neo

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Patient Samples N=501		Positive	Negative
NEO Iris	Positive	272	5
	Negative	0	224
CMV Resolved Results		Galileo Neo / Anti-CMV PA*	
		Positive	Negative
NEO Iris	Positive	272	5
	Negative	0	224
Sensitivity 100.0% (98.7%, 95% 2-sided LCI)			
Specificity 97.8% (95.0%, 95% 2-sided LCI)			

*Only discordant specimens were tested with IgG/IgM Anti-CMV PA.

Reproducibility:

The reproducibility of the Capture-CMV assay was determined using two separate panels of coded samples provided to the trial sites. The reproducibility of Capture-CMV was evaluated on twenty-five (25) coded samples at five (5) test sites comparing the performance of Capture-CMV to a latex agglutination test. The Capture-CMV assay demonstrated a 96.8% agreement (121 of 125 samples) of reproducibility panel test results between test sites. For three (3) donor test sites comparing Capture-CMV to a passive hemagglutination assay, reproducibility was evaluated on identical panels of sixty (60) samples consisting of ten (10) samples of each of six serum pools. The Capture-CMV assay demonstrated a 94.4% agreement (170/180) of reproducibility panel test results between three test sites. The test sites collectively demonstrated a 94.2% (113/120) within-day and a 94.4% (170/180) day-to-day reproducibility. A summary of the reproducibility of the Capture-CMV assay at each donor site on the samples with negative, weakly positive, and moderately positive potencies and the expected frequencies of such potencies in the donor populations are provided in the following table.

Sample Potency	Site #5		Site #6		Site #7	
	*Frequency	Reproducibility	Frequency	Reproducibility	Frequency	Reproducibility
Negative	65.9%	100%	51.4%	100%	69.5%	100%
Weak Positive	1.5%	91.7%	2.2%	83.3%	1.4%	83.3%
Moderately Positive	3.1%	100%	4.4%	100%	2.7%	100%

* Frequency = Expected frequency at the donor site population

NEO Iris Reproducibility

The reproducibility of Capture-CMV assay on the NEO Iris was determined using a panel of ten (10) coded samples, five (5) CMV antibody positive and five (5) CMV antibody negative, at three (3) test sites, two external sites and internally at Immucor, Inc.The samples were tested by two operators, in duplicated on two (2) runs per day for five (5) nonconsecutive days. The summary of reproducibility results by site are presented in the following tables:

(Table 1 of 2):

Concordance by Site				
Site	Total Tests	Expected Positive	Observed Positive	% Concordance (95% LCI)
1	400	200	200	100.0% (98.2%)
2	400	200	200	100.0% (98.2%)
3	400	200	200	100.0% (98.2%)
Total	1200	600	600	100.0% (99.4%)

(Continued) (Table 2 of 2):

Site	Expected Negative	Observed Negative	% Concordance (95% LCI)
1	200	200	100.0% (98.2%)
2	200	200	100.0% (98.2%)
3	200	199	99.5% (97.2%)
Total	600	599	99.8% (99.1%)

Specificity and Crossreactivity:

The following table summarizes Capture-CMV results when testing samples from subjects with the following IgG antibodies:

Category of Specimen	Number	Capture-CMV Positive
EBV (VCA) Epstien-Barr Virus (Viral Capsid Antigen)	16	0
HSV – Herpes Simplex Virus	Type I – 10 Type II – 13 IgM* – 2	0
Hepatitis A	5	1
Parovirus B19	4	0
ANA – Anti-Nuclear Antibodies	11	1
RF – Rheumotoid Factor	10	0
VZ – Varicella Zoster	8	0
Rubella	8	0
Toxoplasma gondii	4	0

*HSV Type not specified

To ensure suitable reactivity and specificity, each assay component lot of the Capture-CMV assay is tested prior to release against sera known to contain specific antibodies to CMV viral proteins, as well as sera known to be free of such antibodies.

For additional information or for technical support, contact Immucor at 855-IMMUCOR (466-8267).

Bibliography:

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