CMV Viral Structure

http://cytomegalovirusproject.wikispaces.com/

CMV Antigens for Commercial Assay Development
Antigens designed for the development of IgG and IgM CMV detection assays

The CMV diagnostics market is growing due to an increased prevalence of CMV infections worldwide and a better awareness of the disease. In the U.S. alone, 60% of the population is carrying CMV and more than 90% of those are in a high risk category (AIDS patients and prenatal babies of CMV infected mothers). Meridian offers a complete panel of native and recombinant CMV antigens that meet the increasing demand for more sensitive and specific CMV serologic assays.

Human cytomegalovirus (CMV) is a member of the herpes virus family and shares the characteristic ability to remain latent within the body for life within an infected individual. Although a CMV infection is typically asymptomatic in healthy persons, immunocompromised individuals such as AIDS patients, organ transplant recipients and newborn infants, are at high-risk of developing life-threatening complications from primary infections and reactivations.

Various diagnostic tests have been developed to detect a CMV infection including viral culture, serological methods, PCR analysis and cytopathology. The p65 antigenemia test, in which a monoclonal antibody against CMV pp65 is used to detect a major CMV matrix protein (pp65) in leukocytes, has the longest history in clinical use but has several weaknesses including subjectivity in reading positive results, time consuming and intricate procedures, difficulty in standardization, and a need for sufficient leukocytes. In contrast, the ELISA IgG/IgM assay has become the most commonly available serologic test and measures antibodies to CMV, specifically CMV IgM, IgG and IgG avidity. The detection of IgM is indicative of an acute or primary infection whereas the detection of IgG is indicative of a past infection, and in the case where both IgM and IgG can be detected, the level of IgG avidity can help distinguish between an acute infection and a past infection. For this reason, newer assays have begun to incorporate the detection of anti-CMV IgM together with determination of the avidity index of anti-CMV IgG.

To improve the sensitivity and specificity of CMV antibody detection, immunogenic CMV proteins have been studied and characterized during the past two decades and over 15 structural polypeptides have been identified in a natural infection. The most suitable proteins are reportedly the matrix phosphoproteins pp150 and pp65, glycoproteins gB and gH, and the major DNA binding protein (pp52). Evidence also suggests that CMV-IgM detection against viral structural proteins (pp 150 and pp38) is a valuable parameter for the early diagnosis of a recurrent CMV infection. The combination of antigens selected is the most critical element affecting assay sensitivity and specificity. Several commercial assays have incorporated a combination of CMV lysate and CMV recombinant proteins to improve their performance.

MERIDIAN CMV ANTIGENS

Native Antigens: prepared from human fibroblast cells infected with CMV AD169

<table>
<thead>
<tr>
<th>No.</th>
<th>Description</th>
<th>EV9268</th>
<th>CMV II Antigen</th>
</tr>
</thead>
<tbody>
<tr>
<td>7504</td>
<td>CMV-G, whole cell extract</td>
<td>CMV II Antigen</td>
<td></td>
</tr>
<tr>
<td>7517</td>
<td>CMV Ext-2, enriched for cell surface glycoprotein antigens</td>
<td>7507</td>
<td>CMV III Antigen, enriched for pp65</td>
</tr>
<tr>
<td>7511</td>
<td>CMV-M Concentrate, nuclear extract and ER antigens</td>
<td>EV7508B</td>
<td>CMV Nucleoprotein (NP) Type I</td>
</tr>
</tbody>
</table>

Recombinant Antigens: expressed in E.coli

<table>
<thead>
<tr>
<th>CMV pp52 (UL44)</th>
<th>CMV pp65 (UL83)</th>
</tr>
</thead>
<tbody>
<tr>
<td>R01565 GST tag, MW 51 kDa</td>
<td>R01566 GST tag, MW 49 kDa</td>
</tr>
<tr>
<td>R01561 His tag, MW 11 kDa</td>
<td>R01562 His tag, MW 52 kDa</td>
</tr>
<tr>
<td>R18062 GST tag, MW 44 kDa</td>
<td>R18412 GST tag, MW 50 kDa</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>CMV pp150 (UL32)</th>
<th>OTHER</th>
</tr>
</thead>
<tbody>
<tr>
<td>R01564 GST tag, MW 35 kDa</td>
<td>CMV pp28 (UL99) GST tag, MW 30 kDa</td>
</tr>
<tr>
<td>R01563 His tag, MW 16 kDa</td>
<td>R18103 CMV pp38 (UL80a) GST tag, MW 55 kDa</td>
</tr>
<tr>
<td>R18113 GST tag, MW 32 kDa</td>
<td>R18512 CMV pp38 (UL80a) GST tag, MW 52 kDa</td>
</tr>
<tr>
<td></td>
<td>R01524 CMV pp72 (UL123) His tag, MW 50 kDa</td>
</tr>
<tr>
<td></td>
<td>R18102 CMV gB (UL55) GST tag, MW 32 kDa</td>
</tr>
</tbody>
</table>
APPLICATIONS
Recommended for:

- **IgG Detection**
CMV IgG antibodies are produced 10-14 days after initial infection and provide protection from subsequent primary infections. Levels of IgG rise during an active infection and then stabilize as the CMV infection resolves and the virus becomes inactive. After a person has been exposed to CMV, measurable amounts of CMV IgG antibody will remain in the body throughout life.

More recently, measurement of CMV-specific IgG avidity has proven to be a powerful tool for distinguishing primary from recurrent CMV infections. A person infected with IgG for the first time produces low avidity IgG antibodies and after approximately 2-4 months begins to produce high-avidity IgG. A CMV IgG avidity test calculates the ratio of total IgG to high-avidity IgG in order to establish the IgG avidity index. Specifically, duplicate measurements with an IgG ELISA assay are compared; the first measurement is for determining the total IgG in a patient’s sample and the second measurement is the IgG value of the same patient sample after treatment with an IgG avidity diluent (such as urea which disrupts the hydrogen bonds and interferes with the binding of low avidity CMV IgG antibodies to the immobilized CMV antigen).

**ANTIGENS FOR IgG DETECTION**
- 7504 Native CMV-G: partially purified viral lysate (whole cell)
- 7517 Native CMV Ext-2: partially purified viral lysate (enriched for cell surface glycoproteins)
- R18102 CMV gB Antigen: membrane glycoprotein gB which is the most abundant component of the viral envelope and a target of neutralizing antibodies
- R18103 CMV pp28: tegument protein essential for virus assembly

- **IgM Detection**
CMV-specific IgM antibodies usually develop within 1-2 weeks after initial infection and are a sensitive marker of a recent or ongoing CMV infection. A CMV IgM test is particularly useful in diagnosing infections in pregnant women and guiding the appropriate treatment options which are aimed to decrease the risk of congenital infection. Typically, IgM antibodies drop to below detectable levels within several months after infection.

**ANTIGENS FOR IgM DETECTION**
- EV9268 Native CMV II: purified surface extract containing pp65, pp52, pp150 and other key proteins
- 7507 Native CMV III: purified surface extract containing pp65, pp52 and other key proteins
- EV7508B Native CMV Nucleoprotein: purified viral lysate containing nuclear extract
- R01564 CMV pp150: tegument protein detectable during both latent and re-activated infections. During primary infection the antibody response to pp150 may be delayed. Also suitable for IgG detection.
- R18113
- R01565 CMV pp52: nonstructural nuclear phosphoprotein which is regarded as an early marker of seroconversion
- R01561
- R18062
- R01566 CMV pp65: major structural protein (lower matrix) and main component of extracellular virus particles.
- R01562 The antibody response is primarily detectable during early infection only
- R18412
- R18512 CMV pp38: structural protein suggested to be an important immunodominant protein in early
- R01567

*Meridian suggests testing a combination of native and recombinant (pp150 & pp52) CMV antigens to develop a sensitive and specific assay that detects both CMV IgG and IgM antibodies*

**FOR RESEARCH OR FURTHER MANUFACTURING USE**

Meridian Life Science, Inc.
5171 Wilfong Rd. / Memphis, TN 38134
www.meridianlifescience.com
Orders/Inquiries: info@meridianlifescience.com / Tel: 901.382.8716 / Fax: 901.333.8223