Value of Cytomegalovirus (CMV) IgG Avidity Index for the Diagnosis of Primary CMV Infection in Pregnant Women

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This study assessed the diagnostic value of the cytomegalovirus (CMV)-specific IgG avidity index (AI) for pregnant women without a history of CMV seroconversion. Sera were studied from 40 women with CMV seroconversion (group I), 70 with past CMV infection (group II), 10 (20 sera) with serologic reactivation (group III), and 41 with CMV-specific IgM without proven seroconversion (group IV). Sera from women in group I collected <14 weeks after seroconversion had a low AI (mean, 30% ± 12%), whereas all sera from women in group II had an AI >60% (mean, 88% ± 9%). Among the 41 babies born to group IV women, only 4 were infected with CMV (all born to mothers with a low [<30%] AI early in pregnancy). These results suggest that AI determination may help to date a primary CMV infection in pregnant women who lack seroconversion history.

Materials and Methods

Study population. Three French laboratories (in Caen, Clamart, and Paris) and one Swiss laboratory (Geneva) participated in the study. Sera from pregnant women were sent from 1992 to 1995 for serologic screening early in pregnancy (Caen, Paris) or for diagnosis of primary CMV infection in women considered at risk (Clamart, Geneva).

Four groups of pregnant women were studied. Group I, 40 women with well-documented CMV seroconversion, had 87 post-seroconversion samples (1–8 samples/woman). Of the samples, 68 were obtained <14 weeks (3 months) after the last seronegative sample, and 19 were obtained ≥14 weeks after that date. Group II comprised 70 women with serologic evidence of prior CMV infection (presence of CMV-specific IgG, absence of CMV-specific IgM). Group III consisted of 10 pregnant women with serologic CMV reactivation (defined by a 4-fold increase in IgG titer with or without detectable CMV-specific IgM). Two serum samples were available for each woman. Group IV comprised 41 women without well-documented seroconversion during the first trimester of pregnancy (each had only 1 serum sample from that period with CMV-specific IgM).

Sixty-six fetuses and infants were tested for CMV infection (25 born to group I mothers and 41 to group IV mothers).

Antibody detection. Serum samples were tested for CMV IgG by commercial indirect enzyme immunoassays (Enzygnost-CMV IgG: Behring, Marburg, Germany; IgG MEIA: Abbott Laboratories, Abbott Park, IL) and for CMV IgM by capture immunoassay (Wellcozyme anti-CMV IgM: Murex Diagnostics, Paris) or by indirect immunoassays (Enzygnost-CMV IgM: Behring; IgM MEIA: Abbott).

Avidity of IgG. For CMV-specific IgG avidity, sera were measured by the Enzygnost-CMV IgG kit according to the manufacturer’s recommendations, except for the wash step after the first antibody incubation, when 8 M urea was added in parallel with PBS– Tween 20, as previously described [4]. The AI was expressed as follows: Percentage of AI = (absorbance result of CMV per well with urea wash/absorbance result of CMV per well without urea wash) × 100.

Virology. When available for groups I and IV, amniotic fluid or infant urine obtained at birth was tested for CMV by culture [6] or polymerase chain reaction [7].

Statistical analysis. AI distributions by group were compared by the nonparametric Wilcoxon test and always expressed with 1
SD and the range. When measurements for the same woman were done at two different time points, we made paired comparisons of the mean percentages. All probability values are two-tailed. The threshold of significance was set at .05.

**Results**

For the 40 women with well-documented seroconversion (group I), 87 sera were obtained after the last seronegative sample. Among these 87 samples, 68 were obtained <14 weeks (3 months) after the last seronegative sample. The mean CMV IgG AI of the first sample was 30% ± 12% (range, 8%–58%). For the 70 pregnant women with a past CMV infection (group II), the mean AI was significantly higher (88% ± 9%; range, 60%–100%; P < .001) than the mean of the first positive sample for women in group I. In the latter group, we observed an increase in AI with time in all serial serum specimens. Figure 1 shows the temporal changes in IgG avidity for the 12 women for whom ≥2 positive samples were available. Of the 68 sera obtained <14 weeks after the negative sample, only 6 had a CMV IgG AI >50% (maximum value, 65%), and 2 of 19 samples obtained after 14 weeks had an AI <50% (43% and 44%). Only 7 women in group I had serum samples available that were positive <3 months after the last seronegative sample and a sample obtained ≥14 weeks later. The mean difference in AIs between these two categories of sera was significantly different from 0 (33% ± 22%; P < .01).

For 25 group I women whose pregnancy outcome was known, 12 fetuses or infants were infected. The mean AI of the first positive sample of women who transmitted virus was 30.3% ± 9.2% (range, 15.5%–43%), not significantly different from that of nontransmitters (33.2% ± 13.8%; range, 8%–53%). The intervals between the last seronegative and the first seropositive samples were 6.5 ± 3.4 and 6.6 ± 3.7 weeks, respectively.

In the 10 cases of reactivation (group III), the mean AIs before and after reactivation were 83% ± 7% (range, 72%–97%) and 92% ± 6% (range, 76%–100%), respectively. The mean difference in AIs between these 2 serum samples was significantly different from 0 (8% ± 5%; P < .01).

Among the 41 serum samples obtained during the first trimester of pregnancy from women who lacked well-documented seroconversion (group IV), 5 had AIs ≤30% (group IVa), 14 had AIs of 40%–70% (group IVb), and 22 had AIs >70% (group IVc). All pregnancy outcomes were known for these 41 women. Four babies were infected with CMV, all born to women with low AIs (≤30%, group IVa).

**Discussion**

Since the overall incidence of symptomatic congenital CMV is low (~0.05%) [1] and risk factors are not well defined, it has been difficult to develop strategies for detection of CMV infection, especially for asymptomatic primary infection in pregnant women. Some obstetricians propose CMV testing of all pregnant women; others screen for CMV antibody only in women considered at risk (e.g., pediatric nurses, teachers, day care workers). Thus, CMV IgM is sometimes detected in the serum of an asymptomatic pregnant woman whose immune status before conception is not known, which raises issues about interpretation of results. For a variety of viral infections, including rubella, varicella-zoster, human herpesvirus, hepatitis C [4, 8–12], and more recently CMV [5, 13], the usefulness...
of specific IgG avidity analysis is recognized for distinguishing primary infection (with low avidity antibody) from past or recurrent infection (in which the specific IgG response is mature and shows high avidity).

To assess the contribution of this method for dating CMV infection in pregnant women with CMV-specific IgM antibody, we studied the CMV AI pattern in women with well-documented primary, past, and secondary CMV infections. Our results demonstrate that pregnant women with past or secondary infection have an AI >60%. In contrast, most pregnant women with CMV primary infection, and whose sera were collected <3 months after the last seronegative sample, have an AI <50%; none had an AI >65%. Thus, an AI >65% is highly suggestive of a non-primary infection. Furthermore, the results obtained with clinical samples from women with specific IgM but lacking well-documented seroconversion (group IV patients) showed that an AI <30% is highly suggestive of a recent primary infection (duration, <3 months). Our results agree with those obtained by Hedman and Rousseau for rubella [4].

References