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1 **Clinical factors associated with congenital cytomegalovirus infection:**

2 **A cohort study of pregnant women and newborns**

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19 **Running title:** Risk factors of congenital CMV infection

1   **Summary of the article's main point:** The presence of maternal fever/flu-like symptoms and

2   threatened miscarriage/threatened premature labor in the second trimester were <u>clinical</u> factors

3   <u>associated with the occurrence of</u> congenital CMV infection of newborns.

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Abstract

Background

The aim of this prospective cohort study was to determine clinical factors associated with the occurrence of congenital cytomegalovirus infection (cCMV) in pregnant women.

Methods

Between March 2009 and November 2017, newborns born at a primary maternity hospital received polymerase chain reaction (PCR) analyses for CMV-DNA in their urine with informed consent of the mothers at a low risk. Clinical data, including age, gravidity, parity, body mass index, occupations, maternal fever/flu-like symptoms, pregnancy complications, gestational weeks at delivery, birth weight, and automated auditory brainstem response (AABR), were collected. Logistic regression analyses were performed to determine clinical factors associated with cCMV.

Results

cCMV was diagnosed by positive PCR results of neonatal urine in 9 of 4,125 pregnancies. Univariate and multivariable analyses revealed that the presence of fever/flu-like symptoms (odds ratio [OR], 17.9; 95% confidence interval [CI], 3.7–86.7; $p<0.001$) and threatened miscarriage/premature labor in the second trimester (OR, 6.0; 95%CI 1.6–22.8; $p<0.01$) were independent clinical factors associated with cCMV. Maternal fever/flu-like symptoms or threatened miscarriage/premature labor in the second trimester had 100%

1   sensitivity, 53.2% specificity, and a maximum Youden index of 0.85.

2   **Conclusions**

3   This cohort study for the first time demonstrated that these clinical factors of pregnant

4   women and newborns were <u>associated with</u> the occurrence of cCMV. This is useful information

5   for targeted screening to assess risks of cCMV in low-risk mothers, irrespective of primary or

6   non-primary CMV infection.

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Introduction

Cytomegalovirus (CMV) is the most common cause of congenital infection in humans. The prevalence rate of congenital CMV infection (cCMV) is 0.2%–2.4% in newborns [1], and 10%–15% of infected newborns are symptomatic at birth. The clinical manifestations of cCMV include fetal growth restriction (FGR), low birth weight, central nervous system and multiple organ involvement with petechiae, hepatomegaly, splenomegaly, jaundice, pneumonia and encephalitis. These are very severe and can cause a high perinatal mortality rate and major neurological sequelae in about 90% of surviving infants with symptomatic cCMV [2]. In addition, 10%–15% of infants with asymptomatic cCMV also develop long-term sequelae, which include progressive sensorineural hearing difficulty and mental retardation [2, 3].

Recently, it was reported that early intervention with antiviral drugs can improve neurological outcomes in children with symptomatic cCMV [4-6]. Prenatal detection of newborns at a high risk for cCMV is clinically important where no universal screening of newborns was performed, because it may enable early diagnoses and therapeutic interventions in affected infants. Generally, the risk of maternal-fetal transmission of CMV is thought to be highest in pregnancies with primary CMV infection. Therefore, maternal serological screening, including blood tests for CMV-specific immunoglobulin (Ig) G, IgM, and IgG avidity index, is considered effective for detecting pregnancies at a high risk for cCMV [7, 8]. Recent observational studies, however, demonstrated that the number and severity of symptoms in

1 infants with cCMV from mothers with non-primary CMV infection during pregnancy were

2 similar to those from mothers with primary infection [9-11].

3   A prospective study of neonatal CMV screening found that socioeconomic factors such

4 as younger, parous mothers born in high resources countries, and higher income were risk

5 factors for cCMV due to maternal primary CMV infection, while younger and unemployed

6 were found to be risk factors for cCMV due to non-primary CMV infection [12].

7   However, no prospective studies have evaluated clinical factors associated with the

8 occurrence of cCMV in pregnant women. The aim of this prospective cohort study was to

9 determine clinical factors predictive of cCMV among low-risk women who delivered at a

10 primary maternity hospital, where pregnant women at a high risk were referred or transferred

11 to regional perinatal medical centers.

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1 **METHODS**

2 **Study Design and Participants**

3   The institutional review board at Kobe University Hospital and the research ethics

4 committee at Nadeshiko Ladies Hospital approved this prospective cohort study (reference

5 number 923). Written informed consent was obtained from all participants. From March 2009

6 to November 2017, newborns who were born at Nadeshiko Ladies Hospital, a primary

7 maternity hospital located in Kobe, Japan, underwent universal screening of polymerase chain

8 reaction (PCR) tests for CMV-DNA in the urine. Congenital infection was diagnosed with the

9 detection of CMV-DNA in their urine. All newborns who had positive results for CMV-DNA

10 in the urine were referred to Kobe University Hospital, and received a workup to identify

11 symptoms of cCMV.

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13 **Procedures**

14   <u>Pregnant women were inquired whether they had symptoms of fever, flu-like illness,</u>

15 <u>genital bleeding, abdominal pain, uterine contraction and other abnormalities at regular prenatal</u>

16 <u>checkup.</u> The obstetricians (A.U., and K.T.) retrospectively collected the clinical data of

17 pregnant women who visited to and gave birth at the maternity hospital, including age, gravidity

18 and parity, body mass index prior to pregnancy, occupation, smoking history, history of assisted

19 reproductive technology (ART) therapy, fever or flu-like symptoms, maternal and obstetric

complications, delivery mode, non-reassuring fetal status (NRFS) during labor, gestational age

at delivery, birth weight, sex of newborns, and abnormality of automated auditory brainstem

response (AABR) screening test performed at 1‑5 days after birth. Maternal and obstetric

complications assessed in this study were as follows: hypertensive disorders of pregnancy

(HDP), thyroid disease, diabetes mellitus/gestational diabetes mellitus, medical disease

requiring immunosuppressive therapy, threatened miscarriage, threatened premature labor,

FGR, preterm delivery, and light-for-date (LFD), and low birth weight (LBW).

In this study, ART therapy included *in vitro* fertilization, intracytoplasmic sperm

injection, and embryo transfer. Fever or flu-like symptoms were defined as the condition that

women have complaints such as fever, nasal mucus, cough and/or sore throat. Thyroid disease

was defined as hyper- and hypothyroidism which required medication. NRFS during labor was

defined as the absence of baseline fetal heart rate variability, the presence of recurrent late

deceleration, recurrent variable deceleration, prolonged deceleration, or sinusoidal pattern

detected *via* continuous cardiotocography (CTG) [13]. Threatened miscarriage and threatened

premature labor in this study were defined as the condition that women have subjective

symptoms of uterine pain, contraction, bleeding, and/or shortening of uterine cervical length,

and therefore require tocolytic agents, including oral administration of β-stimulant or calcium

blocker, and intravenous administration of β-stimulant or magnesium sulfate for one or more

weeks. LFD was defined as a birth weight of less than the 10th percentile for gestational age.

LBW was defined as a birth weight less than 2,500g.

Urine samples were collected from newborns on filter paper within one week after birth and the presence of CMV-DNA was assessed as described previously [14]. The urine-filter based assay used in the present study and different in-vitro diagnostic assays by the regulatory authorities yielded identical results [15]. Liquid urine samples were obtained from CMV positive newborns, and the CMV-DNA copy number determined by real-time quantitative PCR. The presence of cCMV was confirmed by positive PCR results in the liquid urine samples [16]. All newborns with cCMV received a workup to identify the symptoms of congenital infection. Ophthalmoscopy, cerebral ultrasound, physical and neurological examinations, head computed tomography, head magnetic resonance imaging, and repeated AABR tests were performed.

Statistical Analysis

Clinical characteristics were compared between pregnancies with cCMV and those without it. The differences between the two groups were analyzed using the Mann-Whitney U test, Fisher's exact test, and the chi-squared test. Statistical significance was considered present at p-values less than 0.05.

A stepwise approach was used to evaluate clinical factors associated with the occurrence of cCMV among all pregnant women who delivered at the primary maternity clinic. Variables with p-values less than 0.05 in univariate logistic regression analyses were subjected

1   to multivariable logistic regression analyses, and variables with *p*-values less than 0.05 in

2   multivariable logistic regression analyses were determined as <u>clinical factors significantly</u>

3   <u>associated with the occurrence of cCMV</u>. The optimal cutoff value was determined at the

4   maximum Youden index. Sensitivity, specificity, positive predictive value (PPV), negative

5   predictive value (NPV), and accuracy were calculated for the prediction of cCMV. All statistical

6   analyses were performed using SPSS software, version 19 (SPSS Inc., Chicago, Illinois).

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Result

A flow diagram of study participants is shown in Figure 1. During the study period, 6,443 pregnant women visited the primary maternity hospital. Four hundred and fifty-nine pregnancies ended in spontaneous miscarriages, and a total of 1,859 pregnant women were referred to other maternity facilities where they delivered. All women with multiple pregnancies were referred to regional perinatal medical centers. A total of 4,125 pregnant women gave birth at Nadeshiko Ladies Hospital, and their newborns underwent PCR tests for CMV-DNA in the urine. cCMV was diagnosed in nine newborns (0.22%), including one newborn with symptomatic infection and eight with asymptomatic infection.

Table 1 shows clinical characteristics of pregnant women and newborns. The frequencies of the presence of fever/flu-like symptoms ($p<0.05$), threatened miscarriage/premature labor in the second trimester ($p<0.01$), and abnormality of AABR screening test for newborns ($p<0.05$) in mothers who had newborns with cCMV were significantly higher than those in mothers who had newborns without cCMV.

Two of nine mothers who had newborns with cCMV and 201 of 4,116 mothers who had newborns without cCMV received public assistance (p=0.07). None of nine mothers with cCMV newborns had sexually transmitted infections including HBV, HCV, HIV, syphilis, chlamydia, or gonorrhea.

Table 2 shows the clinical characteristics and laboratory findings for nine pregnant

women who had newborns with cCMV. One of the nine newborns was diagnosed with

symptomatic cCMV due to AABR abnormality (Case 1), while the remaining eight newborns

were asymptomatic. The newborn of Case 1 received anti-CMV treatments including

intravenous immunoglobulin infusion 250 mg/kg/day a week 2 times, and valganciclovir 16

mg/kg/day for 6 weeks. At present, he is 8 years and 3 months old without any sequela. The

other eight newborns also had no sequela without anti-CMV treatment. Neurodevelopment of

nine newborns with cCMV was assessed using Kyoto scale of psychological development.

Their developmental quotient (DQ) measured at 2 and/or 3 years old ranged from DQ 81 to DQ

105. Although complement fixation (CF) tests for CMV antibodies were not performed for all

participants with informed consent, 3,193 of a total 4,125 pregnant women received CF tests,

and 71.7% (2,288/3,193) had positive tests. Mothers of the nine newborns with cCMV also

received CF tests. Three mothers tested negative for CF tests, but further antibody tests were

not performed.

Logistic regression analyses of clinical factors associated with the occurrence of

cCMV among the pregnant women were performed. Univariate logistic regression analyses for

findings shown in Table 1 demonstrated that the presence of maternal fever/flu-like symptoms

(odds ratio [OR] 19.8, 95% confidence interval [CI] 4.1–95.7; $p<0.001$) and threatened

miscarriage/premature labor in the second trimester (OR 7.1, 95% CI 1.9–26.7; $p<0.01$) were

associated with the occurrence of cCMV. Multivariable logistic regression analyses of the two

1   factors revealed that the presence of maternal fever/flu-like symptoms (OR 17.9, 95% CI 3.7–

2   86.7; $p<0.001$) and threatened miscarriage/premature labor in the second trimester (OR 6.0,

3   95% CI 1.6–22.8; $p<0.01$) were clinical factors associated with the occurrence of cCMV in

4   pregnant women who delivered at the primary maternity hospital.

5   The optimal predictive factors were estimated using the maximum value of the Youden

6   index which is defined as "sensitivity + specificity -1." As a result, the presence of maternal

7   fever/flu-like symptoms alone and threatened miscarriage/premature labor in the second

8   trimester alone yielded sensitivity of 77.8% and 77.8%, specificity of 85.1% and 61.4%, and

9   the Youden index of 0.63 and 0.39, respectively. Furthermore, combination of the presence of

10   maternal fever/flu-like symptoms or threatened miscarriage/premature labor in the second

11   trimester were determined as optimal predictive factors, showing sensitivity of 100%,

12   specificity of 53.2%, positive predictive value of 0.5%, negative predictive value of 100%,

13   accuracy of 53.3%, and a maximum Youden index of 0.85.

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Discussion

In the present study, nine（0.22%）of the 4,125 women delivered newborns with cCMV. This frequency of cCMV was lower than that of previous reports, which showed 0.31%–0.46% in Japan [14, 17]. The present cohort study found that frequencies of maternal fever/flu-like symptoms during pregnancy, threatened miscarriage/premature labor in the second trimester, and abnormality of AABR screening test for newborns were significantly higher in women who had newborns with cCMV. This study demonstrated for the first time that the presence of maternal fever/flu-like symptoms during pregnancy and threatened miscarriage/premature labor in the second trimester were clinical factors associated with cCMV among pregnant women who delivered at a primary maternity hospital, where pregnant women at a high risk were referred or transferred to regional perinatal medical centers. The high-risk pregnancies included maternal complications, fetal abnormality, severe FGR, HDP, multiple pregnancy, and preterm delivery before 34 gestational weeks. It was likely that maternal fever/flu-like symptoms and threatened miscarriage/premature labor in the second trimester were clinical factors associated with the occurrence of cCMV among women with low-risk pregnancies.

In the present study, not all pregnant women underwent CMV antibody tests, because some did not provide informed consent. However, mothers of the nine newborns with cCMV

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coincidentally underwent CF tests once during pregnancy. Seven of the nine pregnant women with cCMV had fever/flu-like symptoms during pregnancy. Two women (Case 1 and Case 2) had fever/flu-like symptoms in the first trimester and other two (Case 5 and Case 7) in the second trimester. The remaining three with negative CF results (Case 6, Case 8 and Case 9) had fever/flu-like symptoms in the third trimester, suggesting that at least the three had primary CMV infection during pregnancy. Case 1 had fever/flu-like symptoms at 8 GW and a positive for CF test at 21 GW, so she might have primary CMV infection during pregnancy. Case 2, 3, 4, 5, and 7 might have primary infection, reactivation or reinfection of CMV during pregnancy, because they had positive for CF tests in the first trimester. CF tests were known to be less sensitive for detecting CMV IgG antibody than tests using the radioimmunoassay (RIA) or enzyme-linked immunosorbent assay (ELISA) techniques. However, there were few discrepancies (1.5%) of results between the CF tests and tests using RIA or ELISA techniques as the screening tests for CMV antibody [18].

Maternal fever/flu-like symptoms may be associated with primary or reinfection of CMV, causing CMV transmission to their fetuses. If pregnant women have fever/flu-like symptoms during pregnancy, as soon as possible they should receive CMV antibody tests, including CMV IgG/IgM, and IgG avidity measurements, and plasma CMV-DNA analysis to determine whether they have acquired CMV infection, because they may have a substantial risk for cCMV. Plasma CMV-DNA analyses help to diagnose CMV infection.

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1 Threatened miscarriage/premature labor in the second trimester may be caused by

2 intrauterine infection as an effect of cCMV. Alternatively, threatened miscarriage/premature

3 labor may be associated with local inflammation causing reactivation of latent CMV in the

4 uterus and blood circulation. A cohort study also reported that threatened premature labor was

5 a risk factor for cCMV in 1,287 pregnant women with non-primary CMV infection [19]. This

6 study demonstrated that 5 of a total 7 women with cCMV had threatened premature labor during

7 the second and third trimesters, and none had during the first trimester. It is likely that

8 reactivation of latent CMV or re-infection during the second and third trimesters cause cCMV

9 more frequently than during the first trimester in women with non-primary CMV infection. The

10 present study did not find an association between cCMV and NRFS, although a previous study

11 did [20].

12 Observational studies of neonatal CMV screening have found that frequencies of

13 cCMV are 1.3% in very LBW newborns, 1.7%-3.7% in small for gestational age, and 3.0% in

14 preterm delivery [21-23]. Other studies also have shown that frequencies of cCMV are 2.9-

15 3.3% in multiple pregnancy, 0.8-2.0% in threatened premature labor, 1.1-1.4% in maternal

16 fever/flu-like symptoms, 0.8-1.4% in LFD, 1.1-1.4% in LBW and 1.2-1.3% in preterm delivery

17 [17, 19]. A prospective study of 11,715 newborns screened by PCR tests for CMV-DNA in

18 saliva found that socioeconomic factors such as younger (<25 years old), parous mothers born

19 in high resources countries, and higher income were risk factors for cCMV due to maternal

primary CMV infection. On the other hand, younger and unemployed were found to be risk

factors for cCMV caused by non-primary CMV infection [12].

Recent observational studies demonstrated that the number and severity of symptoms

in infants with cCMV from mothers with non-primary CMV infection during pregnancy were

similar to those from mothers with primary infection [9-11]. Furthermore, a majority of

newborns with cCMV were born from mothers with non-primary CMV infection during

pregnancy [24]. A registry-based cohort study also found that a majority of newborns with

symptomatic cCMV were from mothers with non-primary CMV infection during pregnancy

[10]. Recently, a prospective cohort study of CMV screening for 2,193 pregnant women and

their newborns in a perinatal medical center demonstrated that maternal antibody screening

using CMV IgG, IgG avidity index, and IgM could identify pregnancies with cCMV due to

maternal primary CMV infection accounting for 30% of cases; however, it overlooked those

caused by non-primary CMV infection, which accounted for 70% [17]. However, there have

been no cohort studies to assess clinical findings predictive of cCMV in a primary maternity

hospital that usually manages low-risk pregnancies. The present cohort study for the first time

evaluated whether clinical factors of mothers and newborns in low-risk populations were

associated with cCMV, and univariate and multivariable analyses demonstrated that maternal

fever/flu-like symptoms during pregnancy and threatened miscarriage/premature labor in the

second trimester were associated with cCMV. Clinical factors of maternal fever/flu-like

symptoms or threatened miscarriage/premature labor in the second trimester were selected with a maximum Youden index of 0.85, showing 100% sensitivity and 100% negative predictive value. If frequency, severity, morbidity and mortality of newborns with cCMV due to maternal non-primary CMV infection is not different from those with cCMV caused by maternal primary CMV infection, prenatal risk estimation and prediction of cCMV based on manifestation of clinical symptoms on mothers, fetuses and newborns in combination with CMV antibody measurements on that occasion may be more effective than universal antibody screening for pregnant women.

It remains controversial whether universal or targeted screening for cCMV based on PCR assays for CMV-DNA in the saliva or urine of newborns is cost-effective. In targeted neonatal screening approaches, infants who are referred for AABR undergo PCR testing of the urine, but this strategy may overlook infants with asymptomatic cCMV or cases with a delayed onset of hearing loss [25]. In the present study, abnormality of AABR screening test was significantly associated with cCMV. However, only one of the 21 newborns with AABR abnormality had cCMV, while the other eight newborns with cCMV showed no AABR abnormality. The targeted neonatal CMV screening that is based on only results of AABR performed during 1–5 days after birth may not be effective for detecting cCMV.

Universal neonatal screening using the saliva or urine for CMV-DNA can identify almost all newborns with cCMV. If universal neonatal screening cannot be performed, neonatal

CMV screening targeting not only neonates with AABR abnormality but also mothers who have

maternal fever/flu-like symptoms during pregnancy or threatened miscarriage/premature labor

in the second trimester may be an effective method to detect cCMV with a high sensitivity.

Clinical practitioners have to check CMV antibodies for mothers who have these risk factors.

Early diagnoses of cCMV can lead to early commencement of antiviral therapies for infants

with symptomatic cCMV to reduce sequelae.

These results will provide useful information for clinical practitioners to assess risks

of cCMV in low-risk mothers, irrespective of primary or non-primary CMV infection. The

study of the transmission and potential harm after reactivation may be facilitated applying these

clinical markers as well as strain specific serology and characterization of the cCMV strain.

However, the present study had some limitations. Threatened miscarriage or premature labor

may be diagnosed somewhat subjectively depending on practitioners. Tocolytic agents used for

treatments of threatened miscarriage or premature labor were different in the world. Clinical

factors associated with cCMV in high-risk pregnant women who are usually managed in tertiary

maternal-fetal centers might be different. The findings in the present study are valid for low-

risk pregnancies in this population, and have to be followed by further studies in other

population with a higher rate of cCMV. CMV serology, socioeconomic status, and education

of pregnant women may influence the results.

Figure Legends

Figure 1

A flow diagram for the study participants and urine CMV screening of newborns.

1   CMV, cytomegalovirus

2   During the study period, 6,443 pregnant women visited to the maternity hospital for

3   maternity checkup. A total of 4,125 pregnant women were enrolled in this study. Nine (0.22%)

4   of the 4,125 pregnant women had newborns with congenital CMV infection, including one

5   symptomatic infection, and eight asymptomatic infection.

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Table 1. Clinical characteristics of participants and results of logistic regression analyses of clinical factors associated with congenital cytomegalovirus infection

Clinical findings	All women n=4,125	Women who had newborns with congenital CMV infection n=9	Women who had newborns without congenital CMV infection n=4,116	P-value	Univariate logistic regression analysis		Multivariable logistic regression analysis	
					Odd ratio (95% CI)	P-value	Odd ratio (95% CI)	P-value
Clinical findings of pregnant women								
Age, years old	30.4 ± 5.1	28 (16–35)	30 (16–46)	N.S.				
Gravidity	1.9 ± 1.1	2 (1–5)	2 (1–14)	N.S.				
Parity	0.7 ± 0.8	1 (0–4)	1 (0–5)	N.S.				
Assisted reproductive technology therapy	362 (8.8%)	0	362	N.S.				
BMI prior to pregnancy, kg/m2	20.7 ± 2.8	19.8 (18.2–27.5)	20.2 (14.7–41.1)	N.S.				
Smoking history	272 (7.8%)	0	272	N.S.				
Fever or ful-like symptoms	627 (15.2%)	7	620	<0.05	19.8 (4.1–95.7)	<0.001	17.9 (3.7–86.7)	<0.001
Hypertensive disorders of pregnancy	74 (1.8%)	1	73	N.S.				
Thyroid diseases	33 (0.8%)	0	33	N.S.				
Diabetes mellitus gestational diabetes mellitus	74 (1.8%)	0	74	N.S.				
Medical diseases requiring immunosuppressive therapies	4 (0.1%)	0	4	N.S.				
Threatened miscarriage in the first trimester	255 (6.1%)	0	255	N.S.				
Threatened miscarriage or premature labor in the second trimester	618 (15.0%)	5	613	<0.01	7.1 (1.9–26.7)	<0.005	6.0 (1.6–22.8)	<0.01
Threatened premature labor in the third trimester	1101 (26.6%)	4	1097	N.S.				
Preterm delivery	103 (2.4%)	0	103	N.S.				
NRFS during labor	136 (3.4%)	0	136	N.S.				
Gestational weeks at delivery	39.1 ± 1.3	39 (38–41)	39 (33–42)	N.S.				
Cesarean delivery	800 (19.4%)	1	799	N.S.				
Clincal findings of newborns								
Birth weight, g	3,051.2 ± 372.5	2,912 (2,232–3,840)	3,040 (1,936–4,676)	N.S.				
Light-for-date	219 (5.3%)	1	218	N.S.				
Low birth weight	248 (6.0%)	1	247	N.S.				
Male	2125 (51.5%)	5	2120	N.S.				
Abnormality of AABR screening test	22 (0.5%)	1	21	<0.05				

CMV, cytomegalovirus; CI, confidence interval; BMI, body mass index; NRFS, non-reassuring fetal status; AABR, automated auditory brainstem response.

Data are expressed as the average ± standard deviation, median (range), or number.

N.S., not significant

Table 2. Nine pregnant women who had newborns with congenital cytomegalovirus infection

Case	Age, years old	Gravidity /parity	BMI, kg/m2	Occupation	Pregnancy complications (GW)	GW at flu-like symptoms	CF tests, times (GW)	Delivery mode	Gestational age at delivery	Birth weight, g	Sex	Blood gas pH of umbilical artery	Symptoms of newborns	Infant development Age
1	28	5/4	27.3	None	None	8	16 (21)	Cesarean	38w4d	3,160	Male	7.354	AABR abnormality	Normal 8 years and 3 months
2	16	2/0	18.2	None	Threatened miscarriage (19)	8	64 (7)	Vaginal	41w3d	3,620	Female	7.271	None	Normal 7 years and 9 months
3	28	3/1	27.5	None	Threatened premaure labor (24)	None	32 (8)	Vaginal	40w4d	3,840	Male	7.300	None	Normal 7 years and 7 months
4	32	2/1	21.7	None	Threatened premaure labor (23)	None	16 (8)	Vaginal	38w5d	2,606	Female	7.392	None	Normal 6 years and 5 months
5	35	3/2	23	None	Threatened premaure labor (34), HDP (36)	24	16 (8)	Vaginal	39w1d	2,740	Female	7.385	None	Normal 6 years
6	25	2/0	19.8	Child carer	Threatened premaure labor (23)	29	<4 (19)	Vaginal	39w2d	2,768	Male	7.324	None	Normal 5 years and 3 months
7	29	1/0	16.9	Pharmacist	Threatened miscarriage (18)	27	32 (9)	Vaginal	39w3d	2,232	Male	7.349	Light-for-date, low birth weight	Normal 2 years and 9 months
8	28	2/1	19.8	Nurse	None	29	<4 (10)	Vaginal	40w4d	2,912	Male	7.365	None	Normal 2 years and 8 months
9	31	2/1	19.6	None	Threatened premaure labor (34)	32	<4 (9)	Vaginal	38w3d	3,248	Female	7.363	None	Normal 2 years and 5 months

CMV, cytomegalovirus; BMI, body mass index; GW, gestational weeks;

HDP, hepertensive disorders of pregnancy; CF, complement fixation; NRFS, non-reassuring fetal status; AABR, automated auditory brainstem response

Figure 1

Pregnant women who had a maternity checkup
n=6,443

Spontaneous miscarriage
n= 459

Pregnant women who delivered at other facilities
1,736 referred for giving birth at their parents' home
123 transferred to regional perinatal medical centers
n=1,859

Women delivered at the maternity hospital
n=4,125

Newborns underwent PCR tests of CMV-DNA in the urine
n=4,125

Newborns with congenital CMV infection
n= 9

Newborns with no congenital CMV infection
n= 4116

Newborns with
symptomatic infection
n= 1

Newborns with
asymptomatic infection
n= 8