Timing of primary maternal cytomegalovirus infection and rates of vertical transmission and fetal consequences

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OBJECTIVE: Cytomegalovirus infection is the most frequent congenital infection and a major cause of long-term neurologic morbidity. The aim of this meta-analysis was to calculate the pooled rates of vertical transmission and fetal impairments according to the timing of primary maternal infection.

DATA SOURCES: From inception to January 2020, MEDLINE, Scopus, Cochrane Library, and gray literature sources were used to search for related studies.

STUDY ELIGIBILITY CRITERIA: Cohort and observational studies reporting the timing of maternal cytomegalovirus infections and rate of vertical transmission or fetal impairments were included. The primary outcomes were vertical transmission and fetal insult, defined as either prenatal findings from the central nervous system leading to termination of pregnancy or the presence of neurologic symptoms at birth. The secondary outcomes included sensorineural hearing loss or neurodevelopmental delay at follow-up and prenatal central nervous system ultrasonography findings.

STUDY APPRAISAL AND SYNTHESIS METHODS: The pooled rates of the outcomes of interest with their 95% confidence intervals (CI) were calculated for primary maternal infection at the preconception period, periconception period, first trimester, second trimester, and third trimester.

RESULTS: A total of 17 studies were included. The pooled rates of vertical transmission (10 studies, 2942 fetuses) at the preconception period, periconception period, first trimester, second trimester, and third trimester were 5.5% (95% CI, 0.1—10.8), 21.0% (95% CI, 8.4—33.6), 36.8% (95% CI, 31.9—41.6), 40.3% (95% CI, 35.5—45.1), and 66.2% (95% CI, 58.2—74.1), respectively. The pooled rates of fetal insult in case of transmission (10 studies, 796 fetuses) were 28.8% (95% CI, 2.4—55.1), 19.3% (95% CI, 12.2—26.4), 0.9% (95% CI, 0—2.4%), and 0.4% (95% CI, 0—1.5), for maternal infection at the periconception period, first trimester, second trimester, and third trimester, respectively. The pooled rates of sensorineural hearing loss for maternal infection at the first, second, and third trimester were 22.8% (95% CI, 15.4—30.2), 0.1% (95% CI, 0—0.8), and 0% (95% CI, 0—0.1), respectively.

CONCLUSION: Vertical transmission after maternal primary cytomegalovirus infection increases with advancing pregnancy, starting from the preconception period. However, severe fetal impairments are rare after infection in the first trimester of pregnancy.

Key words: cytomegalovirus, magnetic resonance imaging findings, neurodevelopmental delay, neurologic symptoms, primary infection, sensorineural hearing loss, timing, ultrasonography findings, vertical transmission

Introduction

Congenital cytomegalovirus (CMV) infection is the most common congenital infection, with an incidence of 0.2 to 2.2%.[1,2] Vertical transmission to the fetus occurs through the placenta, which is the first fetal organ to be infected. The placenta temporarily acts as a barrier to the virus; however, it also serves as a reservoir for viral replication and is a target organ itself. The virus eventually crosses the placenta, replicates in the tubular epithelium of the kidneys, and has a trophism in the reticuloendothelial and central nervous system; it is the injury of the latter that causes long-term disability. According to the commonly used quotes for counseling purposes, the vertical transmission rate is about 35% to 40%.[3,4] Approximately 10% of newborns with infection are expected to be symptomatic at birth, some with serious impairments, and about 10% of the asymptomatic newborns may develop impairments within the first 6 years of life, mostly hearing loss and less commonly neurodevelopmental delay.[5]

However, there is increasing evidence that both the rate of vertical transmission and the likelihood of impairments change with gestational age at maternal infection, in a fashion similar to other congenital infections like toxoplasmosis or rubella. Thus, several studies indicate that the vertical transmission of CMV is low in the first

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AJOG at a Glance

Why was this study conducted?

This meta-analysis was conducted to explore the temporal correlation between primary maternal infection, vertical transmission, and fetal impairments.

Key findings

The pooled rates of vertical transmission increased from 5.5% at the preconception period to 66.2% at the third trimester. In contrast, the pooled rates of fetal insult in case of transmission were 28.8%, 19.3%, 0.9%, and 0.4%, for maternal infection at the periconception period, first trimester, second trimester, and third trimester, respectively. The pooled rates of sensorineural hearing loss for maternal infection at the first, second, and third trimester were 22.8%, 0.1%, and 0%, respectively.

What does this add to what is known?

Vertical transmission after maternal primary cytomegalovirus infection increases with advancing pregnancy, starting from the preconception period. However, severe fetal impairments are rare after infection in the first trimester of pregnancy.

trimester and progressively increases toward the third trimester.[6–10] Early acquired CMV infections increase the risk of symptomatic neonates at birth.[11–13] This may be even reflected in the pattern of brain lesions, where microcephaly may correspond to infection at <18 weeks' gestation, polymicrogyria between 18 and 24 weeks' gestation, and normal gyration with diffuse white matter heterogeneity at the third trimester.[14] Because the evidence for a temporal trend in both the transmission rates and the risk of fetal impairments is fragmented in mostly small studies, a meta-analysis would be expected to produce more robust results while examining the consistency of the data.

Objectives

The aim of this meta-analysis was to systematically review the association among the timing of maternal infection, vertical transmission, and development of fetal and neonatal impairments and to quantitatively synthesize these data.

Materials and methods

This meta-analysis was performed according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses statement for meta-analyses and registered with the International Prospective Register of Systematic Reviews (CRD42020151679).

Eligibility criteria

Types of studies

Cohort and observational studies reporting the time of CMV infection in the mother and the rate of vertical transmission or fetal impairments were considered eligible for inclusion. Studies assessing the immediate preconception period (3 months) and the duration of pregnancy period were included. No publication date restrictions were imposed, and only studies in European languages were included.

Types of participants

With a timing estimate for the infection available, pregnant women with confirmed primary CMV infection in the immediate preconception period (up to 12 weeks preconception) or during pregnancy were included. Furthermore, fetuses and neonates of these women were included to estimate the vertical transmission rates and the rates of neonates with symptoms at birth.

Types of intervention

There was no intervention.

Types of outcome measures

The main outcomes were the rate of vertical transmission, documented by amniocentesis, and the rate of fetal insult, that is, fetuses that developed impairments because of infection. The latter outcome was assessed in the following 2 ways: (1) overall rate of affected fetuses (including either infants with symptoms at birth or terminations of pregnancy because of the presence of CMV-associated findings in the central nervous system (CNS) on ultrasonography or magnetic resonance imaging [MRI]), and (2) rate of symptomatic infants at birth, defined as the presence of neurologic symptoms in the neonatal period. Secondary outcomes included prenatal CNS findings on ultrasound scans and severe sensorineural hearing loss (SNHL) or neurodevelopmental impairment at follow-up.

Search methods for identification of studies

Eligible studies were identified through a predefined search strategy of electronic databases. We searched the literature (last search January 2020) for cohort and observational studies reporting timing of maternal CMV infection and vertical transmission or fetal impairments. MEDLINE, Scopus, Cochrane Library, and gray literature sources were used to search for related studies. The search and selection criteria were restricted to European languages. The search was carried out using combinations of the terms "cytomegalovirus," "congenital," "fetal," "maternal," "vertical," "transmission," "abnormality," and "termination." Two authors (C.C. and A.S.) independently conducted the literature search, and in case of disagreement, a consensus was reached after discussion. Whenever a consensus could not be reached, a third author offered advice. All studies were carefully screened to avoid inclusion of duplicate or overlapping samples. In case of overlap, the study with the largest number of events was included. There was no limitation concerning the publication dates.

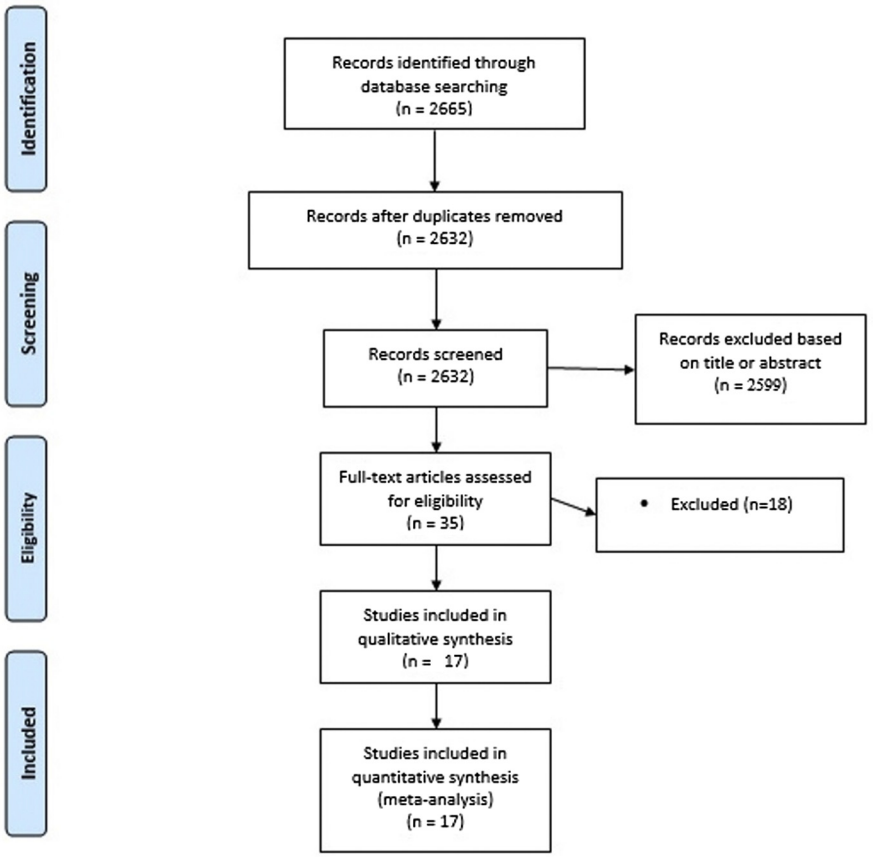
Study selection

Two reviewers independently assessed the eligibility of all identified citations according to the abovementioned criteria. Disagreements between reviewers were resolved by consensus.

Data extraction

Data extraction and assessment of study quality were performed independently

FIGURE 1
Flowchart of the selection process of the included studies



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by 2 authors (C.C. and A.S.). The study characteristics of each included study were assessed according to a predefined data extraction form included in the *Cochrane Handbook for Systematic Reviews*.[15] In case of disagreement, a consensus was reached after discussion between the 2 authors.

The pooled rates of the outcomes of interest were calculated for maternal infection within each of the 5 following periods: (1) preconception period, broadly defined as 3 months before last menstrual period; (2) periconception period, broadly defined as 4 weeks before and 6 weeks after last menstrual period; (3) first trimester; (4) second trimester; and (5) third trimester. As a degree of heterogeneity in the definition of these time periods across studies was anticipated, we recorded and accepted the definitions given in each study. We did so by considering that the impact of this source of heterogeneity would be minimal; in pragmatical terms, the estimation of infection timing is approximate.

Risk of bias in individual studies

Two review authors (C.C. and A.S.) independently assessed the risk of bias in included studies using the Newcastle—Ottawa scale. This scale was developed to assess the quality of nonrandomized studies including cohort studies. Each study was judged on 8 items, categorized into 3 broad groups: the selection of the study groups, the comparability of the groups, and the ascertainment of either the exposure or the outcome of interest.[16] Because all individuals had the same exposure (maternal CMV infection), all included studies received by definition the maximum number of stars for the items "selection of the nonexposed cohort" and "comparability."

Synthesis of the results

The data from each study were extracted, and the number of events in each time period (95% confidence interval [CI]) was estimated for each study in addition to a pooled estimate, weighed by the sample size of each study. This was performed using the Metaprop package in Stata. Metaprop was implemented to perform meta-analyses of proportions in Stata and to perform procedures that were specific to binomial data and allowed computation of exact binomial CIs.[17] Given the nonrandomized design and the anticipated heterogeneity of the studies, we calculated the summary effect sizes using random effects models. The random effects model assumes that the true effect size varies between the studies and that included studies represent a random sample of effect sizes that could have been observed. Therefore, we opted to use this model as it allows for variation not only within studies but also between studies, thus providing a conservative estimate of the summary statistics with wider CIs. Pooled proportions were calculated, and the forest plots were illustrated. The heterogeneity between studies was assessed by the estimation of Cochrane's Q and I^2 statistic.

The unit of analysis (denominator) for the outcome of vertical transmission was exposed fetuses (ie, maternal infections). The corresponding unit for the overall fetal insult was infected fetuses (ie, those in which vertical transmission had already occurred); for the rate of symptoms at birth, it was liveborn fetuses with infection (ie, fetuses with infection after excluding intrauterine losses and terminations of pregnancy).

Additional analyses

A sensitivity analysis was carried out including only studies that were conducted prospectively, had used immunoglobulin G (IgG) avidity assays for the timing of maternal

TABLE 1
Characteristics of the included studies

Study	Patients	Ascertainment of maternal infection	Ascertainment of fetal infection	Ascertainment of presence of symptoms at birth	SNHL and follow-up of the neonates
Bodeus 1999 (retrospective)	Pregnant women with primary cytomegalovirus infection (n=123)	Seroconversion during pregnancy. The women were tested serially (every month). They were assigned to 1 of 3 groups according to the gestational age at the time of seroconversion: seroconversion during the first trimester (25 women), second trimester (49 women), or third trimester (49 women).	Intrauterine infection was assessed by isolating the virus from urine collected within the first 2 weeks of life. The absence of intrauterine infection was assessed by a negative culture.	No information.	No information.
Bodeus 2010 (retrospective)	Pregnant women with seroconversion for cytomegalovirus infection during pregnancy (n=524)	Seroconversion during pregnancy.	Intrauterine infection was assessed by amniocentesis or by isolating the virus from urine or saliva collected within the first 2 weeks of life. The absence of intrauterine infection was assessed by a negative culture.	No information.	No information. Follow-up was obtained in 91% of cases.
Enders 2011 (prospective)	Pregnant women with serologically confirmed primary cytomegalovirus infection (n=248)	(1) Seroconversion of cytomegalovirus-specific IgG and its kinetics; (2) kinetics of cytomegalovirus-specific IgM response; (3) absence or increasing levels of cytomegalovirus neutralizing antibodies; and (4) absence of highly avid cytomegalovirus-specific IgG antibodies or increasing levels of IgG antibody avidity.	In case of spontaneous or therapeutic abortion, congenital infection was either confirmed or excluded by investigation for infectious virus or viral DNA in fetal tissue. Postmortem report.	Presence or absence of clinical abnormalities at birth was systemically recorded for all the liveborn infants.	No information.
Feldman 2011 (prospective)	Pregnant women with primary maternal cytomegalovirus infection (n=508)	Diagnosis of primary maternal CMV infection was exclusively determined by IgG seroconversion (the appearance of de novo–specific IgG antibodies in a previously seronegative patient) or positive specific anti-CMV IgG and IgM antibodies associated with low IgG avidity. Each patient was assigned to the following subgroups: pregestational (12 months to 8 weeks before conception), periconception (between 8 weeks before and 6 weeks after conception), first trimester (up to full 13 weeks' gestation), second trimester (up to full 26 weeks' gestation), and third trimester (gestational age beyond 26 weeks).	Amniocentesis was carried out and neonatal urine was tested after birth for CMV during the first 7 days of life.	All neonates had fundus examination, hearing evaluation, and brain ultrasound scan during the first few days of life.	Hearing evaluation. The parents were referred for long-term follow-up including a physical examination and hearing test in a pediatric infectious diseases unit at a near-home tertiary medical center.

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TABLE 1
Characteristics of the included studies *(continued)*

Study	Patients	Ascertainment of maternal infection	Ascertainment of fetal infection	Ascertainment of presence of symptoms at birth	SNHL and follow-up of the neonates
Feure-Bardon 2019 (prospective)	Pregnant women with primary maternal cytomegalovirus infection (n=255)	Levels of CMV-specific IgM and IgG, seroconversion of cytomegalovirus-specific IgG, levels of IgG antibody avidity.	CMV fetal infection was diagnosed by positive PCR in amniotic fluid. Postmortem report.	Presence or absence of clinical abnormalities at birth was systemically recorded for all the liveborn infants.	Audiological assessment and presence of otitis media. ABRs or audiology. Infected children were followed up including visits at 4, 12, 18, 24, 36, and 48 months.
Foulon 2019 (prospective)	Children with proven congenital CMV infection (n=157)	Maternal seroconversion in CMV IgG occurred during pregnancy. High IgM antibodies and very low IgG antibodies in the beginning of pregnancy.	Isolation of CMV in urine samples, taken within 7 days after birth.	Clinical examination.	The first hearing test was performed during the first month after birth. Hearing evaluation was repeated when possible after 4 and 8 months and at 1 year of age. The tests were repeated annually until the age of 5 years if necessary.
Gindes 2008 (prospective)	All cases of primary cytomegalovirus infection acquired during the third trimester of pregnancy (n=28 pregnant women)	Seroconversion and presence of low avidity anti-CMV IgG later than 25 weeks' gestation.	Fetal diagnosis was made by amniocentesis 6 weeks or more after the maternal seroconversion.	Symptomatic congenital CMV disease was defined by neurologic involvement, including microcephaly, periventricular calcifications, cerebral dysplasia, seizures in an infant with CMV DNA in the cerebrospinal fluid, ventricular and subependymal abnormalities, chorioretinitis, or auditory impairment. All the newborns underwent ophthalmoscopy, cerebral and abdominal ultrasonography, and brainstem auditory evoked responses. Among fetuses with suspected neurologic involvement, cerebrospinal fluid was obtained, and electroencephalography, cerebral CT, and MRI was performed.	By 2 weeks of age, brainstem auditory evoked responses were performed. During the first 2 years of life, brainstem auditory evoked responses were reperformed. Neonates with infection were followed up for a mean of 32 months (range, 6–36 months).

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TABLE 1
Characteristics of the included studies *(continued)*

Study	Patients	Ascertainment of maternal infection	Ascertainment of fetal infection	Ascertainment of presence of symptoms at birth	SNHL and follow-up of the neonates
Hadar 2010 (retrospective)	Pregnant women with periconception cytomegalovirus infection (n=51)	Positive CMV-specific IgM and IgG antibodies along with low IgG avidity or negative IgG antibodies for CMV and positive IgM antibodies for CMV.	Amniocentesis was carried out, and neonatal urine was tested after birth for CMV.	No information.	No information.
Liesnard 2000 (prospective)	Pregnant women with cytomegalovirus infection (n=237)	CMV IgG and IgM antibodies, along with low IgG avidity.	Amniocentesis and fetal blood sample were carried out, and neonatal blood, urine, and saliva samples were tested after birth for CMV. Postmortem report.	Clinical examination, cerebral ultrasounds, ophthalmologic examinations, and hearing evaluations were done.	Hearing evaluation. Pediatric follow-up for at least 2 years.
Lipitz 2010 (prospective)	Pregnant women with cytomegalovirus infection (n=38)	CMV-specific IgG or IgM (or both) in recent serologic assays documented by seroconversion (the de novo appearance of specific IgG and IgM antibodies in a woman who had been seronegative) or a significant rise of IgG antibody titer in the presence of specific IgM antibodies associated with low IgG avidity.	Amniocentesis was carried out, and neonatal urine was tested after birth for CMV. Postmortem report.	During the first 2 weeks, the neonate was evaluated clinically, and the following tests were performed: ocular fundus examination to rule out chorioretinitis and neonatal brain ultrasound scan.	TEOAE and BERA tests. The tests were repeated every 3–4 months until 1 year of age and then twice annually until 3 years of age.
Lipitz 2013 (prospective)	Pregnant women with documented primary CMV infection during the first or second trimester (n=166)	Presence of CMV IgG and IgM antibodies associated with low IgG avidity.	Amniocentesis was carried out, and neonatal urine was tested after birth for CMV. Postmortem report.	During the first 2 postnatal weeks, physical examination, ocular fundus examination, and neonatal brain ultrasound scan were performed.	TEOAE and BERA tests. The tests were repeated every 3–4 months until 1 year of age and then twice annually until 3 years of age.
Pass 2006 (retrospective)	Pregnant with primary CMV infection documented by seroconversion (n=79)	Seroconversion (from serum antibody negative to antibody positive) or detection of serum CMV IgM antibody.	Neonatal urine was tested after birth for CMV.	Physical examination and ocular fundus examination were performed.	Auditory assessment. Infants were followed by the authors in a special clinic that provided serial age-appropriate assessments.
Picone 2013 (retrospective)	Maternal primary CMV infection (n=238)	Serologic criteria: IgG seroconversion or positive IgM with low IgG avidity.	Amniocentesis and fetal blood sample were carried out, and neonatal samples were tested after birth for CMV. Postmortem report.	Physical examination.	No information.
Puhakka 2017 (retrospective)	Patients with symptomatic congenital CMV infection (n=26)	Serologic criteria: IgG seroconversion or positive IgM with low IgG avidity.	No information.	Physical examination and hearing, vision, or neurology examination were performed.	Hearing test. 5 years follow-up.

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TABLE 1
Characteristics of the included studies *(continued)*

Study	Patients	Ascertainment of maternal infection	Ascertainment of fetal infection	Ascertainment of presence of symptoms at birth	SNHL and follow-up of the neonates
Rovello 2002 (retrospective)	Pregnant women with primary CMV infection (n=182)	CMV-specific IgM antibody, increasing levels of IgG antibody avidity, detection of CMV and CMV products in blood.	Amniocentesis and neonatal samples were tested after birth for CMV. Postmortem report.	Physical examination.	No information.
Rovello 2011 (retrospective)	Pregnant women with primary CMV infection (n=735)	Seroconversion (ie, de novo appearance of specific antibodies in a previously seronegative individual), kinetics of CMV-specific IgM and IgG antibody, low IgG avidity index, and detection of CMV and CMV products in blood.	Amniocentesis and culdocentesis. Postmortem report.	No information.	No information.
Stagno 1986 (prospective)	Pregnant women with primary CMV infection (n=96)	Primary CMV infection was defined as seroconversion (de novo appearance of IgG antibodies) during pregnancy.	Isolation of CMV from urine or saliva or both in the first week of life and at 2, 4, or 6 months of age.	No information.	No information.

ABR, auditory brainstem response; *BERA*, brainstem evoked response audiometry; *CMV*, cytomegalovirus; *CT*, computed tomography; *IgG*, immunoglobulin G; *IgM*, immunoglobulin M; *MRI*, magnetic resonance imaging; *PCR*, polymerase chain reaction; *SNHL*, sensorineural hearing loss; TEOAE, transient evoked otoacoustic emissions.

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infection, and had postmortem ascertainment.

Results

Study selection

The electronic search from the databases yielded 2665 potential studies; 2632 of those searches were excluded based on their title or abstract or because they were duplicates, leaving 35 studies for full-text review. After the full-text review, we finally included 17 studies (Figure 1).[6–14,18–26] The excluded studies with reasons are shown in Supplemental Table 1, and the characteristics of the included studies are shown in Table 1.

Overall, 9 studies were conducted prospectively,[12,14,19,20,22,23,27,28] and 11 studies assessed the timing of maternal infection with IgG avidity assays.[7–10,12,14,22,23,25–27] Postmortem examination of the aborted fetuses was carried out in 8 studies.[8,10,12,14,22,23,26,27] Long-term follow-up, up to 5 years for sequential manifestations of infection, was carried out in 7 studies.[12,14,20,22,23,25,29] Notably, 5 studies assessed the outcome of SNHL with the brainstem evoked response audiometry test.[12,14,20,23,29]

Assessment of the risk of bias

The methodological quality of studies was assessed using the Newcastle-Ottawa Scale . The rating of the included studies is listed in Table 2. All studies had adequate outcome assessment methods, ascertainment of exposure, and incident of disease. A total of 7 studies had adequate follow-up for long-term complications.[6,14,19,20,23–25]

Synthesis of results

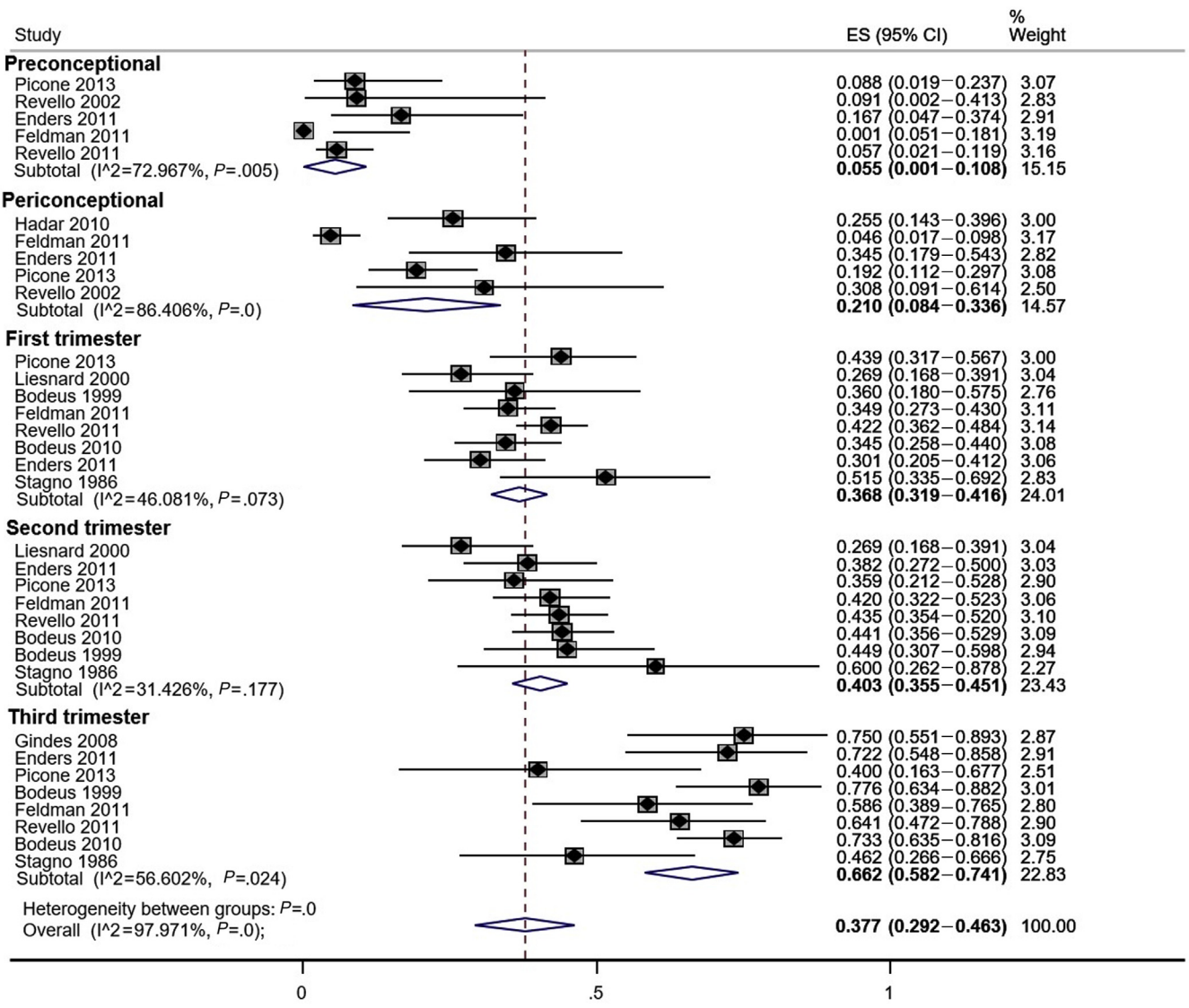
Primary outcomes

Data from 10 studies (N=2942) were used in the evaluation of the maternal to fetal CMV transmission.[6–11,18,22,26,30] Maternal infection in the preconception period was associated with a 5.5% (95% CI, 0.1–10.8; I^2=73%) rate of vertical transmission. In the periconception period, the pooled rate of vertical transmission was 21.0% (95% CI, 8.4–33.6; I^2=86%). In the first trimester, the pooled the rate of vertical transmission was 36.8% (95% CI, 31.9–41.6; I^2=46%). In the second trimester, the pooled rate of vertical transmission was 40.3% (95% CI, 35.5–45.1; I^2=31%). In the third trimester, the pooled rate of vertical transmission was 66.2% (95% CI, 58.2–74.1; I^2=56%) (Figure 2).

Data from 10 studies (N=796) were used to calculate the rate of fetal insult (ie, overall affected fetuses and fetuses with infection), defined as either presence of neurologic symptoms at birth or termination of pregnancy because of CMV-associated findings in the CNS on ultrasonography or magnetic resonance imaging.[7,8,11,12,14,20,22,23,26,30] The pooled rate of fetal insult after maternal infection in the periconception period was 28.8% (95% CI, 2.4–55.1; I^2=80%). In the first trimester, the rate was 19.3% (95% CI, 12.2–26.4; I^2=51%). In the second trimester, the rate was 0.9% (95% CI, 0–2.4; I^2=16%). The affected cases in the second trimester are presented in Supplemental Table 2. There were no accurate timing data for these cases, and second trimester could be anywhere between 14 and 26 weeks gestation. In the third trimester, the rate was 0.4% (95% CI, 0–1.5; I^2=0%) (Figure 3; Table 3).

The pooled rate of symptoms at birth (per fetuses with infection, excluding terminations of pregnancy)

FIGURE 2
Forest plot for the primary outcome of vertical transmission



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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was 1.3% (95% CI, 0−4.6; I^2=0%) for maternal infection in the periconception period, 9.1% (95% CI, 2.7−15.4; I^2=62%) in the first trimester, 0.3% (95% CI, 0−1.1; I^2=0%) in the second trimester, and 0.4% (95% CI, 0−1.6; I^2=0%) in the third trimester (Figure 4; Table 4).

Secondary outcomes

Data from 6 studies (N=1215) were used in the evaluation of the prevalence of CNS ultrasonography findings.[8,9,14,20,22,23] Maternal infection in the periconception period resulted in 30.0% (95% CI, 9.9−50.0) prevalence of CNS findings in fetuses with infection. The pooled prevalence of CNS ultrasound findings was 10.0% (95% CI, 1.0−19.0; I^2=75%) after first-trimester maternal infection, 0.3% (95% CI, 0−1.2; I^2=0%) after second-trimester maternal infection, and 0.5% (95% CI, 0−2.3; I^2=0%) after third-trimester maternal infection (Supplemental Figure 1).

Data from 6 studies (N=772) were used to assess the presence of severe

TABLE 2
Risk of bias assessment for the included studies

	Selection					Outcome		
Study	Representativeness of the exposure (intervention) cohort[a]	Selection of the nonexposed cohort[b]	Ascertainment of exposure[c]	Incident disease[d]	Comparability[e]	Assessment of outcome[f]	Length of follow-up[g]	Adequacy of follow-up[h]
Bodeus 1999	B	A	A	A	A	B	A	D
Bodeus 2010	B	A	A	A	A	B	A	B
Enders 2011	B	A	A	A	A	A	A	D
Feldman 2011	B	A	A	A	A	A	A	D
Feure-Bardon 2019	B	A	A	A	A	A	A	B
Foulon 2019	B	A	A	A	A	A	A	A
Gindes 2008	B	A	A	A	A	A	A	A
Hadar 2010	C	A	A	A	A	A	A	D
Liesnard 2000	A	A	A	A	A	A	A	B
Lipitz 2010	A	A	A	A	A	A	A	A
Lipitz 2013	B	A	A	A	A	A	A	A
Pass 2006	B	A	A	A	A	A	A	C
Picone 2013	B	A	A	A	A	B	A	D
Puhakka 2017	C	A	A	A	A	B	A	A
Rovello 2002	B	A	A	A	A	A	A	D
Rovello 2011	B	A	A	A	A	B	A	D
Stagno 1986	B	A	A	A	A	B	A	D

[a] A, truly representative of the average pregnant woman with CMV infection; B, somewhat representative of the average pregnant woman with CMV infection; C, selected group; D, no description of the derivation of the cohort; [b] A, drawn from the same source as the intervention cohort (concurrent controls); B, drawn from a different source (historical controls); C, no description of the derivation of the nonexposed cohort; [c] A, secure record (eg, hospital records); B, structured interview; C, written self-report; D, no description; [d] Demonstration that outcome of interest was not present at the start of the study: A, yes; B, no; [e] Comparability of cohorts on the basis of the design or analysis: A, study controls for the most important factor; B, study controls for any additional factor; C, not carried out or not reported; [f] A, independent blind assessment; B, record linkage; C, self-report; D, no description; [g] Was follow-up long enough for outcomes to occur? A, yes; B, no; [h] A, complete follow-up; all subjects were accounted for. B, Subjects lost to follow-up were unlikely to introduce bias because small numbers were lost; >90% had follow-up, or description was provided of those lost. C, follow-up rate <90%, and there was no description of those lost. D, no statement.

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SNHL or neurodevelopmental impairment at follow-up according to timing of infection.[12,14,19,23,25,28] Apart from 1 study,[19] all other studies assessed both outcomes. The follow-up in the included studies ranged from 6 months to 5 years. Details about the methods used for the assessment of hearing and neurodevelopment are given in Table 1. Maternal infection in the first trimester resulted in 22.8% (95% CI, 15.4−30.2; I^2=27.6%) pooled prevalence of SNHL or neurodevelopmental impairment. The corresponding pooled rates after second- and third-trimester infection were 0.1% (95% CI, 0−0.8; I^2=24%) and 0% (95% CI, 0−0.1; I^2=0%), respectively (Supplemental Figure 2; Table 5).

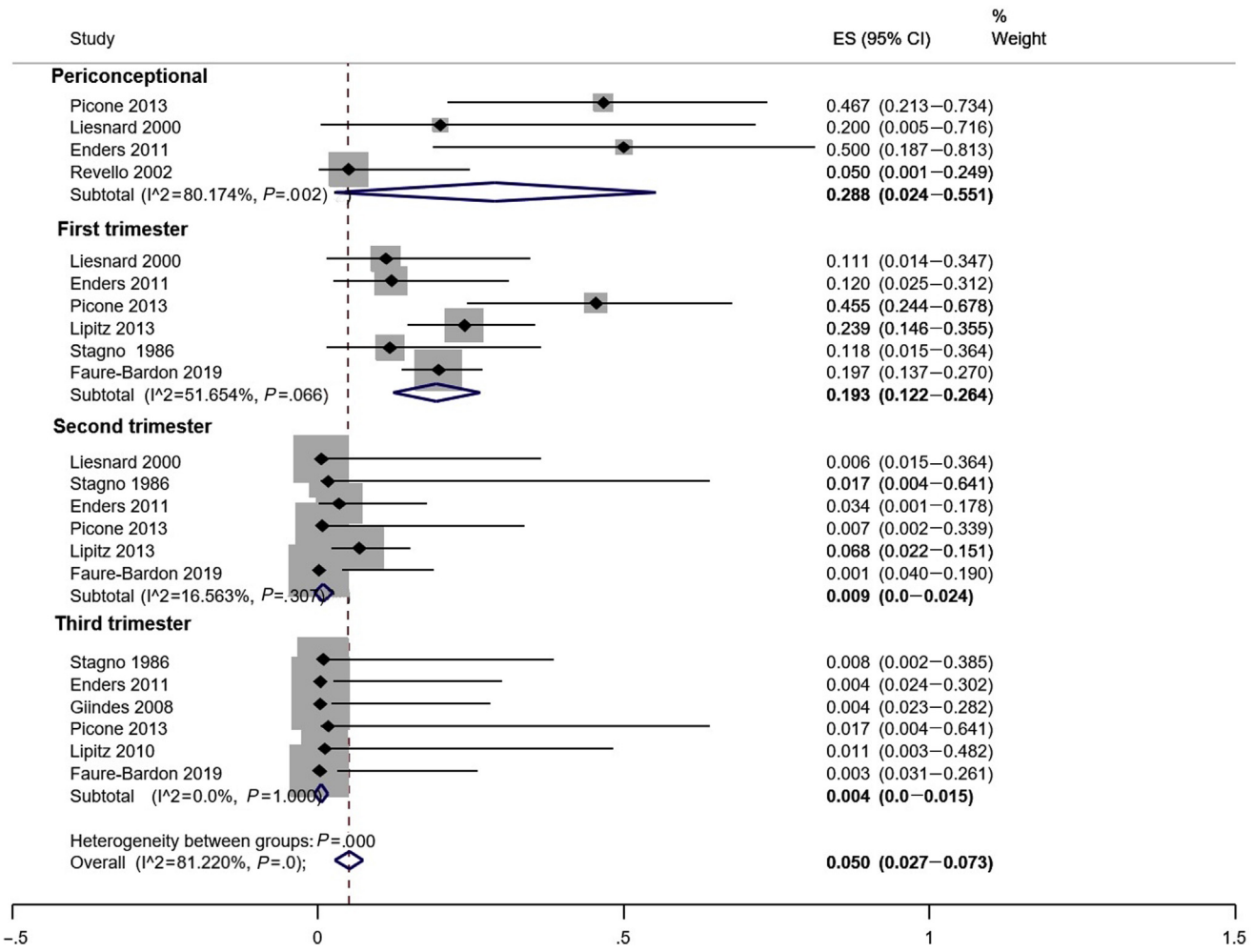
Additional analyses

A post-hoc sensitivity analysis was performed including only studies that were conducted prospectively, had used IgG avidity assays for the timing of maternal infection, and had postmortem ascertainment of prenatal findings. Notably, 7 studies[9,11,12,14,20,22,23] fulfilled these criteria and were subsequently included in the sensitivity analysis (Table 1).

Four studies[9,11,20,22] (N=1018) reported data for the outcome of vertical CMV transmission. The pooled rate was 0.1% (95% CI, 0−0.8) in the preconception period, 5.9% (95% CI, 2.3−9.4) in the periconception period, 31.6% (95% CI, 26.4−36.8) in the first trimester, 35.8% (95% CI, 26.9−44.8) in the second trimester, and 69.5% (95% CI, 60.3−78.8) in the third trimester (Supplemental Figure 3).

Six studies[11,12,14,20,22,23] (N=972) were used for the primary outcome of fetal insult in fetuses with infection. The pooled rate was 36.8% (95% CI, 13.6−60.1) if infection occurred in the periconception period, 19.0% (95% CI, 14.4−23.6) in the first trimester, 1.6% (95% CI, 0−4.1) in the second trimester, and 0.3% (95% CI, 0−1.5) in

FIGURE 3
Forest plot for the primary outcome of fetal insult



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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the third trimester (Supplemental Figure 4).

Six studies[11,12,14,20,22,23] (N=972) were used for the outcome of neurologic symptoms at birth. The pooled rate of the sensitivity analysis was 4.8% (95% CI, 0−19.6) in the periconception period, 7.8% (95% CI, 0.6−15.0) in the first trimester, 0.5% (95% CI, 0−1.8) in the second trimester, and 0.4% (95% CI, 0−1.6) in the third trimester (Supplemental Figure 5).

Five studies (N=977)[9,14,20,22,23] were used for the outcome of CMV-related fetal CNS ultrasonography findings. The pooled rate was 18.1% (95% CI, 0−40.8) in the periconception period, 5.1% (95% CI, 1.3−9.0) in the first trimester, 0.3% (95% CI, 0−1.2) in the second trimester, and 0.5% (95% CI, 0−2.3) in the third trimester (Supplemental Figure 6).

Four studies (N=616)[12,14,23,29] were used for the outcome of severe SNHL or neurodevelopmental delay at follow-up. The median follow-up time in the included studies was 24,[14,23] 31,[29] and 48 months.[12] The pooled rate of severe SNHL or neurodevelopmental impairment was 19.7% (95% CI, 14.3−25.2) in the first trimester, 2.1% (95% CI, 0−5.9) in the second trimester, and 0% (95% CI, 0−0.1) in the third trimester (Supplemental Figure 7).

Finally, we attempted to account for the different definitions of gestational periods at maternal infections by performing a regression analysis, using the median gestational age at each period, in each study, as a central estimate for time. Because this information was commonly

TABLE 3
Risk of CMV congenital infection (transmission) and fetal insult, according to gestational age at maternal primary infection

	Transmission rate	Fetal insult if fetus is infected	Fetal insult if transmission is unknown
Periconception	21% (95% CI, 8.4–33.6)	28.8% (95% CI, 2.4–55.1)	6.0%
First trimester	36.8% (95% CI, 31.9–41.6)	19.3% (95% CI, 12.2–26.4)	7.1%
Second trimester	40.3% (95% CI, 35.5–45.1)	0.9% (95% CI, 0–2.4)	0.4%
Third trimester	66.2% (95% CI, 58.2–74.1)	0.4% (95% CI, 0–1.5)	0.3%

CMV, cytomegalovirus.

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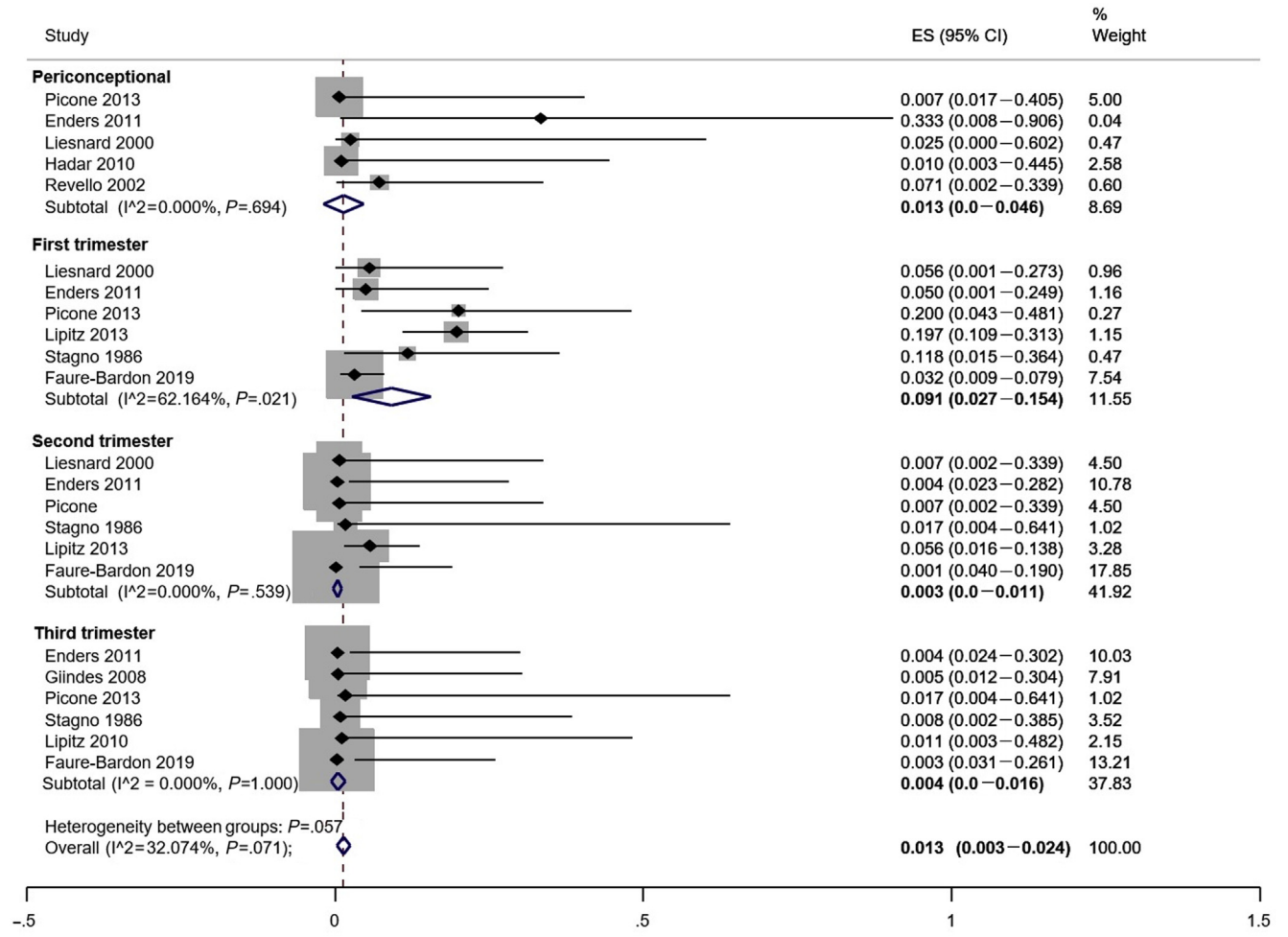
FIGURE 4
Forest plot for the primary outcome of symptoms at birth



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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TABLE 4
Risk of CMV congenital infection (transmission) and symptomatic at birth, according to gestational age at maternal primary infection

	Transmission rate	Symptomatic at birth if fetus infected	Symptomatic at birth if transmission unknown
Periconception	21.0% (95% CI, 8.4–33.6)	1.3% (95% CI, 0–4.5)	0.3%
First trimester	36.8% (95% CI, 31.9–41.6)	9.1% (95% CI, 2.7–15.6)	3.3%
Second trimester	40.3% (95% CI, 35.5–45.1)	0.3% (95% CI, 0–1.1)	0.1%
Third trimester	66.2% (95% CI, 58.2–74.1)	0.4% (95% CI, 0–1.6)	0.3%

CMV, cytomegalovirus.

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not available in the published text, we contacted all authors of the primary studies. Only the authors of 2 studies[25,27] offered additional information; therefore, this analysis could not be implemented.

Comment

Summary of the evidence

Pooling data from 10 observational studies, we found that vertical transmission of CMV infection progressively increases as pregnancy progresses, from 5% in the few weeks preceding conception to 60% in the third trimester. However, severe sequalae are only common when maternal infection occurs in the periconception period and first trimester (29% and 19%, respectively), dropping to approximately 1% after this point.

Interpretation

Vertical transmission and fetal insult rates after maternal CMV infection change with gestational age at infection and are inversely correlated with each other.

The patterns of transmission and impairments are physiologically explained by the evolving structure and function of the placenta and the fetal organs and immunologic systems. The placenta is the first fetal organ to be infected. Virions spread to the cytotrophoblasts in floating and anchoring villi, in the form of low avidity immune complexes, and infect the syncytiotrophoblast.[31,32] Villus CMV infection depends on the expression of functional receptors in distinct populations of progenitor and differentiating cells.[33] Susceptibility to infection increases as cytotrophoblasts proceed along the differentiation pathway,[34] which explains the increased rate of vertical transmission with advancing gestation. Data from nonpregnant populations show that viremia (hence maternal viremia) peaks at approximately 7 weeks and persists for approximately 12 weeks after primary infection,[35] which explains the pooled transmission rate of 20% when maternal infection occurs in the periconception period.

In contrast to transmission trends, the likelihood of fetal impairments is higher when CMV infection happens early in pregnancy. CNS invasion and damage are the cause of chronic disability.[36,37] The human brain begins to develop as a thin sheet of neuroepithelial cells that proliferate rapidly to form the neural tube, as early as 4 weeks gestation.[38] Studies in human embryonic cultures found that at this early stage of development, CMV infection leads to impaired cell proliferation, differentiation, and increased frequency of cell death in neural precursor cells.[39–43] Disruption

TABLE 5
Risk of CMV congenital infection (transmission) and SNHL or neurodevelopmental impairment, according to gestational age at maternal primary infection

	Transmission rate	SNHL or neurodevelopmental impairment if fetus is infected	SNHL or neurodevelopmental impairment if transmission is unknown
First trimester	36.8% (95% CI, 31.9–41.6)	22.8% (95% CI, 15.4–30.2)	8.4%
Second trimester	40.3% (95% CI, 35.5–45.1)	0.1% (95% CI, 0–0.8)	0%
Third trimester	66.2% (95% CI, 58.2–74.1)	0% (95% CI, 0–2.1)	0%

CMV, cytomegalovirus; *SNHL*, sensorineural hearing loss.

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of the CNS development in the early stages could be the cause of the neurologic complication, and possibly, the stage of embryonic development when the congenital infection occurs is the main determinant for the severity of the brain disorder.[44] CMV infections at different gestational ages may have a distinct pattern of cellular and developmental effects on the brain that may ultimately determine the neurologic outcomes.[45] Our pooled proportions for fetal damage in case of vertical transmission progressively decreased, from 28.8% for infection at the periconception period to 19.3% at the first trimester, 0.9% at second trimester, and 0.4% at the third trimester. Specifically, for severe SNHL or neurodevelopmental impairment at follow-up, the likelihood in liveborn fetuses with infection decreased from 22.8% at the first trimester to 0.1% at the second trimester and 0% at the third trimester. Interestingly enough, lesser degrees of SNHL were also reported for infection beyond the first trimester, but severe impairment or deafness was only limited to first-trimester insults.[14,29] Although transmission was possible for maternal infection as far as 3 months before conception (pooled rate 5.5%), the included studies did not report rates of fetal damage so remote from conception.

When maternal CMV infection is diagnosed at the first or early second trimester, the healthcare provider cannot know whether transmission has or will occur, because amniocentesis is performed no earlier than 20 weeks' gestation. Therefore, for counseling purposes, when the status of transmission is unknown, the risk of fetal damage after maternal infection is approximately 6% in the periconception period, reaches a peak of 7.1% at the first trimester, and then progressively declines, reaching a nadir of <1% at the third trimester.

Strengths and limitations

To assess the feasibility of quantitative synthesis, we thoroughly examined the definition criteria for vertical transmission and fetal damage (prenatal or symptoms at birth) in each of the examined studies. Furthermore, studies reporting results for SNHL or neurodevelopmental impairment were evaluated based on the diagnostic methodology that was used and the length of the follow-up (Table 1).

Our analysis had 3 major limitations. First, although CMV infection is the most common congenital infection, the data assessing the relationship between timing of maternal infection and natural history in the fetus are comparatively few. As a result, the CIs were wide and occasionally included zero. Second, accurate timing of maternal infection was not always feasible, because most immunocompetent women were asymptomatic and serologic testing offers only approximate timing. The timing of maternal infection was determined according to the gestational age at the time of seroconversion, and only 3 studies report the median gestation age of the pregnant women at each time period.[7,20,26] Therefore, the homogeneity within the time periods could not be assessed. However, to increase precision, we performed a sensitivity analysis including only studies that were conducted prospectively and had used IgG avidity assays for timing of maternal infection. Finally, it is not feasible to accurately determine the rate and severity of fetal impairments, because prenatal ultrasonography findings do not always correspond to the severity of symptoms at birth and some of the terminated fetuses might not be as severely affected at birth as prenatally expected. We accounted for the latter issue by perusing the individual details of terminated fetuses, when available, and only including those with CMV-associated substantial findings in the CNS on imaging. Studies not reporting individual data of terminated fetuses were excluded.

Conclusions

Vertical transmission after maternal primary CMV infection increases with advancing pregnancy, starting from the preconception period. However, severe fetal impairments are rare after infection in the first trimester of pregnancy. ■

ACKNOWLEDGMENTS

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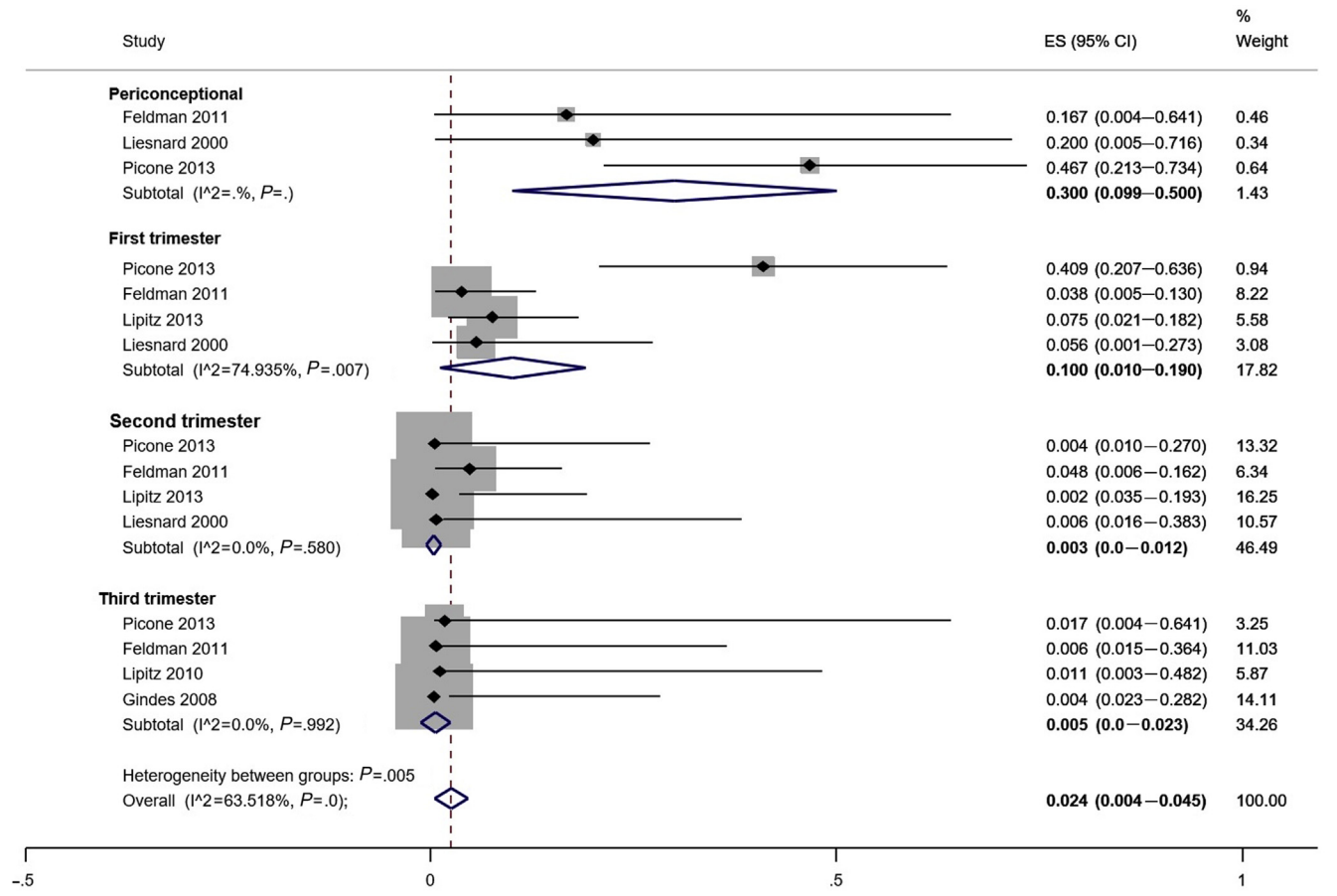
SUPPLEMENTAL FIGURE 1
Forest plot for the secondary outcome of ultrasound findings



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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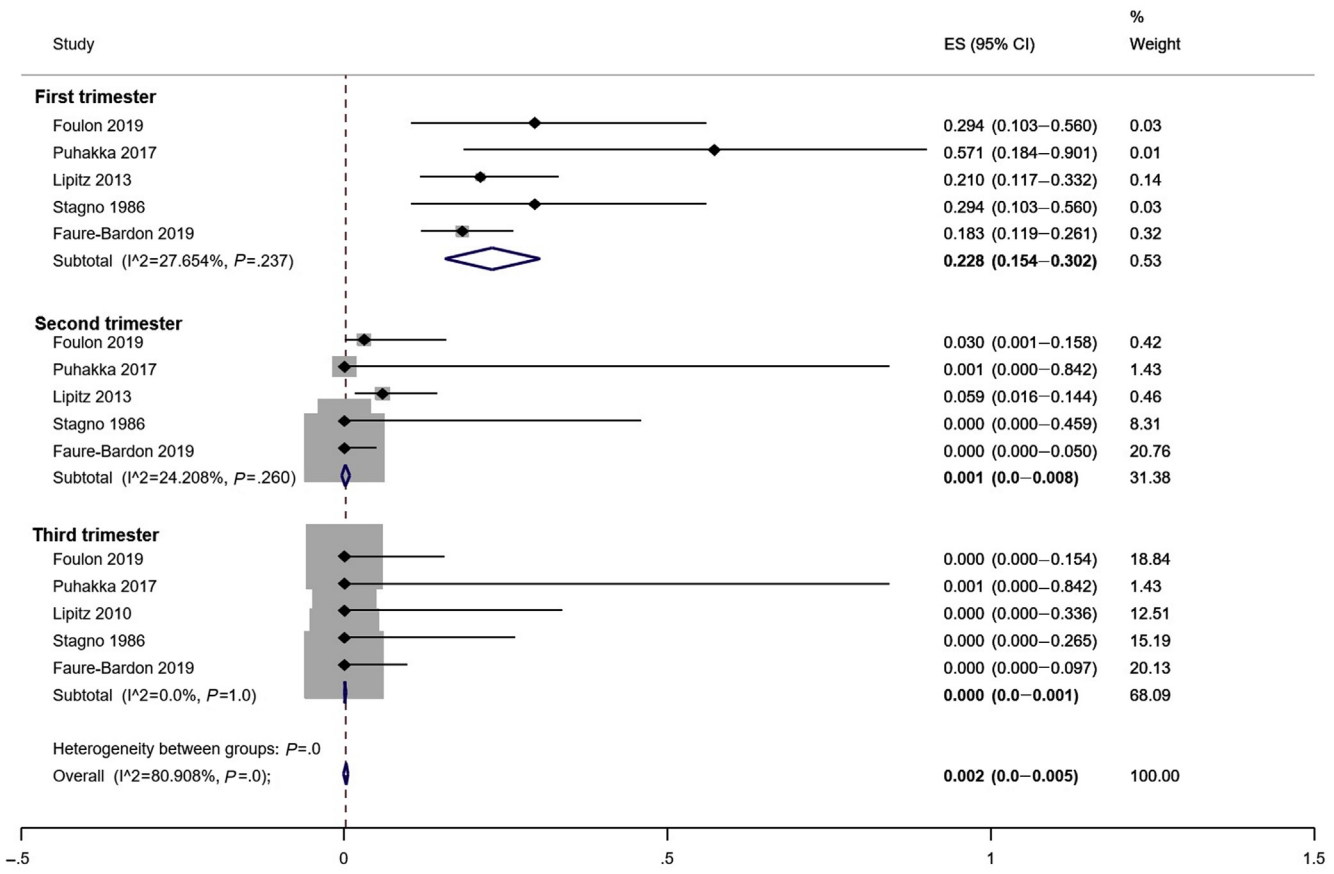
SUPPLEMENTAL FIGURE 2

Forest plot for the secondary outcome of SNHL or neurodevelopmental impairment



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a black diamond, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio; *SNHL*, sensorineural hearing loss.

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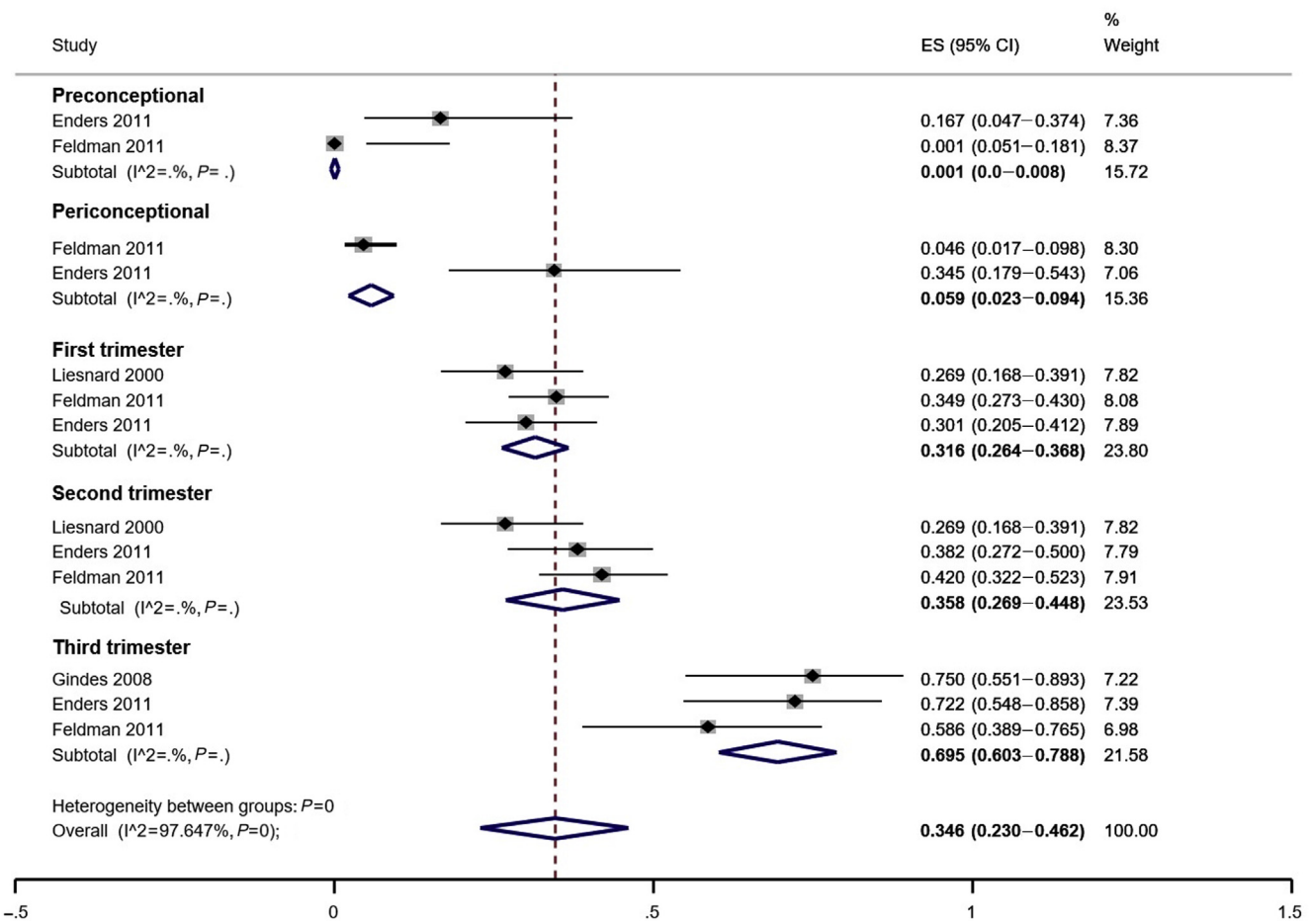
SUPPLEMENTAL FIGURE 3

Forest plot of the sensitivity analysis, for the primary outcome of vertical transmission



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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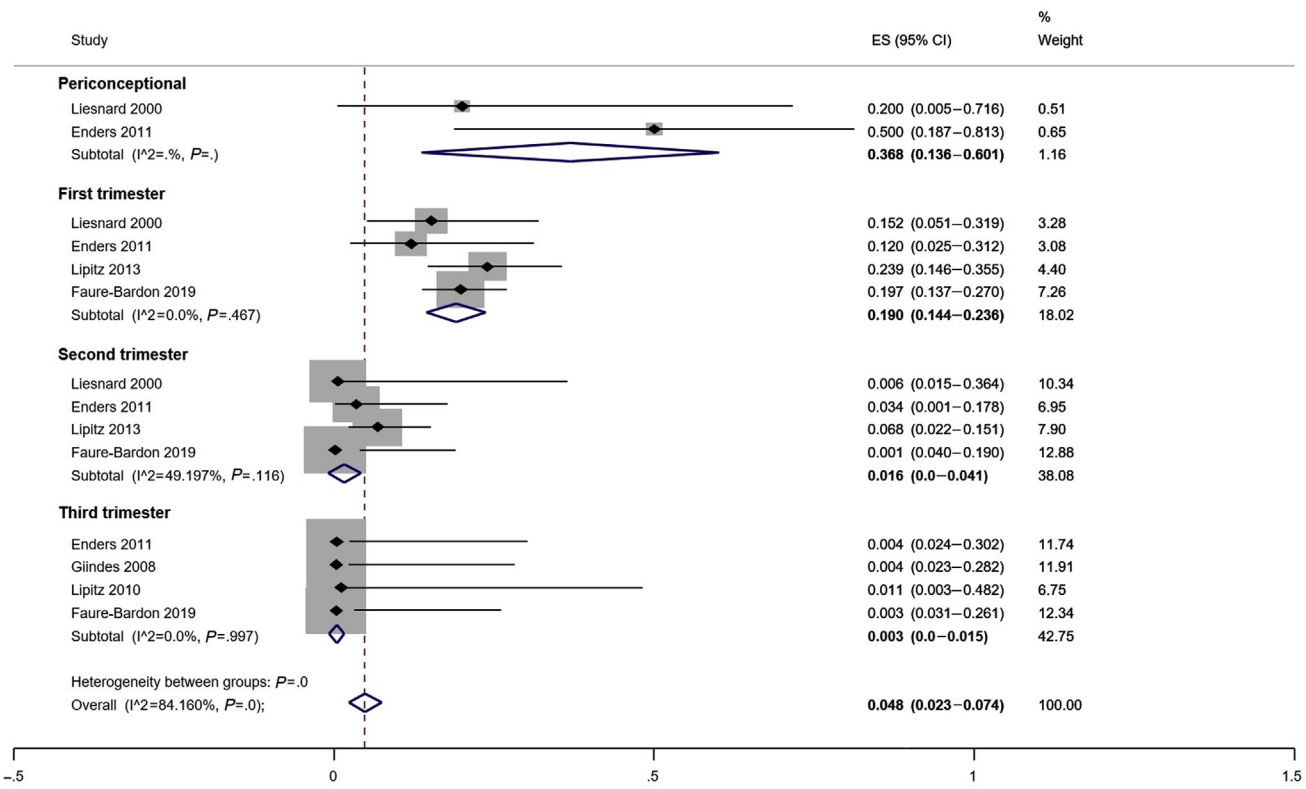
SUPPLEMENTAL FIGURE 4

Forest plot of the sensitivity analysis, for the primary outcome of fetal insult



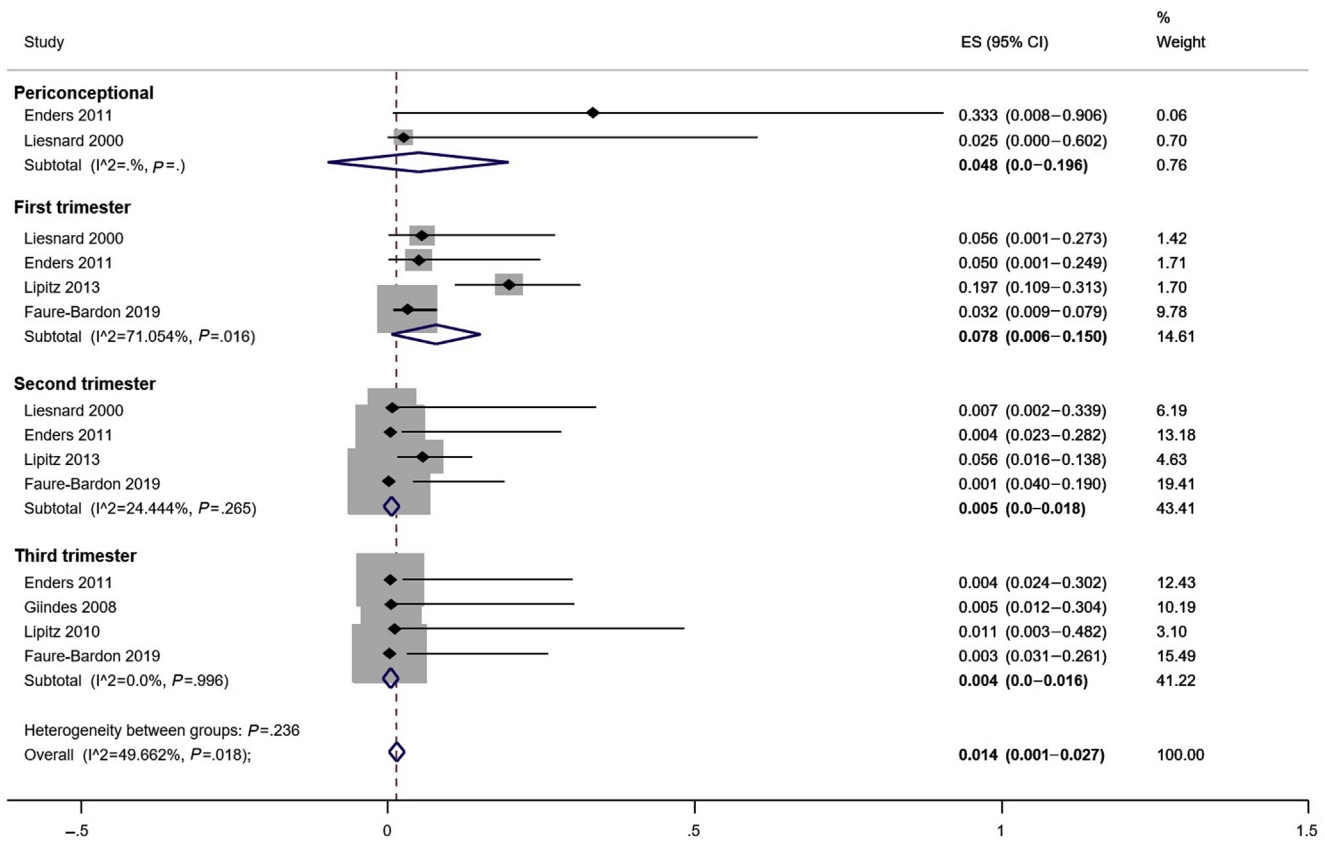
Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

Chatzakis. Timing of primary maternal cytomegalovirus infection and rates of vertical transmission and fetal consequences. Am J Obstet Gynecol 2020.

SUPPLEMENTAL FIGURE 5

Forest plot of the sensitivity analysis, for the primary outcome of symptoms at birth



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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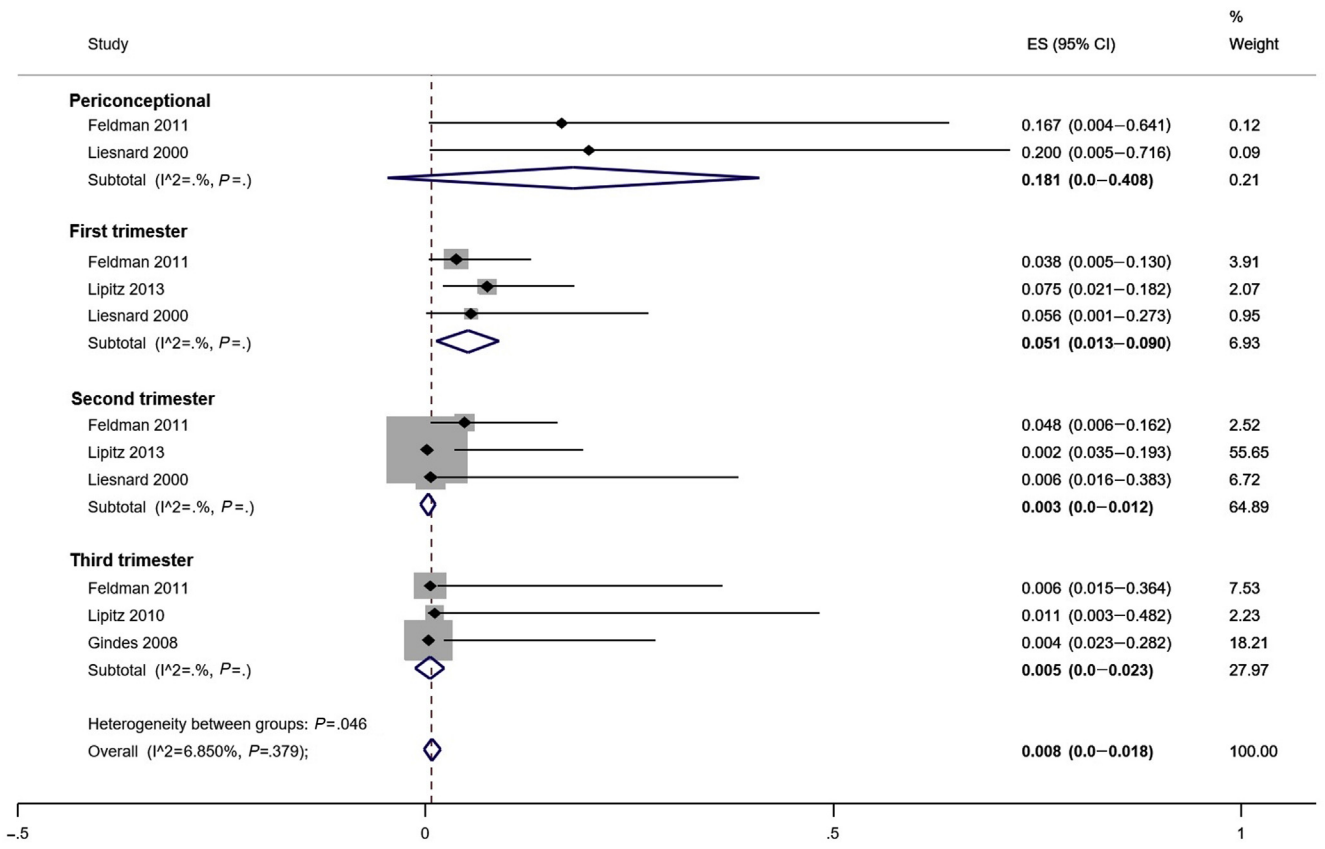
SUPPLEMENTAL FIGURE 6

Forest plot of the sensitivity analysis, for the secondary outcome of ultrasound findings



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio.

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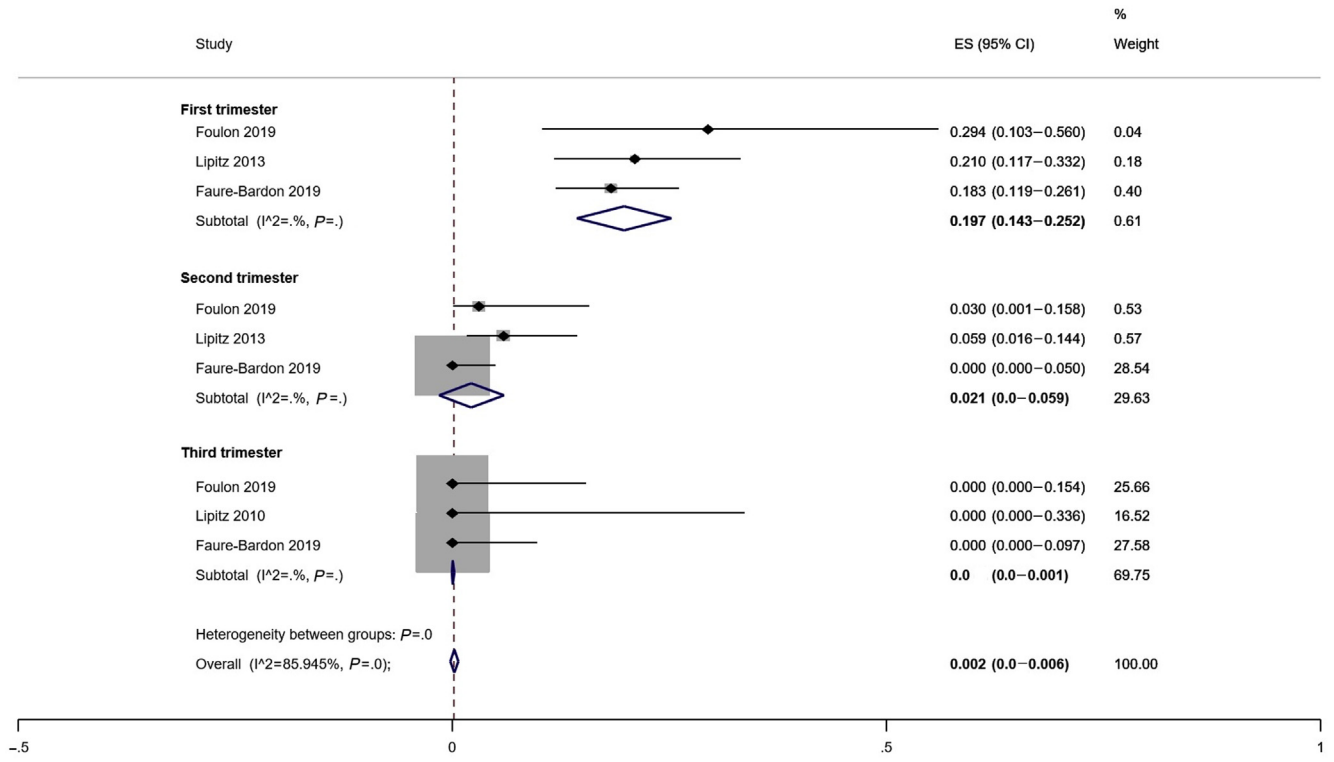
SUPPLEMENTAL FIGURE 7

Forest plot of the sensitivity analysis, for the secondary outcome of SNHL and/or neurodevelopmental impairment



Proportions (95% CIs) for each study are denoted by *black boxes* (*black lines*). The combined proportion estimate for all studies is represented by a *black diamond*, where *diamond* width corresponds to 95% CI bounds. *Box* and *diamond* heights are inversely proportional to precision of the OR estimate. The *P* value for heterogeneity (*P* het) is shown. For each period, there is a separate *diamond* that summarizes the proportion estimate for all studies in that period. At the bottom of the figure, the last *diamond* combines the proportion estimates of all periods.

CI, confidence interval; *OR*, odds ratio; *SNHL*, sensorineural hearing loss.

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SUPPLEMENTAL TABLE 1
Excluded studies with reason

Balcells 2016[1]	Population of interest was preterm infants
Bodeus 2002[2]	Overlap with Bodeus 2011
Daiminger 2005[3]	Overlap with Enders 2011
Enders 2001[4]	Overlap with Enders 2011
Enders 2017[5]	Treatment intervention with intravenous administration of CMV HIG or maternal-fetal valganciclovir
Faure-Bardon 2018[6]	Pool estimates of other studies
Faulon 2008[7]	Overlap with Foulon 2019
Griffiths 1984[8]	Case-control study
Guerra 2008[9]	No information about the time of the transmission
Leruez-Ville 2017[10]	No information about the time of the transmission
Leruez-Ville 2016[11]	Intervention study assessing the efficacy of valacyclovir
Lipitz 2002[12]	No information about the time of the transmission
Preece 1983[13]	Case-control study
Revello 1998[14]	No outcomes of interest
Revello 2002[15]	Not a clinical study, review
Revello 2006[16]	Overlap with Revello 2011
Revello 2014[17]	Treatment intervention with intravenous administration of CMV HIG
Stagno 1982[18]	Overlap with Stagno 1986

CMV, cytomegalovirus; *HIG*, hyperimmune globulin.

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SUPPLEMENTAL TABLE 2
PRISMA 2019 checklist

Section or topic	#	Checklist item	Reported on page #
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	1
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable, background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; and systematic review registration number.	3—4
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	5
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	5
METHODS			
Protocol and registration	5	Indicate if a review protocol exists and if and where it can be accessed (eg, Web address), and, if available, provide registration information including registration number.	5
Eligibility criteria	6	Specify study characteristics (eg, PICOS, length of follow-up) and report characteristics (eg, years considered, language, publication status) used as criteria for eligibility, giving rationale.	6
Information sources	7	Describe all information sources (eg, databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.	6
Search	8	Present full electronic search strategy for at least 1 database, including any limits used, such that it could be repeated.	6—7
Study selection	9	State the process for selecting studies (ie, screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).	7
Data collection process	10	Describe method of data extraction from reports (eg, piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.	7
Data items	11	List and define all variables for which data were sought (eg, PICOS, funding sources) and any assumptions and simplifications made.	7
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level) and how this information is to be used in any data synthesis.	7—8
Summary measures	13	State the principal summary measures (eg, risk ratio, difference in means).	8
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency (eg, I^2) for each meta-analysis.	8

Chatzakis. Timing of primary maternal cytomegalovirus infection and rates of vertical transmission and fetal consequences. Am J Obstet Gynecol 2020. *(continued)*

SUPPLEMENTAL TABLE 2
PRISMA 2019 checklist *(continued)*

Section or topic	#	Checklist item	Reported on page #
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence (eg, publication bias, selective reporting within studies).	8
Additional analyses	16	Describe methods of additional analyses (eg, sensitivity or subgroup analyses, meta-regression), if done, indicating which were prespecified.	9
RESULTS			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	9
Study characteristics	18	For each study, present characteristics for which data were extracted (eg, study size, PICOS, follow-up period) and provide the citations.	9
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).	9
Results of individual studies	20	For all outcomes considered (benefits or harms), present, for each study, (a) simple summary data for each intervention group and (b) effect estimates and confidence intervals, ideally with a forest plot.	9–10
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	9–12
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies (see item 15).	9
Additional analysis	23	Give results of additional analyses, if done (eg, sensitivity or subgroup analyses, meta-regression [see item 16]).	11–12
DISCUSSION			
Summary of evidence	24	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (eg, healthcare providers, users, and policy makers).	12
Limitations	25	Discuss limitations at study and outcome level (eg, risk of bias) and at review level (eg, incomplete retrieval of identified research, reporting bias).	14
Conclusions	26	Provide a general interpretation of the results in the context of other evidence and implications for future research.	15
FUNDING			
Funding	27	Describe sources of funding for the systematic review and other support (eg, supply of data) and role of funders for the systematic review.	15

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