Risk factors for natural hearing evolution in newborns with congenital cytomegalovirus infection

RISK FACTORS FOR NATURAL HEARING EVOLUTION IN NEWBORNS WITH CONGENITAL CYTOMEGALOVIRUS INFECTION

Elise De Cuyper[a], MD, Frederic Acke[a,b], MD, PhD, Annelies Keymeulen[c], MD, PhD, Els De Leenheer[a,b], MD, PhD, Helen Van Hoecke[a,b], MD, PhD, Elizaveta Padalko[d], MD, PhD, An Boudewyns[e,f], MD, PhD, Annick Gilles[e,f,g], MSc, PhD, Marie Muylle[h], MSc, Rudolf Kuhweide[h], MD, Liesbeth Royackers[i], MSc, Christian Desloovere[i], MD, PhD, Margriet Verstreken[j], MD, Isabelle Schatteman[j], MD, Ingeborg Dhooge[a,b], MD, PhD

Affiliations:

a Department of Head and Skin, Ghent University, Ghent, Belgium
b Department of Otorhinolaryngology, Ghent University Hospital, Ghent, Belgium
c Department of Neonatal Intensive Care Unit, Ghent University Hospital, Ghent, Belgium
d Laboratory of Medical Microbiology, Ghent University Hospital, Ghent, Belgium
e Faculty of Medicine and Translational Neurosciences, University of Antwerp, Antwerp, Belgium
f Department of otorhinolaryngology Head and Neck Surgery, Antwerp University Hospital, Antwerp, Belgium
g Department of Education, Health and Social Work, University College Ghent, Ghent, Belgium
h Department of Ear, Nose and Throat, Sint Jan Hospital, Bruges, Belgium
i Department of Otorhinolaryngology, Head and Neck Surgery, University Hospitals of Leuven, Belgium
j Department of Ear, Nose and Throat, GZA hospitals campus Sint Augustinus, Wilrijk, Belgium

Address correspondence to:
Elise De Cuyper
Dept of Head and Skin, Ghent University
Corneel Heymanslaan 10, entrance 69, route 692 (PB NKO)
9000 Ghent
Belgium
elise.decuyper@ugent.be
tel: +329332.23.32

KEY POINTS

Question: Which risk factors are associated with hearing improvement, hearing deterioration, and late-onset hearing loss in newborns with congenital cytomegalovirus (cCMV) infection?

Findings: This cohort study analyzed hearing evolution in 387 untreated children (774 ears) included in the Flemish CMV registry (Belgium). Approximately 70% of the ears with congenital hearing loss deteriorated. Late-onset hearing loss was more prevalent in case of a first trimester seroconversion. None of the 104 ears of children with a third trimester seroconversion developed late-onset hearing loss.

Meaning: These novel insights can aid into parental counselling, patient stratification, and follow-up.

RISK FACTORS FOR NATURAL HEARING EVOLUTION IN NEWBORNS WITH CONGENITAL CYTOMEGALOVIRUS INFECTION

Importance: Congenital cytomegalovirus (cCMV) is the major cause of congenital nonhereditary sensorineural hearing loss in children. Currently, criteria to identify infants at increased risk for unfavourable hearing outcome are lacking.

Objective: To identify risk factors for cCMV-related hearing improvement, hearing deterioration, and late-onset hearing loss.

Design: This cohort study is based on the Flemish CMV registry (Belgium). Data have been collected for 15 years (January 1, 2007, to February 7, 2022) and were analysed September 26, 2022, to January 16, 2023.

Setting: Multicentric study consisting of six secondary and tertiary hospitals.

Participants: Untreated cCMV-infected newborns with minimal 4-year audiological follow-up were included. Patients who presented with other possible causes of sensorineural hearing loss were excluded.

Main Outcomes and Measures: Primary outcome was hearing evolution (per ear analysis; described as stable hearing, improvement, or deterioration). The importance of gestational characteristics, clinical findings, timing of seroconversion, viral load, and hearing status at birth in predicting hearing evolution was investigated using effect sizes (Cramer *V*, odds ratio [OR], or Hedges *g*).

Results: Of the 387 children, 113 (29.2%) were symptomatic and 274 (70.8%) asymptomatic. Ninety percent (701/774) of the ears showed stable hearing (normal hearing or stable hearing loss since birth) over time. Late-onset hearing loss (normal hearing at birth followed by hearing loss) was present in 43 ears (43/683 [6.3%]). Among children with hearing loss present at birth, 70.6% (24/34) of the ears deteriorated and 6.6% (6/91) of the ears improved. Prematurity was associated with a higher chance of hearing improvement (OR, 12.80; 95% CI, 2.03-80.68). Late-onset hearing loss was more prevalent in a first trimester infection (OR, 10.10; 95% CI, 2.90-34.48). None of the 104 ears of children with a third trimester seroconversion developed late-onset hearing loss.

Conclusions and Relevance: Ongoing audiological follow-up for untreated children with congenital hearing loss is important as the majority will have hearing deterioration. The timing of seroconversion impacts the risk of developing late-onset hearing loss. These insights can aid in parental counselling, patient stratification, and follow-up. Future research should focus on the effect of treatment, the influence of determined risk factors, and the study of eventual new risk factors in patients at high risk to develop hearing loss.

1. INTRODUCTION

Affecting 0.7% of all live births, human cytomegalovirus (CMV) is the most common intrauterine infection (1, 2). A congenital CMV infection may entail a continuum of disease expressions including central nervous system malformations (microcephaly, calcification, ventriculomegaly etc.), vestibulocochlear disorders, clinical (petechiae, hepatosplenomegaly etc.) and/or laboratory (leukopenia, thrombocytopenia) abnormalities (3, 4). The majority of the children will be asymptomatic at birth; however, they are still prone to develop sequelae in the first years of life (1, 2). On the long term, the most important sequelae are neurodevelopmental delay, hearing loss, and vestibular disorders (5-7). Being responsible for 15-20% of all hearing loss during childhood, congenital cytomegalovirus (cCMV) is the major cause of congenital, non-hereditary sensorineural hearing loss (SNHL) (8-11). Approximately 50-60% of the symptomatic and 10% of the asymptomatic children will develop hearing loss in the first six years of life (12-15).

Hearing loss caused by a cCMV infection is typically highly variable and unpredictable. Both unilateral and bilateral hearing loss are described with a severity ranging from mild to profound (3, 15, 16). Once present, the hearing loss can improve or deteriorate in the years after birth. Neonates with normal hearing at birth are still at risk of developing late-onset hearing loss (14, 15). All cCMV-infected children should therefore be enrolled in a longitudinal audiovestibular follow-up program until at least four years of age (17, 18).

The variable and unpredictable audiological outcome in cCMV-infected children is cause of parental concern and anxiety, both during pregnancy and childhood. Identifying risk factors to predict hearing evolution may aid in parental counselling, may lead to a more targeted and individualized approach, and may guide treatment decisions (8, 19-21). We recently identified

risk factors for congenital hearing loss (22). However, risk factors for hearing improvement, hearing deterioration, and late-onset hearing loss are still undefined mainly because previous research was limited to small cohort studies (19, 23-27). The purpose of this study was to predict the natural course of cCMV-related hearing loss by identification of risk factors based on a large multicentric database.

2. METHODS

2.1. Data collection and neonatal investigations

This cohort study is based on the Flemish CMV registry (Belgium) (5). Ethical approval was obtained from the Ethical Committee of Ghent University Hospital (EC-2008/247); a written informed consent was obtained from a parent or caregiver of each participant. For 15 years (January 1, 2007, to February 7, 2022), data of children with congenital cytomegalovirus infection have been collected from six secondary and tertiary hospitals (Ghent University Hospital, Antwerp University Hospital, Sint Jan Hospital in Bruges, University Hospitals of Leuven, GZA Sint Augustinus in Antwerp, and GZA Middelheim in Antwerp). All children were subjected to a standardized protocol for diagnosis, treatment, and follow-up. Newborns were tested for cCMV in case of known maternal seroconversion during pregnancy or in case of suggestive symptomatology at birth; universal newborn cCMV screening was not performed. Diagnosis of cCMV was made upon viral isolation and/or PCR on urine/saliva taken within the first 3 weeks of life. A retrospective diagnosis was made by PCR on dried blood spot (DBS). Data concerning pregnancy (timing of seroconversion, gestational age), neonatal period (clinical features at birth, laboratory findings, results of central imaging [cranial ultrasound (US) and/or magnetic resonance imaging (MRI)]), and long-term neurodevelopmental and audiological follow-up have been collected (4, 5). Central imaging, laboratory investigations, and quantification of viral load were performed as earlier described

(22). Inclusion criteria for this study were [A] a confirmed diagnosis of cCMV, [B] a valid hearing assessment at birth and final evaluation at a minimal age of four years, and [C] no administration of antiviral therapy. As antiviral therapy may affect natural hearing evolution, treated children were excluded. Children presenting with other possible causes of SNHL (e.g. cochlear nerve aplasia) were excluded.

2.2. Definitions

The definition of symptomatic cCMV corresponds to the European consensus classification (8). Categorization of children into mildly/moderately/severely symptomatic or asymptomatic cCMV and their eligibility for treatment was based on investigations during the neonatal period (Table 1). Children with isolated SNHL at birth are considered symptomatic (4). As previously described (22), subgroups were made based on the hearing status at birth and the presence or absence of other cCMV-related symptomatology. In case of aberrant clinical, neurological, or laboratory findings, the child was defined as 'having other symptoms at birth'; regardless from hearing loss. Consequently, all newborns were categorized into 4 groups: with congenital hearing loss and other symptoms at birth, without congenital hearing loss and with other symptoms, with congenital hearing loss and without other symptoms (i.e. isolated hearing loss), and without congenital hearing loss or other symptoms (i.e. asymptomatic cCMV). The results for these 4 subgroups can be found in Table 2 and eTable 1.

2.3. Audiological assessment

Children received audiological follow-up from birth until at least the age of 4 years; the audiological protocol is previously described (12). Hearing measurements were excluded if [A] conductive hearing loss could not be excluded or [B] both ears were not measured

separately. The initial hearing status was assessed by click-evoked ABR. A click-threshold ≤ 30 decibels above the normal adult hearing level (dB nHL) was categorized as normal hearing, between 31 and 45 dB nHL as mild hearing loss, between 46 and 70 dB nHL as moderate hearing loss, and between 71 to 90 dB nHL as severe hearing loss. Ears with a click-threshold > 90 dB nHL were classified as profound hearing loss and inherently unable to deteriorate. The final hearing assessment was defined as the most recent audiometric threshold from the age of 4 to 6 years on audiometry. According to the adapted version of the BIAP classification, ears with ≤ 25 decibels hearing level (dB HL) were classified as normal hearing, between 26 and 40 dB HL as mild hearing loss, between 41 and 70 dB HL as moderate hearing loss, between 71 and 90 dB HL as severe hearing loss, and ≥ 91 dB HL as profound hearing loss.

Primary outcome was hearing evolution (per ear analysis) which was described as stable hearing, hearing improvement, or hearing deterioration. Hearing improvement or deterioration was assigned if the absolute difference between first and final hearing assessment was ≥15 dB (28, 29). If no thresholds were found with a click-stimuli ≥ 90 dBnHL on ABR, a threshold of 105 dB nHL – the middle of the profound category - was assigned for statistical reasons. Pure Tone Averages (PTA; the average of thresholds at 500-1000-2000-4000 Hz) were calculated to quantify the final hearing assessment. The results of ABR (first assessment; dB nHL) and audiometry (final assessment; dB HL) were compared without applying a correction factor as zero is included in the confidence interval of the difference (30, 31). Late-onset hearing loss was defined as normal hearing at birth followed by hearing loss. Ears with normal hearing at birth were excluded for the investigation of risk factors for hearing improvement. Following cochlear implantation, the hearing thresholds

prior to implantation were used. The hearing status of the contralateral ear was evaluated as a potential risk factor as well.

2.4. Statistical analysis

Data processing was performed September 26, 2022 to January 16, 2023. Statistical analyses were performed using IBM SPSS, version 28 (IBM Corp, Armonk, NY); all continuous variables were nonparametrically distributed. Risk factors were calculated for hearing improvement, hearing deterioration, and late-onset hearing loss. Differences between groups and risk factors determining hearing evolution were measured using following effect sizes: Cramer *V* (categorical variables with more than 2 subcategories), odds ratio (OR) (categorical variables with 2 subcategories), or Hedges *g* (continuous variables) (32, 33). The Hodges-Lehmann estimator was used to calculate the median difference and 95% CIs. Variance Inflation Factor and Tolerance were used to assess multicollinearity. Bonferroni correction was applied if required.

3. RESULTS

3.1. Baseline characteristics at birth

A total of 387 untreated children with a long-term audiological follow-up of at least 4 years were included. Approximately 47% (180/385) was female (sex was missing for 2 newborns). Seventy-one percent of the newborns was asymptomatic (274/387). None of the children presented with cortical atrophy, cataract, convulsions, abnormal white blood cell count, or intraventricular adhesions or vermis hypoplasia on cranial ultrasound. A considerable number of seroconversions took place in the first trimester (90/218 [41.3%]) (Table 2).

Of the 113 symptomatic children (29.2% [113/387]), 22 children (19.5%) had congenital hearing loss and other symptoms, 50 children (44.2%) had no congenital hearing loss but had other symptoms, and 41 children (36.3%) had isolated hearing loss. Regarding the per ear analysis, 774 ears were included of which 11.8% (91/774) had congenital hearing loss and 88.2% (683/774) normal hearing at birth. Of these, 38.5% (35/91) had unilateral hearing loss and 62.6% (57/91) profound hearing loss (Table 3, eTable 1).

3.2. Natural course of hearing evolution

At final follow-up, 133 ears were diagnosed with cCMV-related SNHL (133/774 [17.2%]); the majority (97/133 [72.9%]) with profound SNHL (Table 3, eTable 1). Finally, 6.1% (47/774) and 11.1% (86/774) had unilateral and bilateral hearing loss respectively. Hearing aids were used in 30 ears (30/772 [3.9%]) and a cochlear implant in 61 ears (61/772 [7.9%]). The median age for cochlear implantation was 18 months (IQR, 12-24 months; minimal and maximal age, 6 and 72 months).

In the entire study population, 90.6% (701/774) of the ears showed stable hearing (including ears with normal hearing since birth and ears with stable hearing loss since birth). Of the ears with normal hearing at birth, 43 (43/683 [6.3%]) presented with late-onset hearing loss being bilateral in 51.2% (22/43). Of the 21 ears with unilateral late-onset hearing loss, a contralateral congenital hearing loss was present in 5 ears (5/43 [11.6%]). The late-onset hearing loss was mild, moderate, severe, and profound in 9.3% (4/43), 23.3% (10/43), 14.0% (6/43), and 53.5% (23/43) respectively. Late-onset hearing loss occurred in 8.9% (12/135) ears of symptomatic and 5.7% (31/548) ears of asymptomatic children. While 61 ears (61/91 [67.0%]) with congenital hearing loss remained stable over time, the hearing of 24 ears (24/34 [70.6%]) deteriorated to a severe (3/24 [12.5%]) or profound (21/24 [87.5%]) hearing

loss. In the entire cohort, six ears (6/91 [6.6%]) improved. Only one ear showed a normalization of pre-existing hearing loss (1/6 [16.7%]) (Table 3, eTable 1).

3.3. Time frame of hearing evolution

The time course of ears with improved, deteriorated, or late-onset hearing loss is shown in Figure 1. Hearing improvement was mainly gradual and occurred before the age of 4 years in all cases (Figure 1A). Hearing deterioration and late-onset hearing loss occurred before the age of 12 months in respectively 90.5% (19/21; exact time point unknown for 3 ears) and 83.3% (15/18; exact time point unknown for 25 ears) (Figure 1B and 1C). Five ears (5/33 [15.2%]; exact time point unknown for 4 ears) showed progressive deterioration between four and six years old, whereas one ear (1/641 [0.2%] of normal-hearing ears at 4 years of age) developed late-onset hearing loss beyond the age of 4 years (for 10/43 ears, the exact timing of late-onset hearing loss could not be determined). One ear had congenital hearing loss and showed persistent deterioration beyond 4 years (1/24 [4.2%]). The other four ears had normal hearing at birth, developed late-onset hearing loss and deteriorated further even after the age of 4 years old (Figure 1D). Twenty-two percent (4/18) of the ears with late-onset hearing loss had persistent hearing deterioration after 4 years of age (10/43 ears already had profound hearing loss before the age of 4 years old so could not further deteriorate; hearing deterioration between 4 and 6 years could not be estimated for 15 ears).

3.4. Risk factors determining hearing evolution

The importance of gestational characteristics, timing of seroconversion, viral load, and hearing status at birth in predicting hearing evolution was investigated in the total group (eTable 2). Prematurity (<37 weeks) and smaller biometric features were associated with a higher chance of hearing improvement (OR prematurity, 12.80; 95% CI, 2.03-80.68 / Hedges

g birth weight, 1.09; 95% CI, 0.24-1.94 / Hedges *g* birth length, 1.99; 95% CI, 0.90-3.06 / Hedges *g* head circumference, 1.59; 95% CI, 0.52-2.65) (Table 4). To correct for multicollinearity, a regression analysis should be performed but the number of ears with hearing improvement (n=6) was too low to prevent model overfitting. None of the investigated factors were associated with hearing deterioration. The risk of late-onset hearing loss was higher in case of a first trimester infection (17/156 [10.9%]) compared to a second or third trimester infection (OR, 10.10; 95% CI, 2.90-34.48) (Cramer *V,* 0.22; 95% CI, 0.14-0.30; as a similar prevalence of late-onset hearing loss was found for a second [2.1%] and third [0%] trimester infection, both groups were combined) (Table 4). Moreover, none of the 104 ears with a third trimester seroconversion developed late-onset hearing loss. Fourteen percent of the children with unilateral hearing loss at birth developed late-onset hearing loss compared to six percent of the children with normal hearing at birth (OR, 2.68; 95% CI 0.98-7.29). The median [IQR] blood viral load of 6 ears developing late-onset hearing loss was 1783.5 [300.0-3309.0] copies/ml compared to 1216.0 [300.0-4098.0] copies/ml for 108 ears with normal hearing since birth; for a median difference of 0.0 (95% CI, -1649.0 to 2468.0) copies/ml (eTable 2C). The viral load of ears with deteriorated or improved hearing was unknown (eTable 2A/B).

To investigate whether particular cCMV-related symptoms were associated with hearing evolution, only the group of children with other symptoms at birth was analysed (eTable 3). Aberrations on clinical examination, US, or MRI were not associated with a higher risk of hearing evolution in this study.

4. DISCUSSION

This multicentric cohort study based on the Flemish CMV registry investigated risk factors determining cCMV-related hearing evolution in untreated children. Premature children had a higher chance of hearing improvement; however, this might be caused by ongoing auditory maturation. Our result suggests a large effect but no definitive conclusions can be made due to the wide confidence interval and imprecision of the estimate. The prevalence of late-onset hearing loss was higher in case of a seroconversion in the first trimester. If unilateral congenital hearing loss was present, the risk of late-onset hearing loss may be increased in the contralateral normal-hearing ear. Particular biometric features were significantly related with hearing improvement which might be caused by multicollinearity. Initial blood viral load was not a predictor of late-onset hearing loss; though confirmation by larger samples is needed. Based on this study, audiological follow-up can likely be ceased if normal hearing is maintained at 4 years of age in children with asymptomatic or mild cCMV given the small risk of late-onset hearing loss (1/641 [0.2%)]) beyond this age.

Identifying risk factors determining hearing evolution contributes to parental counselling (34, 35), patient stratification (23), follow-up, and prognosis (20). Factors associated with hearing improvement can reduce parental anxiety (23). Early identification of neonates at risk for hearing deterioration or late-onset hearing loss can aid in treatment decision making (20). Insight into the time course of late-onset hearing loss and its risk factors can aid in the evaluation of potential costs and benefits of universal screening (17). Moreover, risk factors for late-onset hearing loss could also be used to initiate targeted audiological follow-up (18).

Consequently, identification of risk factors determining hearing evolution – in particular late-onset hearing loss – is urgently needed. However, low sample sizes and the low incidence of cCMV-related hearing evolution make research challenging (24, 25, 36). Foulon et al. found

a higher incidence of late-onset hearing loss after a first trimester seroconversion compared to a seroconversion later in pregnancy (18), similar to our findings. Contrary to Faure-Bardon et al. (37), in our cohort, late-onset hearing loss occurred even in case of a second trimester seroconversion so we advise long-term audiological follow-up also for these children. An interesting observation is that none of the 104 ears of children with a third trimester seroconversion presented with late-onset hearing loss; as noticed in smaller samples by Foulon et al. (18) and Faure-Bardon et al. (37). Clinicians might consider to exclude children with a third trimester seroconversion from long-term audiological follow-up. The risk of late-onset hearing loss seemed to be higher for children with unilateral congenital hearing loss at birth compared to children with bilateral normal hearing at birth; which supports earlier findings (17).

Previous researchers stated that neonates with abnormal central imaging (18) and asymptomatic children with higher initial blood viral loads (25, 36) have a higher risk to develop late-onset hearing loss. Based on this study, clinical presentation at birth, abnormalities on ultrasound or MRI, and initial blood viral load were not associated with a higher risk of hearing improvement, hearing deterioration, or late-onset hearing loss. Despite our larger cohort, we could not relate neonatal clinical findings with progressive hearing loss. Indeed, congenital hearing loss was previously related with neonatal clinical findings (22). These results might suggest a different pathogenic mechanism for progressive hearing loss, such as persistent chronic inflammation (38). In this study, we focused on gestational characteristics and clinical symptoms of disease expression. Future studies investigating the genetic profile of the child may improve our current understanding why a subset of children are prone to develop a particular hearing status (38, 39).

The Flemish CMV registry is – to our knowledge – the largest database worldwide. Nevertheless, the incidence of cCMV-related hearing improvement, deterioration, and late-onset hearing loss is low which complicates analysis. Consequently, particular risk factors (cortical atrophy, cataract, convulsions, abnormal white blood cell count, and intraventricular adhesions or vermis hypoplasia on cranial ultrasound) were not present in the study population so their importance in predicting hearing evolution could not be assessed. Besides, we assume that the association of contralateral congenital hearing loss with late-onset hearing loss only approached statistical significance due to a lack of power as the effect size was rather high. Also note that the absence of risk factors does not guarantee a stable hearing in future. Additionally to previously mentioned limitations (22), results could be subjected to detection bias as hearing status was assessed with various measuring devices by physicians of different centres. Selection of only untreated children could have led to sample selection bias by potential exclusion of children with moderately to severely symptomatic cCMV. The majority of the patients had asymptomatic or mild cCMV; hearing outcomes for children with more severe disease may differ.

5. CONLUSION

Data based on the Flemish CMV registry was used to identify subsets of untreated infants at increased risk for hearing evolution. Late-onset hearing loss was more prevalent in case of a first trimester seroconversion. None of the ears of children infected during the third trimester of pregnancy developed late-onset hearing loss. These novel insights can aid into parental counselling, patient stratification, and follow-up.

REFERENCES

1. Kenneson A, Cannon MJ. Review and meta-analysis of the epidemiology of congenital cytomegalovirus (CMV) infection. Rev Med Virol. 2007;17(4):253-76.
2. Dollard SC, Grosse SD, Ross DS. New estimates of the prevalence of neurological and sensory sequelae and mortality associated with congenital cytomegalovirus infection. Rev Med Virol. 2007;17(5):355-63.
3. Chiopris G, Veronese P, Cusenza F, Procaccianti M, Perrone S, Daccò V, et al. Congenital Cytomegalovirus Infection: Update on Diagnosis and Treatment. Microorganisms. 2020;8(10).
4. Annelies K, Leenheer D, Alexandra C, Veerle C, Sabine L, Ludo M, et al. Results of a multicenter registry for congenital cytomegalovirus infection in Flanders, Belgium: From prenatal diagnosis over neonatal management to therapy. Early Hum Dev. 2021;163:105499.
5. Keymeulen A, De Leenheer E, Goderis J, Dhooge I, Smets K. Congenital cytomegalovirus infection registry in flanders: opportunities and pitfalls. Acta Clin Belg. 2019:1-8.
6. Goderis J, Keymeulen A, Smets K, Van Hoecke H, De Leenheer E, Boudewyns A, et al. Hearing in children with congenital cytomegalovirus infection: Results of a longitudinal study. Journal of Pediatrics. 2016;172:110-5.e2.
7. Dhondt C, Maes L, Rombaut L, Martens S, Vanaudenaerde S, Van Hoecke H, et al. Vestibular Function in Children With a Congenital Cytomegalovirus Infection: 3 Years of Follow-Up. Ear Hear. 2020.
8. Luck SE, Wieringa JW, Blázquez-Gamero D, Henneke P, Schuster K, Butler K, et al. Congenital Cytomegalovirus: A European Expert Consensus Statement on Diagnosis and Management. Pediatr Infect Dis J. 2017;36(12):1205-13.
9. Morton CC, Nance WE. Newborn hearing screening--a silent revolution. N Engl J Med. 2006;354(20):2151-64.
10. Ogawa H, Suzutani T, Baba Y, Koyano S, Nozawa N, Ishibashi K, et al. Etiology of severe sensorineural hearing loss in children: independent impact of congenital cytomegalovirus infection and GJB2 mutations. J Infect Dis. 2007;195(6):782-8.
11. Rosenthal LS, Fowler KB, Boppana SB, Britt WJ, Pass RF, Schmid SD, et al. Cytomegalovirus shedding and delayed sensorineural hearing loss: results from longitudinal follow-up of children with congenital infection. Pediatr Infect Dis J. 2009;28(6):515-20.
12. Goderis J, Keymeulen A, Smets K, Van Hoecke H, De Leenheer E, Boudewyns A, et al. Hearing in Children with Congenital Cytomegalovirus Infection: Results of a Longitudinal Study. The Journal of pediatrics. 2016;172:110-5.e2.
13. Craeghs L, Goderis J, Acke F, Keymeulen A, Smets K, Van Hoecke H, et al. Congenital CMV-Associated Hearing Loss: Can Brain Imaging Predict Hearing Outcome? Ear Hear. 2020.
14. Fletcher KT, Horrell EMW, Ayugi J, Irungu C, Muthoka M, Creel LM, et al. The Natural History and Rehabilitative Outcomes of Hearing Loss in Congenital Cytomegalovirus: A Systematic Review. Otol Neurotol. 2018;39(7):854-64.
15. Goderis J, De Leenheer E, Smets K, Van Hoecke H, Keymeulen A, Dhooge I. Hearing loss and congenital CMV infection: a systematic review. Pediatrics. 2014;134(5):972-82.
16. Liu CC, Parikh SR, Horn DL. Do Antivirals Improve Hearing Outcomes in Neonates With Congenital Cytomegalovirus Infection? Laryngoscope. 2020;130(7):1609-12.
17. Lanzieri TM, Chung W, Flores M, Blum P, Caviness AC, Bialek SR, et al. Hearing Loss in Children With Asymptomatic Congenital Cytomegalovirus Infection. Pediatrics. 2017;139(3).
18. Foulon I, De Brucker Y, Buyl R, Lichtert E, Verbruggen K, Piérard D, et al. Hearing Loss With Congenital Cytomegalovirus Infection. Pediatrics. 2019;144(2).
19. Dimopoulou D, Kourlaba G, Antoniadou A, Mariolis L, Kavatha D, Stoungioti S, et al. Low birth weight and head circumference as potential biomarkers of sensorineural hearing loss in asymptomatic congenitally CMV-infected infants. Journal of Clinical Virology. 2020;129.
20. Bradford RD, Cloud G, Lakeman AD, Boppana S, Kimberlin DW, Jacobs R, et al. Detection of cytomegalovirus (CMV) DNA by polymerase chain reaction is associated with hearing loss in

newborns with symptomatic congenital CMV infection involving the central nervous system. Journal of Infectious Diseases. 2005;191(2):227-33.
21. Rivera LB, Boppana SB, Fowler KB, Britt WJ, Stagno S, Pass RF. Predictors of hearing loss in children with symptomatic congenital cytomegalovirus infection. Pediatrics. 2002;110(4):762-7.
22. De Cuyper E, Acke F, Keymeulen A, De Leenheer EMR, Van Hoecke H, Padalko E, et al. Risk Factors for Hearing Loss at Birth in Newborns With Congenital Cytomegalovirus Infection. JAMA Otolaryngol Head Neck Surg. 2022.
23. Lanari M, Lazzarotto T, Venturi V, Papa I, Gabrielli L, Guerra B, et al. Neonatal cytomegalovirus blood load and risk of sequelae in symptomatic and asymptomatic congenitally infected newborns. Pediatrics. 2006;117(1):e76-83.
24. Ross SA, Novak Z, Fowler KB, Arora N, Britt WJ, Boppana SB. Cytomegalovirus blood viral load and hearing loss in young children with congenital infection. Pediatr Infect Dis J. 2009;28(7):588-92.
25. Lo TH, Lin PH, Hsu WC, Tsao PN, Liu TC, Yang TH, et al. Prognostic determinants of hearing outcomes in children with congenital cytomegalovirus infection. Sci Rep. 2022;12(1):5219.
26. Pinninti SG, Rodgers MD, Novak Z, Britt WJ, Fowler KB, Boppana SB, et al. Clinical Predictors of Sensorineural Hearing Loss and Cognitive Outcome in Infants with Symptomatic Congenital Cytomegalovirus Infection. Pediatr Infect Dis J. 2016;35(8):924-6.
27. Venturini E, Impagnatiello L, Chiappini E, Galli L. Hearing Outcome and Virologic Characteristics of Children With Congenital Cytomegalovirus Infection in Relation to Antiviral Therapy: A Retrospective Cohort Study. Pediatr Infect Dis J. 2023;42(1):52-8.
28. Van Camp G, Smith RJH. Hereditary Hearing Loss - GENDEAF recommendations2009.
29. Royackers L, Rector E, Verhaert N, Desloovere C. Long-term audiological follow-up of children with congenital cytomegalovirus. B-ent. 2013;Suppl 21:57-64.
30. Acke FRE, De Vriese C, Van Hoecke H, De Leenheer EMR. Twelve years of neonatal hearing screening: audiological and etiological results. European Archives of Oto-Rhino-Laryngology. 2021.
31. Stevens JC SG, Wood SA. Guidelines for the early audiological assessment and management of babies referred from the newborn hearing screening programme (v3.1).2013.
32. Cohen J. Statistical Power Analysis for the Behavioral Sciences. . 2nd ed. ed: Erlbaum Associates; 1988.
33. CRAMÉR H. Mathematical methods of statistics, Princeton University Press, 1946. CramérMathematical Methods of Statistics1946. 1946.
34. Boppana SB, Fowler KB, Pass RF, Rivera LB, Bradford RD, Lakeman FD, et al. Congenital cytomegalovirus infection: association between virus burden in infancy and hearing loss. The Journal of pediatrics. 2005;146(6):817-23.
35. Ancora G, Lanari M, Lazzarotto T, Venturi V, Tridapalli E, Sandri F, et al. Cranial ultrasound scanning and prediction of outcome in newborns with congenital cytomegalovirus infection. The Journal of pediatrics. 2007;150(2):157-61.
36. Forner G, Abate D, Mengoli C, Palù G, Gussetti N. High Cytomegalovirus (CMV) DNAemia Predicts CMV Sequelae in Asymptomatic Congenitally Infected Newborns Born to Women With Primary Infection During Pregnancy. J Infect Dis. 2015;212(1):67-71.
37. Faure-Bardon V, Magny JF, Parodi M, Couderc S, Garcia P, Maillotte AM, et al. Sequelae of Congenital Cytomegalovirus Following Maternal Primary Infections Are Limited to Those Acquired in the First Trimester of Pregnancy. Clin Infect Dis. 2019;69(9):1526-32.
38. Ouellette CP, Sánchez PJ, Xu Z, Blankenship D, Zeray F, Ronchi A, et al. Blood genome expression profiles in infants with congenital cytomegalovirus infection. Nat Commun. 2020;11(1):3548.
39. Park AH. Detecting Hearing Loss in a Child With Congenital Cytomegalovirus Infection-Finding the Elusive Needle in the Haystack. JAMA Otolaryngol Head Neck Surg. 2022.

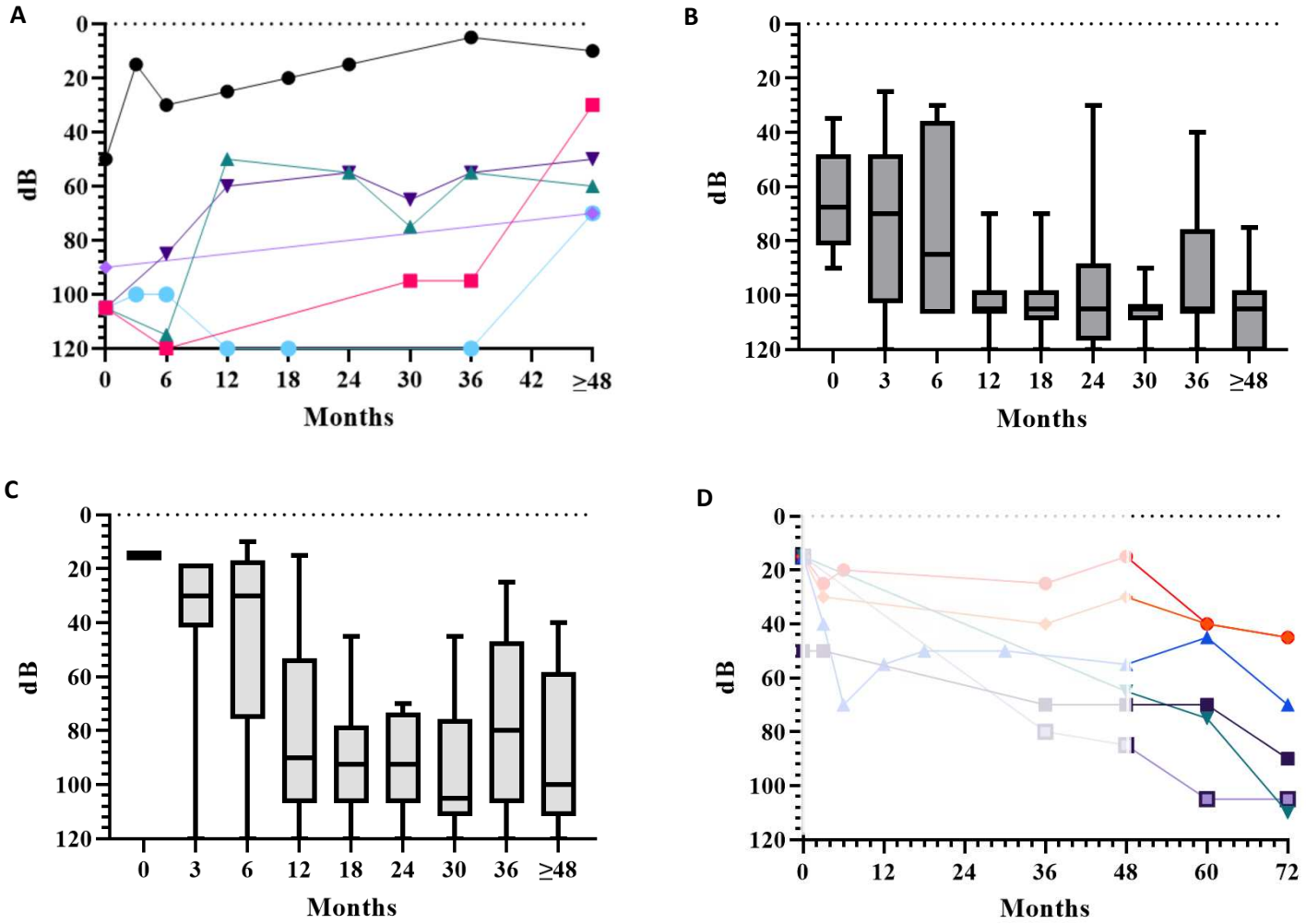
ACKNOWLEDGEMENTS
The authors want to thank the parents for giving their consent and the children for their cooperation. We acknowledge our colleagues of the participating centres for including patients; in particular Cleo Dhondt.

CONFLICT OF INTEREST DISCLOSURES
The authors have no conflicts of interest relevant to this article to disclose.

FUNDING/SUPPORT
EDC is supported by a PhD Fellowship fundamental research (11K6622N) of the Fund for Scientific Research – Flanders, Belgium.

FIGURE

Figure 1. Time course of ears with (A) hearing improvement (n=6), (B) hearing deterioration (n=24), (C) late-onset hearing loss (n=43), and (D) progressive deterioration or late-onset hearing loss beyond 4 years (n=6).



TABLES

Table 1. Categorization of newborns and eligibility criteria for therapy.

Categorization	Definition[a]	Eligibility for therapy
Mildly symptomatic cCMV	Isolated (maximum of 2) clinical nonsignificant or transient findings (e.g., intrauterine growth retardation, petechiae, mild hepatosplenomegaly, mild thrombopenia, anemia, leukopenia, mild elevated aspartate transaminase or alanine aminotransferase, or cholestasis).	Not eligible.
Moderately symptomatic cCMV	More than 2 mild clinical symptoms or with persistent (2 weeks) biological or hematological abnormalities or with mild lesions on central nervous system imaging (e.g., lenticulostriate vasculopathy or isolated cyst).	Might be eligible. Eligibility is decided by an expert; initiation is decided in consultation with the parents after being informed about the possible benefits and short- and long-term side-effects[b].
Severely symptomatic cCMV	- Central nervous system involvement: neurological signs (e.g., convulsions, microcephaly) or chorioretinitis or lesions on central nervous system imaging (e.g., calcifications, moderate to severe ventriculomegaly, multiple cysts, extensive white matter changes, cerebellar or cerebral hypoplasia, hippocampus dysplasia, migration disorders, or polymicrogyria). - Severe single-organ disease (e.g., hepatomegaly with liver failure), severe multiorgan disease, or life-threatening disease. - Isolated hearing loss.	Eligible, therapy is offered. Decision is made in consultation with the parents after being informed about the possible benefits and short- and long-term side-effects[b].
Asymptomatic cCMV	Children without clinical, neurological, or laboratory abnormalities and without hearing loss.	Not eligible.

[a] Investigations during the neonatal period: clinical investigation, laboratory test, cranial ultrasound, MRI, audiological testing, and ophthalmologic investigation.

[b] Treatment protocol: (2007 – 2012) intravenous ganciclovir 6 mg/kg twice daily for 6 weeks
(2012 – end 2017) oral valganciclovir 16 mg/kg twice daily for 6 weeks
(end 2017 – present) oral valganciclovir 16 mg/kg twice daily for 6 months

Table 2. Baseline characteristics of the included children.

	Children, No./total No. (%)				
	Total (n=387)	Symptomatic cCMV			Asymptomatic cCMV
		Children with congenital hearing loss and with other symptoms (n=22/387; 5.7%)	Children without congenital hearing loss and with other symptoms (n=50/387; 12.9%)	Children with congenital hearing loss and without other symptoms (n=41/387; 10.6%)	Children without congenital hearing loss or other symptoms (n=274/387; 70.8%)
Sex					
Male	205/385 (53.2)	13/21 (61.9)	25/50 (50.0)	21/41 (51.2)	146/273 (53.5)
Female	180/385 (46.8)	8/21 (38.1)	25/50 (50.0)	20/41 (48.8)	127/273 (46.5)
Time of seroconversion					
0-13 weeks	90/218 (41.3)	6/8 (75.0)	14/39 (35.9)	10/11 (90.0)	60/160 (37.5)
14-27 weeks	74/218 (33.9)	1/8 (12.5)	19/39 (48.7)	0/11	54/160 (33.8)
>27 weeks	54/218 (24.8)	1/8 (12.5)	6/39 (15.4)	1/11 (9.1)	46/160 (28.8)
Prematurity (<37 wk)					
Yes	17/297 (5.7)	4/20 (20.0)	3/43 (7.0)	2/30 (6.7)	8/204 (3.9)
Method of diagnosis					
PCR on saliva	1/387 (0.3)	0/22	0/49	0/40	1/269 (0.4)
PCR on urine	37/387 (9.6)	4/22 (18.2)	3/49 (6.1)	5/40 (12.3)	25/269 (9.1)
Virus isolation in saliva	4/387 (1.0)	0/22	0/49	0/40	4/269 (1.5)
Virus isolation in urine	294/387 (76.0)	11/22 (50.0)	46/49 (83.9)	12/40 (30.0)	225/269 (82.1)
Dried blood spot	44/387 (11.4)	7/22 (31.8)	0/49	23/40 (57.5)	14/269 (5.1)
Viral load (copies/ml) (only 1 centre)					
No.	57	0	11	0	46
Median (IQR)	1216.0 (300.0-4069.0)	*a*	2417.0 (499.0-6010.0)	*a*	917.5 (133.8-3747.5)

a: Viral load was unknown for all children in this group.

Table 3. Characteristics of congenital hearing loss, final hearing loss, and hearing evolution (per ear analysis).

	Ears, No./total No. (%)	
Severity of hearing loss		
	Hearing status at birth	*Hearing status at final assessment*
Mild hearing loss	5/91 (5.5)	6/133 (4.5)
Moderate hearing loss	18/91 (19.8)	18/133 (13.5)
Severe hearing loss	11/91 (12.1)	12/133 (9.0)
Profound hearing loss	57/91 (62.6)	97/133 (72.9)
Evolution of hearing between birth and final assessment		
Normal hearing at birth	683/774 (88.3)	
Normal hearing since birth	640/683 (93.7)	
Late-onset hearing loss	43/683 (6.3)	
Mild, moderate, or severe hearing loss at birth		
Stable hearing loss since birth	8/34 (23.5)	
Hearing improvement	2/34 (5.9)	
Hearing deterioration	24/34 (70.6)	
Profound hearing loss at birth		
Stable hearing loss since birth	53/57 (93.0)	
Hearing improvement	4/57 (7.0)	
Characteristics of hearing evolution		
Hearing improvement		
Median improvement (IQR; min, max), dB	42.5 (30-60; 20, 75)	
Normalization of hearing loss	1/6 (16.7)	
Hearing deterioration		
Median deterioration (IQR; min, max), dB	40 (25-55; 15, 70)	
Late-onset hearing loss		
Median late-onset (IQR; min, max), dB	80 (45-95; 25, 105)	

Table 4. Significant risk factors for hearing improvement, hearing deterioration, and late-onset hearing loss.

Hearing improvement				
Risk factor		**Ears, No. (%)**		**Difference between the groups, effect size (95% CI)**
		With improved congenital hearing loss (n=6)	**With stable or deteriorated congenital hearing loss (n=85)**	
Prematurity (<37 wk)	yes	3/8 (37.5)	5/8 (62.5)	OR, 12.80 (2.03 to 80.68)
	no	3/67 (4.5)	64/67 (95.5)	
Birth weight				
No.		6	65	
Median (IQR), g		2580.0 (1750.0-3056.3)	3070.0 (2705.0-3450.0)	Hedges *g*, 1.09 (0.24 to 1.94)
Birth length				
No.		4	49	
Median (IQR), cm		43.5 (41.0-47.5)	49.0 (47.3-51.0)	Hedges *g*, 1.99 (0.90 to 3.06)
Head circumference				
No.		4	41	
Median (IQR), cm		28.8 (27.0-32.4)	33.0 (32.0-35.0)	Hedges *g*, 1.59 (0.52 to 2.65)
Hearing deterioration				
No significant risk factors were found.				
Late-onset hearing loss				
Risk factor		**Ears, No. (%)**		**Difference between the groups, effect size (95% CI)**
		With late-onset hearing loss (n=43)	**With normal hearing since birth (n=640)**	
Timing of seroconversion				
First trimester (0-13 wk)		17/156 (10.9)	139/156 (89.1)	OR, 10.10 (2.90 to 34.48)
Second trimester (14-27 wk) or third trimester (> 27 wk)		3/250 (1.2)	247/250 (98.8)	

OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
A Hedges g value around 0.2 indicated a small effect size, around 0.5 a moderate effect size, and around 0.8 a large effect size (32).

eTABLES

eTable 1. Characteristics of congenital hearing loss, final hearing loss, and hearing evolution (per ear analysis).

	Ears, No./total No. (%)									
	Total		Symptomatic cCMV						Asymptomatic cCMV	
			Child with congenital hearing loss and with other symptoms		Child without congenital hearing loss and with other symptoms		Child with congenital hearing loss and without other symptoms		Child without congenital hearing loss or other symptoms	
	Hearing status at birth	*Hearing status at final assessment*	*Hearing status at birth*	*Hearing status at final assessment*	*Hearing status at birth*	*Hearing status at final assessment*	*Hearing status at birth*	*Hearing status at final assessment*	*Hearing status at birth*	*Hearing status at final assessment*
Severity of hearing loss (n=91 at birth, n=133 at final assessment for the total group)										
Mild hearing loss	5/91 (5.5)	6/133 (4.5)	0/36	1/36 (2.8)	NA	1/7 (14.3)	5/55 (9.1)	1/59 (1.7)	NA	3/31 (9.7)
Moderate hearing loss	18/91 (19.8)	18/133 (13.5)	12/36 (33.3)	4/36 (11.1)	NA	1/7 (14.3)	6/55 (10.9)	5/59 (8.5)	NA	8/31 (25.8)
Severe hearing loss	11/91 (12.1)	12/133 (9.0)	3/36 (8.3)	3/36 (8.3)	NA	0/7	8/55 (14.5)	4/59 (6.8)	NA	5/31 (16.1)
Profound hearing loss	57/91 (62.6)	97/133 (72.9)	21/36 (58.3)	28/36 (77.8)	NA	5/7 (71.4)	36/55 (65.5)	49/59 (83.1)	NA	15/31 (48.4)

Evolution of hearing between birth and final assessment					
	Total	Child with congenital hearing loss and with other symptoms	Child without congenital hearing loss and with other symptoms	Child with congenital hearing loss and without other symptoms	Child without congenital hearing loss and without other symptoms
Normal hearing at birth	683/774 (88.3)	8/44 (18.2)	100/100	27/82 (32.9)	548/548
Normal hearing since birth	640/683 (93.7)	7/8 (87.5)	93/100 (93.0)	23/27 (85.2)	517/548 (94.3)
Late-onset hearing loss	43/683 (6.3)	1/8 (12.5)	7/100 (7.0)	4/27 (14.8)	31/548 (5.7)
Mild, moderate, or severe hearing loss at birth					
Stable hearing loss since birth	8/34 (23.5)	1/15 (6.7)	NA	7/19 (36.8)	NA
Hearing improvement	2/34 (5.9)	1/15 (6.7)	NA	1/19 (5.3)	NA
Hearing deterioration	24/34 (70.6)	13/15 (86.7)	NA	11/19 (57.9)	NA

Profound hearing loss at birth					
Stable hearing loss since birth	53/57 (93.0)	17/21 (81.0)	NA	36/36	NA
Hearing improvement	4/57 (7.0)	4/21 (19.0)	NA	0/36	NA
Characteristics of hearing evolution					
Hearing improvement					
Median improvement (IQR; min, max), dB	42.5 (30-60; 20, 75)	45 (40-65: 35, 75)	NA	20[a]	NA
Normalization of hearing loss	1/6 (16.7)	1/5 (20.0)	NA	0/1	NA
Hearing deterioration					
Median deterioration (IQR; min, max), dB	40 (25-55; 15, 70)	40 (30-50; 20,70)	NA	35 (20-65; 15, 70)	NA
Late-onset hearing loss					
Median late-onset (IQR; min, max), dB	80 (45-95; 25, 105)	30[b]	90 (30-100; 25, 105)	80 (65-90; 60,90)	75 (45-95; 25, 105)

NA, not appropriate

[a] Only 1 ear had hearing improvement.

[b] Only 1 ear had late-onset hearing loss.

eTable 2. Risk factors for (A) hearing improvement, (B) hearing deterioration, and (C) late-onset hearing loss in the total group of cCMV-infected children.

A

Risk factor		Ears, No. (%)		Difference between the groups, effect size (95% CI)
		With improved congenital hearing loss (n=6)	With stable or deteriorated congenital hearing loss (n=85)	
Timing of seroconversion				
First trimester (0-13 wk)		1/24 (4.2)	23/24 (95.8)	Cramer *V*, 0.47 (0.06 to 1.0)
Second trimester (14-27 wk)		1/2 (50.0)	1/2 (50.0)	
Third trimester (> 27 wk)		0/4	4/4	
Prematurity (<37 wk)	yes	3/8 (37.5)	5/8 (62.5)	OR, 12.80 (2.03 to 80.68)
	no	3/67 (4.5)	64/67 (95.5)	
Birth weight				
No.		6	65	
Median (IQR), g		2580.0 (1750.0-3056.3)	3070.0 (2705.0-3450.0)	Hedges *g*, 1.09 (0.24 to 1.94)
Birth length				
No.		4	49	
Median (IQR), cm		43.5 (41.0-47.5)	49.0 (47.3-51.0)	Hedges *g*, 1.99 (0.90 to 3.06)
Head circumference				
No.		4	41	
Median (IQR), cm		28.8 (27.0-32.4)	33.0 (32.0-35.0)	Hedges *g*, 1.59 (0.52 to 2.65)
Viral load				
No.		0	0	
Median (IQR), copies/ml		NA	NA	NA
Hearing loss severity at birth				
Mild		0/5	5/5	Cramer *V*, 0.08 (0.07 to 0.33)
Moderate		1/18 (5.6)	17/18 (94.4)	
Severe		1/11 (9.1)	10/11 (90.9)	
Profound		4/57 (7.0)	53/57 (93.0)	
Contralateral congenital hearing loss	yes	4/56 (7.1)	52/56 (92.9)	OR, 1.27 (0.22 to 7.32)
	no	2/35 (5.7)	33/35 (94.3)	

NA, not appropriate
OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
Cramer V ranges from 0 to 1, where 0 to 0.10 suggested a very small association between the variables; 0.10 to <0.20 a small association; 0.20 to <0.50 a medium association; 0.50 to <0.80 a large association; and ≥0.80 a very large association (33).
A Hedges g value around 0.2 indicated a small effect size, around 0.5 a moderate effect size, and around 0.8 a large effect size (32).

B

Risk factor		**Ears, No. (%)**		**Difference between the groups, effect size (95% CI)**
		With deteriorated congenital hearing loss (n=24)	**With stable or improved congenital hearing loss (n=10)**	
Timing of seroconversion				
First trimester (0-13 wk)		4/5 (80.0)	1/5 (20.0)	Cramer *V*, 0.31 (0.08 to 1.00)
Second trimester (14-27 wk)		1/2 (50.0)	1/2 (50.0)	
Third trimester (> 27 wk)		2/4 (50.0)	2/4 (50.0)	
Prematurity (<37 wk)	yes	2/2	0/2	OR, 1.12 (0.96 to 1.30)
	no	17/26 (65.4)	9/26 (34.6)	
Birth weight				
No.		20	7	
Median (IQR), g		3280.0 (2570.0-3488.8)	s	Hedges *g*, 0.44 (-0.41 to 1.28)
Birth length				
No.		14	6	
Median (IQR), cm		49.5 (47.9-51.3)	50.0 (49.5-51.8)	Hedges *g*, 0.20 (-0.72 to 0.12)
Head circumference				
No.		9	4	
Median (IQR), cm		35.0 (32.5-37.5)	34.0 (33.0-35.8)	Hedges *g*, -0.26 (-1.36 to 0.84)
Viral load				
No.		0	0	
Median (IQR), copies/ml		NA	NA	NA
Hearing loss severity at birth				
Mild		3/5 (60.0)	2/5 (40.0)	Cramer *V*, 0.17 (0.05 to 0.56)
Moderate		14/18 (77.8)	4/18 (22.2)	

Severe		7/11 (63.6)	4/11 (36.4)	
Contralateral congenital hearing loss	yes	15/19 (78.9)	4/19 (21.1)	OR, 2.5 (0.55 to 11.33)
	no	9/15 (60.0)	6/15 (40.0)	

NA, not appropriate
OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
Cramer V ranges from 0 to 1, where 0 to 0.10 suggested a very small association between the variables; 0.10 to <0.20 a small association; 0.20 to <0.50 a medium association; 0.50 to <0.80 a large association; and ≥0.80 a very large association (33).
A Hedges g value around 0.2 indicated a small effect size, around 0.5 a moderate effect size, and around 0.8 a large effect size (32).

C

Risk factor		**Ears, No. (%)**		**Difference between the groups, effect size (95% CI)**
		With late-onset hearing loss (n=43)	**With normal hearing since birth (n=640)**	
Timing of seroconversion				
First trimester (0-13 wk)		17/156 (10.9)	139/156 (89.1)	OR, 10.10 (2.90 to 34.48)
Second trimester (14-27 wk) or third trimester (> 27 wk)		3/250 (1.2)	247/250 (98.8)	
Prematurity (<37 wk)	yes	2/26 (7.7)	24/26 (92.3)	OR, 1.20 (0.27 to 5.31)
	no	32/493 (6.5)	461/493 (93.5)	
Birth weight				
No.		30	471	
Median (IQR), g		3395.0 (3000.0-3646.3)	3400.0 (3090.0-3710.0)	Hedges *g*, 0.05 (-0.32 to 0.42)
Birth length				
No.		27	428	
Median (IQR), cm		51.0 (49.0-51.0)	50.0 (49.0-51.0)	Hedges *g*, 0.02 (-0.37 to 0.41)
Head circumference				
No.		24	371	
Median (IQR), cm		34.8 (34.0-35.0)	34.0 (33.0-35.0)	Hedges *g*, 0.09 (-0.32 to 0.50)

Viral load				
No.		6	108	
Median (IQR), copies/ml		1783.5 (300.0-3309.0)	1216.0 (300.0-4098.0)	Hedges *g*, 0.22 (-0.60 to 1.04)
Contralateral congenital hearing loss	yes	5/35 (14.3)	30/35 (85.7)	OR, 2.68 (0.98 to 7.29)
	no	38/648 (5.9)	610/648 (94.1)	

NA, not appropriate
OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
A Hedges g value around 0.2 indicated a small effect size, around 0.5 a moderate effect size, and around 0.8 a large effect size (32).

eTable 3. Risk factors for (A) hearing improvement, (B) hearing deterioration, and (C) late-onset hearing loss in children with clinical, neurological, or laboratory abnormalities.

A

Risk factor		Ears, No. (%) With improved congenital hearing loss (n=5) [a]	Difference between the groups, effect size (95% CI)
Clinical examination at birth			
Dysmaturity	yes	0/8	OR, 0
	no	5/28 (17.9)	
Hepatomegaly	yes	0/5	OR, 0
	no	5/31 (16.1)	
Splenomegaly	yes	0/4	OR, 0
	no	5/32 (15.6)	
Microcephaly	yes	0/3	OR, 0
	no	5/33 (15.2)	
Petechiae	yes	1/4 (25.0)	OR, 2.33 (0.19 to 28.25)
	no	4/32 (12.5)	
Laboratory findings at birth			
Abnormal platelet count	yes	[b]	OR, 0
	no	1/21 (4.8)	
Cranial ultrasound			
Cystic periventricular leukomalacia	yes	0/2	OR, 0
	no	3/20 (15.0)	
Periventricular cysts	yes	2/4 (50.0)	OR, 17.0 (1.02 to 283.01)
	no	1/18 (5.6)	
Striatal vasculopathy	yes	0/5	OR, 0
	no	3/17 (17.6)	
Calcifications	yes	0/1	OR, 0
	no	3/21 (14.3)	
Hyperechogenic caudal pit	yes	0/2	OR, 0
	no	3/20 (15.0)	
Ventriculomegaly	yes	0/4	OR, 0
	no	3/18 (16.7)	
MRI			
Cortical atrophy	yes	[b]	NA
	no	3/30 (10.0)	
Cystic periventricular leukomalacia	yes	[b]	NA
	no	3/30 (10.0)	
Intraventricular adhesions	yes	[b]	NA
	no	3/30 (10.0)	
Periventricular cysts	yes	0/6	OR, 0
	no	3/24 (12.5)	
Gyration disorders	yes	0/1	OR, 0
	no	3/29 (10.3)	
Calcifications	yes	0/1	OR, 0
	no	3/29 (10.3)	
Hyperintensity white matter	yes	2/12 (16.7)	OR, 3.4 (0.27 to 42.44)
	no	1/18 (5.6)	
Ventriculomegaly	yes	0/3	OR, 0
	no	3/27 (11.1)	

NA, not appropriate

MRI, magnetic resonance imaging

OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.

[a] Ears with improved congenital hearing loss were compared to ears with stable or deteriorated congenital hearing loss (n=31).
[b] None of the ears with improved congenital hearing loss had this particular abnormality.

B

Risk factor		Ears, No. (%) With deteriorated congenital hearing loss (n=24) [a]	Difference between the groups, effect size (95% CI)
Clinical examination at birth			
Dysmaturity	yes	4/4	OR, 1.20 (1.00 to 1.44)
	no	20/30 (66.7)	
Hepatomegaly	yes	2/2	OR, 1.09 (0.97 to 1.23)
	no	22/32 (68.8)	
Splenomegaly	yes	2/2	OR, 1.09 (0.97 to 1.23)
	no	22/32 (68.8)	
Microcephaly	yes	2/2	OR, 1.09 (0.97 to 1.23)
	no	22/32 (68.8)	
Petechiae	yes	[b]	NA
	no	24/34 (70.6)	
Laboratory findings at birth			
Abnormal platelet count	yes	[b]	NA
	no	19/28 (67.9)	
Cranial ultrasound			
Cystic periventricular leukomalacia	yes	[b]	NA
	no	17/27 (63.0)	
Periventricular cysts	yes	[b]	NA
	no	17/27 (63.0)	
Striatal vasculopathy	yes	3/4 (75.0)	OR, 1.93 (0.17 to 21.54)
	no	14/23 (60.9)	
Calcifications	yes	[b]	NA
	no	17/27 (63.0)	
Hyperechogenic caudal pit	yes	1/2 (50.0)	OR, 0.56 (0.03 to 10.12)
	no	16/25 (64.0)	
Ventriculomegaly	yes	1/1	OR, 1.06 (0.94 to 1.20)
	no	16/26 (61.5)	
MRI			
Cortical atrophy	yes	[b]	NA
	no	17/25 (68.0)	
Cystic periventricular leukomalacia	yes	[b]	NA
	no	17/25 (68.0)	
Intraventricular adhesions	yes	[b]	NA
	no	17/25 (68.0)	
Periventricular cysts	yes	[b]	NA
	no	17/25 (68.0)	
Gyration disorders	yes	[b]	NA
	no	17/25 (68.0)	
Calcifications	yes	[b]	NA
	no	17/25 (68.0)	
Hyperintensity white matter	yes	5/6 (83.3)	OR, 2.92 (0.28 to 30.30)
	no	12/19 (63.2)	
Ventriculomegaly	yes	1/1	OR, 1.06 (0.94 to 1.20)
	no	16/24 (66.7)	

NA, not appropriate
MRI, magnetic resonance imaging

OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
[a] Ears with deteriorated congenital hearing loss were compared to ears with stable or improved congenital hearing loss (n=10).
[b] None of the ears with deteriorated congenital hearing loss had this particular abnormality.

C

Risk factor		**Ears, No. (%)** **With late-onset hearing loss (n=8)** [a]	**Difference between the groups, effect size (95% CI)**
Clinical examination at birth			
Dysmaturity	yes	0/6	OR, 0
	no	8/102 (7.8)	
Hepatomegaly	yes	0/3	OR, 0
	no	8/105 (7.6)	
Splenomegaly	yes	0/2	OR, 0
	no	8/106 (7.5)	
Microcephaly	yes	0/1	OR, 0
	no	8/107 (7.5)	
Petechiae	yes	0/8	OR, 0
	no	8/100 (8.0)	
Laboratory findings at birth			
Abnormal platelet count	yes	[b]	NA
	no	4/69 (5.8)	
Cranial ultrasound			
Cystic periventricular leukomalacia	yes	0/2	OR, 0
	no	7/100 (0.7)	
Periventricular cysts	yes	2/18 (11.1)	OR, 1.98 (0.35 to 11.09)
	no	5/84 (6.0)	
Striatal vasculopathy	yes	1/35 (2.9)	OR, 0.30 (0.04 to 2.59)
	no	6/67 (9.0)	
Calcifications	yes	2/9 (22.2)	OR, 5.03 (0.82 to 30.77)
	no	5/93 (5.4)	
Hyperechogenic caudal pit	yes	0/2	OR, 0
	no	7/100 (7.0)	
Ventriculomegaly	yes	1/6 (16.7)	OR, 3.0 (0.30 to 29.94)
	no	6/96 (6.3)	
MRI			
Cortical atrophy	yes	1/2 (50.0)	OR, 19.67 (0.98 to 396.90)
	no	3/62 (4.8)	
Cystic periventricular leukomalacia	yes	0/2	OR, 0
	no	4/62 (6.5)	
Intraventricular adhesions	yes	1/4 (25.0)	OR, 6.33 (0.50 to 80.59)
	no	3/60 (5.0)	
Periventricular cysts	yes	0/2	OR, 0
	no	4/62 (6.5)	
Gyration disorders	yes	0/1	OR, 0
	no	4/63 (6.3)	
Calcifications	yes	0/1	OR, 0
	no	4/63 (6.3)	
Hyperintensity white matter	yes	2/16 (12.5)	OR, 3.29 (0.42 to 25.50)
	no	2/48 (4.2)	
Ventriculomegaly	yes	0/3	OR, 0
	no	4/61 (6.6)	

NA, not appropriate
MRI, magnetic resonance imaging
OR, odds ratio. Statistical significance was defined as a 95% CI excluding 1.
[a] Ears with late-onset hearing loss were compared to ears with normal hearing since birth (n=100).
[b] None of the ears with late-onset hearing loss had this particular abnormality.