Received: 29 March 2020 | Revised: 4 October 2020 | Accepted: 25 October 2020

DOI: 10.1111/ppe.12740

ORIGINAL ARTICLE

Paediatric and Perinatal Epidemiology WILEY

Maternal risk factors for congenital limb deficiencies: A population-based case–control study

Johanna Syvänen[1,2] | Yrjänä Nietosvaara[3] | Saija Hurme[4] | Antti Perheentupa[5] | Mika Gissler[6,7] | Arimatias Raitio[1,2] | Ilkka Helenius[1,2,8]

[1]Department of Pediatric Orthopedic Surgery, Turku University Hospital, Turku, Finland

[2]University of Turku, Turku, Finland

[3]Department of Pediatric Orthopedic Surgery, Helsinki University Hospital, Helsinki, Finland

[4]Biostatistics, University of Turku, Turku, Finland

[5]Department of Obstetrics and Gynecology, Turku University Hospital, Turku, Finland

[6]Information Services Department, Finnish Institute for Health and Welfare THL, Helsinki, Finland

[7]Department of Neurobiology, Care Sciences and Society, Karolinska Institute, Stockholm, Sweden

[8]Department of Orthopedics and Traumatology, University of Helsinki and Helsinki University Hospital, Helsinki, Finland

Correspondence
Johanna Syvänen, Department of Pediatric Orthopedic Surgery, Turku University Hospital, Kiinamyllynkatu, Turku, Finland.
Email: johanna.syvanen@fimnet.fi

Funding information
JS received research grants from Turku University Hospital and the Turku University Foundation for the submitted work. IH received scientific funding from industry (Medtronic, Stryker Spine) outside the submitted manuscript. The funding source did not play any role in the investigation.

Abstract

Background: Risk factors for congenital limb deficiencies are poorly understood.

Objective: To investigate risk factors for congenital limb deficiencies.

Methods: We conducted a nationwide population-based case–control (1:5) study in Finland, using national registers on congenital anomalies, births, and induced abortions, cross-linked with data on maternal prescription medicine use obtained from the registers on Reimbursed Drug Purchases and Medical Special Reimbursements. Five hundred and four children with limb deficiencies (241 isolated, 181 syndromic, and 82 other associated anomalies) were identified, and 2,520 controls were matched to cases on residence and year of pregnancy. Non-syndromic cases (n = 323) were subdivided into longitudinal (n = 120), transverse (n = 123), intercalary (n = 24), mixed (n = 18), and unknown (n = 38) deficiencies.

Results: Pregestational diabetes was associated with all limb deficiencies (adjusted odds ratio [OR] 12.71, 95% confidence interval [CI] 2.37, 68.25) and with isolated (OR 11.42, 95% CI 2.00, 64.60) deficiencies. Primiparity was associated with increased risk of congenital limb deficiencies among all cases (OR 1.49, 95% CI 1.15, 1.93), isolated cases (OR 1.46, 95% CI 1.09, 1.96), and among cases with longitudinal (OR 1.90, 95% CI 1.24, 2.90) and transverse deficiencies (OR 1.75, 95% CI 1.13, 2.70). Young maternal age (<25 years) was associated with all congenital limb deficiencies (OR 1.40, 95% CI 1.02, 1.90) and transverse deficiencies (OR 1.76, 95% CI 1.05, 2.96). Advanced maternal age (≥35 years) was associated with syndromic (OR 1.82, 95% CI 1.19, 2.78) and transverse deficiencies (OR 1.94, 95% CI 1.06, 3.57). Maternal antiepileptic medication was associated with all (OR 5.77, 95% CI 1.75, 19.04) and with isolated cases (OR 3.83, 95% CI 1.02, 14.34).

Conclusions: It is important that pregnant women taking medications, especially antiepileptics, or women with pregestational diabetes are carefully monitored with regard to the occurrence and risk of limb deficiencies in the fetus.

KEYWORDS

congenital limb deficiency, diabetes, maternal, population-based, pregnancy, risk factor

1 | INTRODUCTION

Congenital limb deficiencies are rare anomalies resulting in reduced functional ability and quality of life in affected individuals. For cases not associated with chromosomal disorders, single-gene mutations, recognised syndromes, and teratogenic drugs, the cause or causes of limb deficiencies remain mostly unknown.[1] Different types of limb deficiencies (transverse versus longitudinal) may have different aetiologies. Transverse deficiencies are often attributable to the early amnion rupture disruption sequence.[2]

The known teratogen for limb development, thalidomide, came into use after World War II, particularly in the 1950s.[3] In the 1960s and 1970s, several studies also reported an association between maternal sex steroid use and the prevalence of congenital limb anomalies.[4,5] These associations with sex steroids have been shown to be unreliable in a more recent study[6] Other suspected risk factors include low birthweight and short gestation length,[7,8] maternal smoking[9] and alcohol use[7,10] chorionic villus sampling,[11] pre-pregnancy obesity,[12] and infertility treatments.[13,14] Folic acid supplementation and multivitamins containing folic acid have also been reported to show protective effects to prevent congenital limb deficiencies.[15,16] Some reports also show higher risks of limb deficiencies in relation to maternal thyroid problems,[8] preeclampsia,[8] influenza,[17] goiter,[17] and rheumatoid arthritis.[17] Some studies have reported an association between pregestational diabetes and congenital limb deficiencies[8,18-25] but not others.[26] The role of pregestational diabetes as a maternal risk factor for congenital limb deficiencies and its magnitude remains unclear.

We designed a population-based case–control study of congenital limb deficiencies to explore pregnancy and maternal risk factors and to study the effect of maternal medication use immediately prior to pregnancy and during the first trimester in pregnancy. We hypothesised that pregestational diabetes mellitus and first-trimester medication use would be associated with increased risks of congenital limb deficiencies.

Synopsis

Study question

What are the risk factors for congenital limb deficiencies?

What is Already Known

While the causes of limb deficiencies, especially isolated limb deficiencies, remain largely unknown, a few population-based studies have shown an association between exposure to thalidomide, a teratogen, and limb deficiencies. Maternal diabetes has also been reported to be associated with increased risk of congenital limb deficiencies in some, but not all, studies.

What This Study Adds

This large population-based study shows that young maternal age, primiparity, pregestational diabetes, and male sex are associated with increased risk of non-syndromic congenital limb deficiencies. Maternal use of antiepileptics, corticosteroids, progestogens, and topical anti-infective gynaecological drugs was associated with selected subtypes of congenital limb deficiencies.

2 | METHODS

The data on congenital limb deficiencies were collected from the Finnish Register of Congenital Malformations, which is maintained by the Finnish Institute for Health and Welfare. The register contains data on all live births, stillbirths, and fetuses from spontaneous abortions and terminations of pregnancy due to severe fetal anomalies. The register also receives data from other national health registers with the help of the unique personal identification code (PIC) (the Medical Birth Register, the Register of Induced Abortions, the Care Register for Health Care, and the Register of Visual Impairment) and from Statistics Finland cause-of-death statistics. All notified diagnoses are evaluated, classified, and coded by a medical geneticist at the Register. Information on maternal prescription medicine use was obtained from the Register of Reimbursed Drug Purchases and the Register of Medical Special Reimbursements, both maintained by Social Insurance Institution of Finland. These registers receive data from a legally compulsory announcement request and have been validated to confirm accurate data with high coverage.[27-30]

Drug use during the first trimester and one month before pregnancy was analysed. Medication use for maternal long-term diseases (diabetes mellitus, asthma, psychotic mental conditions, depression, epilepsy, and inflammatory bowel diseases) and their connection to limb deficiencies were abstracted from the Register of Reimbursed Drug Purchases and the Register of Medical Special Reimbursements. The frequency of long-term illnesses among pregnant women was studied on the basis of the Special Reimbursement Entitlements granted by the Social Insurance Institution of Finland. The frequency of drug use was determined by examining the number of people who bought drugs. Drug purchases are recorded at all levels of the international Anatomical Therapeutic Chemical (ATC) classification. In Finland, almost all prescription-only drugs necessary for treatment of an illness are reimbursable. Some over-the-counter drugs are also reimbursable when prescribed by a physician. Other risk factors (maternal age, body mass index (BMI), smoking, maternal parity, miscarriages, multiple pregnancy, sex of the fetus/infant, pregestational diabetes, and infertility treatments including in vitro fertilisation) were abstracted from the Medical Birth Register and from the Congenital Malformation Register. BMI was categorised into three groups (<18.5, 18.5-30, ≥30 kg/m^2), as only maternal obesity has been identified as a risk factor for congenital limb deficiencies.[31]

2.1 | Congenital limb deficiency cases and controls

All cases (n = 610) with congenital limb deficiencies born in Finland between 1996 and 2008 were identified. A detailed description of the data collection methods for congenital limb deficiencies has been published previously.[32,33] The study population of upper limb deficiencies was expanded to include the period from 2006 through 2008 and to include elective terminations of pregnancy (n = 63) from 1996 through 2008. We reviewed all cases with ICD-9 codes 75xx and 65xx from 1996 to 2008. Every case with these ICD-9 codes was checked, and cases other than congenital limb deficiencies were excluded. Live births, stillbirths, and fetuses from spontaneous abortions and terminations of pregnancy due to fetal anomalies were included. Diagnoses and medical records of cases with a possible limb deficiency were re-evaluated by a paediatric and orthopaedic surgeon, and a medical geneticist. If the diagnosis was unclear, more information (eg patient records, photographs, X-ray images, specialist opinions) was requested from the hospitals concerned to confirm the diagnosis.

Limb deficiency was defined as total or partial absence or severe hypoplasia of the skeletal structure(s) of limbs, including femoral hypoplasia.[34] The term isolated limb deficiency was used when there were no major malformations involving other organ systems than limbs. However, cases classified as isolated limb deficiencies could have abnormalities in one or more limbs, including the upper and lower extremities. Cases with associated limb deficiencies had major structural anomalies in both limb(s) and non-limb structures (multiple congenital anomalies). The term syndromic or chromosomal was used when a known syndrome or gene disorder was identified. Syndromic cases were analysed separately because the aetiology is usually known. All group included isolated limb deficiencies and those associated with a major anomaly.

Limb deficiencies were categorised into five groups: longitudinal (preaxial, postaxial, and cleft deficiencies), transverse, intercalary, mixed, and unknown deficiencies (cases not amenable to classification).

In cases with syndactylies of the fingers or toes, the limb anomaly was classified as limb deficiency only if at least one bone was clearly missing or severely hypoplastic. Limb deficiency cases with amniotic bands (n = 106) were excluded from the final analytic population because they are generally thought to be part of a sequence. Uncertain cases were discussed by two experienced paediatric orthopaedic surgeons (YN and IH) until a consensus was reached on the type of birth defect.[32,33] The final study population included 504 limb deficiency cases.

For all cases with congenital limb deficiencies, five controls (fetuses/infants and their mothers) without limb deficiencies were randomly selected from the Medical Birth Register (n = 2,520). They were matched according to the university hospital district and the conception year and month to minimise the effect of environmental factors, including events at early pregnancy.

2.2 | Statistical analysis

Maternal risk factors that were analysed included maternal age (<25, 25-34, and ≥35 years), parity (primiparous versus multiparous), pre-pregnancy body mass index (BMI, classified as < 18.5, 18.5-30, ≥30 kg/m^2), smoking (yes/no) defined as active smoking during the first trimester of pregnancy, previous miscarriages (none, 1, 2, or ≥ 3), multiple pregnancy, sex of the fetus/infant, infertility treatment (yes/no), and maternal chronic diseases (maternal medical special reimbursements) (yes/no). The initial analysis of maternal medication (at least 10 exposed mothers) was conducted at the third or fourth level of the Anatomical Therapeutic Chemical (ATC) Classification System by WHO (Appendix 1).

Potential risk factors were considered based on the literature and data availability. We fit conditional logistic regression models (taking into consideration the matching variables) examine potential risk factors for each subgroup separately. Subsequently, a multivariable model was created separately for the different subgroups (Figure 1). Every multivariable model included maternal age, parity, sex of the fetus/infant, and pregestational diabetes, except in longitudinal deficiencies in which diabetes was not included, because zero exposures in control group. We report adjusted odds ratio (OR) with 95% confidence interval (CI) as the effect measure. All analyses were performed using SAS (version 9.4 for Windows; SAS Institute Inc, Cary, NC, USA).

2.3 | Missing data

In cases of elective terminations of pregnancy, no information was available on maternal smoking (95/504 cases, 68/2520 controls), invasive fetal screening (80/504 cases, 0/2520 controls), or infertility treatment (80/504 cases, 0/2520 controls). These cases were excluded from the respective analyses.

The only variable with clearly more than 5% of missing values was BMI as it was only included in the register data from the beginning of 2004. Hence, all BMI values were missing between 1995 and 2003 (373/504 cases and 1683/3024 controls) and 68% of BMI values were missing systematically in whole data. We did multiple imputation (50 imputations) assuming arbitrary missing data pattern and used also maternal age, smoking status, and pregestational diabetes to impute missing BMI. Then, we combined information for 50 imputations and did univariate analysis for all congenital limb deficiencies. Since BMI (following imputations) was not associated with congenital limb deficiencies, we did not consider this model further.

3 | RESULTS

The study included 504 limb deficiency cases and 2,520 matched controls. Isolated limb deficiencies accounted for 241 (47.8%) cases. Eighty-two cases (16.3%) had anomalies in other organ

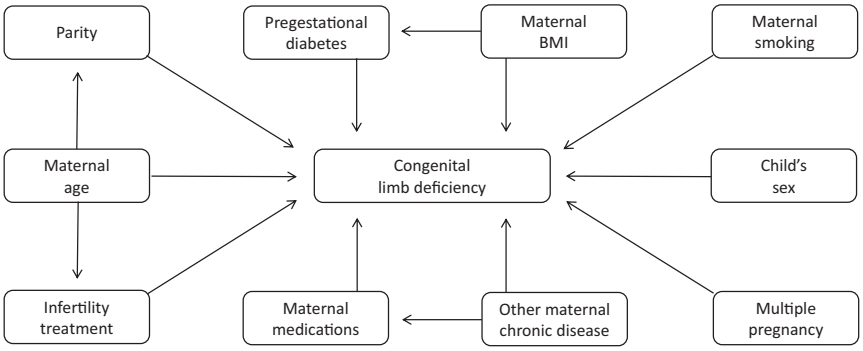

FIGURE 1 DAG: A multivariable model was created separately for the different subgroups (all, isolated, multiple congenital anomaly, syndrome, longitudinal, transverse, intercalary, and mixed defects)

TABLE 1 List of potential and analysed risk factors in univariate analysis for congenital limb deficiencies

	Odds ratio (95% confidence interval)		
Risk factors	**Multiple anomalies (MCA) (n = 82)**	**Syndromic anomalies (n = 181)**	**Transverse anomalies (n = 123)**
Young maternal age (<25 years)	1.85 (1.03, 3.35)	1.10 (0.71, 1.70)	1.85 (1.20, 2.85)
Primiparity	1.77 (1.09, 2.87)	1.12 (0.81, 1.56)	1.67 (1.15, 2.43)
Infertility treatment	4.17 (1.27, 13.65)	1.59 (0.63, 4.03)	0.70 (0.16, 3.19)
Multiple pregnancy	3.31 (1.25, 8.75)	1.98 (1.02, 3.85)	0.89 (0.33, 2.36)
Antiepileptics	10.00 (0.91, 110.28)	7.50 (1.25, 44.89)	2.00 (0.39, 10.31)
Smoking	0.83 (0.37, 1.86)	1.32 (0.81, 2.16)	1.58 (1.00, 2.54)

systems, and 181 cases were syndromic or chromosomal in aetiology (35.9%). Longitudinal deficiencies were identified in 243 (48.2%), transverse in 138 (34.5%), intercalary in 34 (6.7%), mixed in 36 (7.1%), and unknown in 53 (10.5%) cases. Among the 323 non-syndromic cases, there were 120 (37.2%) longitudinal, 123 (38.1%) transverse, 24 (7.4%) intercalary, 18 (5.6%) mixed, and 38 (11.8%) unknown deficiencies. Univariate analyses to identify potential risk factors were performed separately for all subgroups (Table 1).

Multivariable analyses were performed separately to different subgroups. Primiparity was associated with increased risk for congenital limb deficiencies overall, and in isolated cases and also longitudinal and transverse deficiencies. Male sex was associated with increased risk of all, isolated, longitudinal, and mixed cases. Maternal young age (<25 years) had associations with all cases and transverse deficiencies. On the other hand, advanced maternal age (≥35 years) was associated with syndromic and transverse deficiencies. Among maternal illnesses, only pregestational diabetes was associated with all limb deficiencies and with isolated cases (Tables 2-6).

Maternal antiepileptic medication use was associated with all and isolated cases. Corticosteroid use was associated with syndromic cases, sex hormone use was associated with longitudinal deficiencies, and locally used topical anti-infective gynaecological drugs were associated with transverse deficiencies. Tetracycline use showed a suggestive association with transverse deficiencies (Tables 2-6). The multiple anomalies group had an increased association only with multiple pregnancy (aOR 3.37, 95% CI 1.02, 10.91). Also, multiple pregnancy was associated with increased risk for congenital limb deficiencies in mixed deficiencies (aOR 3.42, 95% CI 1.09, 10.77).

3.1 | Comment

3.1.1 | Principal findings

This population-based case–control study found that maternal pregestational diabetes, primiparity, and male sex were associated with congenital limb deficiencies. First-trimester use of antiepileptics, corticosteroids, progestogens, and topical anti-infective gynaecological drugs increased the frequency of congenital limb deficiencies.

3.2 | Strengths of the study

Data on exposures and outcomes were prospectively collected by the universally accessible healthcare system of Finland. The registries used in this study were complete with accurate data.[27-30] The diagnosis of each congenital limb deficiency case was confirmed by reviewing medical records, radiographs, and autopsy reports. The coverage of the data on cases with congenital limb deficiency during the study years is high.[32,33] A case–control design was selected to identify the risk factors for rare clinical conditions with birth prevalence of 2.2 (lower limbs) and 5.6 (upper limbs) per 10,000 births.[32,33] Our study also included stillbirths and terminations of

TABLE 2 Associations of maternal and infant factors and all limb deficiencies

	Number of Events		Odds ratio (95% confidence interval)	
	Cases (n = 323)	Controls (n = 1615)	Unadjusted	Adjusted*
Maternal age (years)				
<25	78 (24.2%)	294 (18.2%)	1.49 (1.11, 2.01)	1.40 (1.02, 1.90)
25-34	196 (60.7%)	1096 (67.9%)	1.00 (Reference)	1.00 (Reference)
≥35	49 (15.2%)	225 (13.9%)	1.22 (0.86, 1.72)	1.27 (0.88, 1.82)
Primiparity	166 (51.6%)	655 (40.6%)	1.56 (1.23, 1.99)	1.49 (1.15, 1.93)
Male sex	184 (57.5%)	778 (48.2%)	1.45 (1.14, 1.85)	1.43 (1.12, 1.83)
Maternal pregestational diabetes	5 (1.6%)	2 (0.1%)	12.50 (2.43, 64.43)	12.71 (2.37, 68.25)
Antiepileptic drugs (N03A)	6 (1.9%)	6 (0.4%)	5.00 (1.61, 15,50)	5.77 (1.75, 19.04)

*Adjusted for maternal age, parity, child's sex, maternal pregestational diabetes, antiepileptic drugs, and progestogens.

pregnancy, which decreases potential for selection bias.[35] We also analysed separately non-syndromic and syndromic cases.

3.3 | Limitations of data

An important limitation of most registry-based studies is incomplete or misclassified data. For example, although a prescription may have been filled for a certain medication, the mother may not have actually taken the medication. Moreover, large percentages of data were missing on maternal body mass index. Our registers do not collect information on the use of illicit drugs or herbal medications during pregnancy. Because we used register-based data, we did not have information on glycated haemoglobin during pregnancy, which is known to influence the pregnancy outcomes in diabetic mothers.[36] Because our study ended in 2008, we may have had incomplete follow-up for the most recent cases. Time period effects are unlikely because maternity care and the guidelines on drug use during pregnancy were similar after 2008. In this study, we did not have information on maternal folic acid or multivitamin supplementation, which may influence the results.[15,16] Overall, over 70% of pregnant mothers in Finland use dietary supplements.[37]

3.4 | Interpretation

In this study, pregestational diabetes was associated with increased risks of limb deficiency in two groups: all and isolated deficiencies. In the Norwegian population-based study, Klungsoyr et al[19] reported a threefold increased risk of limb deficiencies in relation to pregestational diabetes. Correa et al[23] reported a sixfold to sevenfold increased risk with pregestational diabetes for longitudinal limb deficiencies, and Dukhovny et al[24] reported a fivefold increased risk with pregestational diabetes for all limb deficiencies in the National Birth Defects Prevention Study (NBDPS). Tinker et al[25] reported a 10-fold increased risk for longitudinal and twofold increased risk for transverse limb deficiencies with pregestational diabetes from population-based NBDPS study based on multiple registers and a computer-assisted telephone interview. In other population-based studies, maternal diabetes has been associated with congenital limb deficiencies in children.[8,18,20,21] On the other hand, Nielsen et al[26] found no association between pregestational diabetes and limb deficiencies in their population-based case–control study (1:2) based on questionnaires concerning all birth defects. We found primiparous women at increased risk for delivering infants with limb deficiencies. Duong et al[38] found that nulliparous women were more likely to have

TABLE 3 Associations of maternal and infant factors and isolated limb deficiencies

	Number of Events		Odds ratio (95% confidence interval)	
	Cases (n = 241)	Controls (n = 1205)	Unadjusted	Adjusted*
Maternal age (years)				
<25	57 (23.7%)	225 (18.7%)	1.39 (0.99, 1.96)	1.33 (0.93, 1.89)
25-34	149 (61.8%)	815 (67.6%)	1.00 (Reference)	1.00 (Reference)
≥35	35 (14.5%)	165 (13.7%)	1.16 (0.78, 1.74)	1.26 (0.83, 1.90)
Primiparity	124 (51.5%)	501 (41.6%)	1.50 (1.13, 1.98)	1.46 (1.09, 1.96)
Male sex	143 (59.3%)	581 (48.2%)	1.57 (1.18, 2.07)	1.57 (1.18, 2.08)
Maternal pregestational diabetes	4 (1.7%)	2 (0.2%)	10.00 (1.83, 54.60)	11.42 (2.00, 64.60)
Antiepileptic drugs (N03A)	4 (1.7%)	5 (0.4%)	4.00 (1.07, 14.90)	3.83 (1.02, 14.34)

*Adjusted for maternal age, parity, child's sex, maternal pregestational diabetes, and antiepileptic drugs.

TABLE 4 Associations of maternal and infant factors and limb deficiencies associated with syndromes

	Number of Events		Odds ratio (95% confidence interval)	
	Cases (n = 181)	Controls (n = 905)	Unadjusted	Adjusted*
Maternal age (years)				
<25	32 (17.7%)	164 (18.1%)	1.10 (0.71, 1.70)	0.99 (0.61, 1.60)
25-34	109 (60.2%)	611 (67.5%)	1.00 (Reference)	1.00 (Reference)
≥35	40 (22.1%)	130 (14.4%)	1.70 (1.14, 2.54)	1.82 (1.19, 2.78)
Primiparity	76 (42.5%)	361 (39.9%)	1.12 (0.81, 1.56)	1.14 (0.79, 1.66)
Male sex	104 (60.1%)	475 (52.5%)	1.40 (1.00, 1.95)	1.39 (0.99, 1.96)
Maternal pregestational diabetes	4 (2.2%)	7 (0.8%)	2.86 (0.84, 9.76)	2.99 (0.87, 10.34)
Antiepileptic drugs (N03A)	4 (2.2%)	6 (0.7%)	7.50 (1.25, 44.89)	4.17 (1.07, 16.23)

*Adjusted for maternal age, parity, child's sex, maternal pregestational diabetes, corticosteroids, acne medicines, and antiepileptics.

TABLE 5 Associations of maternal and infant factors and longitudinal limb deficiencies

	Number of Events		Odds ratio (95% confidence interval)	
	Cases (n = 120)	Controls (n = 600)	Unadjusted	Adjusted*
Maternal age (years)				
<25	27 (22.5%)	112 (18.7%)	1.22 (0.75, 1.99)	1.07 (0.64, 1.81)
25-34	78 (65.0%)	394 (65.7%)	1.00 (Reference)	1.00 (Reference)
≥35	15 (12.5%)	94 (15.7%)	0.80 (0.44, 1.47)	0.70 (0.36, 1.35)
Primiparity	66 (55.0%)	230 (38.3%)	1.97 (1.32, 2.93)	1.90 (1.24, 2.90)
Male sex	75 (62.5%)	292 (48.7%)	1.82 (1.21, 2.74)	1.88 (1.23, 2.86)
Maternal pregestational diabetes	2 (1.7%)	0 (0.0%)	NA	NA
Sex hormones (G03)	13 (10.8%)	25 (4.2%)	2.72 (1.36, 5.43)	2.57 (1.22, 5.40)

*Adjusted for maternal age, parity, child's sex, and progestogens.

children with limb reduction deficiencies than were to primiparous mothers, and primiparous mothers also had higher risk than multiparous mothers.

There are little data on the associations between antiepileptics and limb deficiencies. Valproic acid therapy has been associated with limb deficiencies.[39,40] Klungsoyr et al[19] reported no association between epilepsy and congenital limb deficiencies. In our study, the use of antiepileptics was associated with increased frequency of all and isolated limb deficiencies by fourfold to sixfold. A population-based study from Sweden reported an association between the use of in vitro *fertilisation* and limb deficiencies (OR 1.86, 95% CI 1.04, 3.07).[13] In the present study, while there was an association between infertility treatment and multiple deficiencies in univariate analysis, the association was attenuated in multivariable analysis.

There are early reports on the maternal use of exogenous sex hormones and associations of various congenital malformations including limb deficiencies.[3,4,41] Other reports have found no appreciable associations.[42] A recent prospective cohort study found no association between the use of oral contraceptives and risk of major birth defects.[43] In the present paper, some subgroup analyses showed positive associations for the use of progestogens and longitudinal deficiencies in multivariable analysis. We could not elucidate whether it is the hormone itself or the technology used, or the maternal and paternal factors related to subfertility, that explain the positive association with congenital limb deficiencies. Previous papers have also experienced the same challenge.[44]

The influence of topical anti-infective gynaecological drugs (G01A) on the risk of congenital limb deficiencies is debatable. These drugs and tetracyclines were associated with transverse deficiencies. In the literature, erythromycins and sulphonamides have been implicated in transverse deficiencies and penicillin in intercalary deficiencies.[45] The number of intercalary cases was small, which reduced our precision and produced chance findings. Another limitation is our inability to determine whether the birth defect is associated with the antibacterial used or the underlying infection. The use of gynaecological anti-infectives is more frequent during all stages of pregnancy than among non-pregnant women.[46] Persistent bacterial vaginosis (BV) is a known risk factor for preterm delivery, infant morbidity, and mortality.[46] Malm et al[46] concluded that there may be a tendency to overestimate the benefits of treating BV and possibly to underestimate the potential risks associated with routine BV treatment during pregnancy.

TABLE 6 Associations of maternal and infant factors and transverse limb deficiencies

	Number of Events		Odds ratio (95% confidence interval)	
	Cases (n = 123)	Controls (n = 615)	Unadjusted	Adjusted*
Maternal age (years)				
<25	35 (28.5%)	116 (18.9%)	1.86 (1.18, 2.94)	1.76 (1.05, 2.96)
25-34	68 (55.3%)	425 (69.1%)	1.00 (Reference)	1.00 (Reference)
≥35	20 (16.3%)	74 (12.0%)	1.64 (0.95, 2.83)	1.94 (1.06, 3.57)
Primiparity	66 (53.7%)	250 (40.7%)	1.70 (1.15, 2.52)	1.75 (1.13, 2.70)
Male sex	65 (52.8%)	302 (49.1%)	1.18 (0.8, 1.76)	1.31 (0.87, 1.99)
Maternal pregestational diabetes	1 (0.8%)	1/615 (0.2%)	5.00 (0.31, 79.94)	5.87 (0.34, 95.96)
Maternal smoking	29 (23.6%)	106 (17.2%)	1.59 (0.99, 2.54)	1.20 (0.70-2.03)
Topical anti-infective gynaecological drugs (G01A)	6 (4.9%)	4 (0.7%)	7.50 (2.12, 26.58)	6.96 (1.73, 28.03)
Tetracyclines (J01A)	3 (2.4%)	5 (0.8%)	3.00 (0.72, 12.55)	4.12 (0.89, 19.21)

*Adjusted for maternal age, parity, child's sex, maternal pregestational diabetes, maternal smoking, topical anti-infective gynaecological drugs, tetracyclines, and nasal decongestants for systemic use.

4 | CONCLUSIONS

Young maternal age, primiparity, male sex, and pregestational diabetes were associated with increased risk of congenital limb deficiency. First-trimester exposure to antiepileptics was associated with limb deficiencies. Ideally, interventions to prevent congenital limb deficiencies should be initiated before conception and include, for example, informing the general public about the risks of maternal medication use during the first trimester of pregnancy.

ORCID

Johanna Syvänen https://orcid.org/0000-0001-6275-5140
Arimatias Raitio https://orcid.org/0000-0001-9114-2204

REFERENCES

1. Bedard T, Lowry RB, Sibbald B, Kiefer GN, Metcalfe A. Congenital limb deficiencies in Alberta-a review of 33 years (1980–2012) from the Alberta Congenital Anomalies Surveillance System (ACASS). *Am J Med Genet A*. 2015;167A(11):2599-2609.
2. Wilcox WR, Coulter CP, Schmitz ML. Congenital limb deficiency disorders. *Clin Perinatol*. 2015;42(2):281-300.
3. Vargesson N. The teratogenic effects of thalidomide on limbs. *J Hand Surg Eur*. 2019;44(1):88-95.
4. Janerich DT, Piper JM, Glebatis DM. Oral contraceptives and congenital limb-reduction defects. *N Engl J Med*. 1974;291(14):697-700.
5. Czeizel A, Keller S, Bod M. An aetiological evaluation of increased occurrence of congenital limb reduction abnormalities in Hungary, 1975–1978. *Int J Epidemiol*. 1983;12(4):445-449.
6. Brent RL. Nongenital malformations following exposure to progestational drugs: The last chapter of an erroneous allegation. *Birth Defects Res A Clin Mol Teratol*. 2005;73(11):906-918. Review.
7. Aro T, Heinonen OP, Saxén L. Risk indicators of reduction limb defects. *J Epidemiol Community Health*. 1983;37(1):50-56.
8. Polednak AP, Janerich DT. Maternal factors in congenital limb-reduction defects. *Teratology*. 1985;32(1):41-50.
9. Caspers KM, Romitti PA, Lin S, Olney RS, Holmes LB, Werler MM. National Birth Defects Prevention Study. Maternal periconceptional exposure to cigarette smoking and congenital limb deficiencies. *Paediatr Perinat Epidemiol*. 2013;27(6):509-520.
10. Martínez-Frías ML, Bermejo E, Rodríguez-Pinilla E, Frías JL. Risk for congenital anomalies associated with different sporadic and daily doses of alcohol consumption during pregnancy: a case-control study. *Birth Defects Res A Clin Mol Teratol*. 2004;70(4):194-200.
11. Firth HV, Boyd PA, Chamberlain PF, MacKenzie IZ, Morriss-Kay GM, Huson SM. Analysis of limb reduction defects in babies exposed to chorionic villus sampling. *Lancet*. 1994;343(8905):1069-1071.
12. Oddy WH, De Klerk NH, Miller M, Payne J, Bower C. Association of maternal pre-pregnancy weight with birth defects: evidence from a case-control study in Western Australia. *Aust N Z J Obstet Gynaecol*. 2009;49(1):11-15.
13. Källén B, Finnström O, Lindam A, Nilsson E, Nygren KG, Otterblad PO. Congenital malformations in infants born after in vitro fertilization in Sweden. *Birth Defects Res A Clin Mol Teratol*. 2010;88(3):137-143.
14. Davies MJ, Moore VM, Willson KJ, et al. Reproductive technologies and the risk of birth defects. *N Engl J Med*. 2012;366(19):1803-1813.
15. Shaw GM, O'Malley CD, Wasserman CR, Tolarova MM, Lammer EJ. Maternal periconceptional use of multivitamins and reduced risk for conotruncal heart defects and limb deficiencies among offspring. *Am J Med Genet*. 1995;59(4):536-545.
16. Botto LD, Olney RS, Erickson JD. Vitamin supplements and the risk for congenital anomalies other than neural tube defects. *Am J Med Genet*. 2004;125C(1):12-21. Review.
17. Aro T. Maternal diseases, alcohol consumption and smoking during pregnancy associated with reduction limb defects. *Early Hum Dev*. 1983;9(1):49-57.
18. Källén B. A prospective study of some aetiological factors in limb reduction defects in Sweden. *J Epidemiol Community Health*. 1989;43(1):86-91.
19. Klungsøyr K, Nordtveit TI, Kaastad TS, Solberg S, Sletten IN, Vik AK. Epidemiology of limb reduction defects as registered in the Medical Birth Registry of Norway, 1970–2016: Population based study. *PLoS One*. 2019;14(7):e0219930.
20. Froster UG, Baird PA. Congenital defects of lower limbs and associated malformations: a population-based study. *Am J Med Genet*. 1993;45(1):60-64.
21. Aberg A, Westbom L, Källén B. Congenital malformations among infants whose mothers had gestational diabetes or preexisting diabetes. *Early Hum Dev*. 2001;61(2):85-95.
22. Becerra JE, Khoury MJ, Cordero JF, Erickson JD. Diabetes mellitus during pregnancy and the risks for specific birth defects: a population-based case-control study. *Pediatrics*. 1990;85(1):1-9.

23. Correa A, Gilboa SM, Besser LM, et al. Diabetes mellitus and birth defects. *Am J Obstet Gynecol*. 2008;199(3):237.e1-e9.
24. Dukhovny S, Van Bennekom CM, Gagnon DR, et al. Metformin in the first trimester and risks for specific birth defects in the National Birth Defects Prevention Study. *Birth Defects Res*. 2018;110(7):579-586.
25. Tinker SC, Gilboa SM, Moore CA, et al. Specific birth defects in pregnancies of women with diabetes: National Birth Defects Prevention Study, 1997–2011. *Am J Obstet Gynecol*. 2020;222(2):176.e1-176.e11.
26. Nielsen GL, Nørgard B, Puho E, Rothman KJ, Sørensen HT, Czeizel AE. Risk of specific congenital abnormalities in offspring of women with diabetes. *Diabet Med*. 2005;22(6):693-696.
27. Pakkasjärvi N, Ritvanen A, Herva R, Peltonen L, Kestilä M, Ignatius J. Lethal congenital contracture syndrome (LCCS) and other lethal arthrogryposes in Finland–an epidemiological study. *Am J Med Genet A*. 2006;140A(17):1834-1839.
28. Leoncini E, Botto LD, Cocchi G, et al. How valid are the rates of Down syndrome internationally? Findings from the International Clearinghouse for Birth Defects Surveillance and Research. *Am J Med Genet A*. 2010;152A(7):1670-1680.
29. Gissler M, Teperi J, Hemminki E, Meriläinen J. Data quality after restructuring a national medical registry. *Scand J Soc Med*. 1995;23(1):75-80.
30. The Social Insurance Institution of Finland. 2020. https://www.kela.fi/tilastojulkaisut_suomen-laaketilasto. Accessed March 25, 2020.
31. Stothard KJ, Tennant PW, Bell R, Rankin J. Maternal overweight and obesity and the risk of congenital anomalies: a systematic review and meta-analysis. *JAMA*. 2009;301(6):636-650.
32. Koskimies E, Lindfors N, Gissler M, Peltonen J, Nietosvaara Y. Congenital upper limb deficiencies and associated malformations in Finland: a population-based study. *J Hand Surg Am*. 2011;36(6):1058-1065.
33. Syvänen J, Nietosvaara Y, Ritvanen A, Koskimies E, Kauko T, Helenius I. High risk for major nonlimb anomalies associated with lower-limb deficiency: a population-based study. *J Bone Joint Surg Am*. 2014;96(22):1898-1904.
34. International Clearinghouse for Birth Defects Surveillance and Research. 2020. http://www.icbdsr.org/. Accessed March 25, 2020.
35. Heinke D, Rich-Edwards JW, Williams PL, et al. National Birth Defects Prevention Study. Quantification of selection bias in studies of risk factors for birth defects among livebirths. *Paediatr Perinat Epidemiol*. 2020;34(6):655-664.
36. Inkster M, Fahey T, Donnan P, Leese G, Mires G, Murphy D. Poor Glycated Haemoglobin Control and Adverse Pregnancy Outcomes in Type 1 and Type 2 Diabetes Mellitus: Systematic Review of Observational Studies. *BMC Pregnancy Childbirth*. 2006;30(6):30.
37. Meinilä J, Koivusalo SB, Valkama A, et al. Nutrient intake of pregnant women at high risk of gestational diabetes. *Food Nutr Res*. 2015;19(59):26676.
38. Duong HT, Hoyt AT, Carmichael SL, et al. National Birth Defects Prevention Study. Is maternal Parity an independent risk factor for birth defects? *Birth Defects Res A Clin Mol Teratol*. 2012;94(4):230-236.
39. Rodríguez-Pinilla E, Arroyo I, Fondevilla J, García MJ, Martínez-Frías ML. Prenatal exposure to valproic acid during pregnancy and limb deficiencies: a case-control study. *Am J Med Genet*. 2000;90(5):376-381.
40. Langer B, Haddad J, Gasser B, Maubert M, Schlaeder G. Isolated fetal bilateral radial ray reduction associated with valproic acid usage. *Fetal Diagn Ther*. 1994;9(3):155-158.
41. Heinonen OP, Slone D, Monson RR, Hook EB, Shapiro S. Cardiovascular birth defects and antenatal exposure to female sex hormones. *N Engl J Med*. 1977;296(2):67-70.
42. Lammer EJ, Cordero JF. Exogenous sex hormone exposure and the risk for major malformations. *JAMA*. 1986;255(22):3128-3132.
43. Charlton BM, Mølgaard-Nielsen D, Svanström H, Wohlfahrt J, Pasternak B, Melbye M. Maternal use of oral contraceptives and risk of birth defects in Denmark: prospective, nationwide cohort study. *BMJ*. 2016;6(352):h6712.
44. Berntsen S, Söderström-Anttila V, Wennerholm UB, et al. The health of children conceived by ART: 'the chicken or the egg?'. *Hum Reprod Update*. 2019;25(2):137-158.
45. Crider KS, Cleves MA, Reefhuis J, Berry RJ, Hobbs CA, Hu DJ. Antibacterial medication use during pregnancy and risk of birth defects: National Birth Defects Prevention Study. *Arch Pediatr Adolesc Med*. 2009;163(11):978-985.
46. Malm H, Martikainen J, Klaukka T, Neuvonen PJ. Prescription drugs during pregnancy and lactation–a Finnish register-based study. Finnish Register-Based Study. *Eur J Clin Pharmacol*. 2003;59(2):127-133.

How to cite this article: Syvänen J, Nietosvaara Y, Hurme S, et al. Maternal risk factors for congenital limb deficiencies: A population-based case–control study. *Paediatr Perinat Epidemiol*. 2021;00:1–9. https://doi.org/10.1111/ppe.12740

APPENDIX 1

List of all analysed ATC drug groups with exposures among case or control mothers.

ATC code	Name of the drug group
A02B	DRUGS FOR PEPTIC ULCER AND GASTRO-OESOPHAGEAL REFLUX DISEASE
A03F	PROPULSIVES
A07E	INTESTINAL ANTI-INFLAMMATORY AGENTS
A10A	INSULINS AND ANALOGUES
B01A	ANTITHROMBOTIC AGENTS
C07A	BETA BLOCKING AGENTS
D01A	ANTIFUNGALS FOR TOPICAL USE
D06B	CHEMOTHERAPEUTICS FOR TOPICAL USE
D07A	CORTICOSTEROIDS, PLAIN
D10A	ANTI-ACNE PREPARATIONS FOR TOPICAL USE
G01A	ANTI-INFECTIVES AND ANTISEPTICS, EXCL. COMBINATIONS WITH CORTICOSTEROIDS
G03C	ESTROGENS
G03D	PROGESTOGENS
G03G	GONADOTROPINS AND OTHER OVULATION STIMULANTS
H01C	HYPOTHALAMIC HORMONES
H02A	CORTICOSTEROIDS FOR SYSTEMIC USE, PLAIN
H03A	THYROID PREPARATIONS
J01A	TETRACYCLINES
J01C	BETA-LACTAM ANTIBACTERIALS, PENICILLINS
J01D	OTHER BETA-LACTAM ANTIBACTERIALS
J01F	MACROLIDES, LINCOSAMIDES AND STREPTOGRAMINS
J01M	QUINOLONE ANTIBACTERIALS
J02A	ANTIMYCOTICS FOR SYSTEMIC USE
L02A	HORMONES AND RELATED AGENTS
M01A	ANTI-INFLAMMATORY AND ANTIRHEUMATIC PRODUCTS, NON-STEROIDS
M03B	MUSCLE RELAXANTS, CENTRALLY ACTING AGENTS
N02B	OTHER ANALGESICS AND ANTIPYRETICS
N02C	ANTIMIGRAINE PREPARATIONS
N03A	ANTIEPILEPTICS
N05A	ANTIPSYCHOTICS
N05B	ANXIOLYTICS
N06A	ANTIDEPRESSANTS
P01A	AGENTS AGAINST AMOEBIASIS AND OTHER PROTOZOAL DISEASES
R01A	DECONGESTANTS AND OTHER NASAL PREPARATIONS FOR TOPICAL USE
R01B	NASAL DECONGESTANTS FOR SYSTEMIC USE
R03A	ADRENERGICS, INHALANTS
R03B	OTHER DRUGS FOR OBSTRUCTIVE AIRWAY DISEASES, INHALANTS
R05D	COUGH SUPPRESSANTS, EXCL. COMBINATIONS WITH EXPECTORANTS
R05F	COUGH SUPPRESSANTS AND EXPECTORANTS, COMBINATIONS
R06A	ANTIHISTAMINES FOR SYSTEMIC USE
S01G	DECONGESTANTS AND ANTIALLERGICS
A10	DRUGS USED IN DIABETES
D01	ANTIFUNGALS FOR DERMATOLOGICAL USE
D07	CORTICOSTEROIDS, DERMATOLOGICAL PREPARATIONS
G03	SEX HORMONES AND MODULATORS OF THE GENITAL SYSTEM
J01	ANTIBACTERIALS FOR SYSTEMIC USE
N06	PSYCHOANALEPTICS
R03	DRUGS FOR OBSTRUCTIVE AIRWAY DISEASES
S01	OPHTHALMOLOGICALS