The risk for four specific congenital heart defects associated with assisted reproductive techniques: a population-based evaluation.

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Title

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Abstract

Study question. Are the risks of hypoplastic left heart syndrome, transposition of great arteries, tetralogy of Fallot (TOF) and coarctation of the aorta increased in infants conceived by different assisted reproductive techniques (ART)?

Study answer. ART, and particularly intracytoplasmic sperm injection (ICSI), are specifically associated with a higher risk of TOF.

What is already known. ART are associated with an increase in the overall risk of birth defects. The risk for congenital heart defects (CHD) associated with ART has been evaluated as a whole but there is limited information on the risks for specific CHD.

Study design, material and methods. We conducted a case-control study using population-based data from the Paris registry of congenital malformations for the period 1987-2009 and a cohort study of CHD (EPICARD) on 1583 cases of CHD and 4104 malformed controls with no known associations with ART. ART included ovulation induction only, IVF and ICSI.

Results. Exposure to ART was significantly higher for TOF than controls (6·6 vs. 3·5%, P=0·002); this was not the case for the other three CHD. ART (all methods combined) were associated with a 2·4-fold higher odds of TOF after adjustment for maternal characteristics, paternal age and year of birth (Adjusted OR 2·4, 95% CI 1·5–3·7) with the highest risk associated with ICSI (Adjusted OR 3·0, 95%CI 1·0-8·9). No statistically significant associations were found for the other CHD.

Limitations. Our study cannot disentangle to what extent the observed associations between risk of TOF and ART are due to causal effects of ART and/or the underlying infertility problems of couples who conceive following ART.

Implications. The developmental basis of the specific association between the risk of TOF and ART ~~and in particular the possible role of neural crest cells in this association~~ need to be further investigated.

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Competing interests. None.

Key Words

Reproductive techniques, assisted

Intracytoplasmic sperm injection

Heart defects, congenital

Tetralogy of Fallot

Epidemiology

Introduction

Assisted reproductive techniques (ART) are known to be associated with a modest increase in the overall risk of congenital anomalies (Wennerholm et al., 2000; Hansen et al., 2002; Koivurova et al., 2002; Hansen et al., 2005; Klemetti et al., 2005; Olson et al., 2005; Schieve et al., 2005). Relatively little specific information exist on the risk of congenital heart defects (CHD) for foetuses conceived following ART (Anthony et al., 2002; Hansen et al., 2002; Katalinic et al., 2004; Lie et al., 2005; Zhu et al., 2006; Reefhuis et al., 2009; Tararbit et al., 2011). Available evidence suggest an overall risk for CHD in relation to ART that is comparable to that found for all congenital anomalies combined (OR~1·4-1·5) (Hansen et al., 2005; Tararbit et al., 2011). Specific associations between different methods of ART and categories of CHD have also been reported by our group (Tararbit et al., 2011) using a subset of the data used in the present study.

However, previous studies have mostly examined the risk of CHD in relation to ART for all CHD combined or for broad categories of CHD rather than for specific CHD. Moreover, the associations between different methods of ART and specific CHD have not been examined. Assessment of such specific associations is important as known teratogens are generally associated with the risk of one or a few specific malformations. Furthermore, specific associations between types of CHD and ART may provide clues about the underlying mechanism of the higher risk of congenital malformations in foetuses conceived following ART.

Using population-based data from the Paris registry of congenital malformations and a cohort study of children with CHD (the EPICARD study), we estimated the risks for four major specific CHD: hypoplastic left heart syndrome (HLHS), transposition of great arteries (TGA), tetralogy of Fallot (TOF), and coarctation of the aorta (CoA) in relation to different methods of ART.

Material and methods

Data sources

Two sources of data were used for this study: 1) the Paris registry of congenital malformations and 2) the EPICARD study (Epidemiological study on the outcomes for congenital heart diseases). These two sources of data are briefly described below.

The Paris registry of congenital malformations

Since 1981, the Paris registry of congenital malformations registers all cases of birth defects and chromosomal anomalies among live-births, still-births (≥ 22 weeks of gestation), and pregnancy terminations. The registry covers the population of women who live in the Greater Paris area (Paris and its surrounding suburb) and deliver or have a termination of pregnancy for foetal anomaly in a Parisian maternity unit. The annual number of deliveries in our population is about 38,000.

The Paris registry is a member of the European network of registries of congenital malformations (European Surveillance of Congenital Anomalies, EUROCAT) and of the International clearinghouse for birth defects surveillance and research (Eurocat special report, 2009; Cocchi et al., 2010; Greenlees et al., 2011; Khoshnood et al.,2011). The registry follows the EUROCAT methodology and quality of data is routinely monitored by both EUROCAT and the French National Committee of Registries. Review of procedures regarding confidentiality of data is overseen by both the National Committee of Registries and the National Committee of Informatics and Freedom (CNIL). Data are based on medical records and are collected from several sources including maternity units, neonatology wards, cytogenetic, and pathology services.

In the present study, data from the registry corresponded to the period January 1st 1987 to December 31st 2009 as the first case of a malformation with exposure to *in vitro* fertilization occurred in 1987 and 2009 was the last year for which data were available at the time of the study.

EPICARD

The EPICARD study is an on-going prospective cohort study of all children with a CHD (Khoshnood et al., 2012) born to women living in the Greater Paris area (Paris and its surrounding suburbs) between 2005 and 2008 regardless of the place of delivery (N = 317,538 births). The principal objectives of the study are to use population-based data from a large cohort of patients with CHD to: i) estimate the total and live birth prevalence, ii) examine timing of diagnosis and assess medical and surgical management of children with CHD, iii) evaluate neonatal mortality and morbidity and neuro-developmental outcomes of children with CHD; and iv) identify the factors associated with their health outcomes, especially the role of events during the neonatal period and of the initial medical and surgical management. All cases (live births, pregnancy terminations, foetal deaths) diagnosed in the prenatal period or up to one year of age in the birth cohorts between May 1st 2005 and April 30th 2008 were eligible for inclusion. The total number of cases of CHD included in the study was 2867, including 2348 live births (82%), 466 pregnancy terminations (16·2%) and 53 foetal deaths (1·8%). Diagnoses were confirmed in specialized paediatric cardiology departments and for the majority of pregnancy terminations and foetal deaths by a foetopathologist examination. For others in which a pathology exam could not be done, the diagnoses were confirmed by consensus by a paediatric cardiologist and a specialist in echocardiography in the study group based on results of prenatal echocardiography examination.

<u>Methods</u>

A case-control study with malformed controls was performed. Cases were foetuses/children with hypoplastic left heart syndrome (HLHS), transposition of great arteries (TGA), tetralogy of Fallot (TOF) and coarctation of the aorta (CoA). Cases included in both the Paris registry and the EPICARD study were counted once. Malformed controls were isolated congenital defects other than CHD for which no evidence of an association with ART was found in literature. As recommended by Hook (1993), we selected a wide spectrum of heterogeneous birth defects as controls in order to decrease the risk of selection bias due to shared etiologic factors between cases and controls (Swan et al., 1992; Lieff et al., 1999). The malformations in the control group comprised cases of club-foot, angioma, skin abnormality, polydactyly, syndactyly and congenital hip dislocation in the Paris registry.

The risk (odds) of each CHD in relation to ART was the main outcome measure. Data on exposure to ART were obtained from medical records. The same procedure for data collection and coding was used for information on ART in the two datasets (Paris registry and EPICARD) used in this study. Exposure to ART included the following categories: ovulation induction (OI) only, IVF, and ICSI. Exposure to ART was assessed as: i) a binary variable (ART yes/no) , ii) a variable in four categories (no ART, OI, IVF, ICSI) and iii) a variable combining IVF and ICSI (IVF + ICSI) in a single category.

Potential confounding factors considered were maternal characteristics (age, occupation and geographic origin), paternal age, and year of birth (or pregnancy termination). Although their exact relations to the risk for specific CHD are not well known, these factors are associated with both exposure to ART and prevalence of birth defects in general (Vrijheid et al., 2000).Maternal occupation was coded in five categories (professional, intermediate, administrative/public service, other, and none) following the French National Institute of Statistics and Economic Studies (INSEE) classification. Geographic origin was coded in four categories: French, North African, Sub-Saharan African, and other countries.

Statistical analysis

The odds of each of the four specific CHD vs. controls in relation to ART was estimated using logistic regression models, after taking into account year of birth, maternal characteristics (age, occupation and geographic origin), and paternal age. Paternal age was missing for 20·6% of the study population. We used multiple imputation (Little and Rubin, 2002) for missing data on paternal age. Paternal age was imputed in twenty sets of data for each CHD separately using the case/control status, exposure to ART, maternal age, and year of birth/termination. The pooled (over the 20 datasets) adjusted ORs for the association between ART and risk of each specific CHD were estimated using the method described by Little and Rubin (2002). In order to explore the possible role of multiple pregnancies in the association between ART and CHD, we also conducted analyses with further adjustment for multiple pregnancies and tested for any interactions effect between multiple pregnancies and ART.

The statistical significance level was set at $\alpha = 0.05$ and all tests were two-sided.

Analyses were done with Stata 11 software (Statacorp, Texas, USA).

Ethics approval

No specific ethical approval was needed for this particular analysis. The French National Committee of Informatics and Freedom (CNIL) has authorised the surveillance and research activities of the registry using anonymous data and has approved the EPICARD study.

Results

Study population

After excluding cases with missing data on ART (3% of cases), the study population comprised 353 cases of HLHS, 444 cases of TGA, 395 cases of TOF and 391 cases of CoA. Approximately 14% of cases of HLHS, 3% of TGA, 20% of TOF and 10% of CoA were associated with chromosomal anomalies. The study population included 4104 malformed controls with complete information on ART, which comprised 1436 with congenital hip dislocation, 824 with club-foot, 782 with polydactyly, 517 with angioma, 381 with skin abnormality, and 164 with syndactyly with complete information on ART; 3% of controls had missing data on ART.

Table 1 summarises the results of the comparison of the maternal, paternal and pregnancy characteristics of cases of CHD (all four specific CHD combined) and controls. Overall, mothers of cases of CHD were older, more likely to be from North Africa and in the occupational category "none" as compared with mothers of controls. Still births and terminations of pregnancy for foetal anomaly were more frequent for cases of CHD than controls.

When comparisons of the characteristics of cases and controls were done for the four defects separately (detailed results not shown – available from authors), for CHD other than TOF, the characteristics of cases and controls were for the most part comparable, except that mothers of cases of CoA were more likely to be from North Africa than controls. Most sociodemographic characteristics were different between cases of TOF and controls. Mothers of cases of TOF were significantly older and more likely to be from North Africa than controls. Mothers of cases of TOF were also more likely to be in the occupational category “none” than controls (data not shown).

Risk of CHD associated with ART

All cases

Exposure to ART (all methods combined, Table 2) was significantly higher for cases of TOF than controls (6·6% vs. 3·5%, p=0·002). Exposure to the different methods of ART (data not shown) was also significantly different between cases of TOF and controls, in particular 2·5% of TOF were born following IVF vs. 1·3% of controls and 1·3% of TOF were born following ICSI vs. 0·3% of controls (p=0·004). Exposure to ART was not associated with a significantly higher risk of other CHD.

Exposure to ART was associated with a 2·4-fold increase in the maternal characteristics and year of birth-adjusted odds of TOF (Adjusted OR= 2·4, 95%CI 1·5– 3·7) (Table 3). ~~The odds of TOF associated with ART remained similar after further adjustment for paternal age~~. In contrast, ART were not associated with statistically significant increases in the risks of HLHS, TGA or CoA and the ORs were generally close to the null value (Table 3). All three methods of ART were associated with significantly higher odds of TOF (Table 4). In particular, ICSI was associated with a three-fold higher odds of TOF after adjustment for maternal characteristics and year of birth (Adjusted OR= 3·0, 95%CI 1·0-8·9). There was no evidence that IVF was associated with a higher odds of TOF as compared with OI (for IVF: Adjusted OR=2·0, 95%CI 1·0 – 4·2; for OI: Adjusted OR= 2·5, 95%CI 1·3– 4·8). For the other three specific CHD, no statistically significant associations were observed. Further adjustment for paternal age using the multiple imputation estimates did not modify appreciably the above estimates (data not shown).

Cases without associated chromosomal anomalies

Tables 3 and 5 show the results of the analyses for the associations between the risks of the four CHD and ART (all methods combined, Table 3) and separately for different methods of ART (Table 5) for the subset of cases without associated chromosomal anomalies. All estimates were essentially the same as those found for all cases combined (i.e. when cases of each specific CHD with and without associated chromosomal anomalies were analysed together).

Results of the analyses, which included further adjustment for multiple pregnancies were essentially the same as those found without adjustment for multiple pregnancies (data not shown). We found no statistically significant interaction effects between ART and multiple pregnancies for any of the four CHD (data not shown).

Discussion

Using population-based data on nearly 1600 cases of specific congenital heart defects (CHD), we assessed the risk of four specific CHD in relation to assisted reproductive techniques (ART). We found that ART (all methods combined) were associated with a 2·4-fold increased risk of tetralogy of Fallot (TOF), after taking into account maternal age, occupation, geographic origin, paternal age, and year of birth. In particular, ICSI was associated with a three-fold higher adjusted odds of TOF. In contrast, we did not find any statistically significant increases in the risk of CHD in relation to ART for the other CHD in our study, i.e., hypoplastic left heart syndrome (HLHS), transposition of the great arteries (TGA), and coarctation of the aorta (CoA). Risk estimates were comparable when cases with chromosomal anomalies were excluded, suggesting that the associations between ART and TOF are not due to the association of the latter with chromosomal anomalies. Further adjustement for multiple pregnancies did not substantially modify our results.

On the basis of our findings, we calculated attributable risk fractions, which would represent the proportion of cases of TOF that may be caused by ART, or equivalently, the proportion of cases of TOF that would be avoided were the exposure to ART removed ceteris paribus, 'if' the association we found between the risk of TOF and ART can be assumed to represent a causal relation (this may of course not be the case in part for reasons that are discussed further below). The attributable risk fraction estimates suggested in particular that around 6.5% of the TOF may have been caused by ART (all methods combined) and 2% by ICSI.

Our study has certain limitations. We had limited power to detect OR lower than two in the association between ART (for all methods combined) and specific CHD and three in case of the different methods of ART. Therefore, our study may have had insufficient power to detect statistically significant associations for other CHD.

The models used to estimate the odds ratios for the different defects in relation to ART were not nested (i.e., were separate models) and we did not formally test the statistical significance of differences in the odds ratios for one defect vs. another. The associations were not statistically significant for any of the defects except for TOF, whereas the numbers of cases for the other CHD were comparable to those of TOF.

A potential source of bias in our study is related to the use of malformed controls (Swan et al., 1992; Lieff et al., 1999). The main advantage of using malformed controls is to reduce the risk of recall or other sources of information bias. But malformed controls may also be a source of selection bias if malformations included as controls are either directly or indirectly associated with ART. Risks could be under (over)-estimated if malformations included in the control group occur more (less) frequently in foetuses conceived following ART. By selecting a heterogeneous group of malformations with no known association with ART, as recommended by Hook (1993), we aimed to minimize such bias. However, the possibility of residual bias due to shared aetiologies between cases and malformed controls cannot be excluded.

A differential misclassification bias for exposure assessment cannot be excluded if exposure to ART is ascertained in a different way for cases and controls. However, we have no reason to believe that ART may have been ascertained differentially for cases of TOF vs. the other CHD examined in our study.

We had a relatively high proportion of missing data on paternal age. The latter is known to be associated with ART and more specifically with ICSI. Estimates for ICSI could therefore be biased if distribution of paternal age was different for subjects with missing data. We used multiple imputations for imputing missing paternal age using case/control status, exposure to ART, maternal age and year of birth and adjustment for paternal age did not appreciably change our results. However, residual bias due to other paternal characteristics cannot be excluded.

The question of multiple pregnancies and its association with both ART and the risk of congenital anomalies is an important issue to consider. There is evidence suggesting that multiple pregnancies may be associated with a higher risk of congenital anomalies (Mastroiacovo, et al., 1999; Glinianaia et al., 2008). This may specifically be the case for CHD, although relatively little, and at times contradictory, information exist on the associations between multiple pregnancies and CHD (Manning et al., 2006; Bahtiyar et al., 2007; Campbell et al., 2009). Moreover, it is not clear to what extent any association between multiple pregnancies and CHD may in fact be due to ART. Our results remained similar after further adjustment for multiple pregnancies and we did not find any statistically significant interaction effects between ART and multiple pregnancies for any of the CHD, although this may have been due to limited power of our study for detecting interaction effects. In any case, none of the above precludes the possibility that multiple pregnancies may be on the causal pathway between ART and CHD. It is worth noting however that the public health impact of ART on the risk for birth defects, including that of TOF found in our study, includes all (singleton and multiple) pregnancies.

Specific associations between ART and certain categories of CHD, particularly the so-called conotruncal defects, which include TOF, have been reported (Reefhuis et al., 2009; Tararbit et al., 2001). In a recent study (Tararbit et al., 2011), the risk of CHD associated with ART was also shown to vary more generally for different methods of ART and categories of CHD defined based on anatomic and clinical criteria (Houyel et al., 2011). In particular, the authors found a stronger association between ICSI and the category “Malformations of the outflow tracts and ventriculoarterial connections” that comprised, among other CHD, the conotruncal defects.

The developmental origins of TOF are complex and not fully understood but they may involve abnormal development of neural crest cells. None of the other three CHD studied is known to be of cardiac neural crest origin. In particular TGA which is a defect of the outflow tract does not belong to the group of the conotruncal defects (Houyel et al., 2011) and migration/proliferation of neural crest cell appear to be normal in this condition (Bajolle et al., 2006). In order to further investigate, the hypothesis of the involvement of neural crest cells in the association between TOF and ART, we assessed the risk for other, rarer CHD thought to be of neural crest origin (TOF with pulmonary atresia, TOF with absent pulmonary valve, and common arterial trunk).

We found an increased overall risk associated with ART (data not shown) but the confidence intervals were wide due to small sample sizes.

Given the uncertainties about both the developmental origins of cardiac defects and possible effects of ART on foetal development, the hypothesis of a potential implication of neural crest cells in the association between ART and TOF must be regarded as very tentative and no more than a reasonable speculation. Future observational and experimental studies using other designs (e.g., animal studies, genetic studies, fundamental research in biology of reproduction / ART as well as additional epidemiological studies) are needed to both further assess our observations and in order to understand the possible underlying mechanisms of the association between the risk of TOF and ART.

In conclusion, we found that cases of TOF were more likely to have been conceived following ART when compared with controls. ART were associated with a 2·4-fold higher risk of TOF after adjustment for maternal age, occupation, geographic origin, paternal age and year of birth; ICSI was specifically associated with a three-fold higher risk of TOF. In contrast, we did not find statistically significant associations between ART and HLHS, TGA or CoA and most ORs were close to the null value. Our study cannot disentangle to what extent the observed associations between risk of TOF and ART may be due to any causal effects of ART and/or the underlying infertility problems of couples who conceive following ART. Nevertheless, the developmental basis of the specific association between risk of TOF and ART, particularly ICSI, and the potential implication of neural crest cells in this association, need to be further investigated.

Role of authors

B. Khoshnood conceived the study. K. Tararbit conducted the main statistical analyses and wrote the first draft of the manuscript with B. Khoshnood. N. Lelong, A-C. Thieulin assisted with statistical analysis. L. Houyel, D. Bonnet and F. Goffinet contributed to the conceptualization of ideas and made suggestions about the required analyses. L. Houyel and D. Bonnet provided expertise as paediatric cardiologists. All of the authors contributed to the interpretation of findings and revisions of the article.

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Conflict of interest

None declared

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Table 1. Associations between predictor variables and case/control status.

	characteristics	controls		cases		p
		n	%[§]	n	%[§]	
Mother	Age (years)					
	mean (SD)	30·4 (5·2)		30·9 (5·5)		
	median (p25-p75)	30 (27 - 34)		31 (27 - 35)		
	<20	59	1·4	22	1·4	0·011
	20 - 29	1809	42·8	654	40·1	
	30 - 34	1434	33·9	531	32·6	
	35 - 39	722	17·1	316	19·4	
	≥ 40	203	4·8	107	6·6	
	missing*	23	0·5	12	0·7	
	Geographic origin					
	France	2412	57·9	882	54·5	<0·001
	North Africa	433	10·4	247	15·3	
	Subsaharan Africa	550	13·2	163	10·1	
	Other	770	18·5	327	20·2	
	missing*	85	2·0	23	1·4	
	Occupation					
	none	1083	26·3	440	29·7	<0·001
	professional	997	24·2	343	23·2	
	intermediate	856	20·8	263	17·8	
	administrative/public service	852	20·7	249	16·8	
	other	330	8·0	185	12·5	
	missing*	132	3·1	162	9·9	
Father	Age (years)					
	mean (SD)	33·9 (6·6)		34·4 (6·7)		
	median (p25-p75)	33 (29 - 38)		33 (30 - 38)		
	<20	5	0·1	3	0·2	0·133
	20 - 29	890	25·8	277	22·6	
	30 - 34	1198	34·7	422	34·4	
	35 - 39	734	21·3	281	22·9	
	≥ 40	623	18·1	244	19·9	
	missing*	800	18·8	415	25·3	
Pregnancy	Multiplicity					
	singletons	2768	96·1	1382	95·8	0·756
	twins	103	3·6	57	4·0	
	triplets	8	0·3	3	0·2	
	Outcome					
	still-births	7	0·2	46	2·8	<0·001
	live-births	4231	99·6	1074	65·4	
	pregnancy terminations	12	0·3	522	31·8	

* % of missing data calculated with the total number of cases or controls as a denominator

§ % calculated with the total of cases or controls without missing data as a denominator

Table 2. Numbers of cases and controls and proportions of fetuses conceived after Assisted Reproductive Technologies (ART).

		N	% exposed to ART	p†
Controls *		4 104	3.5	
All cases	Hypoplastic left heart syndrome	353	2.8	0.491
	Transposition of the great arteries	444	2.7	0.363
	Tetralogy of Fallot	395	6.6	0.002
	Coarctation of the aorta	391	3.3	0.831
Cases without chromosomal anomalies	Hypoplastic left heart syndrome	303	2.6	0.413
	Transposition of the great arteries	430	2.8	0.423
	Tetralogy of Fallot	315	7.3	0.001
	Coarctation of the aorta	350	3.7	0.860

* The following malformations were used as controls: club-foot, angioma, skin abnormality, polydactyly, syndactyly and congenital hip dislocation.

† Comparison of the proportion of children/fetuses conceived after ART between the specific CHD and the malformed controls.

Table 3. Logistic regression analyses of the associations between assisted reproductive technologies (ART, all methods combined) and four specific congenital heart defects (CHD).

	CHD	ART	Unadjusted OR*	95% CI	Maternal Adjusted† OR*	95% CI
All cases	Hypoplastic left heart syndrome	None	1.0	ref.	1.0	ref.
		All methods combined	0.8	0.4 - 1.5	0.8	0.4 - 1.8
	Transposition of the great arteries	None	1.0	ref.	1.0	ref.
		All methods combined	0.8	0.4 - 1.4	0.7	0.4 - 1.4
	Tetralogy of Fallot	None	1.0	ref.	1.0	ref.
		All methods combined	1.9	1.3 - 3.0	2.4	1.5 - 3.7
	Coarctation of the aorta	None	1.0	ref.	1.0	ref.
		All methods combined	0.9	0.5 - 1.7	1.1	0.6 - 2.0
Cases without chromosomal anomalies	Hypoplastic left heart syndrome	None	1.0	ref.	1.0	ref.
		All methods combined	0.7	0.4 - 1.5	0.8	0.3 - 1.7
	Transposition of the great arteries	None	1.0	ref.	1.0	ref.
		All methods combined	0.8	0.4 - 1.4	0.7	0.4 - 1.4
	Tetralogy of Fallot	None	1.0	ref.	1.0	ref.
		All methods combined	2.2	1.4 - 3.4	2.6	1.6 - 4.2
	Coarctation of the aorta	None	1.0	ref.	1.0	ref.
		All methods combined	1.1	0.6 - 1.9	1.2	0.6 - 2.2

* Odds ratios (OR) represent the odds of a birth (including live births, stillbirths and pregnancy terminations) with congenital heart defect (cases) relative to the odds of a birth with one of the malformed controls (club-foot, angioma, skin abnormality, polydactyly, syndactyly and congenital hip dislocation).

†Adjusted for maternal age, geographic origin, occupation and year of birth.

Table 4. Logistic regression analyses of the associations between the different methods of assisted reproductive technologies (ART) and four specific congenital heart defects (CHD).

CHD	ART	Unadjusted OR*	95% CI	Maternal Adjusted† OR*	95% CI
Hypoplastic left heart syndrome	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.7	0.3 - 1.9	0.9	0.3 - 2.5
	IVF	0.6	0.2 - 2.0	0.5	0.1 - 2.3
	ICSI	1.8	0.4 - 7.9	1.6	0.3 - 7.2
	IVF + ICSI	0.8	0.3 - 2.1	0.8	0.3 - 2.3
Transposition of the great arteries	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.6	0.2 - 1.5	0.6	0.2 - 1.7
	IVF	1.2	0.5 - 2.6	1.0	0.4 - 2.5
	ICSI	/	/	/	/
	IVF + ICSI	/	/	/	/
Tetralogy of Fallot	None	1.0	ref.	1.0	ref.
	Ovulation induction only	1.5	0.8 - 2.9	2.5	1.3 - 4.8
	IVF	2.0	1.0 - 3.9	2.0	1.0 - 4.2
	ICSI	4.1	1.5 - 11.6	3.0	1.0 - 8.9
	IVF + ICSI	2.4	1.3 - 4.2	2.3	1.2 - 4.2
Coarctation of the aorta	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.7	0.3 - 1.7	1.0	0.4 - 2.6
	IVF	1.0	0.4 - 2.4	1.1	0.4 - 2.9
	ICSI	2.4	0.7 - 8.5	1.2	0.2 - 5.6
	IVF + ICSI	1.2	0.6 - 2.6	1.1	0.5 - 2.6

* Odds ratios (OR) represent the odds of a birth (including live births, stillbirths and pregnancy terminations) with congenital heart defect (cases) relative to the odds of a birth with one of the malformed controls (club-foot, angioma, skin abnormality, polydactyly, syndactyly and congenital hip dislocation).

†Adjusted for maternal age, geographic origin, occupation and year of birth.

Table 5. Logistic regression analyses of the associations between assisted reproductive technologies (ART) and four specific congenital heart defects (CHD) without associated chromosomal anomalies.

CHD	ART	Unadjusted OR*	95% CI	Maternal Adjusted† OR*	95% CI
Hypoplastic left heart syndrome	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.7	0.3 - 1.9	0.8	0.2 - 2.5
	IVF	0.5	0.1 - 2.0	0.3	0.0 - 2.4
	ICSI	2.1	0.5 - 9.2	1.8	0.4 - 8.4
	IVF + ICSI	0.8	0.3 - 2.2	0.7	0.2 - 2.3
Transposition of the great arteries	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.6	0.2 - 1.5	0.6	0.2 - 1.8
	IVF	1.2	0.5 - 2.7	1.1	0.5 - 2.6
	ICSI	/	/	/	/
	IVF + ICSI	/	/	/	/
Tetralogy of Fallot	None	1.0	ref.	1.0	ref.
	Ovulation induction only	1.6	0.8 - 3.2	2.3	1.1 - 4.8
	IVF	2.2	1.1 - 4.5	2.5	1.2 - 5.2
	ICSI	5.2	1.8 - 14.7	3.7	1.3 - 10.9
	IVF + ICSI	2.8	1.6 - 5.0	2.8	1.5 - 5.2
Coarctation of the aorta	None	1.0	ref.	1.0	ref.
	Ovulation induction only	0.8	0.3 - 1.9	1.1	0.4 - 2.8
	IVF	1.1	0.4 - 2.7	1.3	0.5 - 3.3
	ICSI	2.7	0.8 - 9.6	1.3	0.3 - 6.1
	IVF + ICSI	1.4	0.7 - 2.9	1.3	0.6 - 2.9

* Odds ratios (OR) represent the odds of a birth (including live births, stillbirths and pregnancy terminations) with congenital heart defect (cases) relative to the odds of a birth with one of the malformed controls (club-foot, angioma, skin abnormality,polydactyly, syndactyly and congenital hip dislocation)

†Adjusted for maternal age, geographic origin, occupation and year of birth.

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