In vitro fertilization is associated with an increase in major birth defects

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Objective: To determine the risk of major birth defects in cohorts of children conceived through IVF or through IUI as compared with naturally conceived children.
Design: Retrospective cohort study.
Setting: Academic medical center.
Patient(s): Children conceived by IVF or IUI at the University of Iowa from 1989 through 2002, compared with a matched cohort of naturally conceived children.
Intervention(s): None.
Main Outcome Measure(s): Outcome data were obtained from Iowa state birth and fetal death certificates and from the Iowa Birth Defects Registry.
Result(s): Ninety of 1,462 IVF-conceived children (6.2%) and 17 of 343 IUI-conceived children (5.0%) had a major birth defect, compared with 369 of 8,422 naturally conceived children (4.4%). The adjusted odds ratio of a major birth defect in all IVF-conceived children was 1.30 (95% confidence interval [CI] 1.00–1.67) and 1.11 (95% CI 0.67–1.84) for IUI-conceived children. The birth defect rate was increased after IVF when the analysis was limited to term singletons. Cardiovascular and musculoskeletal defects and known birth defect syndromes were increased after IVF. Among IVF-conceived children, there was no difference in birth defect rates after intracytoplasmic sperm injection (ICSI) or after transfer of cryopreserved embryos.
Conclusion(s): Infants conceived through IVF have a slightly higher rate of major birth defects. More birth defects are noted among children born to infertile couples treated with IUI, although this difference is not statistically significant. Larger studies of infants conceived by infertile couples after all types of infertility treatment are needed to definitively determine whether the increased risk of birth defects is secondary to problems inherent in the infertile couple and/or factors associated with some aspect of the treatment. (Fertil Steril® 2005; 84:1308–15. ©2005 by American Society for Reproductive Medicine.)

Key Words: IVF, infertility, intrauterine insemination, ovulation induction, birth defects, congenital anomaly

The association between IVF and an increased risk of birth defects is controversial. Early studies suggesting that IVF was safe with respect to birth defects are difficult to interpret owing to small size, lack of appropriate controls, and inconsistent methods for detecting birth defects in the treated and control groups (1–5). Several more recent matched cohort studies have demonstrated an increased risk of birth defects in general (6) or cardiovascular birth defects in particular (7) associated with IVF, whereas others have not found an increased risk (8, 9). Two recent studies comparing IVF birth defect rates with national registry data have found increased rates of birth defects in children conceived with IVF, but the difference lost significance when the data were controlled for maternal age, parity, and plurality (10, 11).

Clarifying the possible association between IVF and birth defects is critical because almost 1% of children in the United States are now conceived by IVF. Of equal importance is identifying the mechanism behind this possible association. Some have speculated that epigenetic errors, such as defects in DNA methylation and imprinting, might be caused by the embryo culture that follows IVF. This hypothesis is supported by recent reports of the association between IVF and Beckwith-Wiedemann syndrome (12–14), retinoblastoma (15), and Angelman Syndrome (16, 17), conditions caused by defects in genomic imprinting. Other possible mechanisms of the purported association between IVF and birth defects could include the effects of controlled ovarian hyperstimulation and ovulation induction, in vitro sperm preparation, or an inherent defect in the infertile couple perhaps leading to both the infertility and the birth defect in the resulting child. We sought to determine the risks of birth defects in children conceived after infertility treatment as compared with the risk of defects in a cohort of naturally conceived children born in Iowa. Our study has the benefit of including infants conceived both by IVF with embryo culture and by IUI in which no embryo culture or manipulation occurred. Comparing birth defect rates in these

Received September 16, 2004; revised and accepted March 3, 2005.
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0015-0282/05/$30.00
doi:10.1016/j.fertnstert.2005.03.086

two groups of children with birth defect rates in a cohort of naturally conceived children helps to elucidate whether birth defects result from IVF and embryo culture, from other infertility treatments, or from a factor outside of infertility treatment protocols. Importantly, we have a statewide birth defects registry in which all birth defects are reported in a uniform and unbiased manner.

MATERIALS AND METHODS

The study population included the total cohort of IVF and IUI births (live births and stillbirths delivered at ≥20 weeks' gestational age) conceived at the University of Iowa from 1989 through 2002, excluding children born to couples living outside Iowa and children whose birth certificate did not list Iowa as their state of residence. These children were excluded because we could not be sure that we had accurate birth defect data for them.

For the purpose of this analysis, IVF is defined as any treatment that included retrieval of oocytes and fertilization of those oocytes in vitro (either by intracytoplasmic sperm injection [ICSI] or by culturing oocytes and spermatozoa together) followed by some period of embryo culture. Thus, IVF conceptions include babies born after zygote intrafallopian transfer (ZIFT, culture of embryos for 1 day before transfer to the fallopian tube), after IVF with transcervical transfer of embryos (2, 3, or 5 days in culture), and after transfer of cryopreserved embryos. Intrauterine insemination pregnancies include all pregnancies resulting from IUI, with and without concomitant ovulation induction in the female. However, more than 95% of IUI pregnancies included in this study were conceived with either oral or injectable ovulation-inducing medications in addition to the IUI.

For each infant conceived through infertility treatment, up to five naturally conceived children were selected from a 65-county area (corresponding to the pooled counties of residence of all children conceived through infertility treatments) and were matched on plurality, maternal age at time of delivery (±1 year), year of birth (±2 years), and race (white and other). These couples were not in our infertility database and were assumed to have conceived spontaneously. The resulting children are referred to as "controls" for the rest of this article. Outcome data, including gestational age, birth weight, and pregnancy risk factors and complications were obtained from Iowa birth and death certificates. Included in the analysis are 1,462 children conceived through IVF (476 from ICSI, 335 from cryopreserved em-

TABLE 1

Characteristics of couples in the IVF, IUI, and control groups.

Characteristic	IVF (n = 864)	IUI (n = 270)	Controls (n = 6,374)
Maternal age (y) (mean ± SD)	33.9 ± 4.6[a]	32.4 ± 4.3[a]	33.3 ± 4.3
Paternal age (y) (mean ± SD)	36.1 ± 5.6[a]	34.2 ± 4.9	34.6 ± 5.5
Parity (%)			
0	57[a]	63.4[a]	20
≥1	43[a]	37[a]	80
Maternal race/ethnicity (%)			
Caucasian	97.1	95.2	97.0
Black	0.2	0	0.7
Hispanic	0.9	1.9	1.3
Other	1.7	2.9	1.0
Paternal race/ethnicity (%)			
Caucasian	96.8[a]	94.4[a]	89.3
Black	0.9	0	1.2
Hispanic	1.0	3.0	8.3
Other	1.3	2.6	1.3
Married (%)	99.2[a]	99.0[a]	89.7
Maternal education (y) (median)[b]	16.0[a]	16.0[a]	14.0
Paternal education (y) (median)[b]	15.0[a]	16.0[a]	14.0
Maternal smoking (%)	1.9[a]	4.7[a]	13.3
Maternal alcohol use (%)	0.4[a]	0.7[a]	2.0

Note: All *P* values are for comparison with control group and are noted only if significant.

[a] $P<.005$.

[b] Education variable: values 13–16 = college (1–4 y).

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bryos, and 415 from ZIFT procedures), 343 conceived through IUI, and 8,422 control children. A total of 5,499 singletons (645 IVF, 264 IUI, 4,590 control), 4,248 twins (672 IVF, 64 IUI, 3,512 control), and 480 triplets/quadruplets (145 IVF, 15 IUI, 320 control) were evaluated.

The Iowa Birth Defects Registry (IBDR) database was used to ascertain the presence of birth defects in both children born after infertility treatments and controls. The IBDR was established as an active surveillance system in 1983. Using standardized abstracting forms and searching on ICD9 codes, the IBDR audits all Iowa hospitals for birth defects diagnosed by physicians (most often pediatricians, obstetricians, and family practitioners) in terminated and stillborn deliveries as well as liveborn children through 1 year of age. Trained medical abstractors, using standardized written guidelines, systematically review all medical records, including the child's medical record, the delivery room record, clinic notes, and discharge summaries to ascertain types of defects. If the diagnosis is in question, a pediatric geneticist reviews the records to make a determination, consulting the diagnosing physician for clarification if necessary. For cardiac defects, an echocardiogram is required to confirm the diagnosis. Major and minor birth defects were defined by Centers for Disease Control and Prevention guidelines and were categorized after blinded review of the abstracting forms by a pediatric geneticist (K.K.N.) (18). In general, major malformations are considered to cause functional impairment or require surgical correction, with other birth defects being considered minor. In addition, birth defects were classified by the specific type of defect as well as by affected organ system(s). Major defects were further classified as isolated, multiple, or as a component of an identified chromosomal or other syndrome. This study was approved by the University of Iowa institutional review board.

TABLE 2

Characteristics of infants in the IVF, IUI, and control populations.

Characteristic	IVF (n = 1,462)	*P*	IUI (n = 343)	*P*	Control (n = 8,422)
Mean gestational age (wk) (mean ± SD)					
All	36.5 ± 3.6	<.001	37.5 ± 3.9	.031	37.5 ± 3.0
Singletons	38.7 ± 2.2		38.7 ± 2.3		39.1 ± 1.9
Very low birth weight (<1,500 g)					
All	115 (7.9)	<.001	25 (7.3)	<.001	329 (3.9)
Singletons	12 (1.9)		7 (2.7)		37 (0.8)
Twins	62 (9.2)		9 (14.1)		210 (6.0)
Triplets/quadruplets	41 (28.3)		9 (60.0)		82 (25.6)
Low birth weight (<2,500 g)					
All	500 (34.2)	.002	72 (21.0)	<.001	2,011 (23.9)
Singletons	44 (6.8)		23 (8.7)		195 (4.3)
Twins	326 (48.5)		34 (53.1)		1,520 (43.3)
Triplets/quadruplets	130 (89.7)		15 (100)		296 (92.5)
Preterm delivery (<32 wk)					
All	129 (8.8)	<.001	23 (6.7)	.003	343 (4.1)
Singletons	10 (1.6)		6 (2.3)		36 (0.8)
Twins	70 (10.4)		8 (12.5)		237 (6.8)
Triplets/quadruplets	49 (33.8)		9 (60.0)		70 (21.9)
Multiple births					
Twins	672 (46.0)		64 (18.7)		3,512 (41.7)
Triplets/quadruplets	145 (9.9)		15 (4.4)		320 (3.79)
Male sex					
All	735 (50.3)		179 (52.2)		4,190 (49.8)
Singletons	330 (51.2)		132 (50.0)		2,309 (50.3)
Cesarean delivery					
All	706 (48.3)	<.001	114 (33.2)		3,152 (37.4)
Singletons	198 (30.8)		79 (29.9)		1,086 (23.7)

Note: Data are presented as n (%), unless otherwise noted. Data in this table are for descriptive purposes; no adjustments were made for confounding variables in the analysis. Statistical analyses were performed only for the "all infants" groups and not based on plurality. *P* values are for comparison with control group and are not significant if not shown.

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TABLE 3

Prevalence of major birth defects diagnosed by 1 year of age.

Group	No. of infants	Prevalence (%)	Unadjusted OR (95% CI)	*P*	OR adjusted for plurality and/or parity	*P*
All infants						
Control	8,422	369 (4.4)	Reference		Reference	
IVF	1,462	90 (6.2)	1.44 (1.12–1.85)	.004	1.30 (1.00–1.67)[a]	.048[a]
IUI	343	17 (5.0)	1.14 (0.70–1.87)	.593	1.11 (0.67–1.84)[a]	.679[a]
All singletons						
Control	4,590	171 (3.7)	Reference		Reference	
IVF	645	38 (5.9)	1.62 (1.12–2.34)	.010	1.44 (0.98–2.12)[b]	.061
IUI	264	13 (4.9)	1.33 (0.75–2.37)	.324	1.19 (0.66–2.13)[b]	.568
All term singletons (≥37 wk)						
Control	4,285	148 (3.5)	Reference		Reference	
IVF	581	34 (5.8)	1.74 (1.18–2.56)	.006	1.57 (1.04–2.36)[b]	.031
IUI	231	12 (5.2)	1.53 (0.84–1.79)	.164	1.38 (0.75–2.57)[b]	.298

Note: Logistic regression with the GEE method accounting for correlation between infants from same mother.

[a] Adjusted for plurality and parity.

[b] Adjusted for parity.

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Statistical analysis was performed with commercial software (SAS version 9.0; SAS Institute, Cary, NC). Logistic regression analysis with the method of generalized estimating equations (GEE) was used to test the association of IVF and IUI with major birth defects. The GEE method was used so as to be able to account for the correlation among infants with the same mother. A number of potential risk factors were considered in the model, including infant gestational age and birth weight, maternal and paternal age, education, race, marital status, and maternal alcohol and tobacco use. From this analysis, odds ratios (OR) with 95% confidence intervals (CI) of a major defect for IVF and for IUI relative to natural conception were computed. The infant and parent characteristics of the IVF and IUI groups were compared with the control group by Fisher's exact test for categorical variables and by one-way analysis of variance (ANOVA) with Tukey's procedure for continuous variables. To account for the effect of plurality (singleton, twins, triplets/quadruplets), the Cochran-Mantel-Haenzsel test was used to compare categorical variables, and the two-way ANOVA was used for continuous variables.

RESULTS

Demographic data for identified couples were obtained from vital records and are listed in Table 1. Despite matching on age, the women treated by IVF were slightly older and the women treated by IUI slightly younger than control mothers. The mean age of the fathers in the IVF group was slightly higher than that of men in the IUI or control groups. Women in the treatment groups were more likely to be nulliparous, married, and more highly educated than women in the control groups. Women in the IVF and IUI groups were less likely to use alcohol or tobacco during pregnancy than controls. Racial distribution was similar among the women in the study; however, there was greater variation among the men, with a higher percentage of Hispanic fathers in the control group (Table 1). Children in the treatment groups were more likely to be delivered at an earlier gestational age and to be delivered by cesarean section (Table 2). Our findings regarding birth weight in these populations of children will be the subject of another article and are therefore not reported in detail here. There was no significant difference in gender distribution in children from the different groups. Statistical adjustments for any differences found between the groups were made for evaluations of birth defect rates.

We found that children conceived through IVF had a statistically significant increase in major birth defects as compared with control children (Table 3). The difference was present whether analysis included all infants or just singletons born at term. No statistically significant difference in birth defects was noted in children conceived after IUI procedures as compared with control children (Table 3).

Evaluation of different treatments within the IVF category showed no significant differences in birth defect rates. Children born after ZIFT (1 day of embryo culture followed by ET to the fallopian tube) had the same birth defect rates as children born after more prolonged embryo culture followed by uterine ET (OR 1.10, 95% CI 0.65–1.86 for ZIFT). A comparison of birth defect rates between ICSI- and non–

TABLE 4

Major birth defects by affected organ system.

System	All infants (n = 10,227) Control (n = 8,422)	IVF (n = 1,462)	P	IUI (n = 343)	P	Singletons (n = 5,499) Control (n = 4,590)	IVF (n = 645)	P	IUI (n = 264)	P
CNS	50 (0.6)	9 (0.6)		1 (0.3)		15 (0.3)	2 (0.3)		1 (0.4)	
Cardiovascular	100 (1.2)	33 (2.3)	.002	5 (1.5)		45 (1.0)	16 (2.5)	.003	4 (1.5)	
Ear	29 (0.3)	8 (0.6)		2 (0.6)		12 (0.3)	4 (0.6)		2 (0.8)	
Eye	42 (0.5)	8 (0.6)		2 (0.6)		19 (0.4)	5 (0.8)		0 (0)	
Gastrointestinal	48 (0.6)	8 (0.6)		3 (0.9)		15 (0.3)	4 (0.6)		2 (0.8)	
Genitourinary	86 (1.0)	12 (0.8)		2 (0.6)		39 (0.9)	5 (0.8)		0 (0)	
Musculoskeletal	103 (1.2)	32 (2.2)	.007	9 (2.6)	.042	55 (1.2)	17 (2.6)	.006	7 (2.7)	0.08
Orofacial	43 (0.5)	10 (0.7)		4 (1.2)		27 (0.6)	6 (0.9)		3 (1.1)	
Respiratory	6 (0.1)	3 (0.2)		0 (0)		1 (0.02)	0 (0)		0 (0)	
Skin	19 (0.2)	4 (0.3)		0 (0)		12 (0.3)	4 (0.6)		0 (0)	
Syndrome	30 (0.4)	12 (0.8)	.026	1 (0.3)		17 (0.4)	7 (1.1)	.022	0 (0)	
Tumors	10 (0.1)	3 (0.2)		0 (0)		4 (0.1)	1 (0.2)		0 (0)	
Chromosomal	20 (0.2)	6 (0.4)		0 (0)		13 (0.3)	3 (0.5)		0 (0)	
Other	8 (0.1)	2 (0.1)		0 (0)		0 (0)	1 (0.2)		0 (0)	

Note: Data are presented as n (%). If a child had multiple defects in separate organ systems, the child appears more than once in the table. If a child had more than one unrelated defect affecting the same organ system, the child appears only once in the table. *P* values are comparisons with the control group. *P* values were not significant if not listed. Fisher's exact *t*-test was used. CNS = central nervous system.

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ICSI-conceived children born after IVF procedures showed no differences in birth defect rates for all children (OR 0.86, 95% CI 0.54–1.38 for ICSI) or singletons only (OR 1.06, 95% CI 0.53–2.08 for ICSI) after controlling for age, plurality, and parity.

Cryopreservation of embryos did not seem to have an effect on birth defect rates in singletons (OR 0.4, 95% CI 0.15–1.11 for cryopreserved embryos) as compared with singletons conceived after the transfer of "fresh" embryos with IVF. There was, however, a higher incidence of major birth defects in twins born after transfer of cryopreserved embryos as compared with twins conceived after the transfer of "fresh" embryos (OR 2.11, 95% CI 1.03–4.33, $P=.041$).

Multiple gestations were associated with a higher birth defect rate than singletons when control pregnancies were combined with pregnancies conceived after infertility treatment (OR 1.29, 95% CI 1.05–1.58, $P=.014$ for twins; OR 2.12, 95% CI 1.44–3.11, $P<.0001$ for triplets). A statistically significant increase in defect rate was seen when control twins were compared with control singletons (OR 1.38, 95% CI 1.10–1.73, $P=.005$ for twins), whereas no difference in birth defects was seen in twins conceived with IVF and IUI compared with singletons conceived by the same treatments. Triplets and other higher-order multiple gestations were associated with increased birth defects in both controls (OR 1.94, 95% CI 1.19–3.16, $P=.008$) and IVF-conceived children (OR 2.55, 95% CI 1.28–5.10, $P=.0008$). There were no major birth defects in high-order multiple gestation infants born after IUI.

When major defects were evaluated by affected organ system, there seemed to be a significantly greater proportion of cardiovascular and musculoskeletal defects and syndrome diagnoses among the infants conceived through IVF when compared with control children (Table 4). When the analysis was restricted to singletons, the same defects were found more often in children conceived through IVF. With children conceived after IUI, only the musculoskeletal system was affected more often than in control children. As compared with other children conceived with IVF, no particular organ system involvement was more prevalent in children conceived with ICSI or after transfer of cryopreserved embryos.

Specific defects that were more common in the IVF-conceived children included anotia/microtia, ventricular septal defect, atrial septal defect, tetralogy of Fallot, and upper limb defects. The upper limb defects included a variety of problems, including accessory digits, webbed, fused, or missing fingers, lobster claw hand, and phocomelia of the upper limb. Goldenhar syndrome (craniofacial microsomia) was statistically more common in IUI-conceived children than in control children; however, this is based on a single case. Multiple

defects (two or more major defects affecting different organ systems) were not more prevalent in either the IUI or the IVF group as compared with control children.

Male infants seemed to be at a higher risk for birth defects after IVF. When all children conceived by IVF were analyzed, male infants had an 8.03% rate of major birth defects, compared with a 4.26% rate in female infants (OR 1.96, 95% CI 1.23–3.12, $P=.004$). No similar increase in birth defects among male infants was noted after IUI (OR 1.42, 95% CI 0.52–3.85, $P=.491$) or in control children (OR 1.19, 95% CI 0.96–1.48, $P=.104$). As compared with IVF-conceived female infants, IVF-conceived male infants had a slightly higher rate of birth defects in many organ systems, but the only system that was statistically significant was the genitourinary system—chiefly owing to the diagnosis of hypospadias.

DISCUSSION

To our knowledge, this is the largest American study to evaluate birth defects after infertility treatment as compared with a matched cohort of naturally conceived children. We found a small but significantly increased risk of major birth defects in children conceived by IVF. It is well known that birth defects are more prevalent in children born from multiple gestation pregnancies and that multiples are frequently conceived after infertility treatments. However, this study showed that the prevalence of birth defects was higher even for singletons born after IVF.

Our study supports the findings of some, but not all, previous studies evaluating birth defects after IVF as compared with either a matched cohort or national registry rates after correction for important variables, including maternal age and plurality (6–11). All of these studies detected and reported birth defects in a standardized fashion, yet even these studies cannot be directly compared because birth defects were detected in children for varying lengths of time after birth, and different classification systems for birth defects were used. Our interpretation of published studies combined with our own findings is that IVF is associated with an increase in birth defects but that the effect is small. Findings to date are not likely to dissuade many couples from pursuing infertility treatments.

The cause of an increased rate of birth defects in children born after IVF is unknown. Some have speculated that ICSI might lead to an increased risk of birth defects because the fertilizing spermatozoon is artificially selected and injected into the oocyte and therefore does not have to be competent to bind to the oolemma or activate the oocyte. One study reported an increase in hypospadias with ICSI (19). However, several other studies, including ours, have failed to confirm an increased risk of hypospadias or any other birth defects in children born after ICSI as compared with children born after IVF in which fertilization occurred without ICSI (6, 20–22).

Intuitively, the process of freezing and thawing human embryos seems fraught with opportunities to damage the embryo, thus leading to birth defects. However, we found no increased risk of birth defects associated with cryopreservation of embryos. Although this aspect of IVF has been less extensively examined, several other studies have also found no increased risk of birth defects after transfer of cryopreserved embryos as compared with transfer of fresh embryos during an IVF cycle (23–26).

Recent attention has been directed toward epigenetic errors that might be inherent in the infertile couple or induced as a side effect of the infertility treatment itself. Differential methylation of cytosine leading to expression of only one of two parental alleles is a mechanism of gene regulation known as *genomic imprinting* (27). Defects in imprinting might cause either over- or under-expression of certain genes, leading to birth defects or cancer. Several syndromes caused by imprinting defects, including Beckwith-Wiedemann syndrome and Angelman Syndrome, have been reported to be more prevalent in children born after IVF (12–17). Some have proposed that embryo culture media used in IVF might predispose to imprinting defects in the embryo (28, 29). Recently, an association between reduced sperm concentrations and abnormal genomic imprinting in the spermatozoa has been reported (30). This suggests that imprinting defects and impaired gametogenesis might be linked in men, and this could be the mechanism of imprinting defects in children. A similar process could be involved in female infertility or might be induced with ovulation-inducing medications used for infertility treatment. These syndromes resulting from imprinting defects are exceedingly rare; nevertheless, we did not detect any malformations known to result from imprinting disorders in our study.

Another possible reason for the increase in birth defects with IVF is a genetic problem (other than an imprinting defect) inherent in one or both of the partners, leading to both reduced fertility and subsequent birth defects. Thus, the population of infertile couples might be at risk for having children with birth defects before they undergo any infertility treatment. We hoped to explore this possibility by including children conceived after IUI as well as those conceived by IVF. We suspect, but cannot prove, that our failure to show a statistically significant increase in birth defects in the IUI group was secondary to smaller numbers of children being available for study. If this is the case, and IUI is indeed associated with an increase in birth defects in a larger population of children, this would support the theory that a problem inherent in the infertile couple is responsible for the birth defect outcomes. On the other hand, a large majority of our IUI couples received medications for ovulation induction, so we cannot rule out effects of these medications on the birth defect outcomes. With our current IUI numbers, we have 80% power to detect, at the .05 significance level, an OR for birth defects of $\geq$1.95 and 34% power to detect an OR of $\geq$1.50 in the IUI group. With our current IVF and control children numbers, a sample size of 1,119 IUI children would be needed to detect an OR of $\geq$1.50 at the .05 significance level with 80% power.

Larger studies will be required to further explore this question.

Within the IVF group, we found no difference in birth defect rates when evaluating ICSI, embryo cryopreservation, or even length of embryo culture before transfer. This again supports the theory that the problem might be inherent in the infertile couples or secondary to ovulation induction.

Certain organ systems seem to be disproportionately affected among the treatment groups, with cardiovascular defects more common in the IVF population and musculoskeletal defects more common in both the IVF and IUI groups compared with controls. These results are similar to findings in other recent studies that have reported an increased risk of defects in these organ systems (6, 7, 11). We did not see the increase in urogenital, neural tube, gastrointestinal, genitourinary, or the classic defects associated with imprinting problems that have been reported in other studies (10). When evaluating specific defects within these organ systems, great care must be exercised, owing to the small numbers of infants affected and the multiple statistical comparisons made.

The increased risk of birth defects in male relative to female infants after IVF is difficult to explain. Among IVF-conceived infants, male infants had a higher rate of genitourinary defects, especially hypospadias. This finding might be due to an ascertainment bias because many female genitourinary defects might not be detected by the age of 1 year. This increased risk among male infants after IVF was not found in the other study that specifically examined this question (7).

Birth defect studies can be limited by detection and reporting biases. For example, parents treated with IVF might be more prone to using the health care system for their child and thus be more likely to have birth defects detected in their child. Self-reported birth defect rates have been shown to differ substantially from national registries of defects detected by physicians, indicating another potential bias in studies relying on patient reports of birth defects (11).

We were greatly aided in this study by the presence of a statewide registry that systematically records all birth defects of all children up to the age of 1 year. Although a national reporting system is in place for pregnancy and multiple gestation outcomes from IVF, this registry does not have accurate data regarding birth defects. The presence of our state registry (which is recorded with no knowledge about the methods of conception of a given child) helps to limit detection or reporting bias that might otherwise occur. We cannot completely rule out the possibility of a detection bias if infants conceived after infertility treatments are subjected to closer scrutiny either prenatally or in the year after birth. We think this is unlikely to be a factor because we were only studying major birth defects that cause functional impairment or require surgical correction and are therefore easily diagnosed. Even with this registry, we are limited in our ability to know about all congenital malformations that might have been detected prenatally, with pregnancy terminated as a result.

There are several potential weaknesses to our study. The University of Iowa is the largest of three centers in the state that provide IVF. We only included controls from counties that had women delivering babies from our IVF program, thus restricting controls to our referral area. However, we cannot rule out the possibility that some of the control children were actually conceived after infertility treatment at another practice. If this occurred, it would tend to minimize any difference in birth defect rate that we found. Although we did search the state infant death certificates, it is possible that terminations performed for congenital anomalies could have been missed, especially those performed at <20 weeks outside of the hospital setting. It is also possible that patients who conceived after infertility treatments might receive a higher rate of prenatal testing, leading to the detection of more anomalies. Whether this would result in a higher rate of pregnancy termination among this population is unclear. If it did, the difference in birth defect rates between infants conceived with and without infertility treatments would be minimized. If previously infertile couples are less likely to terminate, then this might explain some of the increased rate seen.

Strengths of this study include the relatively large number of infants studied, the uniform way that birth defects were detected and reported, and the inclusion of infants conceived by infertile patients treated by procedures other than IVF. With the growing numbers of patients undergoing infertility treatments, the systematic evaluation of obstetric and perinatal outcomes, as well as long-term follow-up of these children, has taken on even more importance. Additional multicentered analyses for birth defects after infertility treatment are warranted.

REFERENCES

1. Rizk B, Doyle P, Tan SL, Rainsbury P, Betts J, Brinsden P, et al. Perinatal outcome and congenital malformations in in-vitro fertilization babies from the Bourn-Hallam group. Hum Reprod 1991;6:1259–64.
2. French In Vitro National (FIVNAT) and Instituit National de la Sante et de la Recherché Medicale U292. Pregnancies and births resulting from in vitro fertilization: French national registry, analysis of data 1986 to 1990. Fertil Steril 1995;64:746–56.
3. Olivennes F, Blanchet V, Kerbrat V, Rufat P, Frydman R. Follow-up of a cohort of 422 children aged 6 to 13 years conceived by in vitro fertilization. Fertil Steril 1997;67:284–9.
4. Tanbo T, Dale PO, Lunde O, More N, Åbyholm T. Obstetric outcome in singleton pregnancies after assisted reproduction. Obstet Gynecol 1995;86:188–92.
5. Morin NC, Wirth FH, Johnson DH, Frank LM, Presburg HJ, Van de Water VL, et al. Congenital malformations and psychosocial development of children conceived by in vitro fertilization. J Pediatr 1989;115:222–7.
6. Hansen M, Kurinczuk JJ, Bower C, Webb S. The risk of major birth defects after intracytoplasmic sperm injection and in vitro fertilization. N Engl J Med 2002;346:725–30.
7. Koivurova S, Hartikainen A-L, Gissler M, Hemminki E, Sovio U, Järvelin M-R. Neonatal outcome and congenital malformations in children born after in-vitro fertilization. Hum Reprod 2002;17:1391–8.

8. Dhont M, DeSutter P, Ruyssinck G, Martens G, Bekaert A. Perinatal outcome of pregnancies after assisted reproduction: a case control study. Am J Obstet Gynecol 1999;181:688–95.
9. Westergaard HB, Tranberg Johansen AM, Erb K, Nyboe Anderson A. Danish national in vitro fertilization registry 1994 and 1995: a controlled study of births, malformations and cytogenic findings. Hum Reprod 1999;14:1896–902.
10. Ericson A, Källén B. Congenital malformations in infants born after IVF: A population-based study. Hum Reprod 2001;16:504–9.
11. Anthony S, Buitendijk SE, Dorrepaal CA, Lindner K, Braat DDM, den Ouden AL. Congenital malformations in 4224 children conceived after IVF. Hum Reprod 2002;17:2089–95.
12. DeBaun MR, Niemitz EL, Feinberg AP. Association of in vitro fertilization with Beckwith-Wiedemann Syndrome and epigenetic alterations of LIT1 and H19. Am J Hum Genet 2003;72:156–60.
13. Maher ER, Brueton LA, Bowdin SC, Luharia A, Cooper W, Cole TR, et al. Beckwith-Wiedemann syndrome and assisted reproduction technology. J Med Gen 2003;40:62–5.
14. Gicquel C, Gaston V, Mandelbaum J, Siffroi JP, Glahault A, LeBouc Y. In vitro fertilization may increase the risk of Beckwith-Wiedemann syndrome related to the abnormal imprinting of the KCN1OT gene. Am J Hum Genet 2003;72:1338–41.
15. Moll AC, Imhof SM, Cruysberg JR, Schouten-van Meeteren AY, Boers M, van Leeuwen FE. Incidence of retinoblastoma in children born after in-vitro fertilization. Lancet 2003;361:309–10.
16. Orstavik KH, Eiklid K, van der Hagen CB, Spetalen S, Kierulf K, Skjeldal O, et al. Another case of imprinting defect in a girl with Angelman Syndrome who was conceived by intracytoplasmic sperm injection. Am J Hum Genet 2003;72:218–9.
17. Cox GF, Buerger J, Lip V, Mau UA, Sperling K, Wu BL, et al. Intracytoplasmic sperm injection may increase the risk of imprinting defects. Am J Hum Genet 2002;71:162–4.
18. Rasmussen SA, Olney RS, Holmes LB, Lin AE, Keppler-Noreuil KM, Moore CA, et al. Guidelines for case classification for the National Birth Defects Prevention Study. Birth Defects Res A Clin Mol Teratol 2003;67:193–201.
19. Wennerholm U-B, Bergh C, Hamberger L, Lundin K, Nilsson L, Wikland M, et al. Incidence of congenital malformations in children born after ICSI. Hum Reprod 2000;15:944–8.
20. Bonduelle M, Liebaers I, Deketalaere V, Derde M-P, Camus M, Devroey P, et al. Neonatal data on a cohort of 2889 infants born after ICSI (1991–1999) and of 2995 infants born after IVF (1983–1999). Hum Reprod 2002;17:671–94.
21. Palermo GD, Colombero LT, Schattman GL, Davis OK, Rosenwaks Z. Evolution of pregnancies and initial follow-up of newborns delivered after intracytoplasmic sperm injection. JAMA 1996;276:1893–7.
22. Retzloff MG, Hornstein MD. Is intracytoplasmic sperm injection safe? Fertil Steril 2003;80:851–9.
23. Wennerholm U-B, Albertsson-Wikland K, Bergh C, Hamberger L, Niklasson A, Nilsson L, et al. Postnatal growth and health in children born after cryopreservation as embryos. Lancet 1998;351:1085–90.
24. Sutcliffe AG, Souza SD, Cadman J, Richards B, McKinlay IA, Lieberman B. Minor congenital anomalies, major congenital malformations and development in children conceived from cryopreserved embryos. Hum Reprod 1995;10:3332–7.
25. Aytoz A, Van den Abbeel E, Bonduelle M, Camus M, Joris H, Van Steirteghem A, et al. Obstetric outcome of pregnancies after the transfer of cryopreserved and fresh embryos obtained by conventional in-vitro fertilization and intracytoplasmic sperm injections. Hum Reprod 1999;14:2619–24.
26. Wada I, Macnamee MC, Wick K, Bradfield JM, Brinsden PR. Birth characteristics and perinatal outcome of babies conceived from cryopreserved embryos. Hum Reprod 1994;9:543–6.
27. Reik W, Dean W, Walter J. Epigenetic reprogramming in mammalian development. Science 2001;193:1089–93.
28. Edwards RG, Ludwig M. Are major defects in children conceived in vitro due to innate problems in patients or to induced genetic damage? Reprod Biomed Online 2003;7:131–8.
29. Niemitz EL, Feinberg AP. Epigenetics and assisted reproductive technology: a call for investigation. Am J Hum Genet 2004;74:599–609.
30. Margues CJ, Carvalho F, Sousa M, Barros A. Genomic imprinting in disruptive spermatogenesis. Lancet 2002;363:1700–2.