BMC Medicine

RESEARCH

Open Access

School-age outcomes among IVF and ICSI-conceived children: a causal inference analysis using linked population-wide data

Amber L. Kennedy[1,2,3,4*], Richard J. Hiscock[1,2], Beverley J. Vollenhoven[5,6,7], Catharyn J. Stern[1,2,8], Lyle C. Gurrin[9], Tiki Osianlis[4], Aleah Kink[1], Susan P. Walker[1,2], Jeanie L. Y. Cheong[1,10,11,12], Jon L. Quach[10,13], David Wilkinson[14], John McBain[1,3,8], Mark P. Green[7,15], Jessica A. Atkinson[1,2], Franca Agresta[8], Susan P. Baohm[7,10,14,16], Stephen Tong[1,2], Roxanne Hastie[1,2] and Anthea C. Lindquist[1,2*]

Abstract

Background Use of intracytoplasmic sperm injection (ICSI) continues to increase as the most common mode of oocyte insemination during in vitro fertilisation (IVF), sometimes in the absence of clear indications (i.e. male factor infertility). Several studies suggest an increased risk of congenital abnormalities after ICSI. The association between the ICSI technique and long-term childhood development remains unclear.

Methods Our population-based study included singleton infants conceived via IVF and born between 2005 and 2013. The cohort included state-wide linked maternal and childhood administrative data from Victoria, Australia. The primary exposure was conception via ICSI (without severe male factor infertility), with those born following standard IVF as controls. Childhood development was examined using the Australian Early Development Census (AEDC), a broad assessment of childhood development across five domains of health and neurodevelopment performed in Australian schools every triennium at school entry (age 4–6 years). Our primary outcome used a validated global measure—developmental vulnerability—defined as scoring less than the 10th percentile in two or more of the five developmental domains (DV2). Causal inference methods were used to analyse observational data in a way that emulates a target randomised clinical trial. The adjustment variable set was determined a priori via a modified Delphi procedure. Given the use of observational data, there were missing data and inherent differences in the covariate profile between exposure cohorts. Multiple imputation, bootstrapping and doubly robust inverse probability weighted regression adjustment modelling was utilised to allow a causal interpretation of results.

Results Our cohort ($N=3656$) included 1489 IVF and 2167 ICSI-conceived children. We found no causal effect of ICSI on the risk of AEDC-defined developmental vulnerability at school-entry age compared with children conceived via standard IVF; adjusted risk difference −1.11% (95% CI −4.23 to 2.01%) and adjusted risk ratio 0.90 (95% CI 0.68 to 1.21).

Conclusions Our findings suggest that the use of ICSI in IVF cycles without severe male factor infertility does not increase the risk of early childhood developmental vulnerability among children in their first year of school. These findings provide important reassurance for current and prospective parents and clinicians alike.

Keywords ICSI, IVF, Childhood development, Causal analysis

*Correspondence:
Amber L. Kennedy
amber.kennedy@unimelb.edu.au
Anthea C. Lindquist
anthea.lindquist@unimelb.edu.au
Full list of author information is available at the end of the article

Background

Intracytoplasmic sperm injection (ICSI) is an essential technique in the in vitro fertilisation (IVF) armamentarium. It involves the direct injection of a single spermatozoon into the cytoplasm of an oocyte [1]. This is in contrast to standard insemination where an egg and sperm are simply co-incubated to facilitate fertilisation—a cellular process that is comparable to fertilisation in vivo [2]. The introduction of ICSI to clinical practice in 1992 aimed to improve fertilisation rates in the setting of severe male factor infertility—and indeed it did [3]. The high fertilisation rate associated with ICSI has since led to its increasing use in cases of non-male factor infertility [4, 5].

By 2010, the frequency of use of ICSI in IVF cycles reached 100% in some regions globally [6]. Data from the USA have shown ICSI utilisation increased from 15.4% in 1996 to 66.9% in 2012 for non-male factor indications [7]. As of 2018, reports demonstrated that ICSI rates vary from 53 to 90% across different American states with the national average close to 80% [8]. In Australia, ICSI use peaked in 2016 where it was the fertilisation method of choice in 69% of cycles [4]. Despite this widespread use, there is still no proven benefit on the use of ICSI outside of severe male factor infertility and concerns have evolved about the safety of its widespread use [5, 9].

The actual technique itself—with arbitrary selection of a spermatozoon for injection and bypassing of the zona pellucida—has led to concerns about the transmission of abnormal genetic and epigenetic traits and subsequent implications for the offspring [10, 11]. Multiple studies investigating the risk of congenital malformations associated with ICSI have produced conflicting evidence [5, 12, 13]. A number of studies have also examined longer term developmental outcomes for children conceived via ICSI, with some providing reassurance about childhood outcomes [10, 14–19]. However, past studies have been limited by their sample size, use of historical cohorts, limited accounting for confounding factors and misleading comparator cohorts (using spontaneously conceived children where neither subfertility nor fertility treatments are involved).

Given the rising use of ICSI for non-male factor infertility and the ongoing concerns about its potential harms, we aimed to examine the long-term developmental outcomes for children conceived via ICSI for non-male factor infertility compared with those in which standard IVF was used. Using a state-wide birth cohort and a causal inference framework, our study was designed to emulate a real-world target trial to investigate the causal effect of ICSI for non-male factor infertility compared with standard IVF on the school-age developmental outcomes for children.

Methods

A causal inference framework was employed to answer the research question. This approach attempts to simulate a randomised trial by [1] requiring an a priori statistical analysis protocol that specifies both a hypothetical target randomised control trial and its proposed emulation using observational data; [2] addressing a causal question reflecting the effect of an intervention at a specific clinical decision point on a prespecified outcome; and [3] using inverse probability weighting via a propensity score (PS) model to balance covariate distributions between exposed and control populations, with the aim of producing exchangeable comparison groups and eliminating selection bias [20, 21]. By conducting the study from a causal perspective and adhering to the conditions of a hypothetical target trial, causal conclusions can be made about the association between fertilisation by ICSI and childhood developmental vulnerability, compared with standard IVF.

The first step of the target trial emulation was to describe the ideal hypothetical intervention trial required to answer the research question. Each component of this hypothetical trial was then assessed against the data available in our retrospective cohort (Additional file 1: Target trial emulation). This process helped determine how closely we could emulate the results of the ideal target trial using the observational data available for analysis, and thus minimise as many sources of bias as possible. Limitations to trial emulation were acknowledged and strategies for overcoming them were outlined in our a priori statistical analysis plan (SAP) (Additional file 2: Statistical analysis plan [3, 21–32]).

Study population

The source population included Victorian singleton live births between 2005 and 2013 conceived via assisted reproductive techniques (ART). The three largest ART units operating in Victoria at the time provided data on all cycles that resulted in a live birth during the study period. Linked maternal and child data pairs were obtained using Victorian Perinatal Data Collection data and birth records from the Victorian Births, Deaths and Marriage registry. Data linkage was performed by the Centre for Victorian Data Linkage (CVDL), a third-party government-funded data linkage unit [33]. Post-linkage false matches and duplicates were removed.

Delphi consensus—defining the exposure and covariates

A modified Delphi survey [34] was performed to ensure robustness of our exposure definition and analysis plan, including which covariates to include in our analysis model (Additional file 3: Delphi survey results). This process was necessary since practices vary globally, especially relating to what constitutes a moderate or an absolute indication for ICSI [35, 36]. Fertility specialists, embryology scientists and experts in perinatology, epidemiology and education were surveyed (22 of 30 respondents completed three survey rounds).

Our study sought to examine outcomes for children conceived via ICSI in the absence of any absolute indication for ICSI. Delphi consensus established the absolute indications for ICSI (standard IVF would not be possible), and thus which births to exclude from our analysis. These included surgically extracted sperm (e.g. percutaneous epididymal sperm aspiration, testicular sperm aspiration, microscopic testicular retrieval of sperm), severe male factor infertility (e.g. OAT oligoasthenoteratozoospermia), oocyte thaw cycles, pregestational testing for monogenetic disorders or chromosomal translocations and total motile sperm count less than 2 million on the day of egg collection. In keeping with causal methodology, these cases with Delphi consensus absolute indications for ICSI were excluded since they could not be feasibly be assigned to the non-treatment (standard IVF) group in a target trial.

Exposure was thus defined as conception via in vitro fertilisation (IVF) using intracytoplasmic sperm injection (ICSI) (non-absolute indications) compared with IVF conception using standard insemination. Method of oocyte insemination for the embryo transferred was clearly documented within the IVF database. Singleton pregnancies resulting from double embryo transfer where two different methods were used for insemination, and consequently fertilisation method of the implanted embryo was unable to be known were excluded.

Based on Delphi consensus, the covariates considered to be potential determinates of the exposure were year of oocyte insemination, maternal age (at birth of the child), socioeconomic status, parity, language background other than English (LBOTE), both maternal and paternal highest obtained level of education, as well as laboratory related indices such as number of eggs on day of egg collection, presence of mild-moderate male factor infertility and specific categories of female factor subfertility (e.g. tubal factor, ovulation disorder).

Determinants of the outcome included maternal age (at birth of the child), socioeconomic status, parity, language background other than English (LBOTE), both maternal and paternal highest obtained level of education, census year, child's age in years at assessment and sex of the child. Language background other than English is recorded by the AEDC to identify children from non-English speaking families, which may impact the assessment of language-based aspects of childhood development.

Gestational age at birth, mode of delivery and birthweight were considered mediators on the causal pathway and were therefore not adjusted for in this analysis. Our post-Delphi statistical analysis plan and directed acyclic graph were agreed upon and signed off by all authors prior to the commencement of data analyses (Additional file 2: Statistical analysis plan).

Outcome measure: the Australian Early Development Census

Childhood developmental outcome was assessed using a standardised, national assessment, The Australian Early Development Census (AEDC) [37–42] conducted every 3 years across Australia. All schools, including special schools, are eligible to participate. Children are assessed in their first year of primary school and all children in Victoria start school in February in the year they turn five (by end of March) or six. The AEDC is a teacher-reported measure that assesses broad childhood functional development at school entry (age 4–6) across five domains: physical health and wellbeing, social competence, emotional maturity, language and cognitive skills (school-based) and communication skills and general knowledge. Our primary outcome used a validated global measure—developmental vulnerability—defined as scoring less than the 10th percentile in two or more of the five developmental domains—"DV2" [43]. Secondary outcomes included developmental vulnerability (less than 10th percentile) in each of the five individual domains.

Within the AEDC, teachers are also able to record any diagnosis that may affect development, including vision or hearing impairment, autism spectrum, attention deficit hyperactive disorder or other physical and cognitive conditions. Children with these conditions are noted as "special needs" if a diagnosis has been made prior to school entry or "emerging needs" if a diagnosis if suspected or under assessment. The presence of a special or emerging need was also examined as an outcome in a secondary analysis.

Statistical analyses

Descriptive statistics are presented for each cohort by exposure status, according to type of data.

Missing data

We performed a detailed examination of the patterns of missing data and its frequency by exposure status. For the AEDC outcome data, the major mechanism for

missing outcome was considered "informative" or "missing not at random" (MNAR), where a child was flagged as having "special needs", the domain score of the metric is arbitrarily deemed invalid by the AEDC, and was not provided to us in the dataset, resulting in missing outcome data. Estimation bias related to missing outcomes due to "special needs" status was managed conservatively by deterministically imputing all missing outcomes as "developmentally vulnerable" in DV2 and the individual domains. Rarely, the reason for missing outcome data was considered "non-informative" that is the mechanism was considered "missing at random" (MAR) conditional on measured covariates. For example, where a teacher was unable to sufficiently complete response tool due to time constraints. If the frequency of this outcome MAR missingness was both of small magnitude and did not differ by exposure status so its exclusion is very unlikely to result in biased inference. Consequently, there were 13 children that were excluded from analysis.

We considered that covariate data were likely to be MAR and imputation was performed using fully conditional specification that included birth mother to provide standard errors adjusted for maternal clustering [31, 32, 44]. Provided the imputation model is correctly specified and data are missing at random, multiple imputation methods can provide least biased estimates even with high proportion of covariate missingness [45].

All imputation models included exposure; both overall and domain outcomes; potential analysis model covariates including interaction terms; and auxiliary variables (see Additional file 4: Missing data diagnostics—for details). Standard diagnostics were performed (Additional file 4). To obtain analysis model standard errors that account for the variability induced by imputation, we used the two-stage method detailed by von Hippel and Bartlett [35]. This involved creating 1000 bootstrapped samples from the original dataset; performing two imputations on each of these 1000 samples and then running the analysis model on each of these 2000 datasets, allowing us to obtain pooled point estimates and associated standard errors using one-way ANOVA.

Average treatment effect modelling

The target estimand for the primary and secondary outcomes is the difference in the potential outcome means between exposure groups for AEDC defined vulnerability at school entry. These estimands are presented as the ATE (average treatment effect) relative risk (RR) and risk difference (RD) point estimates and 95% confidence intervals (CIs), based upon the potential outcomes framework for causal inference [46]. Each estimator required three sequential stages (i) bootstrap and impute datasets (vide supra), (ii) run an augmented doubly robust inverse-probability-weighted regression adjustment (AIPW model [47, 48] on each of the 2000 datasets and (iii) combine estimates using one way ANOVA to compute the ATE RD and RR estimates. Initial modelling included a selection model with 16 terms including two interaction terms (child age at Feb 1st in test year with exposure and gender) and one quadratic term (child age at Feb 1st in test year) and a regression adjustment model with 12 terms.

Prespecified sensitivity modelling using Targeted Maximum Likelihood Estimation (TMLE) that accounts for both interaction and non-linear relationships between exposure and covariates was also undertaken for both the cohort excluding and the cohort including donor egg, donor sperm and IVF laboratory (see Additional file 5: Sensitivity analysis). Standard regression diagnostics were used to assess the adequacy of covariate balance (Additional file 6: Covariate balance). All statistical analyses were performed using Stata MP version 18.0, including the *t*-effects and multiple imputation suites of commands.

Secondary analysis

An exploratory secondary analysis was performed to determine the association between male subfertility and childhood developmental outcome. This was an analysis that did not attempt to generate an effect estimate with a causal interpretation, comparing cases of ICSI with male factor subfertility to cases of ICSI without male factor indication/subfertility; all cases of standard insemination were excluded.

Results

The total original cohort of ART conceived children included 16,054 singleton births in Victoria between 2005 and 2013. Among this cohort, 4700 were linked to AEDC outcome data. As the AEDC is conducted triennially, the biggest determinate of successful linkage was year of birth (Fig. 1 and Table 1). To remove the potential of confounding by indication and ensure consistency with the causal framework, we excluded cases with documented severe male factor infertility ($n=630$), donated gametes ($n=379$), egg thaw cycles ($n=11$) and missing exposure status ($n=9$). This left a final study cohort of 3656 children including 1489 IVF-conceived and 2167 ICSI-conceived children (Fig. 1).

The baseline characteristics of the IVF-conceived and ICSI-conceived populations were similar. The frequency of infant sex at birth differed between exposure groups, with 45% female after IVF conception and 51% female after ICSI. There were minimal differences across other demographic variables between the two groups with similar maternal age, education, socioeconomic status

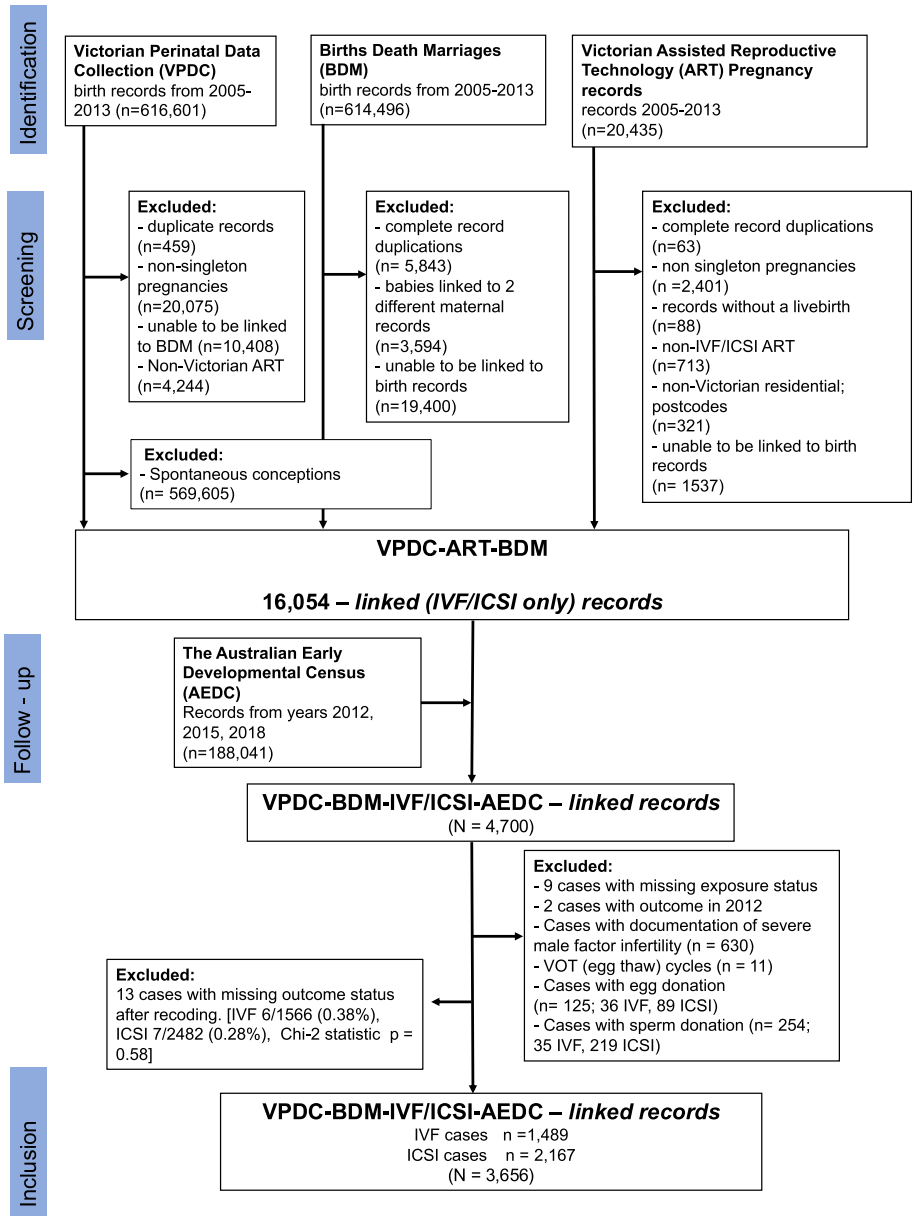

Fig. 1 Participant flow chart. Graphical representation of study population illustrating datasets used in data linkage

indicator and parity, as well as second parent level of education (Table 1).

The laboratory-based characteristics were also similar between the two exposure groups, excluding the frequency of documented male factor subfertility. The frequency of mild-moderate male factor subfertility was 4.6% in the IVF group compared with 40.7% in the ICSI group. Female factor subfertility was grouped into board categories; tubal factor subfertility was more common in the IVF than ICSI group (14.0% vs 6.0%) (Table 1).

Unadjusted rates of developmental vulnerability were similar between exposure cohorts for the primary outcome (DV2; IVF with standard—10.6%, versus ICSI—9.8%) and secondary outcomes (individual domains) (Table 2).

Causal analysis—primary outcome (DV2)

Our findings support the null hypothesis of no causal effect of mode of oocyte fertilisation on developmental vulnerability at school entry (less than 10th percentile in more than 2 of 5 domains of the AEDC), with 11.06%

Table 1 Descriptive statistics—baseline demographics of primary analysis population

	Whole cohort	IVF with standard oocyte insemination	Intracytoplasmic sperm injection
N	**3656**	**1489**	**2167**
Child baseline data			
Sex (% female)	48.7	44.9	51.3
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Year of birth (*n*)			
2005	23	6	17
2006	869	370	499
2007	188	83	105
2008	37	16	21
2009	925	408	517
2010	170	84	86
2011	27	12	15
2012	1173	431	742
2013	244	79	165
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Age of assessment (median) [IQR]	5.50 [5.19 to 5.73]	5.52 [5.20 to 5.74]	5.49 [5.18 to 5.72]
(number) % missing	(1) 0.03	(1) 0.07	(0) 0.0
Language background other than English (%)	13.8	12.8	14.4
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Aboriginal/Torres Strait Islander (%)	0.4	0.4	0.4
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Birthweight in grams (mean (SD))	3349 (± 571)	3344 (± 570)	3353 (± 573)
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
% SGA (< 10th centile for gestation)	0.8	0.7	0.8
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Gestational at delivery (median) [IQR]	39.1 [38.1 to 40.1]	39.0 [38.0 to 40.1]	39.1 [38.3 to 40.1]
(number) % missing	(7) 0.19	(5) 0.34	(2) 0.09
Method of delivery (% of each category)			
Normal vaginal birth	33.7	33.0	34.1
Instrumental vaginal birth	20.2	20.3	20.2
Caesarean section	46.0	46.5	45.7
(number) % missing	(2) 0.05	(1) 0.07	(1) 0.05
Maternal baseline data			
Maternal age (median) [IQR]	35.6 [32.8 to 38.4]	35.7 [33.0 to 38.4]	35.4 [32.7 to 38.4]
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
Maternal high school education level (% for each category)			
Year 9 or below	0.5	0.5	0.6
Year 10	2.7	2.3	2.9
Year 11	4.0	4.0	4.0
Year 12 and above	61.1	59.6	62.0
(number) % missing	(1160) 31.7	(500) 33.6	(660) 30.5
Maternal post-school education level (% for each category)			
No post-school education	7.6	6.7	8.3
Certificate (including trade)	10.2	9.7	10.6
Advanced diploma	9.9	8.6	10.8
Bachelor degree or above	39.3	40.0	38.7
(number) % missing	(1207) 33.0	(521) 35.0	(686) 31.7

Table 1 (continued)

	Whole cohort	IVF with standard oocyte insemination	Intracytoplasmic sperm injection
N	**3656**	**1489**	**2167**
Parity (% for each category)			
0	62.7	62.9	62.7
1	31.0	31.2	31.0
2	5.0	4.8	5.2
3 +	1.3	1.1	1.1
(number) % missing	(0) 0.0	(0) 0.0	(0) 0.0
SEIFA (Socio-Economic Index for Areas) quintile			
1 (most disadvantaged)	5.7	4.9	6.3
2	10.7	10.2	11.0
3	18.6	18.1	18.9
4	29.3	31.4	27.9
5 (least disadvantaged)	35.5	35.2	35.8
(number) % missing	(6) 0.16	(2) 0.13	(4) 0.18
Second parent high school education level (% for each category)			
Year 9 or below	1.2	0.9	1.3
Year 10	4.7	3.5	5.6
Year 11	5.9	6.2	5.7
Year 12 and above	52.2	51.3	52.8
(number) % missing	(1316) 36.0	(566) 38.0	(750) 34.6
Post-school education (% for each category)			
No post-school education	7.8	6.9	8.4
Certificate (including trade)	14.6	14.5	14.6
Advanced diploma	8.6	7.7	9.3
Bachelor degree or above	31.6	31.2	31.8
(number) % missing	(1369) 37.4	(591) 39.7	(778) 35.9
Laboratory based variables			
Year of fertilisation			
2004–2005	16.8	17.4	16.4
2006–2007	12.9	13.6	12.4
2007–2008	30.1	33.6	27.8
2009–2010	39.1	34.7	42.2
(number) % missing	(38) 1.04	(12) 0.81	(26) 1.20
Number of eggs at egg collection			
0–1	4.1	4.4	3.9
3–5	7.9	8.0	7.8
6–10	16.7	16.6	16.7
10–20	33.0	31.1	34.2
> 20	11.2	11.8	10.8
(number) % missing	(995) 27.2	(419) 28.1	(576) 26.6
Male factor			
Mild-moderate	26.0	4.6	40.7
Severe	Excluded		
Female factors[a]			
Tubal	9.2	14.0	6.0
Polycystic ovary syndrome	17.1	16.9	17.2
Endometriosis	6.8	7.8	6.1
Other[b]	6.8	10.6	7.3
Specified as "unexplained subfertility"	19.4	19.2	19.5

Table 1 (continued)

	Whole cohort	IVF with standard oocyte insemination	Intracytoplasmic sperm injection
N	**3656**	**1489**	**2167**
Poor prognosis female[c]	5.4	5.6	5.4
Specified as "no female factor"	17.4	13.8	19.8
(number) % missing	(656) 17.9	(249) 16.7	(407) 18.8

Severe male factor (see Additional file 7 for detailed demographics of this population), egg thaw cycles, donor egg and sperm cases excluded from this population baseline demographics

[a] As recorded in IVF database, primary diagnosis only

[b] Smaller categories combined, e.g. genetic disorders, fibroids, single sex, non-PCOS ovulation defects

[c] Advanced age, low ovarian reserve, previous failed IVF

of ICSI-conceived children predicted to be developmentally vulnerable in two or more AEDC domains compared with 9.95% of IVF-conceived children. The ATE RD was − 1.11% (95% CI: − 4.23 to 2.01%), and the ATE RR was 0.90 (95% CI: 0.68 to 1.21) consistent with no causal effect at the population level of method of oocyte insemination for children who were conceived by ICSI compared with children born after standard IVF (Table 2).

Causal analysis—secondary outcomes (individual domains and special needs status)

We examined each of the five AEDC domains individually as secondary outcomes, in addition to special and emerging needs status. The unadjusted and causal model results for each individual domain are reported in Table 2. There were no differences between ICSI and IVF-conceived children in adjusted risk difference for any of the individual AEDC domains nor special and emerging needs status.

Missing data

There were two cases with missing exposure status and seven cases of double embryo transfer with different fertilisation methods where we were unable to tell which embryo implanted. These nine cases (0.25% of the cohort) were excluded. Outcome data were missing for 256 (5.5%) of the AEDC-linked cohort. The majority (95.0%) of these missing cases were children with special needs ($n = 243$, 5.2% of overall cohort) and these were coded as developmentally vulnerable. The 13 remaining cases with missing outcome data were considered as MAR as there was no evidence of association with exposure status (χ^2-test $p = 0.58$) and they were removed from analysis.

Most covariates had minimal or no missing data (< 1.0%). Maternal education level was missing for 31.7% and maternal post-school education was missing for 33.0%. The number of eggs collected at ovum pick up (OPU) was missing in 27.2% of cases. Missing data for documented female factor fertility was 17.9%. Exposure and outcome distributions for these cases are presented in Table 1 and Additional file 4. The imputed values for missing covariate data, in particular the four covariates with greater than 15% missingness (number of oocytes, type of female factor and maternal levels of school and post-school education), produced plausible values and data distributions were consistent with a MAR assumption (Additional file 4).

Model construction and sensitivity analyses

We found that an AIPW model that included both donor egg, sperm and IVF laboratory were computationally unstable due to perfect prediction errors; consequently the primary AIPW estimator models were run on a cohort without both (Table 2) and then containing one or the other (Additional file 5: Sensitivity analysis). Further, the addition of the two interaction terms and/or the non-linear term to each of these models also resulted in perfect prediction errors. The final primary DV2 outcome model was linear with ten covariates in the selection model and nine in the outcome model (see Table 2 legend). The influence of interactions and non-linear terms on the estimates was assessed using alternate TMLE estimators, including a cohort containing donor egg, donor sperm and IVF laboratory. The results did not meaningfully differ from the findings of the primary analysis (Additional file 5: Sensitivity analysis).

Secondary analysis—ICSI with male factor infertility versus ICSI without male factor

An exploratory analysis was performed to determine the association between ICSI indication and childhood outcome and assess the marginal effect of [1] ICSI with documented male factor compared with ICSI performed without documented male factor and [2] ICSI in the setting of severe male factor compared with ICSI with mild or mild-moderate male factor. The baseline population characteristics between exposure groups were similar

Table 2 Unadjusted results and adjusted casual analysis results—IVF with standard insemination versus IVF with ICSI

	Unadjusted analysis				Adjusted causal analysis			
	Observed proportions (%)		Unadjusted effect estimates		Estimated proportions (%)		Treatment effect estimates	
	IVF with standard	IVF with ICSI	Relative risk (95% CI)	Risk difference (%) (95% CI)	IVF with standard	IVF with ICSI	ATE relative risk (95% CI)	ATE risk difference (%) (95% CI)
Primary outcome								
Developmental vulnerability (DV2) (< 10th in ≥ 2 domains)	10.6	9.8	0.93 (0.75 to 1.11)	− 0.79 (− 2.90 to 1.32)	10.79	10.12	0.90 (0.68 to 1.21)	− 1.11 (− 4.23 to 2.01)
Secondary outcomes								
Individual domains								
Physical health and wellbeing	10.9	9.9	0.91 (0.74 to 1.09)	− 0.92 (− 3.04 to 1.20)	12.11	10.65	0.81 (0.61 to 1.09)	− 2.37 (− 5.75 to 1.02)
Social competence	10.9	9.9	0.91 (0.74 to 1.09)	− 0.90 (− 3.06 to 1.26)	11.05	10.21	0.91 (0.68 to 1.22)	− 1.02 (− 4.20 to 2.17)
Emotional maturity	11.3	10.1	0.89 (0.72 to 1.06)	− 1.26 (− 3.30 to 0.77)	11.37	10.61	0.90 (0.70 to 1.56)	− 1.21 (− 4.04 to 1.64)
Language and cognitive skills (school-based)	8.6	6.9	0.81 (0.62 to 0.99)	− 1.57 (− 3.45 to 0.31)	8.70	6.90	0.79 (0.58 to 1.08)	− 1.82 (− 4.35 to 0.70)
Communication skills and general knowledge	8.7	7.5	0.86 (0.67 to 1.05)	− 1.17 (− 3.01 to 0.68)	8.81	7.53	0.86 (0.64 to 1.18)	− 1.19 (− 3.76 to 1.38)
Special and emerging needs	17.7	15.0	0.85 0.72 to 0.97	− 2.17 (− 5.92 to − 0.34)	18.0	15.5	0.86 (0.71 to 1.05)	− 2.48 (− 5.75 to 0.79)

IVF in vitro fertilisation, *IVF with standard* IVF with standard oocyte insemination, *ICSI* intracytoplasmic sperm injection, *95% CI* 95% confidence interval

AIPW adjustment model (linear in covariate terms and no interaction terms):

Treatment assignment—year of oocyte insemination, maternal age (at birth of the child), socioeconomic status, parity, language background other than English (LBOTE), maternal school education and maternal highest obtained level of education, number eggs on day of egg collection, presence of mild-moderate male factor infertility and categories of female factor subfertility

Outcome adjustment model—maternal age (at birth of the child), socioeconomic status, parity, language background other than English (LBOTE), maternal school education and maternal highest obtained level of education, AEDC census year, child's age in years at assessment and sex of the child

Table 3 Unadjusted results and adjusted analysis—ICSI with no male factor versus ICSI with male factor

	Unadjusted analysis				Adjusted analysis			
	Observed proportions (%)		Unadjusted effect estimates		Estimated proportions (%)		Marginal effect estimates	
	ICSI with no male factor	ICSI with male factor	Relative risk (95% CI)	Risk difference (%) (95% CI)	ICSI with no male factor	ICSI with male factor	Adjusted relative risk (95% CI)	Adjusted risk difference (%) (95% CI)
Primary outcome								
Developmental vulnerability (DV2) (< 10th in ≥ 2 domains)	9.6	10.0	1.05 (0.81 to 1.28)	0.4 (−1.8 to 2.7)	9.8	9.7	0.99 (0.78 to 1.26)	−0.1 (−2.4 to 2.3)
Secondary outcomes								
Individual domains								
Physical health and wellbeing	9.6	10.1	1.05 (0.81 to 1.28)	0.4 (−1.8 to 2.7)	9.6	10.2	1.07 (0.84 to 1.35)	0.6 (−1.7 to 3.0)
Social competence	9.7	9.7	1.00 (0.77 to 1.23)	0.0 (−2.2 to 2.2)	9.9	9.5	0.96 (0.75 to 1.21)	−0.4 (−2.7 to 1.9)
Emotional maturity	9.9	9.2	0.93 (0.71 to 1.14)	−0.7 (−2.9 to 1.5)	9.9	9.1	0.93 (0.74 to 1.17)	−0.7 (−2.9 to 1.5)
Language and cognitive skills (school-based)	6.1	7.8	1.28 (0.92 to 1.64)	1.7 (−0.2 to 3.6)	6.3	7.7	1.21 (0.91 to 1.62)	1.3 (−0.7 to 3.4)
Communication skills and general knowledge	6.9	8.0	1.15 (0.84 to 1.46)	1.1 (−0.9 to 3.0)	10.1	9.0	0.89 (0.70 to 1.13)	−1.1 (−3.4 to 1.1)
Special and emerging needs	15.8	14.9	0.94 (0.78 to 1.11)	−0.9 (−3.6 to 1.8)	16.0	14.8	0.93 (0.77 to 1.12)	−1.2 (−4.1 to 1.7)

ICSI intracytoplasmic sperm injection, *95% CI* 95% confidence interval

Adjustment model: outcome regression adjustment model—maternal age (at birth of the child), socioeconomic status, parity, language background other than English (LBOTE), both maternal and paternal highest obtained level of education, child's age in years at assessment and sex of the child

(Additional file 7: Male factor cohort). Both unadjusted (RR 1.05, 95% CI: 0.81 to 1.28) and adjusted (aRR 0.99, 95% CI: 0.78 to 1.26) results found little difference between the comparative groups in outcomes of DV2, the five individual AEDC domains or special and emerging needs (Table 3).

Discussion

Using population level data and statistical methods with a formal framework for causal inference, our findings suggest that ICSI for non-absolute indications does not confer an altered risk of childhood developmental vulnerability in two or more domains of the AEDC, compared with IVF with standard insemination. While there is growing evidence of a modest increase in congenital abnormalities associated with ICSI (e.g. hypospadias [13]), it appears that the mechanism for this difference does not extend to broader neurodevelopmental concerns.

Given the limitations of previous research and the rising use of ICSI globally, our study has generated much needed evidence. The strengths of our study lie in the use of state-wide birth and IVF data, deployment of record linkage between birth and longer-term childhood data and our rigorous application of formal methods for causal inference.

Many previous studies in this field were published over two decades ago, when ICSI was used almost exclusively for severe male factor subfertility or previous failed fertilisation with standard IVF, and the practice and technique of ICSI had not yet been refined to its current standard [15, 17, 18]. Additionally, past prospective observational cohort studies looking at detailed health and neurodevelopmental assessments were likely too underpowered (e.g. $n=76$ to 201 ICSI-conceived cases) to detect small yet clinically relevant effects [15, 16, 30, 49]; have focused on early perinatal outcomes such as congenital abnormalities; have relied upon simplistic, coarse or low-prevalence binary outcome measures such as cerebral palsy or autism diagnosis [13, 29, 50–53]; or have, where differences were detected, suggested that these were largely explained by parental factor like maternal age, parity,

socioeconomic status, as well as parental education and parents' cognitive ability [54–56].

Uniquely, our study was able to delineate between indications for ICSI—we were able to assess outcomes for ICSI cases were no severe male factor was evident. Previous studies have clustered all children conceived by ICSI together, without separating ICSI for severe male factor infertility from ICSI for non-essential indications. This is a crucial difference because ICSI is widely considered essential for fertilisation in cases of severe male factor infertility but for other more elective indications, and thus modifiable as an exposure. The ability to exclude cases of severe male factor infertility is an integral element of our target trial emulation. These cases could never be randomised to standard IVF in an intervention trial and thus should not contribute data in a randomised trial context.

By designing our study cohort in the context of a potential clinical trial, our study has arguably provided a stronger conclusion that an association analysis of observational data would not. We argue that existing evidence in this field is currently limited by the frequent use of the spontaneously conceived children as the comparison group. A 2008 large systematic review by Middelburg et al. evaluated the impact of IVF and ICSI conception on infant neurodevelopmental outcomes [14]. Reassuringly, they found no increased risk of adverse development, in children conceived by standard IVF or ICSI compared with children conceived without assistance. We were able to further echo this reassurance with a recent analysis making the same comparison [21]. The spontaneously conceived children and their parents are a vastly different population to that of the IVF-conceived. Correct comparison of vastly different exposed and unexposed population groups (even in studies where data for a rich set of covariates is available for adjustment [54]) requires complex methodology to appropriately account for these differences, rather than traditional regression methods [21, 57]. However, comparison with spontaneous conception represents a different research question and fails to tease out the specific impact of ICSI over and above that of IVF as a whole and subfertility as a condition.

To achieve this comparison, we have implemented a target trial emulation, which by explicitly defining inclusion criteria reduces observational cohort size, but if well-emulated leads to a closer estimation of causal effect.

In assessing baseline characteristics, our study found notable differences in the sex proportions of babies born after IVF with standard insemination versus ICSI. This phenomenon, with a higher incidence of female babies conceived via ICSI, has been described previously in the literature [58, 59]. As sex is a known determinant of performance in developmental assessments, sex at birth was included as a covariate to ensure this difference between exposure groups was accounted for.

There are data-based limitations inherent in all studies. In our study, as in others, documentation of male factor infertility was highly variable within and between the data from different IVF laboratories. Severe male factor infertility was documented as [1] surgical sperm extraction, [2] free text data which included entries such as "oligoasthenoteratozoospermia" or "OAT", "CBAVD" and "azoospermia" and [3] clear abnormalities on the laboratory analysis parameters on day of egg collection. The conclusions drawn from this study depend on the reliability and accuracy of data on which they are based. We recognise that even the most sophisticated analysis techniques cannot compensate for inaccuracies or inconsistencies in the data itself.

Due to the nature of record-linkage studies and the AEDC as a metric, our study may be limited by selection bias. A small fraction of children who did not attend mainstream school due to severe disability or home schooling were not captured. In Australia, the majority of children with a disability attend mainstream school, with only approximately 1% of the population attending specialist schools [60] and 0.5% home-schooled [61]. All Victorian schools including specialist schools are invited to participate in the AEDC but can elect not to. However, our study was not designed to assess severe disability or developmental delay, but rather an overall measure of global development and school readiness—within our study cohort of 2468 ICSI cases, no increased risk of developmental vulnerability was identified.

Finally, as with all observational studies it is possible that unmeasured confounders may have led to bias in estimating the treatment effects. To the best of our knowledge, the important covariates have been identified, measured and accounted for.

Conclusions

Within a causal framework, the developmental outcomes at school entry for children conceived by ICSI, without severe male factor infertility, are equivalent to those conceived via standard IVF. Furthermore, conception via ICSI in the setting of male factor subfertility is not associated with an increased risk of childhood developmental vulnerability compared with conception via ICSI in the absence of male factor subfertility.

Our findings suggest that empirical use of ICSI does not affect early childhood developmental outcomes. These findings provide further important reassurance for current and prospective parents and clinicians alike.

Abbreviations

ART	Assisted reproductive technology, umbrella term for fertility treatments that involved the handling of human gametes
IVF	In vitro fertilisation—the process of surgically collecting eggs, fertilising eggs in a laboratory, culturing embryos and performing an embryo transfer with the aim of achieving pregnancy
Standard IVF	IVF with standard oocyte insemination, i.e. the coincubation of cumulus-oocyte complexes with a prepared droplet of ~ 100,000 spermatozoa in a well to facilitate fertilisation
ICSI	Intracytoplasmic sperm injection, the alternative method of oocyte fertilisation and the exposure of interest of this study
AEDC	Australian Early Development Census
CVDL	Centre for Victorian Data Linkage
VPDC	Victorian Perinatal Data Collection
LBOTE	Language background other than English
ATE	Average treatment effect
RD	Risk difference
RR	Risk ratio
OPU	Ovum pick up
OAT	Oligoasthenoteratozoospermia, low number of sperm, poor sperm motility and abnormal sperm morphology
DV2	Developmental vulnerability in two or more domains of the AEDC—primary outcome of this study
IPWRA	Inverse-probability-weighted regression adjustment, a doubly robust analysis model
MAR	Missing at random
MNAR	Missing not at random
95% CI	95% Confidence interval

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12916-025-03963-w.

Additional file 1: Target trial emulation.

Additional file 2: Statistical analysis plan.

Additional file 3: Delphi…Results_04Sep2023 (short title: Delphi survey results).

Additional file 4: Missing data diagnostics.

Additional file 5: Sensitivity analyses.

Additional file 6: Covariate balance after IPW (short title: covariate balance).

Additional file 7: Male factor cohort.

Acknowledgements

This paper uses data from the Australian Early Development Census (AEDC). The AEDC is funded by the Australian Government Department of Education. The findings and views reported are those of the author(s) and should not be attributed to the Department or the Australian Government. We are grateful for the provision of data by the AEDC.

We are grateful to CCOPMM for providing access to the data used for this project and for the assistance of the staff at Safer Care Victoria. The conclusions, findings, opinions and views or recommendations expressed in this paper are strictly those of the author(s). They do not necessarily reflect those of CCOPMM.

We are thankful for contribution of Victorian in vitro fertilization (IVF) providers, Melbourne IVF, Monash IVF and City Fertility Centre to this research. We acknowledge the significant amount of work undertaken on behalf of this project and appreciate the opportunity to work with staff from each unit.

We also acknowledge the contributions of local Victorian IVF specialists, embryologists and research scientists who generously gave up their time to contribute to the Delphi survey enabling us to conduct this analysis having the strength of an accurate DAG. Those who contributed to the Delphi survey outside of the core authorship team include Dr Sameer Jatkar (Deputy Medical Director – Newlife IVF, Melbourne), Dr Genia Rozen (Fertility Specialist, Genea, Melbourne), Professor Alex Polyakov (Medical Director – Genea, Melbourne), Michael Chan (Scientific Director—The Women's Hospital, Melbourne), Dr Lauren Hicks (Fertility Specialist—Newlife IVF, Melbourne), A/Professor Martin Healey (Fertility Specialist—Newlife IVF, Melbourne), Fatima Figueiredo (Embryologist and Research Assistant – The Women's Hospital Melbourne), Professor Deirdre Zander-Fox (Chief Scientific Officer – Monash IVF, Melbourne), Melinda Dear (Laboratory Supervisor – Monash IVF, Melbourne), Dr Wan Tinn Teh (Fertility specialist, Co-Director Public Fertility Services – Melbourne IVF/The Women's Hospital) and Dr Natasha Pritchard (Clinician researcher and obstetrics and gynaecology registrar – The University of Melbourne/Mercy Hospital for Women, Melbourne).

Authors' contributions

AK1 had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. All authors qualify for authorship by contributing substantially to this article. AK1, AL, SW, ST, BV, KS, JM and DW conceived the study. AK1 completed the ethic applications, coordinated with the data custodians and linkage agency, performed the literature review, the data cleaning, analysis, wrote the manuscript and is the guarantor for this publication. AK2 assisted AK1 with extensive data cleaning. AK1 is the corresponding author. AL, RH1 and ST acted as co-supervisors. RH2 assisted in planning and performing the statistical analysis and contributed substantially to the STATA coding required for the analysis and performed sensitivity analysis. LG provided advice and guidance for causal inference theory and analysis. JQ and JC provided advice with education and paediatric expertise required to inform the research question. AL, RH1, LG and ST assisted with preparation and writing of the manuscript. FA, SB, MG and TO facilitated the data extraction from respective IVF units, performed some of the data cleaning and assisted with ethics applications. JA developed and administered the online survey platform for the Delphi consenus. BV, KS, JM, DW and MG provided expert guidance in the field of IVF and supported engagement in the study within each respective IVF unit. TO provided lab-based subject matter expertise and guidance. RH1 and SW provided intellectual support and guidance. All authors have contributed to critical discussion, agreed upon the prespecified statistical analysis plan and read and approved the final manuscript for publication.

Authors' Twitter handles

https://x.com/PerinatalEpi/status/1768393853789454411?s=20.

Funding

Appreciation is given to the Australian Federal Government for funding of the graduate research scheme and to Mercy Perinatal who both provided a student stipend, followed by ongoing salary which supported the work undertaken in this study. Additionally, we thank Ferring Pharmaceutics who provided a small unconditional research grant in support of this study.

Data availability

The data that support the findings of this study are available from the corresponding author but restrictions apply to the availability of these data, which were used under license for the current study, and so are not publicly available. Data are however available from the authors upon reasonable request and with permission of the individual data custodians; Monash IVF, Melbourne IVF, City Fertility Centre, The Consultative Counsel of Perinatal Mortality and Morbidity and the Australian Early Development Census.

Declarations

Ethics approval and consent to participate

Each data custodian provided contractual approval for data access and linkage. Given the retrospective and deidentified nature of this study, individual participant informed consent was not required. Ethical approval for the project was obtained from Mercy, Monash Health and Melbourne IVF Health Human Research Ethics Committees.

Consent for publication

Not applicable.

Competing interests

All authors have completed the ICMJE uniform disclosure form (http://www.icmje.org/disclosure-of-interest/). Most authors have declared that no

competing interests exist. A.L.K has received various education sponsorship and honoraria from Merck PTY LTD and Organon Pharma PTY LTD unrelated to this project. B.V has a paid role as a member of the Therapeutic Good Administration. T.O, B.V, F.A and K.S own shares in respective IVF companies (Newlife IVF, Monash IVF, Virtus Health and Melbourne IVF). The funders had no role in the study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Author details
[1]Department of Obstetrics, Gynaecology and Newborn Health, University of Melbourne, Parkville, Melbourne 3052, Australia. [2]Mercy Perinatal, Mercy Hospital for Women, Heidelberg, VIC, Australia. [3]Reproductive Services Unit, The Royal Women's Hospital, Parkville, VIC, Australia. [4]Newlife IVF, Box Hill, VIC, Australia. [5]Department of Obstetrics and Gynaecology, Monash University, Clayton, VIC, Australia. [6]Women's and Newborn Program, Monash Health, Clayton, VIC, Australia. [7]Monash IVF, Cremorne, VIC, Australia. [8]Melbourne IVF, East Melbourne, VIC, Australia. [9]Melbourne School of Population and Global Health, University of Melbourne, Melbourne, Australia. [10]Murdoch Children's Research Institute, Parkville, VIC, Australia. [11]Newborn Research, The Royal Women's Hospital, Parkville, VIC, Australia. [12]Department of Paediatrics, University of Melbourne, Melbourne, VIC, Australia. [13]Faculty of Education, University of Melbourne, Carlton, VIC, Australia. [14]City Fertility Centre, Melbourne, VIC, Australia. [15]School of BioSciences, University of Melbourne, Parkville, VIC, Australia. [16]Centre for Health Analytics, Melbourne Children's Campus, Melbourne, Australia.

Received: 5 July 2024 Accepted: 20 February 2025
Published online: 01 April 2025

References

1. Palermo GD, Cohen J, Alikani M, Adler A, Rosenwaks Z. Development and implementation of intracytoplasmic sperm injection (ICSI). Reprod Fertil Dev. 1995;7(2):211–7 discussion 7-8.
2. Choe J, Archer JS, Shanks AL. In vitro fertilization. Treasure Island (FL: StatPearls Publishing; 2022 [updated 2022 May 1. Available from: https://www.ncbi.nlm.nih.gov/books/NBK562266/.
3. Palermo G, Joris H, Devroey P, Van Steirteghem AC. Pregnancies after intracytoplasmic injection of single spermatozoon into an oocyte. Lancet. 1992;340(8810):17–8.
4. The National Perinatal Epidemiology and Statistics Unit (NESPU). The Australia and New Zealand Assisted Reproduction Database (ANZARD) [Available from: https://npesu.unsw.edu.au/data-collection/australian-new-zealand-assisted-reproduction-database-anzard.
5. Wang Y, Li R, Yang R, Zheng D, Zeng L, Lian Y, et al. Intracytoplasmic sperm injection versus conventional in-vitro fertilisation for couples with infertility with non-severe male factor: a multicentre, open-label, randomised controlled trial. Lancet. 2024;403(10430):924–34.
6. Dyer S, Chambers GM, de Mouzon J, Nygren KG, Zegers-Hochschild F, Mansour R, et al. International Committee for Monitoring Assisted Reproductive Technologies world report: assisted reproductive technology 2008, 2009 and 2010. Hum Reprod. 2016;31(7):1588–609.
7. Boulet SL, Mehta A, Kissin DM, Warner L, Kawwass JF, Jamieson DJ. Trends in use of and reproductive outcomes associated with intracytoplasmic sperm injection. JAMA. 2015;313(3):255.
8. Zagadailov P, Cho KS, Seifer DB. Differences in ICSI utilization rates among states with insurance mandates for ART coverage. Reprod Biol Endocrinol. 2021;19(1):174.
9. Quaas AM. ICSI for non-male factor: do we practice what we preach? J Assist Reprod Genet. 2021;38(1):125–7.
10. Palermo GD, Neri QV, Takeuchi T, Squires J, Moy F, Rosenwaks Z. Genetic and epigenetic characteristics of ICSI children. Reprod Biomed Online. 2008;17(6):820–33.
11. Ludwig M, Katalinic A. Malformation rate in fetuses and children conceived after ICSI: results of a prospective cohort study. Reprod Biomed Online. 2002;5(2):171–8.
12. Marino JL, Moore VM, Willson KJ, Rumbold A, Whitrow MJ, Giles LC, et al. Perinatal outcomes by mode of assisted conception and sub-fertility in an Australian data linkage cohort. PLoS ONE. 2014;9(1):e80398.
13. Venetis C, Choi SKY, Jorm L, Zhang X, Ledger W, Lui K, et al. Risk for congenital anomalies in children conceived with medically assisted fertility treatment: a population-based cohort study. Ann Intern Med. 2023;176(10):1308–20.
14. Middelburg KJ, Heineman MJ, Bos AF, Hadders-Algra M. Neuromotor, cognitive, language and behavioural outcome in children born following IVF or ICSI-a systematic review. Hum Reprod Update. 2008;14(3):219–31.
15. Bowen JR, Gibson FL, Leslie GI, Saunders DM. Medical and developmental outcome at 1 year for children conceived by intracytoplasmic sperm injection. Lancet. 1998;351(9115):1529–34.
16. Bonduelle M, Joris H, Hofmans K, Liebaers I, Van Steirteghem A. Mental development of 201 ICSI children at 2 years of age. Lancet. 1998;351(9115):1553.
17. Bonduelle M, Wilikens A, Buysse A, Van Assche E, Devroey P, Van Steirteghem AC, et al. A follow-up study of children born after intracytoplasmic sperm injection (ICSI) with epididymal and testicular spermatozoa and after replacement of cryopreserved embryos obtained after ICSI. Hum Reprod. 1998;13(Suppl 1):196–207.
18. Bonduelle M, Bergh C, Niklasson A, Palermo GD, Wennerholm UB, Collaborative Study Group of Brussels G, et al. Medical follow-up study of 5-year-old ICSI children. Reprod Biomed Online. 2004;9(1):91–101.
19. Ponjaert-Kristoffersen I, Bonduelle M, Barnes J, Nekkebroeck J, Loft A, Wennerholm UB, et al. International collaborative study of intracytoplasmic sperm injection-conceived, in vitro fertilization-conceived, and naturally conceived 5-year-old child outcomes: cognitive and motor assessments. Pediatrics. 2005;115(3):e283–9.
20. Hernan MA. Methods of public health research - strengthening causal inference from observational data. N Engl J Med. 2021;385(15):1345–8.
21. Kennedy AL, Vollenhoven BJ, Hiscock RJ, Stern CJ, Walker SP, Cheong JLY, et al. School-age outcomes among IVF-conceived children: a population-wide cohort study. PLoS Med. 2023;20(1):e1004148.
22. Lie RT, Lyngstadaas A, Orstavik KH, Bakketeig LS, Jacobsen G, Tanbo T. Birth defects in children conceived by ICSI compared with children conceived by other IVF-methods; a meta-analysis. Int J Epidemiol. 2005;34(3):696–701.
23. Bonduelle M, Wilikens A, Buysse A, Van Assche E, Wisanto A, Devroey P, et al. Prospective follow-up study of 877 children born after intracytoplasmic sperm injection (ICSI), with ejaculated epididymal and testicular spermatozoa and after replacement of cryopreserved embryos obtained after ICSI. Hum Reprod. 1996;11 Suppl 4:131–55 discussion 56-9.
24. Sutcliffe AG, Taylor B, Saunders K, Thornton S, Lieberman BA, Grudzinskas JG. Outcome in the second year of life after in-vitro fertilisation by intracytoplasmic sperm injection: a UK case-control study. Lancet. 2001;357(9274):2080–4.
25. Norrman E, Petzold M, Bergh C, Wennerholm UB. School performance in children born after ICSI. Hum Reprod. 2020;35(2):340–54.
26. Rosenbaum PR, Rubin DB. The central role of the propensity score in observational studies for causal effects. Biometrika. 1983;70(1):41–55.
27. Hernan MA, Robins JM. Using big data to emulate a target trial when a randomized trial is not available. Am J Epidemiol. 2016;183(8):758–64.
28. Austin PC. Variance estimation when using inverse probability of treatment weighting (IPTW) with survival analysis. Stat Med. 2016;35(30):5642–55.
29. Davies MJ, Moore VM, Willson KJ, Van Essen P, Priest K, Scott H, et al. Reproductive technologies and the risk of birth defects. N Engl J Med. 2012;366(19):1803–13.
30. Agarwal P, Loh SK, Lim SB, Sriram B, Daniel ML, Yeo SH, et al. Two-year neurodevelopmental outcome in children conceived by intracytoplasmic sperm injection: prospective cohort study. BJOG. 2005;112(10):1376–83.
31. von Hippel PT, Bartlett JW. Maximum likelihood multiple imputation: faster imputations and consistent standard errors without posterior draws. Stat Sci. 2021;36(3):400–20, 21.
32. Bartlett JW, Hughes RA. Bootstrap inference for multiple imputation under uncongeniality and misspecification. Stat Methods Med Res. 2020;29(12):3533–46.
33. A guide for data integration projects involving commonwealth data for statistical and research purposes Australian Government - National Statistical Service [Available from: https://statisticaldataintegration.abs.gov.au/topics/applying-the-separation-principle.
34. Barrett D, Heale R. What are Delphi studies? Evid Based Nurs. 2020;23(3):68–9.

35. Hamberger L, Lundin K, Sjogren A, Soderlund B. Indications for intracytoplasmic sperm injection. Hum Reprod. 1998;13(Suppl 1):128–33.
36. Babayev SN, Park CW, Bukulmez O. Intracytoplasmic sperm injection indications: how rigorous? Semin Reprod Med. 2014;32(4):283–90.
37. Bell MF, Bayliss DM, Glauert R, Harrison A, Ohan JL. Children of parents who have been hospitalised with psychiatric disorders are at risk of poor school readiness. Epidemiol Psychiatr Sci. 2019;28(5):508–20.
38. Bell MF, Bayliss DM, Glauert R, Ohan JL. Developmental vulnerabilities in children of chronically ill parents: a population-based linked data study. J Epidemiol Community Health. 2019;73(5):393–400.
39. Falster K, Hanly M, Banks E, Lynch J, Chambers G, Brownell M, et al. Maternal age and offspring developmental vulnerability at age five: a population-based cohort study of Australian children. PLoS Med. 2018;15(4):e1002558.
40. Hanly M, Falster K, Banks E, Lynch J, Chambers GM, Brownell M, et al. Role of maternal age at birth in child development among Indigenous and non-Indigenous Australian children in their first school year: a population-based cohort study. Lancet Child Adolesc Health. 2020;4(1):46–57.
41. Pearce A, Scalzi D, Lynch J, Smithers LG. Do thin, overweight and obese children have poorer development than their healthy-weight peers at the start of school? Findings from a South Australian data linkage study. Early Child Res Q. 2016;35:85–94.
42. Simpson A, Sarkic B, Enticott JC, Richardson Z, Buck K. Developmental vulnerability of Australian school-entry children with hearing loss. Aust J Prim Health. 2020;26:70–5.
43. The Australian Early Development Census. 'About the AEDC' 2022 [Available from: https://www.aedc.gov.au/about-the-aedc.
44. Bartlett JW, Seaman SR, White IR, Carpenter JR. Alzheimer's Disease Neuroimaging I. Multiple imputation of covariates by fully conditional specification: accommodating the substantive model. Stat Methods Med Res. 2015;24(4):462–87.
45. Madley-Dowd P, Hughes R, Tilling K, Heron J. The proportion of missing data should not be used to guide decisions on multiple imputation. J Clin Epidemiol. 2019;110:63–73.
46. Hernán MA, Robins JM. Causal inference: what if: Boca Raton: Chapman & Hall/CRC; 2020.
47. Williamson EJ, Forbes A, Wolfe R. Doubly robust estimators of causal exposure effects with missing data in the outcome, exposure or a confounder. Stat Med. 2012;31(30):4382–400.
48. Desai RJ, Franklin JM. Alternative approaches for confounding adjustment in observational studies using weighting based on the propensity score: a primer for practitioners. BMJ. 2019;367:l5657.
49. Bonduelle M, Ponjaert I, Steirteghem AV, Derde MP, Devroey P, Liebaers I. Developmental outcome at 2 years of age for children born after ICSI compared with children born after IVF. Hum Reprod. 2003;18(2):342–50.
50. Hvidtjorn D, Grove J, Schendel DE, Vaeth M, Ernst E, Nielsen LF, et al. Cerebral palsy among children born after in vitro fertilization: the role of preterm delivery–a population-based, cohort study. Pediatrics. 2006;118(2):475–82.
51. Lazaraviciute G, Kauser M, Bhattacharya S, Haggarty P, Bhattacharya S. A systematic review and meta-analysis of DNA methylation levels and imprinting disorders in children conceived by IVF/ICSI compared with children conceived spontaneously. Hum Reprod Update. 2014;20(6):840–52.
52. Sandin S, Nygren KG, Iliadou A, Hultman CM, Reichenberg A. Autism and mental retardation among offspring born after in vitro fertilization. JAMA. 2013;310(1):75–84.
53. Kissin DM, Zhang Y, Boulet SL, Fountain C, Bearman P, Schieve L, et al. Association of assisted reproductive technology (ART) treatment and parental infertility diagnosis with autism in ART-conceived children. Hum Reprod. 2015;30(2):454–65.
54. Knoester M, Helmerhorst FM, Vandenbroucke JP, van der Westerlaken LA, Walther FJ, Veen S, et al. Cognitive development of singletons born after intracytoplasmic sperm injection compared with in vitro fertilization and natural conception. Fertil Steril. 2008;90(2):289–96.
55. Leslie GI, Gibson FL, McMahon C, Cohen J, Saunders DM, Tennant C. Children conceived using ICSI do not have an increased risk of delayed mental development at 5 years of age. Hum Reprod. 2003;18(10):2067–72.
56. Goldbeck L, Gagsteiger F, Mindermann I, Strobele S, Izat Y. Cognitive development of singletons conceived by intracytoplasmic sperm injection or in vitro fertilization at age 5 and 10 years. J Pediatr Psychol. 2009;34(7):774–81.
57. Lee BK, Lessler J, Stuart EA. Weight trimming and propensity score weighting. PLoS ONE. 2011;6(3):e18174.
58. Du T, Xie Q, Ye J, Wang X, Qiu J, Yan Z, et al. Factors affecting male-to-female ratio at birth in frozen-thawed embryo transfer cycles: a large retrospective cohort study. Front Endocrinol (Lausanne). 2023;14:1188433.
59. Hentemann MA, Briskemyr S, Bertheussen K. Blastocyst transfer and gender: IVF versus ICSI. J Assist Reprod Genet. 2009;26(8):433–6.
60. Australian Institute of Health and Welfare. People with disability in Australia - web report: Canberra, ACT; 2022 [DIS 72:[Available from: https://www.aihw.gov.au/reports/disability/people-with-disability-in-australia/formats.
61. Chapman S. Australia homeschooling trends over the last decade 2021 [Available from: https://hslda.org/post/australia-homeschooling-trends-over-the-last-decade.

Publisher's Note