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Fresh versus frozen embryo transfers in assisted reproduction (Review)

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[Intervention Review]

Fresh versus frozen embryo transfers in assisted reproduction

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ABSTRACT

Background

In general, in vitro fertilisation (IVF) or intracytoplasmic sperm injection (ICSI) implies a single fresh and one or more frozen-thawed embryo transfers. Alternatively, the 'freeze-all' strategy implies transfer of frozen-thawed embryos only, with no fresh embryo transfers. In practice, both strategies can vary technically including differences in freezing techniques and timing of transfer of cryopreservation, that is vitrification versus slow freezing, freezing of two pro-nucleate (2pn) versus cleavage-stage embryos versus blastocysts, and transfer of cleavage-stage embryos versus blastocysts.

In the freeze-all strategy, embryo transfers are disengaged from ovarian stimulation in the initial treatment cycle. This could avoid a negative effect of ovarian hyperstimulation on the endometrium and thereby improve embryo implantation. It could also reduce the risk of ovarian hyperstimulation syndrome (OHSS) in the ovarian stimulation cycle by avoiding a pregnancy.

We compared the benefits and risks of the two treatment strategies.

Objectives

To evaluate the effectiveness and safety of the freeze-all strategy compared to the conventional IVF/ICSI strategy in women undergoing assisted reproductive technology.

Search methods

We searched the Cochrane Gynaecology and Fertility Group Trials Register, the Cochrane Central Register of Studies (CRSO), MEDLINE, Embase, PsycINFO, CINAHL, and two registers of ongoing trials in November 2016 together with reference checking and contact with study authors and experts in the field to identify additional studies.

Selection criteria

We included randomised clinical trials comparing a freeze-all strategy with a conventional IVF/ICSI strategy which includes fresh transfer of embryos in women undergoing IVF or ICSI treatment.

Data collection and analysis

We used standard methodological procedures recommended by Cochrane. The primary review outcomes were cumulative live birth and OHSS. Secondary outcomes included other adverse effects (miscarriage rate).

Main results

We included four randomised clinical trials analysing a total of 1892 women comparing a freeze-all strategy with a conventional IVF/ICSI strategy. The evidence was of moderate to low quality due to serious risk of bias and (for some outcomes) serious imprecision. Risk of bias was associated with unclear blinding of investigators for preliminary outcomes of the study, unit of analysis error, and absence of adequate study termination rules.

There was no clear evidence of a difference in cumulative live birth rate between the freeze-all strategy and the conventional IVF/ICSI strategy (odds ratio (OR) 1.09, 95% confidence interval (CI) 0.91 to 1.31; 4 trials; 1892 women; I^2 = 0%; moderate-quality evidence). This suggests that if the cumulative live birth rate is 58% following a conventional IVF/ICSI strategy, the rate following a freeze-all strategy would be between 56% and 65%.

The prevalence of OHSS was lower after the freeze-all strategy compared to the conventional IVF/ICSI strategy (OR 0.24, 95% CI 0.15 to 0.38; 2 trials; 1633 women; I^2 = 0%; low-quality evidence). This suggests that if the OHSS rate is 7% following a conventional IVF/ICSI strategy, the rate following a freeze-all strategy would be between 1% and 3%.

The freeze-all strategy was associated with fewer miscarriages (OR 0.67, 95% CI 0.52 to 0.86; 4 trials; 1892 women; I^2 = 0%; low-quality evidence) and a higher rate of pregnancy complications (OR 1.44, 95% CI 1.08 to 1.92; 2 trials; 1633 women; low-quality evidence). There was no difference in multiple pregnancies per woman after the first transfer (OR 1.11, 95% CI 0.85 to 1.44; 2 trials; 1630 women; low-quality evidence), and no data were reported for time to pregnancy.

Authors' conclusions

We found moderate-quality evidence showing that one strategy is not superior to the other in terms of cumulative live birth rates. Time to pregnancy was not reported, but it can be assumed to be shorter using a conventional IVF/ICSI strategy in the case of similar cumulative live birth rates, as embryo transfer is delayed in a freeze-all strategy. Low-quality evidence suggests that not performing a fresh transfer lowers the OHSS risk for women at risk of OHSS.

PLAIN LANGUAGE SUMMARY

Fresh versus frozen embryo transfers for assisted reproduction

Review question

We reviewed the evidence about the effectiveness and safety of a 'freeze-all' strategy for women undergoing in vitro fertilisation (IVF) or intracytoplasmic sperm injection (ICSI) compared to a conventional IVF/ICSI strategy, in terms of cumulative live birth rate and risk of ovarian hyperstimulation syndrome (OHSS).

Background

Embryo transfer in IVF/ICSI can be performed using either fresh or frozen-thawed embryos. There are therefore two embryo transfer strategies in IVF: 1) the conventional IVF/ICSI strategy with a single transfer of fresh and one or more transfers of frozen-thawed embryos, and 2) the 'freeze-all' strategy with transfer of frozen-thawed embryos only, and no fresh embryo transfer. Differences in freezing technique and timing of cryopreservation and transfer exist within both transfer strategies. In the freeze-all strategy, embryo transfers are disengaged from ovarian stimulation in the ovarian stimulation cycle. This strategy may be beneficial, as the ovarian hyperstimulation is suggested to have a negative effect on the receptivity of the endometrium for embryo implantation. The freeze-all strategy would lower the risk of OHSS since pregnancies do not occur in the cycle with ovarian stimulation.

Study characteristics

We included four studies comparing a freeze-all strategy with a conventional IVF/ICSI strategy in a total of 1892 women undergoing assisted reproductive technology. The evidence is current to November 2016.

Key results

We found evidence showing seemingly no difference between the strategies in cumulative live birth rate per woman. Our findings suggest that if the cumulative live birth rate is 58% following a conventional IVF/ICSI strategy, the rate following a freeze-all strategy would be between 56% and 65%. Time to pregnancy was not reported as an outcome in in the included studies, but it can be assumed to be shorter using a conventional IVF/ICSI strategy including fresh transfer in the case of similar cumulative live birth rates, as embryo

transfer is delayed in a freeze-all strategy. Not performing a fresh transfer (freeze-all strategy) lowers the OHSS risk for women at risk of OHSS. Our findings suggest that if the OHSS rate is 7% following a conventional IVF/ICSI strategy, the rate following a freeze-all strategy would be between 1% and 3%.

Quality of the evidence

The evidence was of moderate to low quality due to serious risk of bias and (for some outcomes) serious imprecision. Risk of bias was associated with unclear blinding of investigators for preliminary outcomes of the study, unit of analysis error, and absence of adequate study termination rules.

SUMMARY OF FINDINGS FOR THE MAIN COMPARISON [Explanation]

Fresh versus frozen embryo transfers in assisted reproduction

Patient or population: women undergoing assisted reproduction
Setting: assisted reproduction clinic
Intervention: frozen embryo transfers only
Comparison: fresh and frozen embryo transfers

Outcomes	Anticipated absolute effects* (95% CI)		Relative effect (95% CI)	№ of participants (studies)	Quality of the evidence (GRADE)	Comments
	Risk with fresh and frozen embryo transfers	Risk with frozen embryo transfers only				
Live birth rate cumulatively for all embryo stages of transfer	579 per 1000	600 per 1000 (556 to 643)	OR 1.09 (0.91 to 1.31)	1892 (4 RCTs)	⊕⊕⊕◯ MODERATE [1]	
Ovarian hyperstimulation syndrome per cycle with ovarian hyperstimulation	70 per 1000	18 per 1000 (11 to 28)	OR 0.24 (0.15 to 0.38)	1633 (2 RCTs)	⊕⊕◯◯ LOW [1,2]	
Multiple pregnancy per woman after first ET	161 per 1000	176 per 1000 (141 to 217)	OR 1.11 (0.85 to 1.44)	1630 (2 RCTs)	⊕⊕◯◯ LOW [1,2]	
Miscarriage per woman after first ET	184 per 1000	131 per 1000 (105 to 162)	OR 0.67 (0.52 to 0.86)	1892 (4 RCTs)	⊕⊕◯◯ LOW [1,2]	
Pregnancy complications per woman after first ET	110 per 1000	151 per 1000 (118 to 191)	OR 1.44 (1.08 to 1.92)	1633 (2 RCTs)	⊕⊕◯◯ LOW [1,2]	
Time to pregnancy	Not reported in any of the included studies					

*The risk in the intervention group (and its 95% confidence interval) is based on the mean risk in the comparison group and the relative effect of the intervention (and its 95% CI).

CI: confidence interval; **ET:** embryo transfer; **OR:** odds ratio; **RCT:** randomised clinical trial

GRADE Working Group grades of evidence

High quality: We are very confident that the true effect lies close to that of the estimate of the effect.

Moderate quality: We are moderately confident in the effect estimate: The true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different.

Low quality: Our confidence in the effect estimate is limited: The true effect may be substantially different from the estimate of the effect.

Very low quality: We have very little confidence in the effect estimate: The true effect is likely to be substantially different from the estimate of effect

[1]Downgraded one level due to serious risk of bias associated with lack of power calculation (unclear what determined end of study) and/or use of interim analysis that was calculated per transfer (unit of analysis error) with absence of adequate stopping rules (possible overestimation of treatment effect).

[2]Downgraded one level due to serious imprecision: event rate < 300.

BACKGROUND

Description of the condition

Subfertility is defined as the failure to conceive after one year of unprotected intercourse (Van Voorhis 2007). One in six couples experience subfertility at least once during their reproductive lifetime, and approximately 10% of couples worldwide are subfertile (ESHRE 2010; CDC 2011). Common causes of subfertility include poor semen quality, obstruction of the fallopian tubes, absence of ovulation, and endometriosis (Hull 1985). Poor semen quality can manifest itself as low sperm concentration, low motility, or low numbers of sperm with normal morphology. Fallopian tubes can be blocked or damaged by infection, or there can be adhesions of the tubes or ovaries caused by surgery, chlamydia, or endometriosis. Couples who fail to conceive naturally are diagnosed as having unexplained infertility if no cause can be found after standard fertility tests.

Description of the intervention

Assisted reproductive technology has rapidly evolved as an intervention to improve pregnancy rates. It involves the handling of gametes and embryos outside the human body and consists of in vitro fertilisation (IVF) with or without intracytoplasmic sperm injection (ICSI). After fertilisation, fresh transfer of the morphologically best embryo(s) into the uterine cavity is performed. Embryos suitable for transfer, but not transferred fresh, are frozen for future use.

Even so, many women fail to achieve a pregnancy after transfer of one or more fresh embryos. Recent technical improvements in cryopreservation have led to increased chances of embryo survival after thawing and subsequently increased pregnancy rates per frozen-thawed embryo transfer (CDC 2011; Wong 2014). In fact, pregnancy rates after frozen-thawed embryo transfer are now almost equal to pregnancy rates after fresh transfer when calculated per transfer. This has fuelled the call for a new IVF/ICSI strategy where no fresh embryo transfer is conducted and all available embryos are cryopreserved, thawed, and transferred in a subsequent cycle. This would reduce any residual chance of ovarian hyperstimulation syndrome (OHSS) and possibly increase the cumulative live birth rates (Mastenbroek 2011; Maheshwari 2013).

How the intervention might work

In contrast to the conventional strategy, in a 'freeze-all' strategy there are no fresh embryo transfers in the cycle with ovarian stimulation, but only frozen-thawed embryo transfers in subsequent cycles without ovarian stimulation. This avoids possible adverse effects of ovarian stimulation. The underlying reason here is the claim that ovarian stimulation reduces endometrial receptivity for the implanting embryo (Kolibianakis 2002; Bourgain 2003). Studies on the molecular level comparing stimulated with unstimulated endometrium samples have shown distinct gene-expression profiles between the two conditions (Haouzi 2009). Transfer of frozen-thawed embryos only would thus circumvent a possible negative effect of gonadotropins on the endometrium in the cycle with ovarian stimulation, and consequently increase live birth rates, the main outcome of interest to subfertile couples.

Ovarian stimulation with exogenous gonadotropins in IVF also increases the risk of OHSS when a pregnancy occurs in such a cycle with ovarian stimulation. Avoiding a pregnancy in the cycle with ovarian stimulation by only transferring frozen-thawed embryos in subsequent unstimulated cycles would eliminate the residual risks of OHSS, and OHSS would therefore be self limiting. Mild OHSS symptoms can still occur as a result of the human chorionic gonadoptropin trigger in the hyperstimulated cycle in the freeze-all strategy, but OHSS in its severe form should be rare.

Why it is important to do this review

Nowadays, an increasing number of clinics apply the freeze-all strategy as a standard treatment strategy in their practice. However, the relative effectiveness and safety of IVF treatment with the freeze-all strategy compared to the conventional IVF/ICSI strategy is unclear. A previous non-Cochrane systematic review reported that a freeze-all strategy was associated with higher ongoing and clinical pregnancy rates, and lower miscarriage rates than the conventional IVF/ICSI strategy (Roque 2013). However, this review did not report live birth or safety outcomes. This review aimed to provide a systematic, up-to-date summary of reliable evidence of the benefits and risks of a freeze-all strategy.

OBJECTIVES

To evaluate the effectiveness and safety of the freeze-all strategy compared to the conventional IVF/ICSI strategy in women undergoing assisted reproductive technology.

METHODS

Criteria for considering studies for this review

Types of studies

We included published randomised clinical trials and excluded quasi- and pseudo-randomised clinical trials. We excluded trials published only as abstract. We planned to include cross-over trials

for completeness, but would only pool the data from the first phase in the meta-analysis (Vail 2003).

Types of participants

All women undergoing IVF or ICSI.

Types of interventions

Trials comparing the freeze-all strategy with transfer of frozen-thawed embryos only versus the conventional IVF/ICSI strategy with transfer of fresh and subsequent frozen-thawed embryos until a live birth occurred or until all embryos from the initial cycle were transferred.

Types of outcome measures

Primary outcomes

1. Effectiveness: cumulative live birth rate per randomised woman, i.e. the rate of live birth following the transfer of all (fresh or frozen-thawed) embryos available from the stimulated cycle.
2. Safety: OHSS per randomised woman.

Secondary outcomes

1. Cumulative ongoing pregnancy rate, defined as the number of ongoing pregnancies per woman randomised (demonstrated by the presence of a gestational sac with fetal heartbeat on ultrasound at $\geq$ 12 weeks of gestation).
2. Clinical pregnancy, defined as the cumulative number of clinical pregnancies per woman randomised (demonstrated by a pregnancy confirmed by ultrasonographic visualisation of one or more gestational sacs.
3. Time to pregnancy, defined as the time between the first day of the last menstrual period and clinical pregnancy.
4. Multiple-pregnancy rate, defined as the number of multiple pregnancies per woman.
5. Miscarriage rate, defined as the number of miscarriages per woman.
6. Pregnancy complications (including ectopic pregnancy, foetal growth disorders, preterm birth < 37 weeks, pregnancy-induced hypertension, (pre-) eclampsia, women with haemolysis, elevated liver enzymes, and low platelets in the blood (HELLP syndrome) per woman.
7. Birth weight of babies born per baby.
8. Congenital disorders, defined as the number of congenital abnormalities at birth per live-born children plus number of foetuses therapeutically terminated.

We also calculated multiple pregnancy, miscarriage, pregnancy complications, and birth weight per clinical pregnancy in a secondary analysis.

Search methods for identification of studies

We searched for all published randomised clinical trials on the freeze-all strategy, without language restriction and in consultation with the Cochrane Gynaecology and Fertility Group (CGF) Information Specialist.

Electronic searches

We searched the following electronic databases, trial registers, and websites from inception to 14 November 2016 without language restriction and in consultation with the CGF Information Specialist: Cochrane Gynaecology and Fertility Group Specialised Register, Cochrane Central Register of Studies (CENTRAL CRSO), MEDLINE, Embase, PsycINFO, and CINAHL. These search strategies are presented in Appendix 1; Appendix 2; Appendix 3; Appendix 4; Appendix 5; Appendix 6.

Other electronic sources of trials included:

- trial registers for ongoing and registered trials: ClinicalTrials.gov, a service of the US National Institutes of Health (clinicaltrials.gov/ct2/home) and the World Health Organization International Clinical Trials Registry Platform (WHO ICTRP) (www.who.int/trialsearch/Default.aspx); see Appendix 7; Appendix 8.
- DARE (Database of Abstracts of Reviews of Effects) in the Cochrane Library for reference lists from relevant non-Cochrane reviews (onlinelibrary.wiley.com/o/cochrane/cochrane_cldare_articles_fs.html);
- conference abstracts in the Web of Knowledge (wokinfo.com/);
- OpenGrey (System for Information on Grey Literature in Europe) (www.opengrey.eu/);
- PubMed (www.ncbi.nlm.nih.gov/pubmed/).

Searching other resources

We examined the reference lists of eligible articles and contacted study authors where necessary to obtain additional relevant data. We also handsearched relevant journals and conference abstracts that were not covered in the CGF Register.

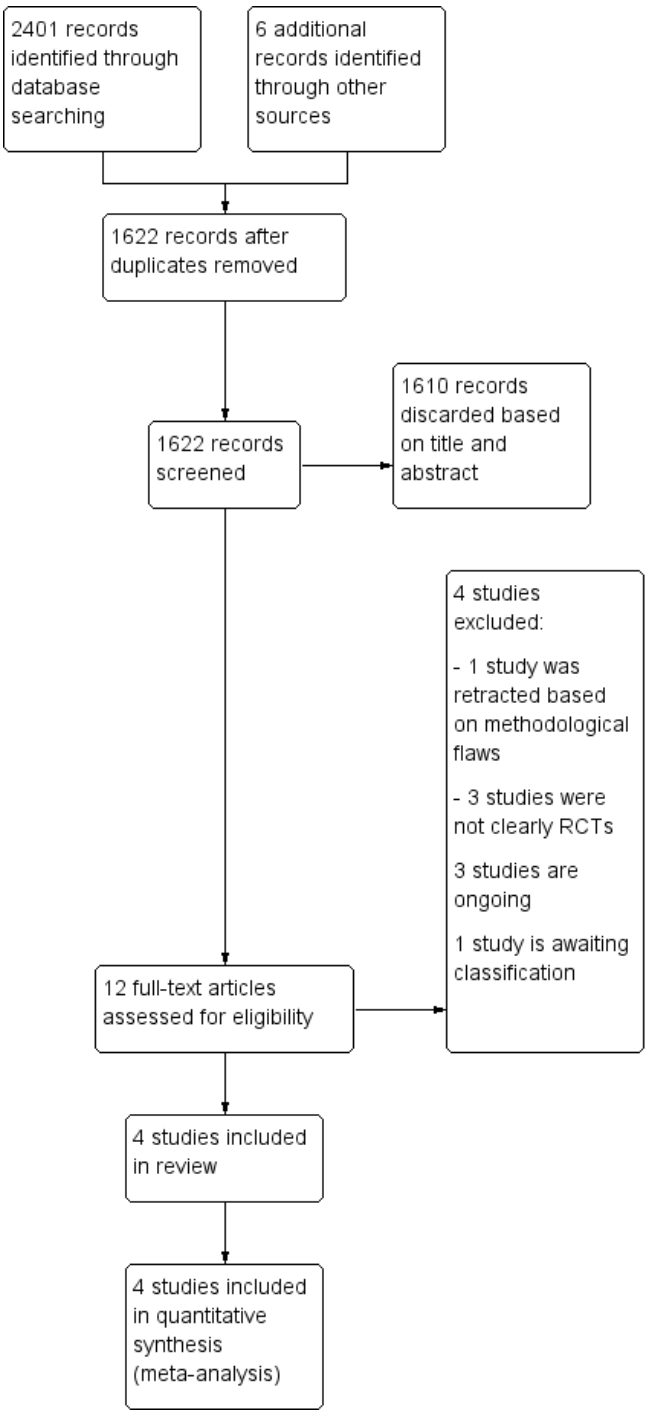
Data collection and analysis

Selection of studies

Two review authors (KMW and SM) screened the titles and abstracts retrieved by the search and retrieved the full texts of all potentially eligible studies. We independently examined these full-text articles for compliance with the inclusion criteria and selected studies eligible for inclusion in the review. We corresponded with study investigators as required to clarify study eligibility. Disagreements as to study eligibility were resolved by discussion or by con-

sulting a third review author (SR). We documented the selection process with a PRISMA flow chart (Figure 1).

Figure 1. Study flow diagram.



Data extraction and management

Two review authors (KMW and SM) independently extracted data from the eligible studies using a data extraction form designed and pilot-tested by the authors. Any discrepancies were resolved by discussion. The data extraction forms included methodological quality and allocation information. We included this information in the review and presented it in the Characteristics of included studies and Characteristics of excluded studies tables.
We corresponded with study investigators to request further data on methods or results, or both, as required.

Assessment of risk of bias in included studies

Two review authors (KMW and SM) independently assessed the included studies for risk of bias using the Cochrane 'Risk of bias' assessment tool for the following domains (Higgins 2011).

Sequence generation

We allocated a low risk of bias if the investigators described a random component in the sequence generation process, such as:

- using a computerised random number generator;
- using a random numbers table.

Allocation concealment

We allocated a low risk of bias if the participants and investigators enrolling participants could not foresee assignment because one of the following, or an equivalent method, was used to conceal allocation:

- central computer randomisation;
- serially numbered, sealed, opaque envelopes.

Blinding

We allocated a low risk of bias if blinding of participants, scientists, and clinicians or nurses had been ensured. However, in this study design it was ethically not possible to blind participants and clinicians. Lack of blinding may not increase the risk of bias if follow-up is complete and outcomes are unequivocal (live birth).

Completeness of outcome data

We allocated a low risk of bias if there were no missing data, which meant live birth rate and length of follow-up were stated, loss to follow-up was accounted for, and an intention-to-treat analysis had been carried out.

Selective outcome reporting

We allocated a low risk of bias if all of the study's primary, secondary, and additional outcomes that were of interest in the review had been reported in a prespecified way.

Other sources of bias

We allocated a low risk of bias if the study:

- was free of commercial funding;
- reported multiple-pregnancy rate in the case of an embryo transfer policy of multiple embryos per treatment cycle;
- had no other source of bias identified (e.g. imbalance in prognostic factors at baseline).

Two review authors (KMW and SM) assessed these domains and resolved any disagreements by consensus or by consulting a third review author (SR). We described the judgements and presented the conclusions in the 'Risk of bias' figures. We took into account all judgements in the interpretation of review findings.

Measures of treatment effect

For dichotomous data (e.g. live birth rates), we used the numbers of events in the freeze-all strategy and in the conventional IVF/ICSI strategy group of each study to calculate Mantel-Haenszel odds ratios (ORs) with 95% confidence intervals (CI). We used Peto ORs where the event was very rare (less than 1%) or in the case of zero cell counts. For continuous data (e.g. birth weight), we calculated mean difference (MD) between treatment groups provided that the same measure was used. We reversed the direction of effect of individual studies if required to ensure consistency across trials. We treated ordinal data as continuous data. Where data to calculate ORs or MDs were not available, we utilised the most detailed numerical data available that would facilitate similar analyses of included studies (e.g. test statistics, P values). We compared the magnitude and direction of effect reported by studies with how they were presented in the review, taking into account legitimate differences.
We planned to analyse the outcome 'time to pregnancy' using hazard ratios (HRs).

Unit of analysis issues

We performed the analyses with data per woman randomised, apart from birth weight, which we analysed per baby. If data of the primary analysis were reported per embryo, per oocyte, per cycle, or per transfer, we contacted the authors of the studies for per-woman data for completeness.
We counted reported multiple live births as one live birth event.

We planned to include only first-phase data from cross-over trials. We also performed secondary analyses for multiple pregnancy, miscarriage, pregnancy complications, and birth weight per clinical pregnancy since these conditions only occur in pregnant women.

Dealing with missing data

We analysed the data on an intention-to-treat basis, and contacted the authors of three included studies, Shapiro 2011a, Shapiro 2011b, and Ferraretti 1999, and one excluded study, Absalan 2013, for missing data. We queried the study authors about these missing data and about bias (e.g. randomisation and blinding). One author did not reply to our request for information (Absalan 2013). The remaining authors very kindly responded to our request for additional information, and we were able to include these data in our analysis.

We assumed that live births had not occurred in women without a reported outcome. If studies reported sufficient detail to calculate MDs, but provided no information on associated standard deviations (SD), we assumed that the outcome had a SD equal to the highest SD from other studies within the same analysis.

Assessment of heterogeneity

We considered heterogeneity when the clinical and methodological characteristics of the included studies were sufficiently similar for a meta-analysis to provide a clinically meaningful summary. We performed statistical analyses in accordance with the guidelines developed by Cochrane (Higgins 2003; Higgins 2011). We assessed heterogeneity between the results of different studies by the I^2 statistic, considering an I^2 value greater than 50% to indicate substantial heterogeneity (Higgins 2003; Higgins 2011).

Assessment of reporting biases

We aimed to minimise the potential impact of publication and reporting biases by performing a comprehensive search for eligible studies and looking for duplication of data. We planned to perform a funnel plot to investigate the possibility of small-study effects if 10 or more studies were included in an analysis.

If included studies reported neither the primary outcome measure of live birth nor interim outcomes such as clinical pregnancy, we undertook informal assessment as to whether studies reporting the primary outcome measures reflected typical findings for the interim outcomes. We considered within-study reporting bias by looking at the protocols.

We addressed the assessment of reporting biases in the Risk of bias in included studies section of the Results.

Data synthesis

We used Review Manager 5 software to perform the meta-analyses with a fixed-effect model to calculate pooled ORs and 95% CIs (RevMan 2014). To aid interpretation, we translated findings for primary outcomes to absolute risks, expressed as percentages based on the 95% CIs. We combined results for continuous outcomes using MDs.

Prospectively, we planned to present the analyses as:

- cumulative live birth rates for IVF/ICSI cycles with frozen-thawed embryo transfers until live birth was achieved or when all frozen embryos originating from the cycle with ovarian stimulation were transferred in the freeze-all strategy versus IVF/ICSI cycles with fresh and subsequent frozen-thawed embryo transfers until live birth was achieved or when all frozen embryos originating from the cycle with ovarian stimulation were transferred in the conventional IVF/ICSI strategy;
- pregnancy and live birth rates for one IVF/ICSI cycle with the first frozen-thawed embryo transfer in the freeze-all strategy versus one IVF/ICSI cycle with the first fresh embryo transfer in the conventional IVF/ICSI strategy (as an additional table).

Subgroup analysis and investigation of heterogeneity

We had planned to perform subanalyses on timing of cryopreservation (e.g. day of embryo development) and method of cryopreservation (e.g. slow freezing or vitrification). However, data were insufficient to conduct all planned subgroup analyses. Should more data become available in the future, we will conduct additional subgroup analyses in later updates of this review.

Sensitivity analysis

We conducted sensitivity analyses for the primary outcome. These analyses included consideration of whether the review conclusions would have differed if:

1. eligibility was restricted to studies without high risk of bias;
2. a random-effects model had been adopted;
3. the summary effect measure was risk ratio rather than OR.

Overall quality of the body of evidence: 'Summary of findings' table

We prepared a 'Summary of findings' table using GRADEpro software and Cochrane methods (GRADEpro GDT 2014; Higgins 2011). This table evaluates the overall quality of the body of evidence for the main review outcomes. Two review authors independently evaluated the overall quality of the evidence for the outcomes (live birth, OHSS, multiple pregnancy, miscarriage, pregnancy complications and time to pregnancy) using GRADE criteria (study limitations such as risk of bias, consistency of effect, imprecision, indirectness, and publication bias). We justified, documented, and took into account judgements about evidence quality (high, moderate, low, or very low) in the results for each outcome.

RESULTS

Description of studies

Results of the search

Our searches on 14 November 2016 revealed 2401 reports, of which 785 were duplicates, leaving 1622 reports. After screening the title and abstract, we found 12 reports to be potentially eligible, and retrieved these reports in full text.

We excluded four studies: one randomised women to a different intervention that was not clear from the abstract (Boostanfar 2016); two were considered not properly randomised (Absalan 2013; Yang 2015); and one has been retracted (Aflatoonian 2010).

Three studies were ongoing trials and awaiting data (ACTRN12612000422820; NCT02148393; NTR3187).

One study did not clearly report the methods used; it has been classified as awaiting classification and will be reassessed in the next iteration of this review (Chandel 2016).

We included four studies in the review.

See the study flow diagram (Figure 1) and study tables (Characteristics of included studies; Characteristics of excluded studies; Characteristics of ongoing studies).

Included studies

Study design and setting

We included four parallel-design randomised clinical trials (RCTs) in the review. Three were single-centre studies, conducted in reproductive medical centres in Italy and the United States (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b), and one was a multicentre trial conducted in 14 reproductive medical centres throughout China (Chen 2016).

Participants

The four studies enrolled a total of 1892 women, with 934 women undergoing the freeze-all strategy and 958 women undergoing the conventional IVF/ICSI strategy. The inclusion criteria of the two studies of Shapiro and colleagues were based on the number of antral follicles observed at baseline ultrasound examination: Shapiro 2011a included normal responders (8 to 15 antral follicles), and Shapiro 2011b included high responders (> 15 antral follicles). Ferraretti 1999 included women at risk of developing OHSS, based on level of oestradiol (E2) and number of retrieved eggs (≥ 15 oocytes). In all trials, the baseline characteristics were similar between the two strategies. Chen 2016 included women with polycystic ovary syndrome. The ages of the women included by Shapiro ranged from 18 to 41 years. The mean age for the women included in Ferraretti 1999 ranged from 31.6 to 31.4 years. Women in Chen 2016 were between the ages of 20 and 34 years. For details, see Characteristics of included studies.

Interventions

All four studies compared the freeze-all strategy versus the conventional IVF/ICSI strategy.

Women in the Ferraretti 1999 study received a down-regulation protocol with gonadotropin-releasing hormone (GnRH) analogue (0.3 mg subcutaneous buserelin acetate (Suprefact) two times a day) and ovarian stimulation with urinary gonadotropin (4 ampoules of follicle-stimulating hormone (FSH) on the first and second days of treatment, and 2 ampoules of FSH plus 2 ampoules of human menopausal gonadotropins (hMG) on the third and fourth treatment days, followed by an adjusted dosage of gonadotropins according to the individual response measured by plasma concentration of E2 and follicular growth assessed by ultrasound) (Ferraretti 1996). All women received 7500 IU of human chorionic gonadotropin (hCG) 34 to 36 hours before follicle aspiration followed by 20 g of intravenous albumin. Embryos were frozen at the pronuclear stage. All embryos were transferred at the early cleavage stage (day 3) in artificial cycles. The scheme included oral administration of oestradiol valerate, 2 mg daily for the first 5 days of the cycle; 4 mg/day from day 6 to day 10; 6 mg/day from day 11 to day 13; then 4 mg/day from day 14 onward. On day 15 of the cycle, 50 mg of progesterone in oil was administered daily, and on day 17 the dose was increased to 100 mg/day.

In Shapiro 2011a and Shapiro 2011b, women received down-regulation with a GnRH antagonist and a combination of recombinant FSH and highly purified urinary FSH. Human chorionic gonadotropin (5 to 15 IU per pound body weight (11 to 33 IU/kg)) was administered 34 to 36 hours prior to follicle aspiration. In those women with greater ovarian response, 4 mg leuprolide acetate was added concomitant to the hCG. Embryos were vitrified at the pronuclear stage. All embryos were transferred as blastocysts in artificial cycles. Women with fresh embryo transfers received 6.0 mg daily E2 and daily progesterone injections (typically 100 mg), with progesterone supplementation beginning one to two days after follicle aspiration and E2 initiated as needed. Women with frozen-thawed embryo transfers were down-regulated with leuprolide acetate in a subsequent cycle and received oral 6.0 mg daily E2 and E2 patches as needed starting 10 to 14 days before thawing to achieve a target endometrial thickness of at least 8 mm. Daily progesterone injections (typically 100 mg) were started the day before thawing. In both groups, E2 and progesterone supplements were adjusted as needed to sustain serum levels of at least 200 pg/mL and 15 ng/mL, respectively, until increasing serum levels indicated placental production, typically at 9 to 10 weeks' gestation.

In Chen 2016, women received recombinant FSH at a daily dose of 112.5 IU for those weighing less than 60 kg and 150 IU for those weighing over 60 kg starting on day 2 or 3 of the men-

strual cycle. This was adjusted following ovarian response. Human menopausal gonadotropin could be added when considered to appropriate. On the day of oocyte retrieval, women had to have more than 3 and fewer than 30 oocytes with a low risk of OHSS to be randomised. Intramuscular progesterone at a daily dose of 80 mg was administered for luteal-phase support in the fresh-transfer group. Embryos were cryopreserved at day 3 of development. Oral oestradiol valerate was used for endometrial preparation on day 2 or 3 of the second menstrual cycle after oocyte retrieval. Intramuscular progesterone (80 mg/day) was added when endometrial thickness reached 8 mm or more or at the physician's discretion. On day 4 of progesterone administration, two day 3 frozen embryos were thawed and transferred. Luteal-phase support with oestradiol valerate and intramuscular progesterone for endometrium preparation continued until 10 weeks after conception.

Outcomes

Data were extracted from study reports or provided by authors for the following outcomes.

Primary outcomes

1. Effectiveness: Cumulative live birth per woman. Two studies did not report on live birth in their published article (Shapiro 2011a; Shapiro 2011b), but we were able to obtain these data by personal communication with the authors. One study did not report on live birth rate after the first embryo transfer (Ferraretti 1999), but we were able to obtain these data by personal communication with the authors. Chen 2016 reported these data.
2. Safety: OHSS. One study reported OHSS per woman if hospitalisation was required (Ferraretti 1999). Two studies did not report on OHSS (Shapiro 2011a; Shapiro 2011b), but we were able to obtain these data by personal communication with the authors. However, we did not include the data from these two studies in the analysis, as women with high risk of OHSS were excluded and standardly received the freeze-all strategy. Chen 2016 reported these data.

Secondary outcomes

1. Two studies reported ongoing pregnancy rate determined at 10 weeks of gestational age (Shapiro 2011a; Shapiro 2011b).
2. Three studies reported clinical pregnancy rate (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b), but only one study reported this outcome cumulative per woman (Ferraretti 1999).
3. None of the studies reported time to pregnancy or the results for each menstrual cycle following randomisation.
4. Two studies reported multiple-pregnancy rate (Shapiro 2011b; Chen 2016).
5. All four studies reported the number of miscarriages (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b; Chen 2016).
6. One study reported on congenital disorders (Chen 2016).
7. Two studies reported pregnancy complications (Ferraretti 1999; Chen 2016).
8. One study reported birth weight of the newborn (Chen 2016).

Excluded studies

We excluded four potentially eligible studies from the review, for the following reasons.

- Aflatoonian 2010, as this study was retracted.
- Absalan 2013, as it was unclear whether it was truly a RCT. This study compared the clinical and delivery rates between the freeze-all strategy and the conventional strategy in women at risk for OHSS. In their abstract it was stated that women with OHSS were randomly divided into two groups, with fresh embryo transfer and with frozen transfer. However, nothing is mentioned in the methods section about the method of randomisation (sequence generation or allocation concealment) or which method was used to divide women into the two groups. Nothing was reported on the occurrence of OHSS in these women. The authors did not respond to our request for additional information.
- Yang 2015, as one-third of all randomised women chose to be in group 3 (fresh transfer of a day 3 embryo followed by frozen-thawed embryos) after randomisation. We did not consider the study to be a properly randomised RCT.
- Boostanfar 2016 randomised women to a different intervention that was not clear from the abstract.

Awaiting classification

Chandel 2016 is awaiting classification (see Characteristics of studies awaiting classification); we await more information from the study authors.

Ongoing studies

We identified 12 ongoing studies from trial registers that may have results for inclusion in future versions of this review (ACTRN12612000422820; ACTRN12616000643471; ISRCTN61225414; NCT02000349; NCT02133950; NCT02148393; NCT02471573; NCT02570386; NCT02681367; NCT02712840; NCT02746562; NTR3187). Note that studies that were registered in the trial registers but that were not started or that were withdrawn or stopped were not included in this review.

Risk of bias in included studies

See the 'Risk of bias' summary (Figure 2) and graph (Figure 3) for the four included trials. See also Characteristics of included studies.

Figure 2. Risk of bias summary: review authors' judgements about each risk of bias item for each included study.

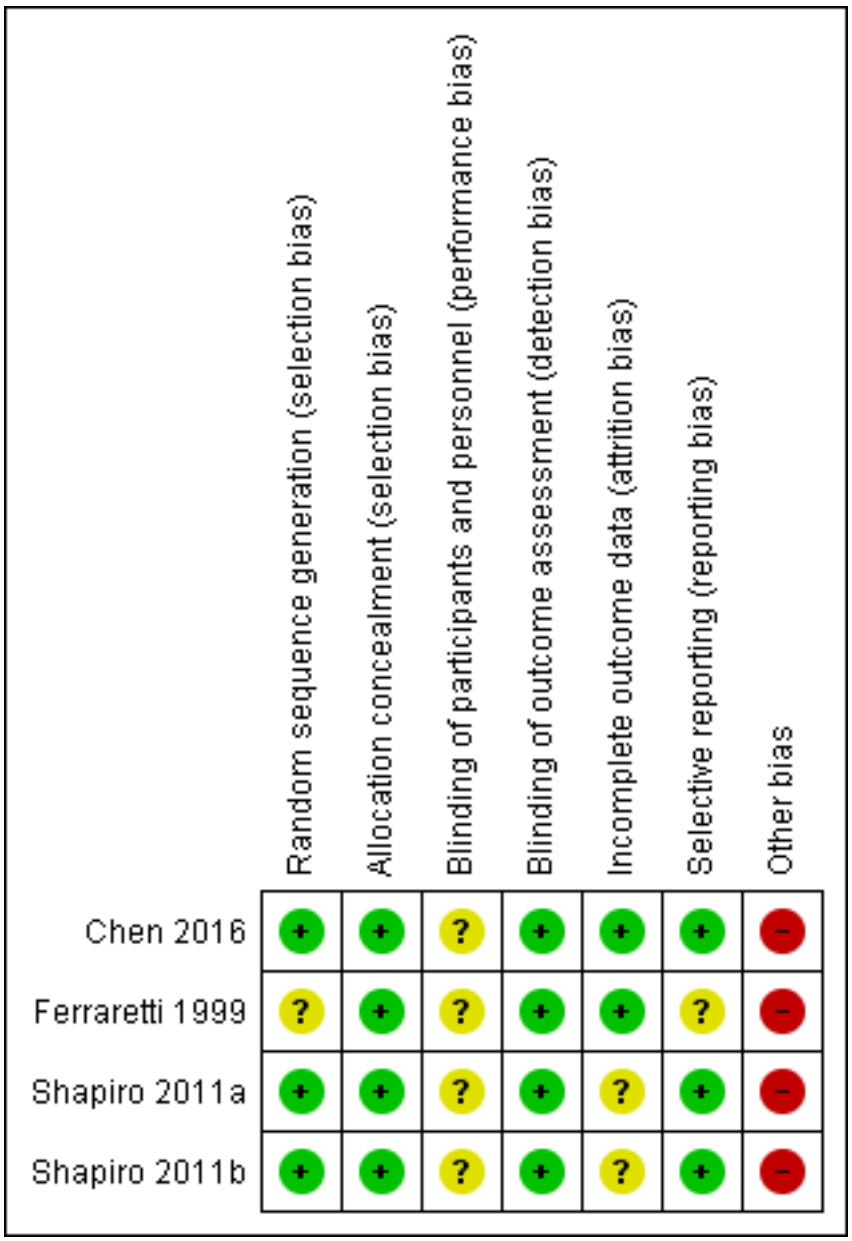
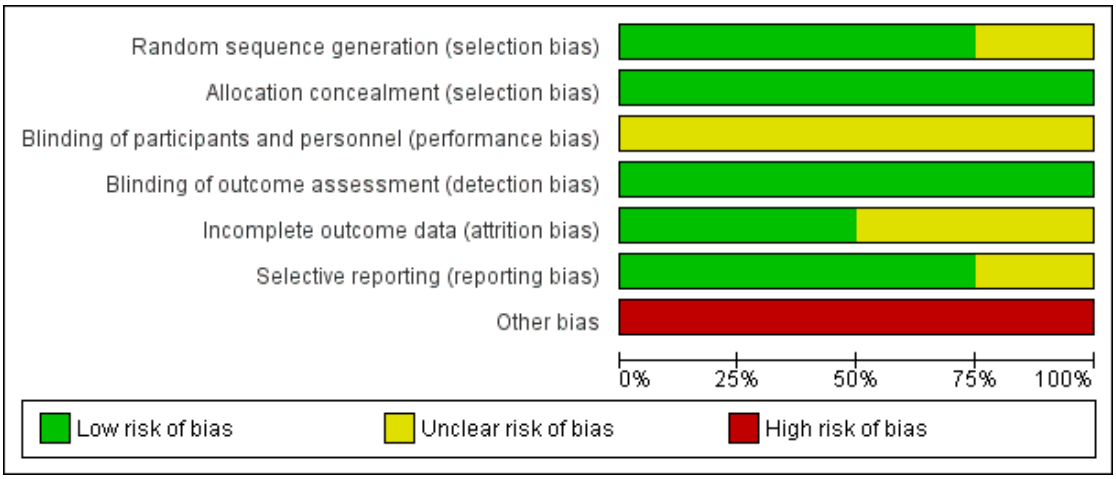


Figure 3. Risk of bias graph: review authors' judgements about each risk of bias item presented as percentages across all included studies.



Allocation

Sequence generation

The randomisation procedure was well described in Shapiro 2011a. In the second study of Shapiro 2011b, the authors referred to the randomisation procedure in Shapiro 2011a. These two studies used randomly chosen envelopes, and we judged these two studies to be at low risk of selection bias related to sequence generation. Ferraretti 1999 did not describe the method of randomisation in the published article, but replied in a personal communication that the method of randomisation was performed with random sealed envelopes; we judged this study to be at unclear risk of this bias, as random sequence was used but it was unclear whether envelopes were opaque and sequentially numbered. Randomisation in Chen 2016 was well described; an online central randomisation system was used. We considered risk of selection bias related to sequence generation to be low.

Allocation concealment

Shapiro 2011a and Shapiro 2011b performed allocation concealment by using identical, opaque, unmarked, sealed envelopes, and we therefore judged both studies to be at low risk of selection bias related to allocation concealment. The first author of the Ferraretti 1999 study provided additional information on allocation concealment. This study performed participant allocation by sealed envelopes, and we therefore judged it to be at low risk of bias for this domain. There was low risk of selection bias related to allocation concealment in Chen 2016 due to the use of an online central randomisation system.

Blinding

As described in the Methods section, blinding of the participant or the clinician is technically not possible due to the nature of the intervention in this study design. We felt that lack of blinding was not likely to influence findings for the primary outcomes live birth or OHSS. However, blinding for the primary outcome was not reported for the investigators of the four studies, which could have influenced the decision to terminate a trial. The risk of performance bias was unclear in all four studies.

Incomplete outcome data

Three studies did not report intention-to-treat analysis in the methodological or analysis sections (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b), while one study did report intention-to-treat analysis (Chen 2016).

We judged the studies by Shapiro 2011a and Shapiro 2011b to be at high risk of attrition bias. These studies did not take into account withdrawals or exclusions of randomised women in the reported analyses. Both studies also analysed the outcomes per embryo transferred instead of per woman. However, sufficient data were available for analysis per woman in meta-analysis. We prespecified ongoing pregnancy as a viable pregnancy at 12 weeks' gestation. These two studies defined ongoing pregnancy at 10 weeks' gestation, which could slightly overestimate the results for this outcome. Taken these issues into account, we considered the risk of bias to be unclear in these two studies.

Ferraretti 1999 and Chen 2016 did analyse all randomised women. Risk of attrition bias was low.

Selective reporting

One study was registered in a prospective trial register under number NCT01841528, including an automatically indexed link on the published report on the study, and the study protocol was published beforehand (Chen 2016). Prespecified outcomes were generally reported, although some prespecified outcomes (e.g. time to pregnancy) were missing from the report. Considering this, we judged this study to be at low risk of reporting bias. Two studies were registered in a prospective trial register with the respective trial numbers NCT00963625 and NCT00963079 (Shapiro 2011a; Shapiro 2011b). Data on the follow-up of the studies were available in the trial register. The prespecified outcomes of interest were reported in the two studies, and we judged these studies to be at low risk of this bias. We could not assess reporting bias for Ferraretti 1999, as trial registers did not exist at that time, therefore the risk of reporting bias for this study was unclear.

Other potential sources of bias

Three of the studies did not clearly report their prespecified criteria for early termination of their trial. Ferraretti 1999 did not prespecify rules as to when to terminate the study. In the two studies by Shapiro 2011a and Shapiro 2011b, an interim analysis was planned after 100 completed blastocyst transfers. While women were randomised, the interim analyses were based on completed blastocyst transfers (unit of analysis error). They did not report whether the interim analysis was performed by an independent committee that was blinded for the primary outcome. In addition, Shapiro 2011b pre-terminated the study after an interim analysis based on differences in embryo quality between the two strategies. This reason was not mentioned as one of the criteria to terminate the study. All three studies cryopreserved embryos at the two pronucleate (2pn) stage with slow freezing, which is not currently a common freezing protocol in IVF centres.

After freezing and thawing, the four studies transferred embryos at a different developmental stage: Ferraretti 1999 and Chen 2016 transferred cleavage embryos, and Shapiro 2011a and Shapiro 2011b transferred blastocysts. None of the four studies reported time to pregnancy or (separate or incremental) data per subsequent menstrual or cryo-transfer cycle (relevant for time-to-pregnancy comparison). The difference in technical protocols (some of which are not common practice) between studies in day of cryopreservation and embryo developmental stage of transfer, together with the differences in study population, complicates the comparison between freeze-all and conventional IVF/ICSI strategies and could introduce heterogeneity between studies. We therefore judged all studies to be at high risk of this bias.

Effects of interventions

See: **Summary of findings for the main comparison** Fresh versus frozen embryo transfers in assisted reproduction

We included four studies involving 1892 women in this review. See Summary of findings for the main comparison.

1. Comparison of the freeze-all strategy versus the conventional IVF/ICSI strategy

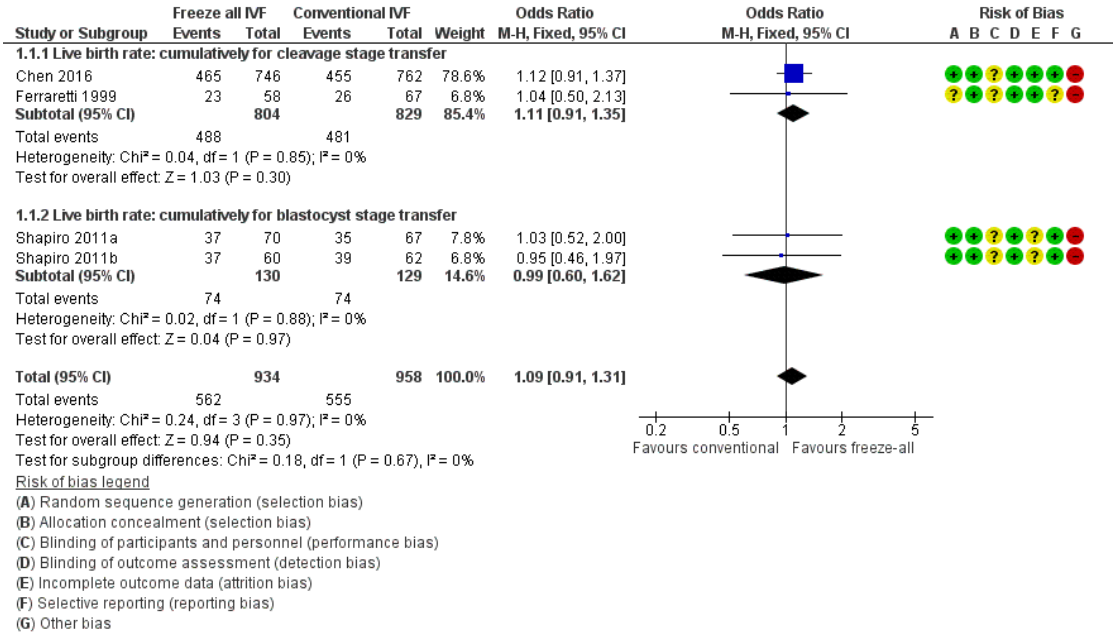
Primary outcomes

1.1 Effectiveness: Cumulative live birth per woman

All studies collected data on cumulative live birth rates (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b; Chen 2016). There was no clear evidence of a difference between the freeze-all strategy and the conventional IVF/ICSI strategy in cumulative live birth rates (OR 1.09, 95% CI 0.91 to 1.31; 4 trials; 1892 women; I^2 = 0%; moderate-quality evidence).

It was unclear whether there was any difference between the two strategies in cumulative live birth rate when the studies were analysed per cleavage stage (OR 1.11, 95% CI 0.91 to 1.35; 2 trials; 1633 women; low-quality evidence) or blastocyst transfer stage (OR 0.99, 95% CI 0.60 to 1.62; 2 trials; 259 women; low-quality evidence) (Analysis 1.1; Figure 4).

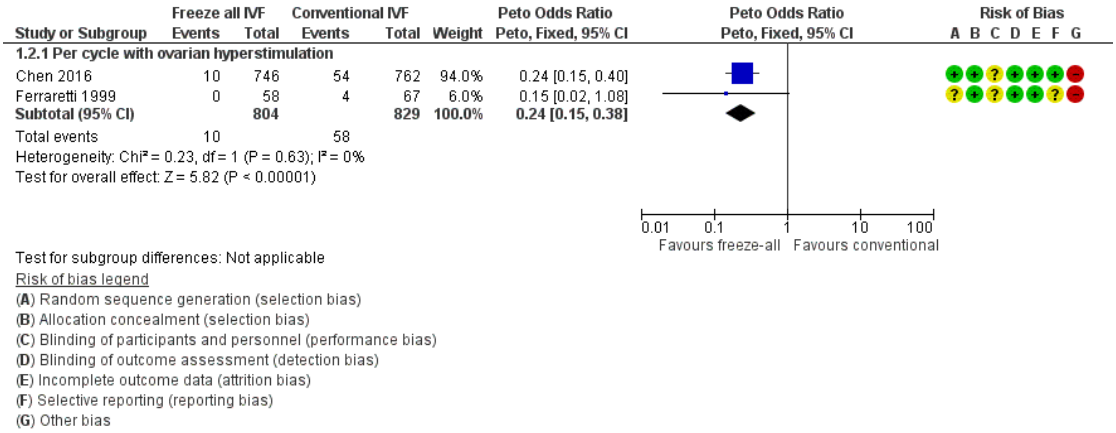
Figure 4. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.1 Live birth rate.



1.2 Safety: Ovarian hyperstimulation syndrome per woman

One study reported OHSS per woman if hospitalisation was required (Ferraretti 1999). Two studies did not report on OHSS, but we were able to obtain these data by personal communication with the authors (Shapiro 2011a; Shapiro 2011b). However, we did not include the data from these two studies in the analysis as women with high risk of OHSS were excluded and standardly received the freeze-all strategy. Chen 2016 reported these data. The prevalence of OHSS was lower after the freeze-all strategy compared to the conventional IVF/ICSI strategy (OR 0.24, 95% CI 0.15 to 0.38; 2 trials; 1633 women; I^2 = 0%; low-quality evidence) (Analysis 1.2; Figure 5).

Figure 5. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.2 OHSS.

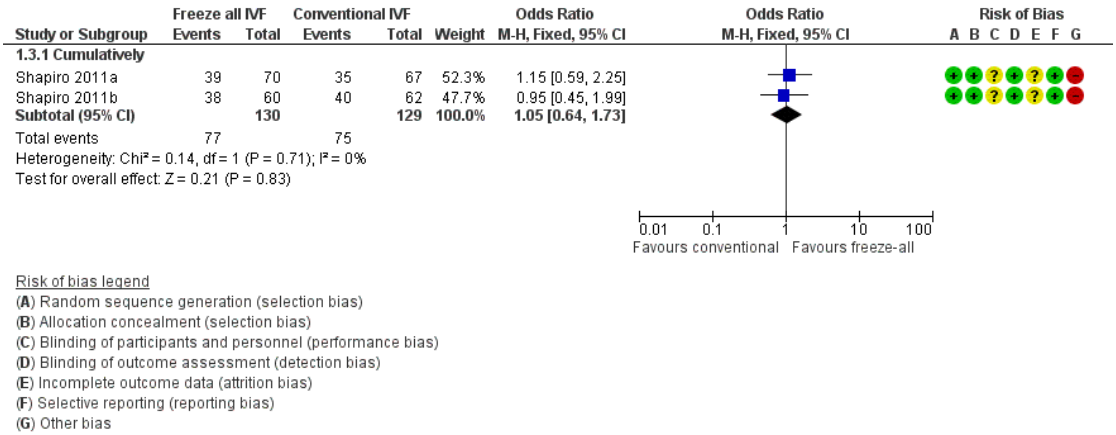


Secondary outcomes

1.3 Ongoing pregnancy rate per woman

Two studies reported on the cumulative ongoing pregnancy rates (Shapiro 2011a; Shapiro 2011b). There was no evidence of a difference between the two strategies in the cumulative ongoing pregnancy rate (OR 1.05, 95% CI 0.64 to 1.73; 2 trials; 259 women; I^2 = 0%; low-quality evidence) (Analysis 1.3; Figure 6).

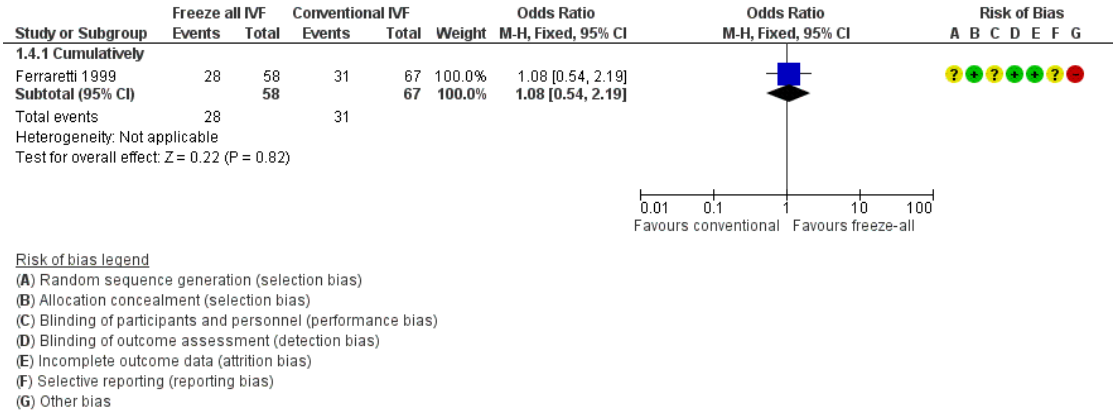
Figure 6. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.3 Ongoing pregnancy rate.



1.4 Clinical pregnancy rate per woman

One study reported the cumulative clinical pregnancy rates (Ferraretti 1999), therefore pooling was not possible. There was no evidence of a difference between the two strategies in clinical pregnancy rate (OR 1.08, 95% CI 0.54 to 2.19; 1 trial; 125 women; low-quality evidence) (Analysis 1.4; Figure 7).

Figure 7. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.4 Clinical pregnancy rate.



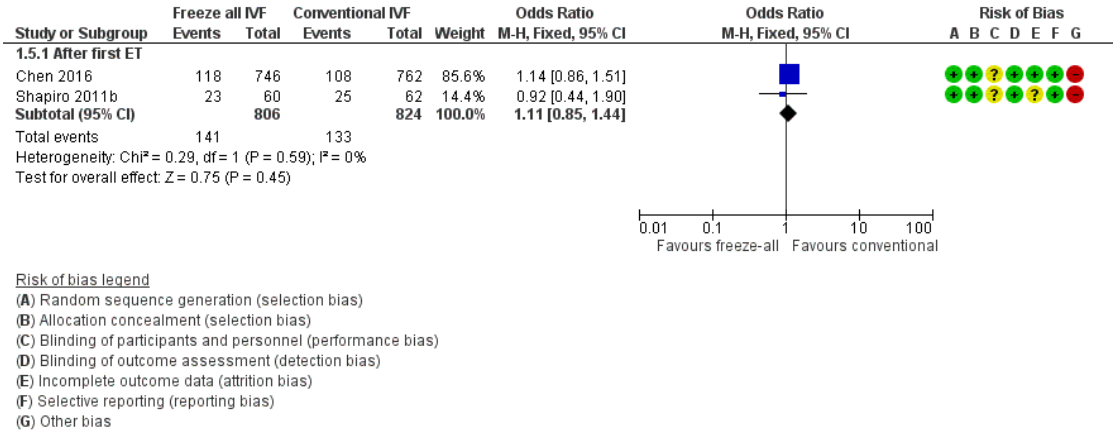
1.5 Time to pregnancy

No study reported the time to pregnancy or (separate or incremental) data per subsequent menstrual or cryo-transfer cycle (relevant for time-to-pregnancy comparison).

1.6 Multiple-pregnancy rate

Two studies reported on the multiple-pregnancy rate (Shapiro 2011b; Chen 2016). There was no evidence of a difference between the two strategies in multiple-pregnancy rate (OR 1.11, 95% CI 0.85 to 1.44; 2 trials; 1630 women; low-quality evidence) (Analysis 1.5; Figure 8).

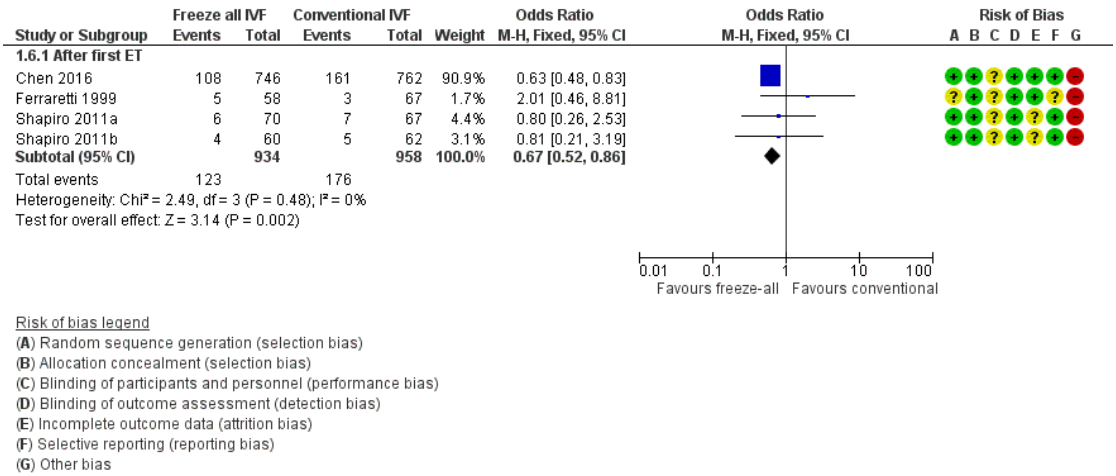
Figure 8. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.5 Multiple pregnancy rate.



1.7 Miscarriage rate

All studies reported the miscarriage rate (Ferraretti 1999; Shapiro 2011a; Shapiro 2011b; Chen 2016). Miscarriage rate was lower in the freeze-all group (OR 0.67, 95% CI 0.52 to 0.86; 4 trials; 1892 women; I^2 = 0%, low-quality evidence) (Analysis 1.6; Figure 9).

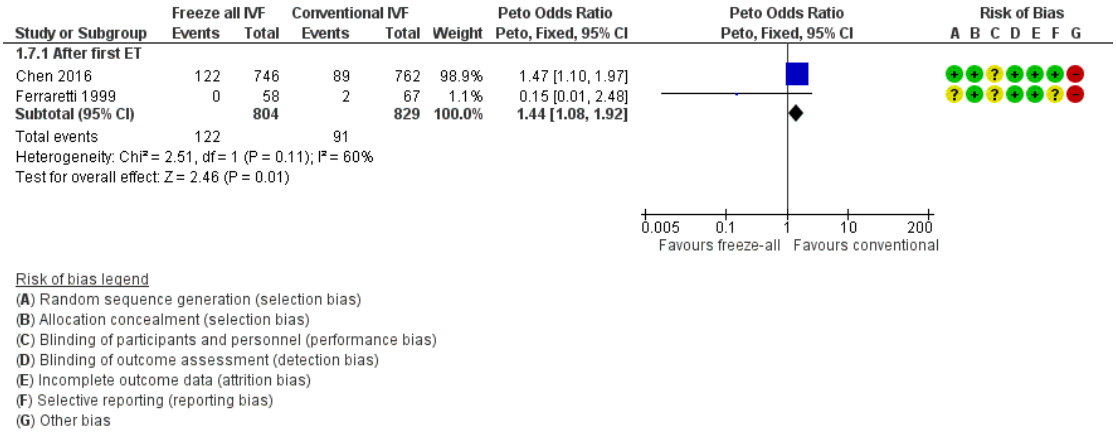
Figure 9. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.6 Miscarriage rate.



1.8 Pregnancy complications

Two studies reported on pregnancy complications (Ferraretti 1999; Chen 2016). There were more pregnancy complications in the freeze-all group (OR 1.44, 95% CI 1.08 to 1.92; 2 trials; 1633 women; low-quality evidence) (Analysis 1.7; Figure 10).

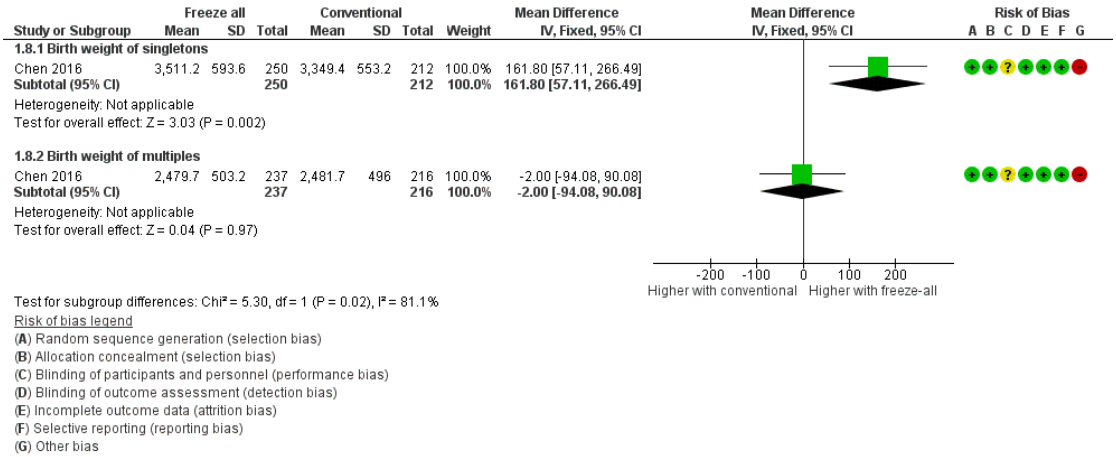
Figure 10. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.7 Pregnancy complications.



1.9 Birth weight

One study reported on birth weight (Chen 2016). A higher birth weight of singleton babies born was reported for the freeze-all strategy (MD 161.8 g, 95% CI 57.1 to 266.5; 1 trial; 462 singletons; low-quality evidence). Birth weight of multiples was similar between strategies (MD -2.00 g, 95% CI -94.08 to 90.08; 1 trial; 453 multiples; low-quality evidence) (Analysis 1.8; Figure 11).

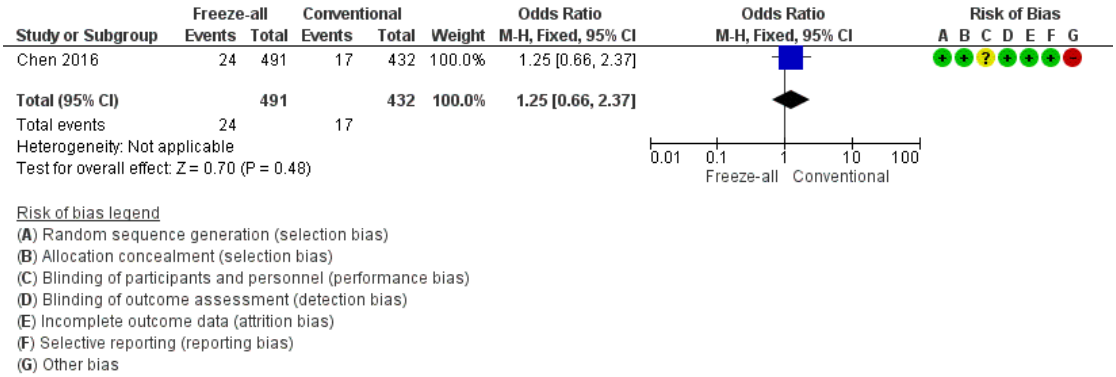
Figure 11. Forest plot of comparison: 1 Freeze-all vs conventional IVF, outcomes per woman, outcome: 1.8 Birth weight of babies born.



1.10 Congenital abnormalities

One study reported on congenital abnormalities (Chen 2016). There was no evidence of a difference between the two strategies for congenital abnormalities per live-born children plus number of fetuses therapeutically terminated (OR 1.25, 95% CI 0.66 to 2.37; 1 trial; 923 live-born children plus number of fetuses therapeutically terminated; low-quality evidence) (Analysis 3.1; Figure 12).

Figure 12. Forest plot of comparison: 3 Freeze-all vs conventional IVF, congenital abnormalities per live-born children plus number of foetuses therapeutically terminated, outcome: 3.1 Congenital abnormalities.



Other analyses

We also analysed the adverse events multiple pregnancy, miscarriage and pregnancy complications per clinical pregnancy (Analysis

2.1, Analysis 2.2, Analysis 2.3). There was no evidence of a difference between the two strategies for multiple pregnancy per clinical pregnancy after the first transfer (OR 1.02, 95% CI 0.77 to 1.37; 2 trials; 939 clinical pregnancies) (Analysis 2.1). Miscarriage rate was lower in the freeze-all group per clinical pregnancy after the first transfer (OR 0.56, 95% CI 0.41 to 0.77; 4 trials; 1058 clinical pregnancies) (Analysis 2.2).There were more pregnancy complications in the freeze-all group per clinical pregnancy after the first transfer (OR 1.43, 95% CI 1.05 to 1.95; 2 trials; 914 clinical pregnancies) (Analysis 2.3).

Sensitivity analysis

We did not undertake sensitivity analysis by risk of bias because all studies in the analyses were at high risk of bias in at least one domain. We undertook sensitivity analyses of the primary outcome using 1) adoption of a random-effects model and 2) using the summary effect measure of risk ratio (RR) rather than OR. Neither of the sensitivity analyses made any material difference to the findings (Table 1).

DISCUSSION

Summary of main results

There was no clear evidence of a difference between the freeze-all strategy and the conventional IVF/ICSI strategy in cumulative live birth rates per woman, but the prevalence of OHSS appears to be lower after the freeze-all strategy. The freeze-all strategy appears to be associated with fewer miscarriages and a higher birth weight of singleton babies (MD 161.80 g, 95% CI 57.11 to 266.49), but also with an increased rate of pregnancy complications.

Overall completeness and applicability of evidence

All trials provided data on the primary outcome live birth rate, but for OHSS we could use data from only two studies.

Three out of four included studies involved a small number of women. All studies had specific and differing technical protocols, and studies had differing inclusion criteria leading to the inclusion of select groups of women ('normal responders', 'high responders', women with polycystic ovary syndrome, women with high OHSS risk). No study included women with low ovarian response.

Quality of the evidence

We rated the quality of evidence using GRADE methods and judged it to be moderate to low, due to serious risk of bias and (for some outcomes) serious imprecision. Risk of bias was associated with unclear blinding of investigators for preliminary outcomes of the study, unit of analysis error, and absence of adequate study termination rules.

The four included studies involved a total of 934 women undergoing the freeze-all strategy and 958 women undergoing the conventional IVF/ICSI strategy. Varying protocols between studies (some not common in routine practice), varying study population (select groups of women undergoing IVF), one study without power calculation reported (unclear what determined the end of study), and two studies with interim analysis that was calculated per transfer (unit of analysis error) with absence of adequate stopping rules (possible overestimation of treatment effect) resulted in an overall judgement of the evidence as low quality.

Our searches identified 12 ongoing studies. We anticipate that the evidence from these will provide a more definitive answer on the relative effectiveness and safety of a freeze-all strategy.

Potential biases in the review process

We tried to reduce potential bias in the review process to a minimum by identifying all eligible trials for inclusion in this meta-analysis. We were able to retrieve additional information on three included trials where required, which helped us in providing accurate study outcomes.

Agreements and disagreements with other studies or reviews

Three out of four included studies reported higher pregnancy or live birth rates for the freeze-all strategy than for conventional IVF/ICSI treatment including fresh transfer in the published manuscripts (Shapiro 2011b; Shapiro 2011a; Chen 2016), while our review concluded that there was no difference in live birth rates between the strategies. This discrepancy in conclusion is attributed to the fact that these publications focussed on outcomes that were reported per first transfer, whereas in our review we focused on the cumulative live birth rate per woman randomised. The live birth rate calculated per first transfer possibly shows differences in outcome for a stimulated and an unstimulated uterus, although this does not take into account the number of embryos that were thawed for transfer. But for women undergoing treatment, the live birth rate per first transfer is less relevant since at the same time of first transfer in a freeze-all strategy, they would already have received the second transfer (in case of sufficient number of embryos) in a conventional strategy including fresh transfer. We therefore used the cumulative live birth rate as a primary outcome. In case cumulative live birth rates are comparable, as found in our review, then the difference between strategies could be time to pregnancy. Unfortunately, this outcome was not reported in any of the included studies, but by design time to pregnancy is

shorter in the conventional strategy than in the freeze-all strategy when the cumulative live birth rate is comparable. For illustrative purposes we also calculated and presented the live birth rate per first transfer (Table 2).

The same difference in primary outcome (cumulative live birth rate versus live birth rate per first transfer) explains the difference from previous reviews that found improved IVF/ICSI outcomes with the freeze-all strategy, such as Roque 2013, although the included studies also differed. Roque and colleagues did not include the study of Ferraretti 1999 in their analysis, for reasons that are unclear. The authors did include the retracted study of Aflatoonian 2010 in their analysis. The Chen 2016 study was not yet published when this review was written.

Although we reported pregnancy and live birth rates only cumulatively for the above reasons, for other outcomes, such as the number of multiples and the number of miscarriages, we did report the numbers per first transfer, as cumulative rates for these outcomes were not available from any of the studies.

The lower rate of OHSS found in our review is in agreement with previous studies, and is to be expected, as avoiding a pregnancy in the initial cycle with ovarian stimulation by only transferring frozen-thawed embryos in subsequent unstimulated cycles would eliminate the residual risks of OHSS, and OHSS would therefore be self limiting. Mild OHSS symptoms can still occur as a result of the hCG trigger in the hyperstimulated cycle in the freeze-all strategy, but OHSS in its severe form should be rare.

Implications for practice

Moderate-quality evidence does not show one strategy to be superior to the other in terms of cumulative live birth rates. Time to pregnancy was not reported, but can be assumed to be shorter using a conventional IVF/ICSI strategy in case of similar cumulative live birth rates, as embryo transfer is delayed in a freeze-all strategy. Low-quality evidence suggests that not performing a fresh transfer lowers the OHSS risk for women at risk of OHSS.

Implications for research

Well-designed RCTs reporting on cumulative live birth rate and OHSS per hyperstimulated cycle are required. Participant characteristics (e.g. women with good prognosis versus poor prognosis), treatment characteristics (e.g. number of available embryos, number of embryos transferred, results for first and every subsequent transfer, time to pregnancy), and protocols used (e.g. timing and method of cryopreservation) should be properly reported, as these are relevant for future meta-analyses. Subanalyses of RCTs with data on freezing and transferring frozen-thawed embryos at the same developmental phase as in the conventional IVF/ICSI strategy would be a better way to compare the two strategies. These RCTs should be performed with well-described randomisation and allocation concealment methods, and should include intention-to-treat analyses. Outcome measures should be expressed as cumulative live birth rate per woman rather than per first transfer.

AUTHORS' CONCLUSIONS

ACKNOWLEDGEMENTS

We would like to acknowledge the team at the Cochrane Gynaecology and Fertility Group for their assistance, and especially Information Specialist Marian Showell for the literature search.

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NCT02148393 *{unpublished data only}*
NCT02148393. Implantation enhancement by elective cryopreservation of all viable embryos (ICE). clinicaltrials.gov/ct2/show/NCT02148393 (first received 16 May 2014).

NCT02471573 *{unpublished data only}*
NCT02471573. Freeze all protocol versus fresh embryo transfer in women undergoing in-vitro fertilization (IVF). clinicaltrials.gov/ct2/show/NCT02471573 (first received 11 June 2015).

NCT02570386 *{unpublished data only}*
NCT02570386. Clinical effectiveness of frozen thawed embryo transfer compared to fresh embryo transfer. clinicaltrials.gov/ct2/show/record/NCT02570386 (first received 29 September 2015).

NCT02681367 *{unpublished data only}*
NCT02681367. Management of recurrent implantation failure (RIF). clinicaltrials.gov/ct2/show/NCT02681367 (first received 8 February 2016).

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NCT02712840. Comparing pregnancy outcomes in good prognosis patients between fresh and 'freeze-all' single blastocyst transfers. clinicaltrials.gov/ct2/show/NCT02712840 (first received 9 September 2015).

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* *Indicates the major publication for the study*

CHARACTERISTICS OF STUDIES

Characteristics of included studies *[ordered by study ID]*

Chen 2016

Methods	Multicentre randomised controlled trial 14 reproductive medical centres throughout China Enrolment: June 2013 to May 2014 Power calculation: stated Randomisation: an online central randomisation system (www.medresman.org) was used. Nature of intervention: day 3 embryo cryopreservation by means of vitrification. Local investigators had the option to transfer day 2 embryos if there were fewer than 3 embryos on day 2 Follow-up: cumulative live birth (including all frozen-embryo transfers performed within 12 months after the initial transfer)
Participants	1508 women (746 freeze-all, 762 control) Inclusion criteria: • Polycystic ovary syndrome, using modified Rotterdam criteria (which included menstrual abnormalities (irregular uterine bleeding, oligomenorrhoea, or amenorrhoea) combined with either hyperandrogenism or polycystic ovaries) • First IVF cycle Exclusion criteria: history of unilateral oophorectomy, recurrent spontaneous abortion (defined as 3 or more previous spontaneous pregnancy losses), congenital or acquired uterine malformations, abnormal results on parental karyotyping, or medical conditions that contraindicated assisted reproductive technology or pregnancy
Interventions	For women who were assigned to the fresh embryo group, on day 3, 2 high-quality embryos were picked out for fresh transfer and supernumerary embryos were transferred by means of vitrification For women who were assigned to the frozen embryo group, there was no fresh transfer as all day 3 embryos were cryopreserved for later transfer. Local investigators had the option to transfer day 2 embryos if there were fewer than 3 embryos on day 2. In cycles following the menstrual cycle with ovum pickup, on day 4 of the progesterone regimen, 2 day 3 frozen embryos were thawed and transferred
Outcomes	Primary outcome was a live birth, defined as delivery of any viable infant at 28 weeks or more of gestation during the first embryo transfer. Prespecified secondary outcomes included biochemical pregnancy, clinical pregnancy, ongoing pregnancy, singleton live birth, cumulative live birth (including subsequent frozen embryo transfer), pregnancy loss, moderate or severe OHSS, ectopic pregnancy, pregnancy and neonatal complications, and congenital anomalies
Notes	

Risk of bias

Bias	Authors' judgement	Support for judgement

Chen 2016 (*Continued*)

Random sequence generation (selection bias)	Low risk	An online central randomisation system (www.medresman.org) was used to automatically generate the assignment sequence
Allocation concealment (selection bias)	Low risk	Assignment sequence was unknown to the clinical investigators
Blinding of participants and personnel (performance bias) All outcomes	Unclear risk	Blinding of doctors and participants was not possible due to the nature of the intervention. Blinding of doctors to interim analyses of outcomes of the study was not reported. Blinding of investigators was not reported (which is relevant for determining end of study), therefore judged to be unclear risk
Blinding of outcome assessment (detection bias) All outcomes	Low risk	Outcome assessor blinding was not reported, however primary outcome is not likely to be influenced by lack of blinding
Incomplete outcome data (attrition bias) All outcomes	Low risk	Data were analysed for all randomised women.
Selective reporting (reporting bias)	Low risk	All registered outcomes reported.
Other bias	High risk	(Cumulative) data per subsequent menstrual or cryo-transfer cycle not reported (relevant for time-to-pregnancy comparison and the related comparison of results after first transfer in frozen group vs results after first 2 transfers in fresh group)

Ferraretti 1999

Methods	Single-centre randomised controlled trial Conducted: in Italy, from January 1996 until July 1997 Power calculation: not reported Randomisation: allocation was performed with sealed envelopes, timing of randomisation was not reported Nature of intervention: slow freezing Follow-up: until no cryopreserved embryos were left or delivery of child
Participants	125 women (58 freeze-all, 67 control) Inclusion criteria: all women with a high level of oestradiol the day of hCG administration (oestradiol ≥ 1500 pg/mL or ≥ 5.500 mmol/mL (conversion factor to SI unit 53.671)) and a high number of retrieved eggs (≥ 15 oocytes)

Ferraretti 1999 (*Continued*)

Interventions	Intervention: zygotes were cryopreserved, 3 or 4 zygotes were thawed and cultured for 36 to 40 h before embryo transfer. If 2 or more zygotes did not cleave 24 h after being cultured, 1 or 2 additional zygotes were thawed Control: zygotes were cultured for a subsequent 48 h, 3 or 4 fresh embryos were transferred, surplus embryos were cryopreserved
Outcomes	Clinical pregnancies: gestational sac and foetal heartbeat by ultrasound
Notes	Funding was not reported. Additional information was obtained from the authors by email

Risk of bias

Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Unclear risk	Random sequence was used, but it is unclear whether envelopes were opaque and sequentially numbered
Allocation concealment (selection bias)	Low risk	Allocation concealment was performed with sealed envelopes.
Blinding of participants and personnel (performance bias) All outcomes	Unclear risk	Blinding of doctors and participants was not possible due to the nature of the intervention. Blinding of doctors to interim analyses of outcomes of the study was not reported. Blinding of investigators was not reported (which is relevant for determining end of study), therefore judged to be unclear risk
Blinding of outcome assessment (detection bias) All outcomes	Low risk	Outcome assessor blinding was not reported, however primary outcome is not likely to be influenced by lack of blinding
Incomplete outcome data (attrition bias) All outcomes	Low risk	Data were analysed for all randomised women.
Selective reporting (reporting bias)	Unclear risk	No protocol available.
Other bias	High risk	No power calculation reported. Unclear what determined the end of study (Cumulative) data per subsequent menstrual or cryo-transfer cycle not reported (relevant for time-to-pregnancy comparison and the related comparison of results after first transfer in frozen group vs results after first 2 transfers in fresh group)

Shapiro 2011a

Methods	Single-centre randomised controlled trial Conducted: in the United States, from October 2007 until October 2010 Power calculation: stated. However, study was prematurely terminated after interim analysis Randomisation: performed after retrieval by random drawing among identical, opaque, unmarked sealed envelopes Nature of intervention: slow freezing Follow-up: clinical pregnancy after first embryo transfer
Participants	137 women (70 freeze-all, 67 fresh transfer) Inclusion criteria: • Women must be undergoing her first IVF cycle • Cycle day 3 FSH < 10 IU/L • 8 to 15 antral follicles observed on baseline ultrasound scan Exclusion criteria: genetic testing of embryos was excluded.
Interventions	Intervention: 2pn oocytes were frozen, and entire cohorts of frozen 2pn oocytes were thawed and subsequently cultured to the blastocyst stage. The morphologically best 1 or 2 blastocysts were transferred on the first day on which at least 1 good expanded blastocyst appeared. Supernumerary expanded blastocysts of high quality were cryopreserved Control: fresh blastocysts transfer
Outcomes	• Pregnancy: serum hCG levels within 10 days after blastocyst transfer • Clinical pregnancy: foetal heart motion at 7 weeks' gestation • Ongoing pregnancy: foetal heart motion at 10 weeks' gestation • Implantation rate: proportion of transferred blastocysts that resulted in foetal heart motion (monozygotic twins with foetal heart motion counted as single implantations) • Early pregnancy losses: pregnancies that did not become ongoing pregnancies
Notes	Funding: research grant from the Investigator-Initatiated trial research grant from Ferring Pharmaceuticals, Parsippany, NJ. Medications for this study were also provided by Ferring Pharmaceuticals Time period was obtained from trial register. Additional information was obtained from authors by email.

Risk of bias

Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Drawing randomly among identical, opaque, unmarked, sealed envelopes
Allocation concealment (selection bias)	Low risk	Drawing randomly among identical, opaque, unmarked, sealed envelopes
Blinding of participants and personnel (performance bias) All outcomes	Unclear risk	Blinding of doctors and participants was not possible due to the nature of the intervention. Blinding of doctors to interim

Shapiro 2011a (*Continued*)

		analyses of outcomes of the study was not reported. Blinding of investigators was not reported (which is relevant for determining end of study), therefore judged to be unclear risk
Blinding of outcome assessment (detection bias) All outcomes	Low risk	Outcome assessor blinding was not reported, however primary outcome is not likely to be influenced by lack of blinding
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Data were not reported for all women randomised, but per transfer. Dropouts and loss to follow-up were not accounted for in the analysis. No ITT analysis was performed. Sufficient data available for analysis per woman in meta-analysis. Ongoing pregnancy was determined at 10 weeks' gestation instead of 12 weeks' gestation
Selective reporting (reporting bias)	Low risk	All registered outcomes reported.
Other bias	High risk	Trial was pre-terminated after interim analysis. Interim analysis was preplanned, but calculated per transfer (unit of analysis error) with a P value of 0.03, overestimating possible effects (Cumulative) data per subsequent menstrual or cryo-transfer cycle not reported (relevant for time-to-pregnancy comparison and the related comparison of results after first transfer in frozen group vs results after first 2 transfers in fresh group)

Shapiro 2011b

Methods	Single-centre randomised controlled trial Conducted: in the United States, from July 2007 until July 2010 Power calculation: Power calculation: stated (referred to Shapiro 2011a). However, study was terminated because of differing embryo quality between the 2 groups Randomisation: performed after retrieval by random drawing among identical, opaque, unmarked, sealed envelopes Nature of intervention: slow freezing Follow-up: clinical pregnancy after 1 embryo transfer
Participants	122 women (60 freeze-all, 62 control) Inclusion criteria: • First cycle • Cycle day 3 FSH < 10 IU/L

Shapiro 2011b (*Continued*)

	• > 15 antral follicles observed on baseline ultrasound examination Exclusion criteria: genetic testing of embryos was excluded.
Interventions	Intervention: 2pn oocytes were frozen, and entire cohorts of frozen 2pn oocytes were thawed and subsequently cultured to the blastocyst stage. The morphologically best 1 or 2 blastocysts were transferred on the first day on which at least 1 good expanded blastocyst appeared. Supernumerary expanded blastocysts of high quality were cryopreserved Control: fresh blastocysts transfer
Outcomes	• Pregnancy: serum hCG levels within 10 days after blastocyst transfer • Clinical pregnancy: foetal heart motion at 6 to 7 weeks' gestation • Ongoing pregnancy: foetal heart motion at 10 weeks' gestation • Implantation rate: ratio of the number of observed foetal hearts to the number of transferred blastocysts • Early pregnancy losses: pregnancies that did not become ongoing pregnancies
Notes	Funding: research grant from the Investigator-Initatiated Studies Program of Merck Sharp & Dohme Time period was obtained from trial register. Additional information was obtained from authors by email.

Risk of bias

Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	Drawing randomly among identical, opaque, unmarked, sealed envelopes
Allocation concealment (selection bias)	Low risk	Drawing randomly among identical, opaque, unmarked, sealed envelopes
Blinding of participants and personnel (performance bias) All outcomes	Unclear risk	Blinding of doctors and participants was not possible due to the nature of the intervention. Blinding of doctors to interim analyses of outcomes of the study was not reported. Blinding of investigators was not reported (which is relevant for determining end of study), therefore judged to be unclear risk
Blinding of outcome assessment (detection bias) All outcomes	Low risk	Outcome assessor blinding was not reported, however primary outcome is not likely to be influenced by lack of blinding
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Data were not reported for all women randomised, but per transfer. Dropouts and loss to follow-up were not accounted for in the analysis. No ITT analysis was performed. Sufficient data available for anal-

Shapiro 2011b (*Continued*)

		ysis per woman in meta-analysis. Ongoing pregnancy was determined at 10 weeks' gestation instead of 12 weeks' gestation
Selective reporting (reporting bias)	Low risk	All registered outcomes reported.
Other bias	High risk	Trial was pre-terminated after interim analysis. Interim analysis was preplanned, but calculated per transfer (unit of analysis error) with a P value of 0.03, overestimating possible effects. Stopping rules for interim analysis (embryo quality) were unclear (Cumulative) data per subsequent menstrual or cryo-transfer cycle not reported (relevant for time-to-pregnancy comparison and the related comparison of results after first transfer in frozen group vs results after first 2 transfers in fresh group)

2pn: 2 pro-nucleate
FSH: follicle-stimulating hormone
hCG: human chorionic gonadotropin
ITT: intention-to-treat
IVF: in vitro fertilisation
OHSS: ovarian hyperstimulation syndrome

Characteristics of excluded studies *[ordered by study ID]*

Study	Reason for exclusion
Absalan 2013	Unclear from report whether trial was a RCT, and the authors did not respond to our request for further information
Aflatoonian 2010	This article has been retracted from the literature at the request of the Editor and the ASRM Publications Committee
Boostanfar 2016	Randomised a different intervention
Yang 2015	One-third of participants chose to be in group 3 after randomisation. Not considered a properly randomised RCT

ASRM: American Society for Reproductive Medicine
RCT: randomised controlled trial

Characteristics of studies awaiting assessment *[ordered by study ID]*

Chandel 2016

Methods	Single-centre randomised controlled trial Conducted: in India from September 2013 until September 2014 Power calculation: not reported Randomisation: computer-based sequence generation, remote allocation Nature of intervention: day 3 frozen embryo transfer (FET), using rapid freezing Follow-up: unclear (see notes)
Participants	Inclusion criteria: • Inferrtile women • Male factor infertility, testicular sperm aspiration • Women who had developed ovarian hyperstimulation syndrome (OHSS) in a previous in vitro fertilisation cycle • Women known to be at high risk of OHSS • All women with ≥ 2 stimulated eggs/follicles, with E2 ≥ 2000 Exclusion criteria: • Donor embryos • Poor responders with < 4 stimulated follicles • Previous history of uterine curettage, endocrine disorders (diabetes mellitus, hypothyroidism) • Embryo transfer performed in a natural cycle
Interventions	1. Day 3 FET 2. Day 3 fresh embryo transfer Controlled ovarian stimulation was achieved mainly using the gonadotropin-releasing hormone (GnRH) antagonist for pituitary suppression and recombinant follicle-stimulating hormone. Women underwent pituitary desensitisation with the use of GnRH antagonist. Immediately after the ovum pick-up, intracytoplasmic sperm injection was performed for all the oocytes. The day 3 embryos were either transferred in the same cycle or were frozen using vitrification technique and transferred in the next cycle
Outcomes	Conception/pregnancy: definition unclear
Notes	Attempts to contact authors unsuccessful to date (January 2017) Unclear whether follow-up continued until a live birth occurred or until all embryos from the initial cycle were transferred Poor reporting of results: both groups labelled as FET in study tables, no clear definition of pregnancy

Characteristics of ongoing studies *[ordered by study ID]*

ACTRN12612000422820

Trial name or title	A randomized study of IVF patients to assess whether freezing all of the embryos and transferring them in a later natural, unstimulated cycle results in a higher pregnancy rate than transferring an embryo 5 days after egg collection
Methods	RCT Target enrolment: 200

ACTRN12612000422820 (*Continued*)

Participants	Included: • Females of infertile couples for whom controlled ovarian stimulation and IVF with or without ICSI is indicated • Age 20 to 38 years at the time of screening, regular menstrual cycles with a range of 24 to 33 days, BMI 18 to 28, AMH 5 to 20 Excluded: • Previous IVF treatment cycle that resulted in < 6 follicles on day 8 ultrasound • More than 2 previous unsuccessful stimulated cycles • History of or current endocrine abnormality such as polycystic ovary syndrome or evidence of ovarian dysfunction • Any clinically significant abnormal laboratory value (TSH, PRL, SHBG test) • Any ovarian or abdominal abnormality, or both, that would interfere with adequate ultrasound investigation of at least 1 ovary • Only 1 ovary • Contraindications for the use of gonadotropins • Alcohol or drug abuse, or history thereof, within the 12 months preceding signing informed consent • Smokers
Interventions	Both study groups will undertake a stimulated IVF cycle. The first (intervention) group will have all embryos cryostored electively for transfer in a later natural menstrual cycle. The second group will have the best-quality embryo transferred to the endometrial cavity fresh and all remaining embryos cryostored. The protocol for the second group is standard practice today. Both groups will undertake the same drug regimen, therefore there is no difference in drug intervention
Outcomes	• Live birth • Cumulative clinical pregnancy: a foetal heartbeat seen on ultrasound at 7 weeks • Perinatal complications • Blastulation anomalies
Starting date	1 May.2012
Contact information	Mark Livingstone: ecosse@ihug.com.au
Notes	www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=362361

ACTRN12616000643471

Trial name or title	Comparison of the probability of live birth after elective freezing of all embryos versus standard fresh embryo transfer in patients undergoing in-vitro fertilisation (IVF)
Methods	RCT Target enrolment: 400
Participants	Women aged 18 to 39 with indication for COS and IVF or ICSI with autologous gametes Key inclusion criteria: • Age: 18 to 39 years

ACTRN12616000643471 (*Continued*)

	• BMI: 18 to 32 kg/m^2 • Presence of both ovaries • Normal menstruating cycles: 21 to 35 days • Cycle where prevention of premature LH rise is achieved using a GnRH antagonist • 8 to 19 follicles ≥ than 10 mm in mean diameter on the day of triggering Key exclusion criteria: • Endometriosis stage > II • Indication for PGD/PGS • History of OHSS • Previous participation in the RCT • > 3 previous unsuccessful stimulated cycles • History of hypothalamic dysfunction or history of inadequate pituitary response to GnRH agonist triggering
Interventions	
Outcomes	• Live birth after the transfer of the first embryo: delivery of a live baby after the 20th week of gestation • Ongoing pregnancy diagnosed by ultrasonography as presence of foetal heart activity at 10 to 12 weeks of gestation • Clinical pregnancy diagnosed by ultrasound as presence of foetal heart activity at 6 to 8 weeks of gestation • First trimester miscarriage, defined as a biochemical pregnancy (assessed by serum hCG) at 11 to 16 days after embryo transfer but no foetal heart activity at 10 to 12 weeks of gestation as assessed by ultrasonography • Occurrence of severe OHSS • Preterm labour (defined as delivery < 37 weeks of gestation) • Mode of delivery (normal vaginal delivery, assisted vaginal delivery, Caesarean section) • Neonatal birth weight • Stillbirth • Neonatal mortality • Death within the first 28 days of life • Intrauterine growth restriction • Hypertensive disorders of pregnancy (including gestational hypertension, pre-eclampsia, eclampsia) • Gestational diabetes mellitus
Starting date	May 2016
Contact information	Christos Venetis: c.venetis@unsw.edu.au
Notes	www.anzctr.org.au/Trial/Registration/TrialReview.aspx?ACTRN=12616000643471

ISRCTN61225414

Trial name or title	Freezing of embryos in assisted conception: a randomized controlled trial evaluating the clinical and cost-effectiveness of a policy of freezing embryos followed by thawed frozen embryo transfer, compared with a policy of fresh embryo transfer in women undergoing in-vitro fertilization
Methods	RCT Estimated enrolment: 1086

ISRCTN61225414 (*Continued*)

Participants	Inclusion criteria: • Female partner is between 18 and 42 years of age at the start of treatment (i.e. start of ovarian stimulation) • Couples who are undergoing their first cycle of IVF/ICSI treatment • Both partners are resident in the United Kingdom • Both partners are able to provide written informed consent Exclusion criteria: Couples in whom: • donor gametes are used; • pre-implantation genetic diagnosis is performed; • elective freezing of all embryos is preferred or clinically indicated (e.g. severe risk of OHSS).
Interventions	Intervention arm: All good-quality embryos will be frozen and couples will undergo frozen-thawed embryo transfer within 3 months of the egg collection process. Couples will attend a clinic visit and additional monitoring visits before frozen embryo transfer is performed Standard-care arm: Women will undergo fresh embryo transfer on day 3 or 5 (after egg collection)
Outcomes	• Live birth • Clinical pregnancy: ultrasonic visualisation of 1 or more gestational sacs or definitive clinical signs of pregnancy; ectopic counts as clinical pregnancy; multiple gestational sacs count as 1 clinical pregnancy • Ongoing pregnancy (pregnancy with presence of foetal heartbeat) • OHSS • Miscarriage rate • Gestational diabetes mellitus • Multiple pregnancy • Hypertensive disorders of pregnancy (comprising pregnancy-induced hypertension, pre-eclampsia and eclampsia) • Antepartum haemorrhage • Preterm delivery (defined as delivery at < 37 completed weeks) • Very preterm delivery (defined as delivery at < 32 completed weeks) • Low birth weight (defined as weight < 2500 g at birth) • Very low birth weight (defined as < 1500 g at birth) • Large for gestational age (defined as birth weight > 90th centile for gestation, based on standardised charts) • Small for gestational age (defined as < 10th centile for gestational age at delivery) • Congenital anomaly (all congenital anomalies identified will be included) • Perinatal mortality (late as well as early neonatal deaths, up to 28 days after birth)
Starting date	1 March 2015
Contact information	Christina Cole: christina.cole@npeu.oxa.c.uk
Notes	www.isrctn.com/ISRCTN61225414

NCT02000349

Trial name or title	Comparison of frozen-thawed embryo transfers and fresh embryo transfers with whole chromosome analysis using next generation sequencing
Methods	RCT Estimated enrolment: 186
Participants	Women aged 18 to 42 Inclusion criteria (pre-stimulation): • Age up to 42 years Exclusion criteria (pre-stimulation): • MESA and TESE patients • At least 1 partner carrier of a chromosomal abnormality • Egg donor cycle (sperm donor is acceptable) • Gender selection cycles • Thaw cycles • Any woman who cannot have a fresh embryo transfer • FSH above 12 or AMH less than 1
Interventions	Frozen embryo transfer with PGD: All embryos will be hatched on day 3. Women will have hatching blastocysts* biopsied on day 5 or day 6, embryos will then be vitrified, analysed by NGS, and women will have 1 or 2 euploid embryo(s) thawed and transferred on a FET cycle, before noon. *If more than 2 euploid blastocysts are available, the one(s) to be transferred will be selected based on morphology Fresh embryo transfer with PGD: All embryos will be hatched on day 3. Women will have hatching blastocysts* biopsied on day 5, analysed by NGS, and will have 1 or 2 euploid embryo(s) transferred on day 6, in the a.m. Any morulas developing to hatching blastocyst on day 6 will be also analysed but vitrified for use in a future cycle. *If more than 2 euploid blastocysts are available, the one(s) to be transferred will be selected based on morphology
Outcomes	• Implantation rate • Correlation of mitochondrial DNA and implantation
Starting date	September 2013
Contact information	Study director: Santiago Munne, Reprogenetics
Notes	clinicaltrials.gov/ct2/show/NCT02000349

NCT02133950

Trial name or title	Efficacy study of segmentation of PGD treatment
Methods	RCT Estimated enrolment: 240
Participants	Women aged 20 to 40 Inclusion criteria: • 1st, 2nd, or 3rd cycle of PGD in which embryo transfer was performed • Indications for PGD: monogenic indications and X-linked disorders with a 25% to 50% risk of transmission and that are not associated with reduced ovarian response

NCT02133950 (*Continued*)

	• Normal ultrasound scan, i.e. presence of both ovaries, without evidence of abnormality within 6 months prior to randomisation • Regular menstrual cycles of 21 to 35 days, presumed to be ovulatory Exclusion criteria: • Polycystic ovary syndrome (Rotterdam criteria) • Poor responders (Bologna criteria) • Endocrine or metabolic abnormalities (pituitary, adrenal, pancreas, liver, or kidney) • Anticipated high response: AMH > 5.0 ng/mL or AFC > 20 • Endometriosis ≥ grade 3 • Age > 40 years and 364 days
Interventions	Elective cryopreservation of available embryos after PGD PGD and elective fresh embryo transfer plus cryopreservation of supernumerary available embryos after PGD
Outcomes	Cumulative live birth rate of a single PGD treatment
Starting date	May 2014
Contact information	Willem Verpoest, Centre for Reproductive Medicine UZ Brussel
Notes	

NCT02148393

Trial name or title	Implantation enhancement by elective cryopreservation of all viable embryos (ICE)
Methods	RCT Estimated enrolment: 212
Participants	Women aged 18 to 40 Inclusion criteria: • First or second IVF/ICSI cycle • High response to ovarian stimulation (defined as presence of ≥ 18 follicles of ≥ 11 mm on the day of GnRH triggering) • GnRH antagonist down-regulation • Signed informed consent • Women can be included only once in the trial • Planned replacement of 1 or 2 blastocysts Exclusion criteria: • Other known reasons for impaired implantation (i.e. hydrosalpinx, fibroid distorting the endometrial cavity, Asherman's syndrome, thrombophilia, or endometrial tuberculosis) • Oocyte/embryos donation acceptors • Embryos planned to undergo pre-implantation genetic diagnosis/screening • BMI ≥ 35 or ≤ 18 • Women who have previously enrolled in the trial • Those unable to comprehend the investigational nature of the proposed study

NCT02148393 (*Continued*)

Interventions	Intervention: Elective vitrification with subsequent-cycle embryo thawing/transfer (Cryo Bio System) will be performed. Hence, no luteal-phase support will be provided immediately after oocyte retrieval. Instead, women will wait for a subsequent cycle before starting exogenous hormone therapy for endometrial preparation. On the day of embryo transfer, blastocyst(s) will be warmed one by one until 1 or 2 blastocysts are suitable for transfer. Embryo transfer to the uterine cavity will be performed under ultrasound guidance whenever possible Control: Following oocyte retrieval, intensified luteal-phase support for fresh embryo transfer (with hCG (Pregnyl), progesterone (Utrogestan), and oestradiol valerate (Progynova)) will be performed. Fresh embryo transfer in the uterine cavity will be performed on the 5th day of embryo development at blastocyst stage under ultrasound guidance whenever possible
Outcomes	• Clinical pregnancy rate • OHSS incidence
Starting date	May 2014
Contact information	Samuel Santos-Ribeiro: samuel.ribeiro@uzbrussel.be
Notes	clinicaltrials.gov/ct2/show/NCT02148393

NCT02471573

Trial name or title	Freeze all protocol versus fresh embryo transfer in women undergoing in-vitro fertilization (IVF)
Methods	RCT Estimated enrolment: 780
Participants	Women aged 18 to 42 Inclusion criteria: • Undergoing IVF treatment • Number of previously failed embryo transfers ≤ 2 • Permanent living in Vietnam • Ovarian hyperstimulation with GnRH antagonist protocol • Eligible for embryo transfer on day 3 • Having at least 1 top-quality embryo on day 3 • Number of embryos transferred ≤ 2 • Willing to participate in the study • Not concurrently participating in another IVF study Exclusion criteria: • In vitro maturation cycles • Oocyte donation cycles • Using GnRH agonist for triggering
Interventions	Freeze-all protocol: Embryos are selected for cryopreservation using vitrification technique. 2 vitrified embryos will be warmed and transferred in subsequent cycle Fresh transfer protocol: 2 embryos are selected and transferred fresh in the same cycle

NCT02471573 (*Continued*)

Outcomes	• Live birth: a live newborn delivered • Ongoing pregnancy: a pregnancy with positive heartbeat beyond 12 weeks of gestation • Clinical pregnancy: presence of a gestational sac seen by transvaginal sonography 7 weeks after embryo placement • Multiple pregnancy: 2 or more foetal heart rates by transvaginal sonography 7 weeks after embryo placement
Starting date	June 2015
Contact information	Lan TN Vuong: drlan@yahoo.com.vn Vinh Q Dang: bsvinh.dq@myduchospital.vn
Notes	clinicaltrials.gov/ct2/show/record/NCT02471573

NCT02570386

Trial name or title	Clinical effectiveness of frozen thawed embryo transfer compared to fresh embryo transfer
Methods	RCT Estimated enrolment: 800
Participants	Women aged 18 to 42 Inclusion criteria: • Women under 42 years of age • Presence of at least 3 embryos suitable to freeze on day 2 or 3 following fertilisation based on the centre's criteria • Written informed consent Exclusion criteria: • Women using donor eggs/donor sperm • Women undergoing pre-implantation genetic diagnosis • Women with abnormal uterine cavity shown on hysterosalpingogram or saline infusion sonogram • Women with hydrosalpinges shown on scanning and not treated • Women with excessive ovarian response at risk of ovarian hyperstimulation where elective freeze is already planned • Women with serum progesterone level on day of hCG > 1.5 ng/mL or 5 nmol/L • Women whose embryos have previously not survived freeze-thawing • Fresh transfer is planned, e.g. women with endometriosis or adenomyosis who have received prolonged down-regulation • Only frozen transfer is planned, e.g. women receiving ovarian stimulation regimens that may adversely impact the endometrium
Interventions	Intervention: Fresh embryo transfer will not be undertaken in this group. Embryos will be frozen by vitrification or slow freezing at cleavage or blastocyst stage according to standard agreed local protocols. Women will be contacted after 4 weeks and arrangements made for frozen embryo transfer Control: Women allocated to the control arm will either undergo fresh embryo transfer at cleavage stage or extended culture and transfer at blastocyst stage according to local policy. A maximum of 2 embryos or blastocysts will be replaced according to the standard protocol under transabdominal ultrasound guidance. Luteal-phase support is given according to local protocols

NCT02570386 (*Continued*)

Outcomes	• Cumulative live birth: within 6 months of ovarian stimulation from the fresh and frozen-thawed embryo transfer • Live birth: a baby born alive after 20 weeks' gestation • Miscarriage: miscarriage before 20 weeks' gestation • Clinical pregnancy: presence of at least 1 gestational sac on ultrasound at 6 weeks • Ovarian hyperstimulation: classified according to Royal College of Obstetrics and Gynaecology in the United Kingdom • Complications of pregnancy
Starting date	October 2015
Contact information	Ernest HY Ng: nghye@hku.hk
Notes	clinicaltrials.gov/ct2/show/record/NCT02570386

NCT02681367

Trial name or title	Management of recurrent implantation failure (RIF)
Methods	RCT Estimated enrolment: 200
Participants	Women aged 20 to 39 Inclusion criteria: • GnRH agonist and recombinant FSH in a long protocol cycle • Female partners were under 40 years of age • History of recurrent implantation failure, i.e. failed to achieve a clinical pregnancy after at least 3 fresh ICSI cycles where at least 8 good embryos had been transferred • Embryos were pushed to day 5 resulting in blastocysts, and undergo embryo transfer day 5 or vitrification on day 5 • Endometrial thickness $\geq$ 7 mm • Selected embryos for embryo transfer were blastocysts Exclusion criteria: Couples with testicular or epididymal sperm were excluded
Interventions	Fresh embryo transfer: ICSI cycle followed by day 5 fresh embryo transfer Freeze-all: ICSI cycle, all embryos were cryopreserved at day 5 and transferred in a consecutive natural cycle
Outcomes	• Clinical pregnancy: foetal cardiac activity on ultrasound examination 4 weeks after embryo transfer • Ongoing pregnancy: pregnancies with visible foetal heart motion over the number of transferred embryos at 10 weeks' gestation
Starting date	February 2012
Contact information	Yasmin Magdi, Research and Development Department Director, TopLab Company for ART Laboratories Consultation and Training
Notes	clinicaltrials.gov/ct2/show/record/NCT02681367

NCT02712840

Trial name or title	Comparing pregnancy outcomes in good prognosis patients between fresh and 'freeze-all' single blastocyst transfers
Methods	RCT Estimated enrolment: 118
Participants	Women aged 18 to 35 Inclusion criteria: • First IVF cycle • Normal ovarian reserve parameters (antral follicle count > 12, FSH < 10 IU/L, AMH (if measured) > 15 pmol/L) • Infertility cause due to tubal factor, male factor with ejaculated sperm, or unexplained • 3 or more fresh-transfer or cryopreservation-quality blastocysts on day 5 post-oocyte-retrieval • GnRH antagonist or long GnRH agonist cycles Exclusion criteria: • Evidence of (or evidence for significant risk of) OHSS on post-oocyte-retrieval day 5 (in which the standard protocol is not to perform a fresh embryo transfer, but rather to freeze all blastocysts for future frozen embryo transfers) • Use of a GnRH agonist trigger for ovulation and resulting intensive luteal-phase support protocol • Women requiring automatic freeze-all approaches (such as for pre-implantation genetic testing or cryopreservation for fertility preservation) • Female infertility causes that may adversely affect implantation, such as severe endometriosis, fibroids, Müllerian abnormalities, or prior uterine procedures resulting in a potentially compromised endometrial cavity • In vitro maturation of oocytes • Oocyte donation cycles
Interventions	Freeze-all protocol: All good morphologic quality blastocysts are vitrified on day 5 or 6. The best-quality vitrified blastocyst frozen on day 5 will be warmed and transferred in a subsequent cycle Fresh protocol: Women receive fresh embryo transfer of best morphologic quality blastocyst on day 5 and vitrification of all good-quality supernumerary blastocysts
Outcomes	• Live birth rate per blastocyst transfer • Cumulative ongoing pregnancy rate for all blastocysts transferred from the same ovarian hyperstimulation IVF cycle: a pregnancy with a positive heartbeat beyond 12 weeks of gestation • Cumulative clinical pregnancy rate for all blastocysts transferred from the same ovarian hyperstimulation IVF cycle: presence of a gestational sac seen by transvaginal ultrasonography 4 to 5 weeks after embryo transfer
Starting date	September 2015
Contact information	Stephanie Jewell: sjewell@mtsinai.on.ca Jason E Elliott: jelliott@mtsinai.on.ca
Notes	Contact: Jason E Elliott, MD, MSc

NCT02746562

Trial name or title	Study comparing outcomes between conventional IVF and a "freeze-all"-strategy in assisted reproductive technology
Methods	RCT Estimated enrolment: 424
Participants	Women aged 18 to 39 Inclusion criteria: • AMH > 6.28 pmol/L (Roche Elecsys assay) • Female age 18 years to less than 40 years • 1, 2, or 3 IVF/ICSI cycle with oocyte aspiration • Regular menstrual cycle between 24 and 35 days • BMI between 18 and 35 • 2 ovaries • Can and will sign informed consent Exclusion criteria: • Endometriosis stage III to IV • Ovarian cysts with diameter > 30 mm at day of start of stimulation • Submucosal fibroids • Women with severe comorbidity (IDDM, NIDDM, gastrointestinal, cardiovascular, pulmonary, liver, or kidney disease) • Dysregulation of thyroid disease • Not Danish or English speaking • Contraindications or allergies to use of gonadotropins or GnRH antagonists • TESA • Oocyte donation • Previous inclusion in the study
Interventions	Freeze-all: transfer of a frozen-thawed blastocyst in a subsequent natural menstrual cycle Fresh embryo transfer: standard procedure
Outcomes	• Cumulative live birth: measured after 1 stimulated cycle with oocyte retrieval and after use of all frozen blastocysts or after at least 1 year of follow-up • Time to pregnancy: from start of ovarian stimulation to positive hCG • Ongoing pregnancy per transfer of the first blastocyst, per oocyte pick-up, per start of ovarian stimulation, and per randomised woman • Live birth after the first blastocyst transfer calculated per randomised woman, per started ovarian stimulation, per oocyte pick-up, and per transfer • Preterm birth • Low birth weight • Small for gestational age • Large for gestational age • Perinatal mortality • Pre-eclampsia • Placental rupture • Miscarriages, ectopic pregnancies
Starting date	May 2016

NCT02746562 (*Continued*)

Contact information	Sacha Stormlund: sacha.stormlund.01@regionh.dk Anja Pinborg: anja.bisgaard.pinborg@regionh.dk
Notes	clinicaltrials.gov/ct2/show/record/NCT02746562

NTR3187

Trial name or title	A single-center non-blinded randomised controlled trial on the effect of ovarian hyperstimulation on endometrial receptivity
Methods	RCT Target enrolment: 193
Participants	Women aged < 43 years Inclusion criteria: • Subfertile couples with female age < 43 undergoing IVF or ICSI Exclusion criteria: • Couples undergoing a PGD cycle • Couples for whom IVF/ICSI is used to prevent the transmission of HIV • Couples undergoing a modified natural cycle • Women with borderline or invasive ovarian cancer • Women with contraindications for IVF/ICSI treatment such as cardiovascular-pulmonary disease, severe diabetes, bleeding disorders, immunodeficiency, and morbid obesity • Women with premature ovarian failure • Women with severe psychopathology, severe anxiety, and inability to cope with treatment • Not able or willing to provide informed consent
Interventions	Experimental arm: All embryos will be cryopreserved for subsequent transfer in artificial cycles. Ovarian hyperstimulation, oocyte retrieval, and oocyte fertilisation will be performed using standard procedures. Control arm: 1 or 2 fresh embryo(s) will be transferred in the same cycle with cryopreservation of all supernumerary embryos and subsequent transfer of frozen-thawed embryos in artificial cycles if pregnancy is not achieved after fresh transfer
Outcomes	• Cumulative ongoing pregnancy rate per cycle after 12 months of treatment • Embryo quality • Clinical pregnancy rate • Miscarriage rate • Live birth rate • Time to pregnancy • Birth weight • Percentage of children with congenital abnormalities
Starting date	January 2013
Contact information	Sebastian Mastenbroek: S.Mastenbroek@amc.uva.nl
Notes	www.trialregister.nl/trialreg/admin/rctview.asp?TC=3187

AFC: antral follicle count
AMH: anti-Müllerian hormone
BMI: body mass index
COS: controlled ovarian stimulation
FET: frozen embryo transfer
FSH: follicle-stimulating hormone
GnRH: gonadotropin-releasing hormone
hCG: human chorionic gonadotropin
ICSI: intracytoplasmic sperm injection
IDDM: Insuline-Dependent Diabetes Mellitus
IVF: in vitro fertilisation
LH: luteinising hormone
MESA: microsurgical epididymal sperm aspiration
NGS: next-generation sequencing
NIDDM: Non-Insuline-Dependent Diabetes Mellitus
OHSS: ovarian hyperstimulation syndrome
PGD: pre-implantation genetic diagnosis
PGS: pre-implantation genetic screening
PRL: prolactine
RCT: randomised clinical trial
SHBG: sex hormone-binding globulin
TESA: testicular sperm aspiration
TSH: thyroid-stimulating hormone

DATA AND ANALYSES

Comparison 1. Freeze-all versus conventional IVF, outcomes per woman

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Live birth rate	4	1892	Odds Ratio (M-H, Fixed, 95% CI)	1.09 [0.91, 1.31]
1.1 Live birth rate: cumulatively for cleavage stage transfer	2	1633	Odds Ratio (M-H, Fixed, 95% CI)	1.11 [0.91, 1.35]
1.2 Live birth rate: cumulatively for blastocyst stage transfer	2	259	Odds Ratio (M-H, Fixed, 95% CI)	0.99 [0.60, 1.62]
2 OHSS	2		Peto Odds Ratio (Peto, Fixed, 95% CI)	Subtotals only
2.1 Per cycle with ovarian hyperstimulation	2	1633	Peto Odds Ratio (Peto, Fixed, 95% CI)	0.24 [0.15, 0.38]
3 Ongoing pregnancy rate	2		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
3.1 Cumulatively	2	259	Odds Ratio (M-H, Fixed, 95% CI)	1.05 [0.64, 1.73]
4 Clinical pregnancy rate	1		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
4.1 Cumulatively	1	125	Odds Ratio (M-H, Fixed, 95% CI)	1.08 [0.54, 2.19]
5 Multiple pregnancy rate	2		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
5.1 After first ET	2	1630	Odds Ratio (M-H, Fixed, 95% CI)	1.11 [0.85, 1.44]
6 Miscarriage rate	4		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
6.1 After first ET	4	1892	Odds Ratio (M-H, Fixed, 95% CI)	0.67 [0.52, 0.86]
7 Pregnancy complications	2		Peto Odds Ratio (Peto, Fixed, 95% CI)	Subtotals only
7.1 After first ET	2	1633	Peto Odds Ratio (Peto, Fixed, 95% CI)	1.44 [1.08, 1.92]
8 Birth weight of babies born	1		Mean Difference (IV, Fixed, 95% CI)	Subtotals only
8.1 Birth weight of singletons	1	462	Mean Difference (IV, Fixed, 95% CI)	161.80 [57.11, 266.49]
8.2 Birth weight of multiples	1	453	Mean Difference (IV, Fixed, 95% CI)	-2.0 [-94.08, 90.08]

Comparison 2. Freeze-all versus conventional IVF, adverse events per clinical pregnancy

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Multiple pregnancy	2		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
1.1 After first ET	2	939	Odds Ratio (M-H, Fixed, 95% CI)	1.02 [0.77, 1.37]
2 Miscarriage	4		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
2.1 After first ET	4	1058	Odds Ratio (M-H, Fixed, 95% CI)	0.56 [0.41, 0.77]
3 Pregnancy complications	2		Peto Odds Ratio (Peto, Fixed, 95% CI)	Subtotals only
3.1 After first ET	2	914	Peto Odds Ratio (Peto, Fixed, 95% CI)	1.43 [1.05, 1.95]

Comparison 3. Freeze-all versus conventional IVF, congenital abnormalities per live-born children plus number of foetuses therapeutically terminated

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Congenital abnormalities	1	923	Odds Ratio (M-H, Fixed, 95% CI)	1.25 [0.66, 2.37]

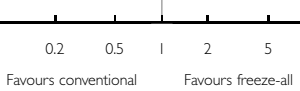
Analysis 1.1. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 1 Live birth rate.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 1 Live birth rate

Study or subgroup	Freeze all IVF n/N	Conventional IVF n/N	Odds Ratio M-H,Fixed,95% CI	Weight	Odds Ratio M-H,Fixed,95% CI
1 Live birth rate: cumulatively for cleavage stage transfer					
Chen 2016	465/746	455/762		78.6 %	1.12 [0.91, 1.37]
Ferraretti 1999	23/58	26/67		6.8 %	1.04 [0.50, 2.13]
Subtotal (95% CI)	**804**	**829**		**85.4 %**	**1.11 [0.91, 1.35]**
Total events: 488 (Freeze all IVF), 481 (Conventional IVF)					
Heterogeneity: Chi² = 0.04, df = 1 (P = 0.85); I² =0.0%					
Test for overall effect: Z = 1.03 (P = 0.30)					
2 Live birth rate: cumulatively for blastocyst stage transfer					
Shapiro 2011a	37/70	35/67		7.8 %	1.03 [0.52, 2.00]
Shapiro 2011b	37/60	39/62		6.8 %	0.95 [0.46, 1.97]
Subtotal (95% CI)	**130**	**129**		**14.6 %**	**0.99 [0.60, 1.62]**
Total events: 74 (Freeze all IVF), 74 (Conventional IVF)					
Heterogeneity: Chi² = 0.02, df = 1 (P = 0.88); I² =0.0%					
Test for overall effect: Z = 0.04 (P = 0.97)					
Total (95% CI)	**934**	**958**		**100.0 %**	**1.09 [0.91, 1.31]**
Total events: 562 (Freeze all IVF), 555 (Conventional IVF)					
Heterogeneity: Chi² = 0.24, df = 3 (P = 0.97); I² =0.0%					
Test for overall effect: Z = 0.94 (P = 0.35)					
Test for subgroup differences: Chi² = 0.18, df = 1 (P = 0.67), I² =0.0%					

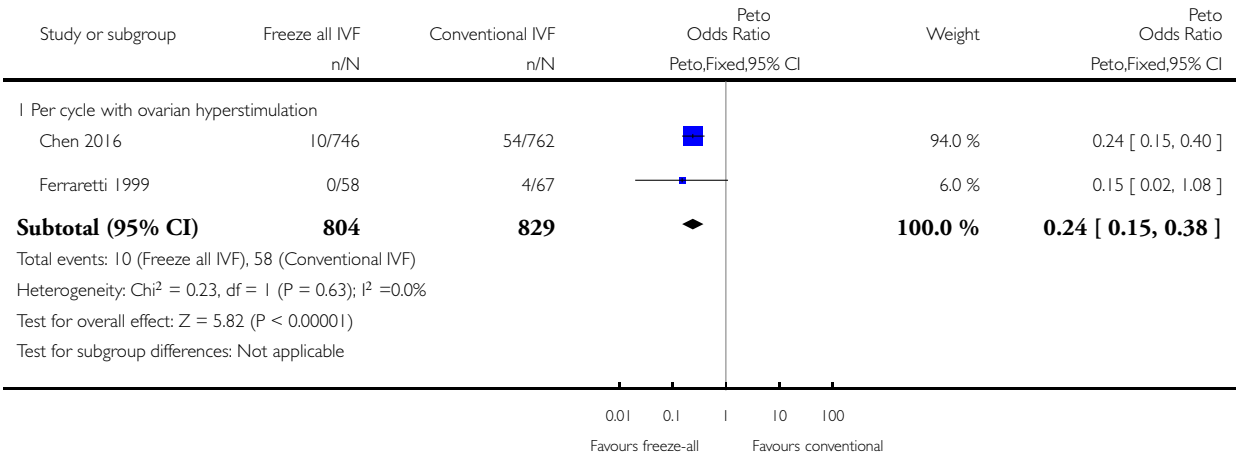


Analysis 1.2. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 2 OHSS.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 2 OHSS

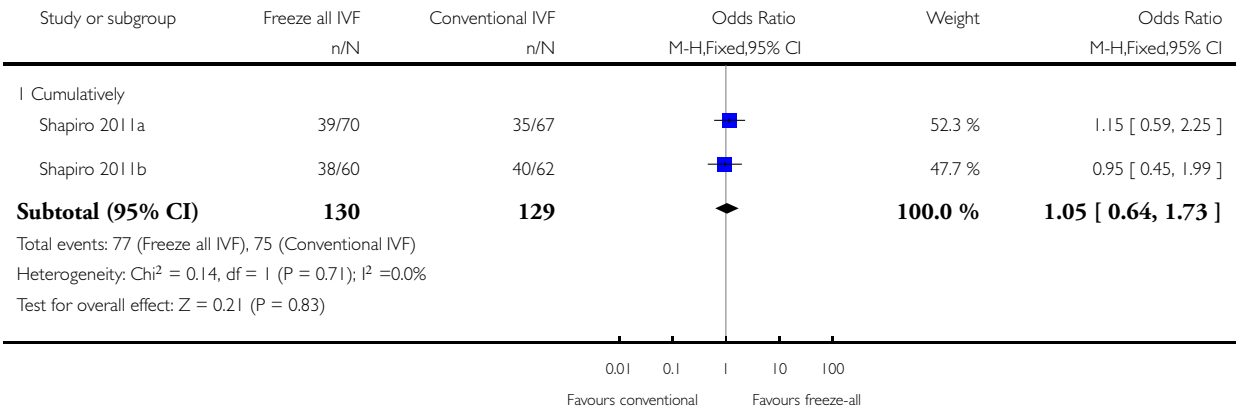


Analysis 1.3. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 3 Ongoing pregnancy rate.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 3 Ongoing pregnancy rate

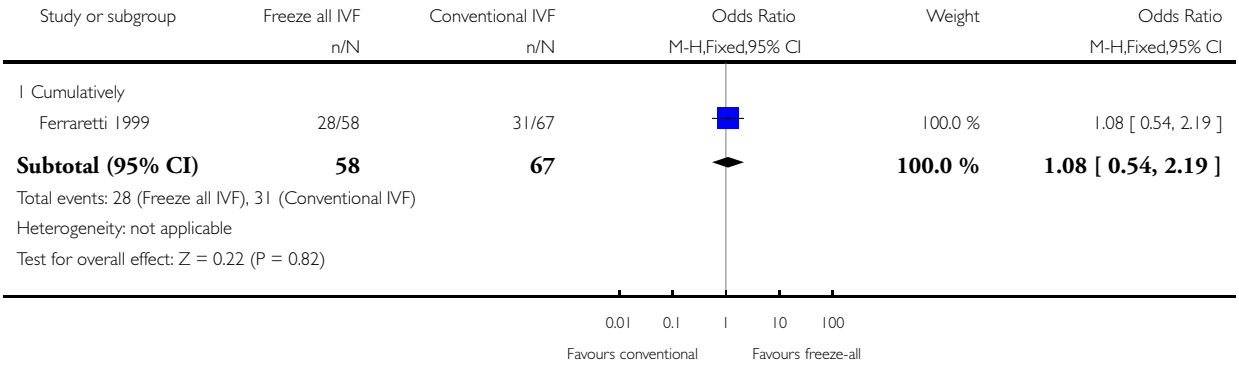


Analysis 1.4. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 4 Clinical pregnancy rate.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 4 Clinical pregnancy rate

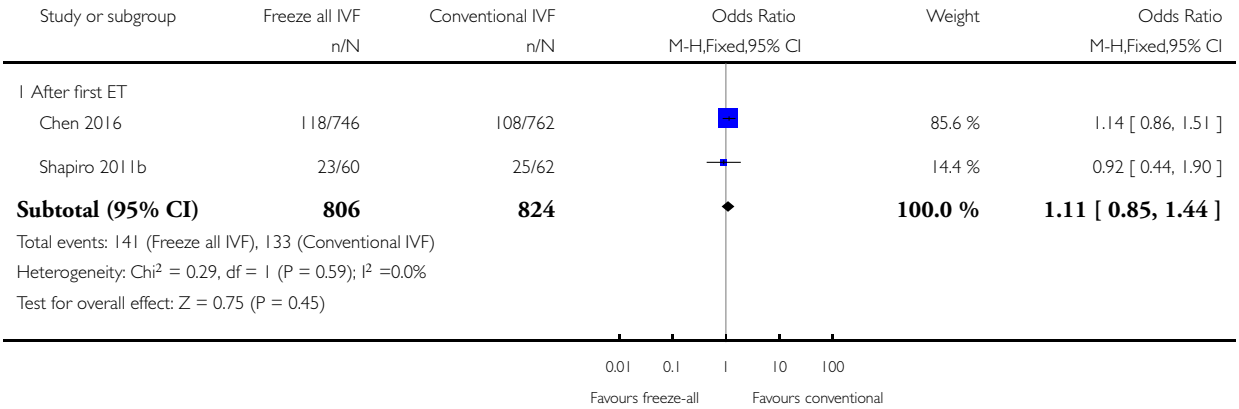


Analysis 1.5. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 5 Multiple pregnancy rate.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 5 Multiple pregnancy rate

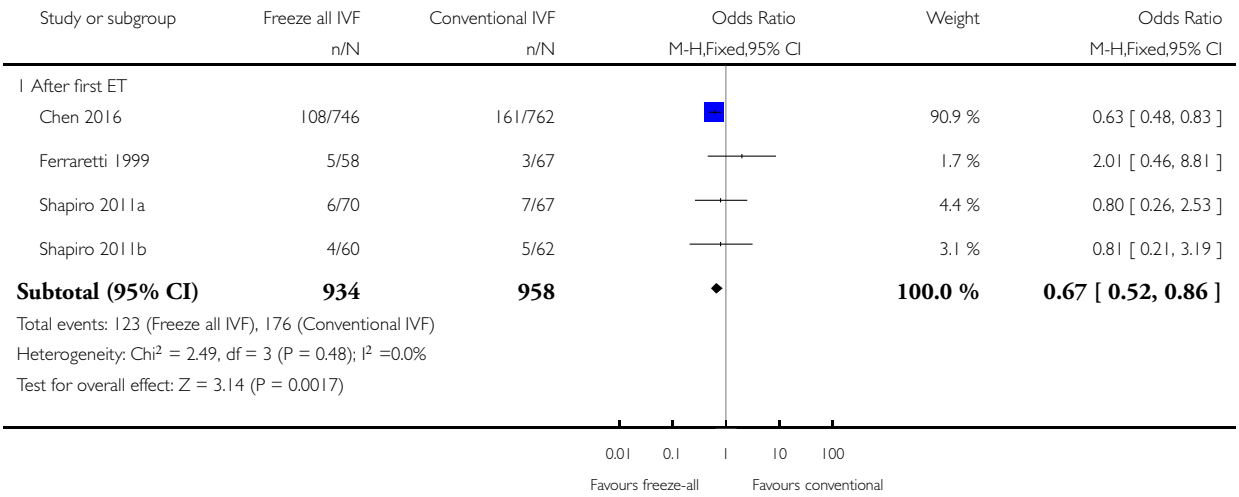


Analysis 1.6. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 6 Miscarriage rate.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 6 Miscarriage rate

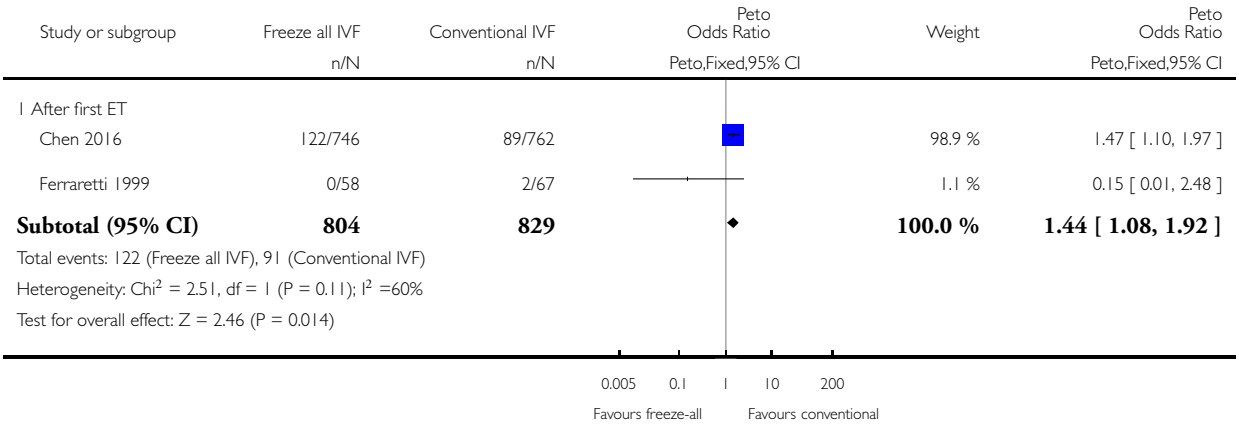


Analysis 1.7. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 7 Pregnancy complications.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 7 Pregnancy complications

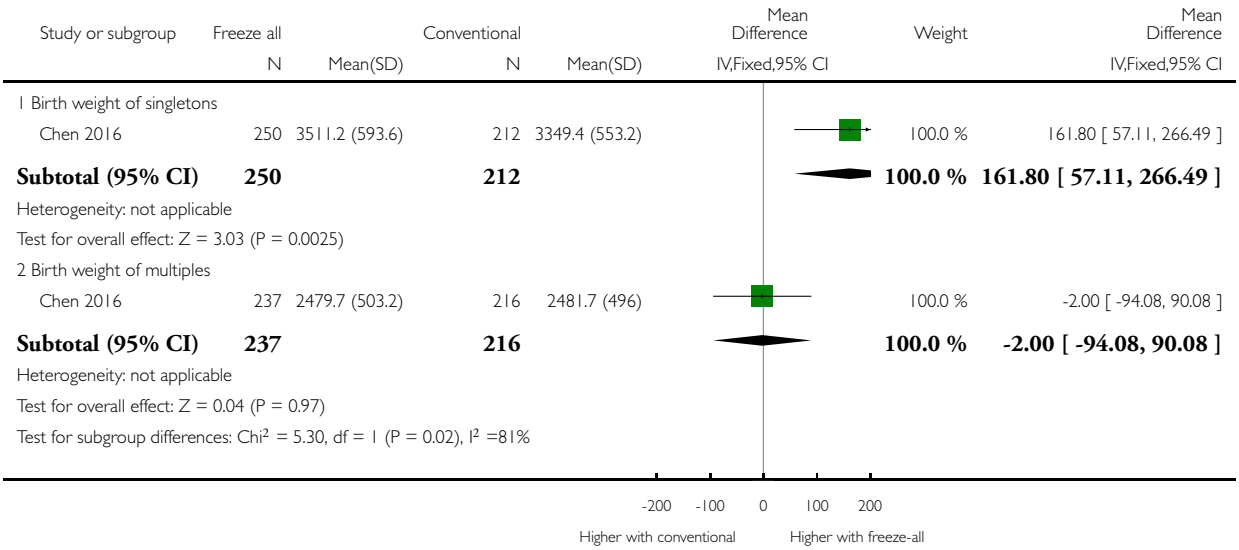


Analysis 1.8. Comparison 1 Freeze-all versus conventional IVF, outcomes per woman, Outcome 8 Birth weight of babies born.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 1 Freeze-all versus conventional IVF, outcomes per woman

Outcome: 8 Birth weight of babies born

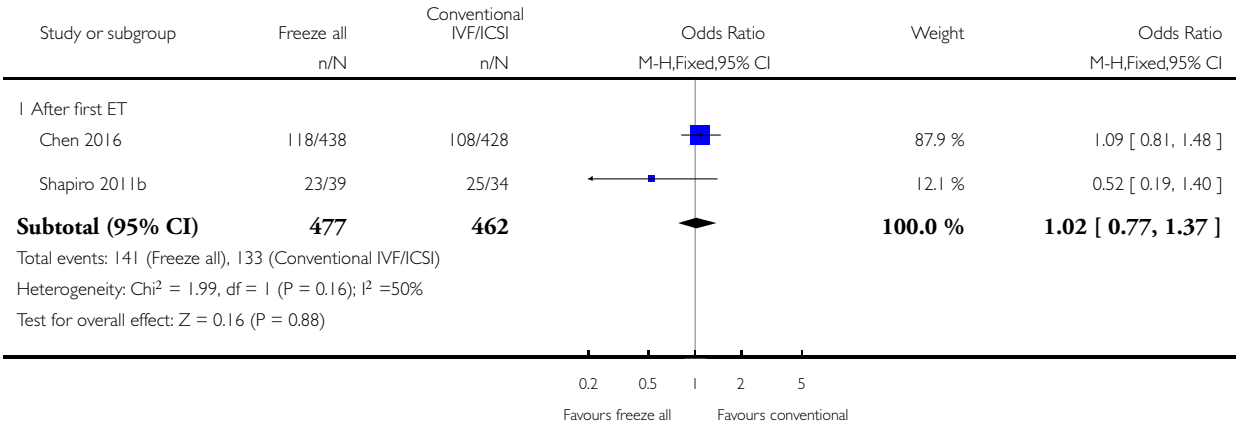


Analysis 2.1. Comparison 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy, Outcome 1 Multiple pregnancy.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy

Outcome: 1 Multiple pregnancy



Analysis 2.2. Comparison 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy, Outcome 2 Miscarriage.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy

Outcome: 2 Miscarriage

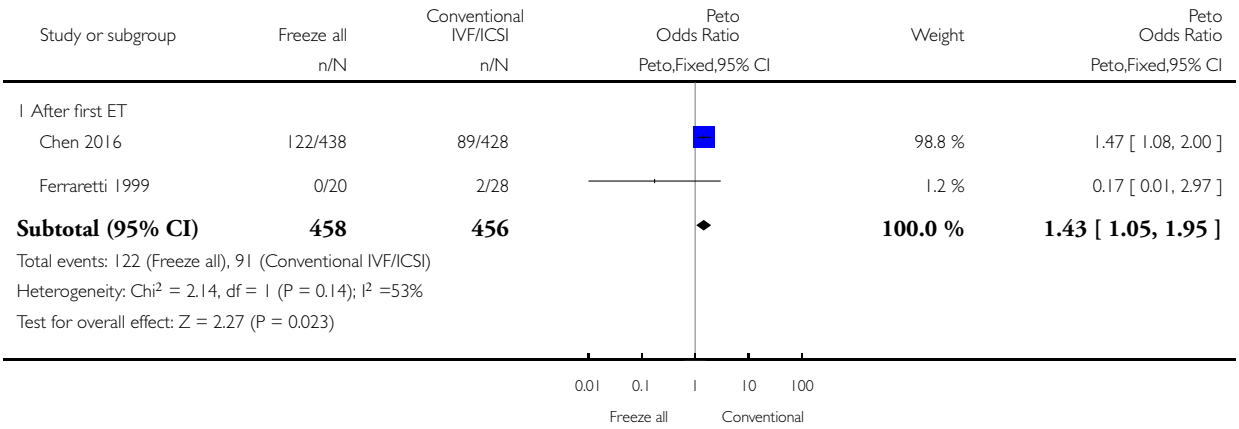
Study or subgroup	Freeze all n/N	Conventional IVF/ICSI n/N	Odds Ratio M-H,Fixed,95% CI	Weight	Odds Ratio M-H,Fixed,95% CI
1 After first ET					
Chen 2016	64/438	107/428		87.0 %	0.51 [0.36, 0.72]
Ferraretti 1999	5/20	3/28		1.8 %	2.78 [0.58, 13.32]
Shapiro 2011a	6/42	7/29		6.7 %	0.52 [0.16, 1.76]
Shapiro 2011b	4/39	5/34		4.5 %	0.66 [0.16, 2.70]
Subtotal (95% CI)	**539**	**519**		**100.0 %**	**0.56 [0.41, 0.77]**
Total events: 79 (Freeze all), 122 (Conventional IVF/ICSI)					
Heterogeneity: Chi^2 = 4.32, df = 3 (P = 0.23); I^2 =31%					
Test for overall effect: Z = 3.62 (P = 0.00029)					

Analysis 2.3. Comparison 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy, Outcome 3 Pregnancy complications.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 2 Freeze-all versus conventional IVF, adverse events per clinical pregnancy

Outcome: 3 Pregnancy complications

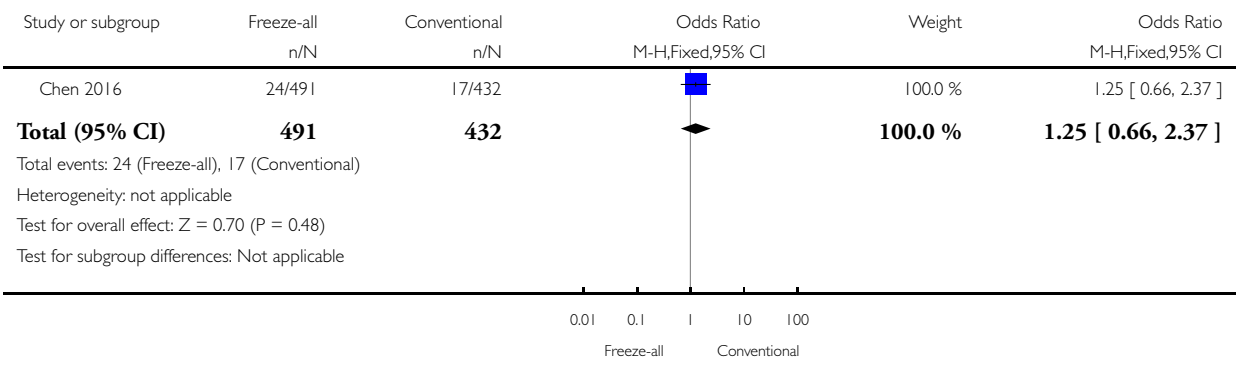


Analysis 3.1. Comparison 3 Freeze-all versus conventional IVF, congenital abnormalities per live-born children plus number of foetuses therapeutically terminated, Outcome 1 Congenital abnormalities.

Review: Fresh versus frozen embryo transfers in assisted reproduction

Comparison: 3 Freeze-all versus conventional IVF, congenital abnormalities per live-born children plus number of foetuses therapeutically terminated

Outcome: 1 Congenital abnormalities



ADDITIONAL TABLES

Table 1. Sensitivity analysis for cumulative live birth rate

Studies, number of participants	OR, 95% CI, fixed effect	OR, 95% CI, random effect	RR, 95% CI, fixed effect	RR, 95% CI, random effect
Ferraretti 1999 (n = 125) Shapiro 2011a (n = 103) Shapiro 2011b (n = 122) Chen 2016 (n = 1508)	1.09 (0.91, 1.31)	1.09 (0.91, 1.31)	1.04 (0.96, 1.12)	1.04 (0.96, 1.12)

CI: confidence interval
OR: odds ratio
RR: risk ratio

Table 2. Live birth rate after first transfer

Outcome	Number of studies	Number of participants	Analysis method	OR
Live birth rate after first embryo transfer for all embryo stages of transfer	4	1892	Odds ratio (Mantel-Haenszel, fixed, 95% confidence interval)	1.34 (1.12, 1.61)
Live birth rate after first transfer with cleavage-stage embryos	2	1633	Odds ratio (Mantel-Haenszel, fixed, 95% confidence interval)	1.31 (1.08, 1.59)
Live birth rate after first transfer with blastocyst-stage embryo	2	259	Odds ratio (Mantel-Haenszel, fixed, 95% confidence interval)	1.54 (0.94, 2.52)

Live birth rate calculated per first transfer is added for illustrative purposes as this comparison is often reported in the literature. It possibly shows differences in outcome for a stimulated and an unstimulated uterus, although this does not take into account the number of embryos that were thawed for transfer. This outcome is less relevant for women undergoing treatment since at the same time of first transfer in a freeze-all strategy, they would already have received the second transfer (in case of sufficient number of embryos) in a conventional strategy that includes fresh transfer.

APPENDICES

Appendix 1. Cochrane Gynaecology and Fertility Group specialised register search strategy

From inception to 27 July 2016
PROCITE platform
Keywords CONTAINS "cryopreservation"or "frozen embryo transfer" or "frozen embryos" or "frozen-thawed cycle" or "frozen-thawed embryo transfer" or "frozen-thawed embryos" or "FET" or "cryopreserved embryos" or "cryopreserved-thawed embryos" or "vitrified"or "vitrification"or"fresh v cryopreserved" or "freeze all" or Title CONTAINS "cryopreservation"or "frozen embryo transfer" or "frozen embryos" or "frozen-thawed cycle" or "frozen-thawed embryo transfer" or "frozen-thawed embryos" or "FET" or "cryopreserved embryos" or "cryopreserved-thawed embryos" or "vitrified"or "vitrification"or "fresh v cryopreserved"or "freeze all"
AND
Keywords CONTAINS "fresh"or "fresh blastocyst transfer"or "fresh cycle"or "fresh embryos"or "fresh v cryopreserved"or "fresh versus frozen" or Title CONTAINS "fresh"or "fresh blastocyst transfer"or "fresh cycle"or "fresh embryos"or "fresh v cryopreserved"or "fresh versus frozen" (89 hits)

Appendix 2. CENTRAL CRSO search strategy

From inception until 14th November 2016
CRSO Web platform
#1 MESH DESCRIPTOR Embryo Transfer EXPLODE ALL TREES 917
#2 MESH DESCRIPTOR Fertilization in Vitro EXPLODE ALL TREES 1782
#3 MESH DESCRIPTOR Sperm Injections, Intracytoplasmic EXPLODE ALL TREES 445
#4 embryo*: TI,AB,KY 3959
#5 (vitro fertili?ation):TI,AB,KY 1873
#6 ivf:TI,AB,KY 2946
#7 icsi:TI,AB,KY 1307
#8 (intracytoplasmic sperm injection*):TI,AB,KY 1000
#9 blastocyst*:TI,AB,KY 518
#10 #1 OR #2 OR #3 OR #4 OR #5 OR #6 OR #7 OR #8 OR #9 6032
#11 MESH DESCRIPTOR Cryopreservation EXPLODE ALL TREES 447
#12 MESH DESCRIPTOR Vitrification EXPLODE ALL TREES 23
#13 ((cryopreservat* or cryofixation or cryonic suspension)):TI,AB,KY 558
#14 (freez* or frozen):TI,AB,KY 2815
#15 Vitrif*:TI,AB,KY 193
#16 Thaw*:TI,AB,KY 494
#17 #11 OR #12 OR #13 OR #14 OR #15 OR #16 3332
#18 #10 AND #17 625
23 10 and 22 (451)

Appendix 3. MEDLINE search strategy

From 1946 until 14th November 2016
Ovid platform
1 exp Cryopreservation/ (33440)
2 exp Freezing/ (23063)
3 (cryopreservat$ or cryofixation or cryonic suspension).tw. (13490)
4 freez$.tw. (60485)
5 thaw$.tw. (22148)
6 exp Vitrification/ (970)
7 Vitrif$.tw. (4029)
8 froze$.tw. (71374)

9 disengage$.tw. (4166)
10 or/1-9 (158372)
11 exp embryo transfer/ or exp fertilization in vitro/ or exp sperm injections, intracytoplasmic/ or exp ovulation induction/ (44353)
12 embryo$.tw. (344559)
13 blastocyst$.tw. (20065)
14 vitro fertili?ation.tw. (20204)
15 ivf.tw. (20159)
16 icsi.tw. (6895)
17 intracytoplasmic sperm injection$.tw. (6056)
18 ovulation induc$.tw. (3841)
19 (ovar$ adj3 hyperstim$).tw. (4654)
20 (ovar$ adj3 stimulat$).tw. (6873)
21 exp Superovulation/ or Superovulat$.tw. (3796)
22 or/11-21 (382656)
23 10 and 22 (12133)
24 randomized controlled trial.pt. (469524)
25 controlled clinical trial.pt. (95062)
26 randomized.ab. (403302)
27 placebo.tw. (196678)
28 clinical trials as topic.sh. (189460)
29 randomly.ab. (284766)
30 trial.ti. (178345)
31 (crossover or cross-over or cross over).tw. (75863)
32 or/24-31 (1176650)
33 exp animals/ not humans.sh. (4668056)
34 32 not 33 (1083487)
35 23 and 34 (550)

Appendix 4. Embase search strategy

From 1980 until 14th November 2016
Ovid platform
1 exp Cryopreservation/ (33582)
2 exp Freezing/ (33013)
3 (cryopreservat$ or cryofixation or cryonic suspension).tw. (17116)
4 freez$.tw. (64880)
5 thaw$.tw. (26477)
6 exp Vitrification/ (4378)
7 Vitrif$.tw. (5937)
8 froze$.tw. (87650)
9 disengage$.tw. (4337)
10 or/1-9 (182265)
11 exp embryo transfer/ or exp fertilization in vitro/ or exp intracytoplasmic sperm injection/ (63530)
12 in vitro fertili?ation.tw. (24581)
13 icsi.tw. (12341)
14 intracytoplasmic sperm injection$.tw. (7704)
15 (blastocyst adj2 transfer$).tw. (1593)
16 ivf.tw. (31218)
17 exp superovulation/ (2550)
18 superovulat$.tw. (3423)
19 exp ovulation induction/ (12957)
20 blastocyst$.tw. (23859)

21 embryo$.tw. (351069)
22 vitro fertili?ation.tw. (24607)
23 ovulation induc$.tw. (5059)
24 (ovar$ adj3 stimulat$).tw. (9775)
25 (ovar$ adj3 hyperstim$).tw. (6460)
26 or/11-25 (404447)
27 10 and 26 (18175)
28 Clinical Trial/ (990930)
29 Randomized Controlled Trial/ (460474)
30 exp randomization/ (83533)
31 Single Blind Procedure/ (27015)
32 Double Blind Procedure/ (136735)
33 Crossover Procedure/ (53739)
34 Placebo/ (321486)
35 Randomi?ed controlled trial$.tw. (149041)
36 Rct.tw. (22285)
37 random allocation.tw. (1626)
38 randomly allocated.tw. (26549)
39 allocated randomly.tw. (2206)
40 (allocated adj2 random).tw. (843)
41 Single blind$.tw. (18625)
42 Double blind$.tw. (172648)
43 ((treble or triple) adj blind$).tw. (645)
44 placebo$.tw. (247067)
45 prospective study/ (385364)
46 or/28-45 (1770419)
47 case study/ (92640)
48 case report.tw. (322778)
49 abstract report/ or letter/ (985370)
50 or/47-49 (1391670)
51 46 not 50 (1720179)
52 27 and 51 (1488)

Appendix 5. PsycINFO search strategy

From 1806 to 14th November 2016
Ovid platform
1 (cryopreservat$ or cryofixation or cryonic suspension).tw. (66)
2 freez$.tw. (3756)
3 thaw$.tw. (124)
4 Vitrif$.tw. (11)
5 froze$.tw. (1321)
6 disengage$.tw. (5591)
7 or/1-6 (10628)
8 exp reproductive technology/ (1573)
9 icsi.tw. (61)
10 intracytoplasmic sperm injection$.tw. (47)
11 (blastocyst adj2 transfer$).tw. (4)
12 assisted reproduct$.tw. (730)
13 ovulation induc$.tw. (26)
14 (ovari$ adj2 stimulat$).tw. (55)
15 COH.tw. (86)

16 superovulat$.tw. (6)
17 infertil$.tw. (2923)
18 subfertil$.tw. (77)
19 (ovari$ adj2 induction).tw. (6)
20 ivf.tw. (466)
21 vitro fertili?ation.tw. (630)
22 (ovar$ adj3 hyperstimulat$).tw. (11)
23 or/8-22 (4438)
24 7 and 23 (98)
25 random.tw. (47258)
26 control.tw. (365925)
27 double-blind.tw. (19960)
28 clinical trials/ (9713)
29 placebo/ (4602)
30 exp Treatment/ (656349)
31 or/25-30 (1011580)
32 24 and 31 (24)

Appendix 6. CINAHL search strategy

From inception to 14th November 2016
Ebsco platform

#	Query	Results
S32	S19 AND S31	98
S31	S20 OR S21 OR S22 OR S23 OR S24 OR S25 OR S26 OR S27 OR S28 OR S29 OR S30	954,451
S30	TX allocat* random*	4,243
S29	(MH “Quantitative Studies”)	13,282
S28	(MH “Placebos”)	9,173
S27	TX placebo*	33,620
S26	TX random* allocat*	4,243
S25	(MH “Random Assignment”)	38,985
S24	TX randomi* control* trial*	85,907
S23	TX ((singl* n1 blind*) or (singl* n1 mask*)) or TX ((doubl* n1 blind*) or (doubl* n1 mask*)) or TX ((tripl* n1 blind*) or (tripl* n1 mask*)) or TX ((trebl* n1 blind*) or (trebl* n1 mask*))	763,614
S22	TX clinic* n1 trial*	170,899

(*Continued*)

S21	PT Clinical trial	77,668
S20	(MH "Clinical Trials+")	186,062
S19	S17 AND S18	436
S18	S8 OR S9 OR S10 OR S11 OR S12 OR S13 OR S14 OR S15 OR S16	13,480
S17	S1 OR S2 OR S3 OR S4 OR S5 OR S6 OR S7	7,170
S16	TX ovulation induc*	574
S15	TX icsi	251
S14	TX ivf	1,142
S13	TX vitro fertili?ation	2,914
S12	TX blastocyst*	603
S11	TX embryo*	10,541
S10	TX intracytoplasmic sperm injection*	234
S9	(MM "Fertilization in Vitro")	1,445
S8	(MM "Embryo Transfer")	261
S7	TX disengage*	917
S6	TX frozen	3,308
S5	TX Vitrif*	72
S4	TX thaw*	576
S3	TX freez*	2,163
S2	TX (cryopreservat* or cryofixation or cryonic suspension)	1,111
S1	(MH "Cryopreservation+")	1,143

Appendix 7. ClinicalTrials.gov search string

search terms https://clinicaltrials.gov/
(IVF OR ICSI OR embryo transfer) AND (freeze-all OR frozen thawed embryo transfer OR cryopreservation OR disengage)

Appendix 8. WHO ICTRP search string

search terms who.int/trialsearch
(IVF OR ICSI OR embryo transfer) AND (freeze-all OR frozen thawed embryo transfer OR cryopreservation OR disengage)

CONTRIBUTIONS OF AUTHORS

Kai Mee Wong and Sebastiaan Mastenbroek wrote the review. Kai Mee Wong, Sjoerd Repping, and Sebastiaan Mastenbroek developed the concept of the study. Madelon van Wely, Femke Mol, and Sjoerd Repping provided feedback on the review.

DECLARATIONS OF INTEREST

Kai Mee Wong: none known

Madelon van Wely: none known

Femke Mol: none known

Sjoerd Repping: none known

Sebastiaan Mastenbroek is principal investigator of one of the ongoing studies.

SOURCES OF SUPPORT

Internal sources

- None, Other.

External sources

- None, Other.

DIFFERENCES BETWEEN PROTOCOL AND REVIEW

We added a method of analysing time to pregnancy (by hazard ratios), as this was not reported in the protocol; in the event, no data were available for this outcome.

We performed a subgroup analysis by timing of embryo transfer for the primary outcome of cumulative live birth.

We changed the unit of analysis for birth weight (from per woman to per baby).

We added some details to the section specifying our plans for the summary of findings table.

Congenital disorders, defined as the number of congenital abnormalities at birth, were reported per live-born children plus number of foetuses therapeutically terminated in stead of per all clinical pregnancies.

INDEX TERMS

Medical Subject Headings (MeSH)

*Cryopreservation; *Embryo, Mammalian; Abortion, Spontaneous [epidemiology]; Embryo Transfer [*methods]; Live Birth [epidemiology]; Ovarian Hyperstimulation Syndrome [epidemiology; prevention & control]; Pregnancy Complications [epidemiology]; Pregnancy Rate; Pregnancy, Multiple [statistics & numerical data]; Randomized Controlled Trials as Topic

MeSH check words

Female; Humans; Pregnancy