PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	The effectiveness and safety of in vitro maturation of oocytes versus in vitro fertilisation in women with high antral follicle count: study protocol for a randomised controlled trial
AUTHORS	Vuong, Lan; Ho, Vu; Ho, Tuong; Dang, Vinh; Phung, Tuan; Giang, Nhu; Le, Anh; Pham, Toan; Wang, Rui; Norman, Robert; Smitz, Johan; Gilchrist, Robert; Mol, Ben

VERSION 1 – REVIEW

REVIEWER	Waljit Dhillo Imperial College London
REVIEW RETURNED	14-Apr-2018

GENERAL COMMENTS	1. This is an interesting study, with appropriate design, although one would suspect that IVM is unlikely to be as effective as IVF. 2. The primary outcome should be more clearly defined to read 'live birth rate', rather than 'ongoing pregnancy rate leading to live birth' as this could be confusing to the reader. Similarly page 9 line 42-48 requires clarification. 3. It should be specified that the comparison is between IVM and 'IVF with segmentation'. 4. Will patients be routinely assessed for OHSS, if so at which timepoint for each group? How will the RCOG guidance be used to grade OHSS? The guidance suggests moderate OHSS if ultrasound evidence of ascites. What threshold for ascites present on ultrasound will be used? How many features from each category will be used to assign patients- For example, the guidance suggests that all patients with at least one feature from severe category e.g. hyponatremia <135 will be categorised as severe. Some of these are biochemical and will only be identified if patients are routinely screened. If a variation of these criteria is to be used, this is fine, but the parameters for categorisation should be clearly delineated. 5. Will the decision to have 1 or 2 embryos transferred be decided by patient preference? 6. Regarding the sample size calculation, what is the current live birth rate for IVM at the centre? Is a non-inferiority design chosen as IVM is not expected to achieve similar outcomes to IVF, in which case is there sufficient evidence for clinical equipoise to conduct the trial? Minor:

	page 18, some formatting remains visible. Some of the formatting for table 1 is not aligned correctly. page 11 line 20 'participantss'. Some of the initials are not obvious based on the authorship list e.g. TDP same as TPD?

REVIEWER	Enrico Papaleo IRCCS San Raffaele, Milan, Italy
REVIEW RETURNED	17-May-2018

GENERAL COMMENTS	The primary end-point should be modified since the cumulative pregnancy rate better reflect the efficacy of two different strategies in term of live birth per single ovum retrieval.

REVIEWER	Fan QU Zhejiang University, China
REVIEW RETURNED	30-May-2018

GENERAL COMMENTS	None

REVIEWER	Karice Hyun University of Sydney
REVIEW RETURNED	11-Jul-2018

GENERAL COMMENTS	This would be an informative study to compare the effectiveness of in vitro maturation and in vitro fertilisation in women with high PCOS, given that PCOS is a very common condition in women. I have a few comments: 1. The protocol states that the study is a randomised block design, however, it is unclear what the block is. 2. Re page 13 lines 12-16, will there be categorical baseline data to be presented, and if so, how? 3. For page 12 line 18, do you mean "The rate of live birth and the associated 95% confidence interval (CI) will be estimated 'and compared' for each group using the exact method for binomial proportion"? 4. Please report the model that will be used for the comparison of the secondary outcome variables (lines 29-31). 5. Please report which program will be used for the analysis at the end of the statistical analysis section. 6. Under data handling, it is reported that Microsoft Excel will be used to record and store data, however, this is strongly not recommended as it is very easy to make errors and accidentally edit data that could affect the results. A more structured database, such as Redcap (which is also free) is recommended to collect data in even if the analysis will be done in excel.

VERSION 1 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 1
1. This is an interesting study, with appropriate design, although one would suspect that IVM is unlikely to be as effective as IVF.

Thank you for the comments. At our centre, we achieve a good success rate with IVM, which is relatively comparable to that with IVF. We have been doing IVM since 2006 and perform about 300 IVM cycles per year. Therefore, we have extensive experience with IVM, which could contribute to the good success rates achieved. Our live birth rate after IVM is 31.7% per cycle, which is similar to that of IVF cycles (33.8% for frozen transfer and 31.5% for fresh transfer; Vuong et al. NEJM 2018). The current RCT has been designed to investigate the comparative effectiveness of IVM and IVF. Given that this research question has not yet been addressed in an RCT, RCTs are needed on this topic.

2. The primary outcome should be more clearly defined to read 'live birth rate', rather than 'ongoing pregnancy rate leading to live birth' as this could be confusing to the reader.
Similarly page 9 line 42-48 requires clarification.
We appreciate the reviewer's perspective. However, the "ongoing pregnancy leading to live birth" endpoint was chosen deliberately. As a dichotomous outcome, ongoing pregnancy leading to live birth will have the same number of women with a live birth, and participants will still followed until live birth. The advantage of ongoing pregnancy resulting in live birth endpoint is that in the time-to-event analysis that we plan there is little delay from randomization till the event. Live birth as an outcome has the disadvantage that it requires conception, and then a period of 9 months waiting the time before the actual event occurs. Given that ongoing pregnancy leading to live birth was defined in the original study protocol, we prefer to leave it this way.

3. It should be specified that the comparison is between IVM and 'IVF with segmentation'.
We thank the reviewer for this comment. We have modified the manuscript text in multiple places to make this clear.

4. Will patients be routinely assessed for OHSS, if so at which timepoint for each group?
Yes, routine assessments for OHSS will be performed on the day of oocyte retrieval and on the day of embryo freezing (day 3 during IVF and day 5 during IVM). At other times, OHSS will be evaluated if symptoms were reported by the patient. Text describing assessments for OHSS has been added to the bottom on page 10 in the revised manuscript.

How will the RCOG guidance be used to grade OHSS? The guidance suggests moderate OHSS if ultrasound evidence of ascites. What threshold for ascites present on ultrasound will be used? How many features from each category will be used to assign patients- For example, the guidance suggests that all patients with at least one feature from severe category e.g. hyponatremia <135 will be categorised as severe. Some of these are biochemical and will only be identified if patients are routinely screened. If a variation of these criteria is to be used, this is fine, but the parameters for categorisation should be clearly delineated.
The RCOG guidelines were used for assessing OHSS, as described above by the reviewer and shown in Table 1. We have added a sentence at the bottom of page 10 to reinforce this, and cite the RJOG guidelines document.

5. Will the decision to have 1 or 2 embryos transferred be decided by patient preference?
Yes, this is correct. A sentence stating that transfer of one or two embryos was based on patient preference has been added to the revised manuscript (page 9, line 17 of the revised manuscript).

6. Regarding the sample size calculation, what is the current live birth rate for IVM at the centre? Is a non-inferiority design chosen as IVM is not expected to achieve similar outcomes to IVF, in which case is there sufficient evidence for clinical equipoise to conduct the trial?
The current live birth rate for IVM with hCG priming at our center is 31.7%. More recently, we have performed IVM without hCG priming and have used a new culture system for IVM that resulted in live birth rates of up to 40% while the live birth rate for IVF at our centre is 45%, as stated in the manuscript. The non-inferiority design was chosen because IVM has a number of other advantages

over IVF, including lower cost (about half that of IVF), and avoidance of complications such as OHSS, ovarian torsion and venous thromboembolism. In the future, multiple IVM cycles could be performed compared with one IVF cycle.

Minor:
page 18, some formatting remains visible.
The manuscript has been checked and amended.

Some of the formatting for table 1 is not aligned correctly.
The manuscript has been checked and amended.

page 11 line 20 'participantss'.
The manuscript has been checked and amended.

Some of the initials are not obvious based on the authorship list e.g. TDP same as TPD
TDP stands for Toan D Pham; TPD stands for Toan P Duong. They are for the same person. The author name has been corrected.

Reviewer: 2
The primary end-point should be modified since the cumulative pregnancy rate better reflect the efficacy of two different strategies in term of live birth per single ovum retrieval.
The study was designed with ongoing pregnancy resulting in live birth as the primary outcome because the main aim was to assess the effectiveness of one IVM treatment versus IVF (see response to comment 2 by reviewer 1 above). This is in line with the Harbin consensus meeting (Hum Reprod 2014;29(10):2075-82). Cumulative pregnancy rate is included as a secondary outcome.

Reviewer: 3
No comments to address.

Reviewer: 4
This would be an informative study to compare the effectiveness of in vitro maturation and in vitro fertilisation in women with high PCOS, given that PCOS is a very common condition in women. I have a few comments:

1. The protocol states that the study is a randomised block design, however, it is unclear what the block is.
The block size has been mentioned in the manuscript in the last paragraph on page 7, line 1.

2. Re page 13 lines 12-16, will there be categorical baseline data to be presented, and if so, how?
Categorical baseline data will be presented as number (%). A sentence has been added to the end of this paragraph to provide this information.

3. For page 12 line 18, do you mean "The rate of live birth and the associated 95% confidence interval (CI) will be estimated 'and compared' for each group using the exact method for binomial proportion"?
Yes, that is correct. The rate of live birth and the associated 95% confidence interval (CI) will be estimated and compared for each group using the exact method for binomial proportion. The text has been updated to clarify this.

4. Please report the model that will be used for the comparison of the secondary outcome variables (lines 29-31).

Differences between groups in secondary outcome variables will be analysed using Student t-test or Wilcoxon signed-rank test for normally distributed or skewed variables and Fisher's exact test for categorical variables. The text has been updated to clarify this.

5. Please report which program will be used for the analysis at the end of the statistical analysis section.
We will use R version 3.5.0 (Copyright (C) 2018 The R Foundation for Statistical Computing); text has been added to show this.

6. Under data handling, it is reported that Microsoft Excel will be used to record and store data, however, this is strongly not recommended as it is very easy to make errors and accidentally edit data that could affect the results. A more structured database, such as Redcap (which is also free) is recommended to collect data in even if the analysis will be done in excel.
Thank you for the comment. We acknowledge the limitations, and have now changed to using REDCap software. The manuscript text has been updated to reflect this (page 15, line 11-12).

VERSION 2 – REVIEW

REVIEWER	Professor Waljit S Dhillo Imperial College London
REVIEW RETURNED	09-Aug-2018

GENERAL COMMENTS	Remaining Queries: It is preferable to use live birth rate rather than ongoing pregnancy rate, however if ongoing pregnancy rate at 12 weeks has been selected, then it should be referred to as 'ongoing pregnancy rate' rather than 'ongoing pregnancy rate leading to live birth' as it is not possible to assess at 12 weeks gestation whether this pregnancy will indeed certainly lead to live birth. Screening for OHSS should ideally be carried out on the same days in both groups (perhaps day 3 post oocyte retrieval) otherwise a systematic bias could be introduced between the groups. The RCOG guidance provides some symptoms and signs of OHSS compatible with different grading of OHSS, but lacks the specificity and objectivity to accurately grade OHSS. It would be preferable to clearly state in the methods the criteria for how the RCOG guideline would be used in an objective manner such that the grading could be repeated in other centers in the same manner if desired. This is especially important as blinding is not readily possible in this study.

REVIEWER	Karice Hyun University of Sydney
REVIEW RETURNED	06-Sep-2018

GENERAL COMMENTS	No further comments

VERSION 2 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 1

Reviewer Name: Professor Waljit S Dhillo

Institution and Country: Imperial College London

Please state any competing interests or state ‘None declared’: No relevant competing interests. Expert in kisspeptin use in IVF treatment.

Please leave your comments for the authors below

Remaining Queries:

It is preferable to use live birth rate rather than ongoing pregnancy rate, however if ongoing pregnancy rate at 12 weeks has been selected, then it should be referred to as ‘ongoing pregnancy rate’ rather than 'ongoing pregnancy rate leading to live birth’ as it is not possible to assess at 12 weeks gestation whether this pregnancy will indeed certainly lead to live birth.

RESPONSE: Thank you for the comment. As a dichotomous outcome, ongoing pregnancy leading to live birth is the same as live birth, i.e. we will only count ongoing pregnancies that result in live birth while ongoing pregnancies not resulting in live birth (e.g. late pregnancy loss) will not be counted. Therefore, this outcome is different from ongoing pregnancy. The reason we chose it as the primary outcome was that when performing time-to-event analysis, we will take the moment that ongoing pregnancy has confirmed (12 weeks) as the date that the endpoint occurred. A comment was already made on page 10 of the original manuscript. We propose no changes.

Screening for OHSS should ideally be carried out on the same days in both groups (perhaps day 3 post oocyte retrieval) otherwise a systematic bias could be introduced between the groups. The RCOG guidance provides some symptoms and signs of OHSS compatible with different grading of OHSS, but lacks the specificity and objectivity to accurately grade OHSS. It would be preferable to clearly state in the methods the criteria for how the RCOG guideline would be used in an objective manner such that the grading could be repeated in other centers in the same manner if desired. This is especially important as blinding is not readily possible in this study.

RESPONSE: The reviewer makes valid points. We will assess OHSS on the day of oocyte retrieval (day 3) in both groups. Methods text (page 11, paragraph 1) has been updated to reflect this, and now reads as follows: “Routine assessments for OHSS were performed on day of oocyte retrieval (day 3) in both groups.”

Regarding the accurate grading of OHSS, we agree that a different method would be more objective and detailed. Therefore, we will now use (and reference) the flow diagram developed by Humaidan et al for use in clinical trial settings. The manuscript text and Table 1 have been updated accordingly. The new methods text (page 11, paragraph 1) now reads as follows:

“OHSS was classified using the flow diagram developed by Humaidan and colleagues for use in clinical trial settings.”

VERSION 3 – REVIEW

REVIEWER	Waljit S Dhillo

	United Kingdom
REVIEW RETURNED	23-Oct-2018

GENERAL COMMENTS	It would still be more reasonable to describe your primary outcome as 'live birth' rather than 'ongoing pregnancy' that will in the future lead to live birth, as this is a more accurate description of what you are using, but then state in the methods that 12weeks gestation was used as the timepoint for 'time to event' analyses. The current description is unnecessarily counterintuitive.

VERSION 3 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 1
Reviewer Name: Waljit S Dhillo
Institution and Country: United Kingdom
Please state any competing interests or state ‘None declared’: None declared.

Please leave your comments for the authors below

It would still be more reasonable to describe your primary outcome as 'live birth' rather than 'ongoing pregnancy' that will in the future lead to live birth, as this is a more accurate description of what you are using, but then state in the methods that 12weeks gestation was used as the timepoint for 'time to event' analyses. The current description is unnecessarily counterintuitive.

RESPONSE: Thank you for the comment. The primary outcome has been changed to ‘live birth’. The new text (page 10, paragraph 1) now reads as follows: “The primary endpoint is live birth after the first embryo transfer of the started treatment cycle”.