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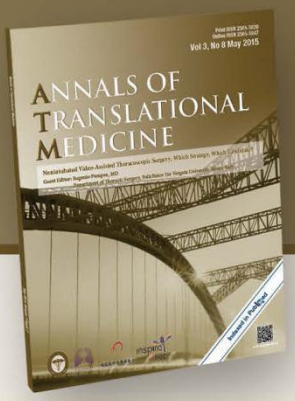


**Peer Review File**

Article information: http://dx.doi.org/10.21037/atm-20-3623

**Reviewer A**

Although this is a study with a large number of patients, it does not provide much new clinical information. There have been many studies in the literature.
And the design of retrospective research does not provide strong information
In addition, when submitting documents, cluttered revision messages make it difficult for reviewers to distinguish between important and unimportant messages.

Reply: We thank the reviewer for his/her valuable comments and apologize for the inconvenience caused by the mistakes in uploading the files. We found that in previous research conclusions, the influence of ECF on pregnancy outcome is still controversial. A small amount of ECF was not considered to have an adverse effect on pregnancy outcome in previous studies. Our study found that even a small amount of ECF can have an adverse effect on pregnancy outcome, but this effect will not continue to the long-term obstetric outcome. These will play a guiding role in the future clinical treatment of uterine effusion.

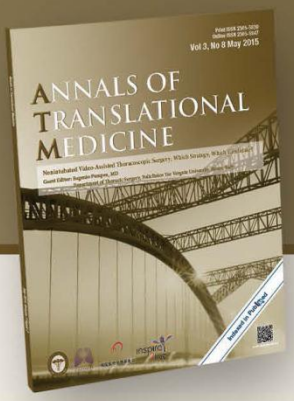
**Reviewer B**

The manuscript has a good concept but needs some valid explanations justifying the results. The comments have been stated in the separate document.

Comments:

Line 68:
The numbers show that the patients without ECF had lower %.
Reply：The inaccurate representation has been revised.
Changes in the text: line 64.

Line 93 – 95:
Please rephrase the sentence for better understanding.
Reply: The sentence has been revised to make it easier to understand.
Changes in the text: line 88-91.




Line 96:
(COH): plz mention the full form when using for the first time.
Reply: Thanks, modified.
Changes in the text: line 92.

Line 109:
ERC number needs to be mentioned.
Reply: the ERC number is provided.

Line 115:
Word “diagnosed”, to be replaced by “detected an measured”.
Reply: Thanks, modified.
Changes in the text: line 112.

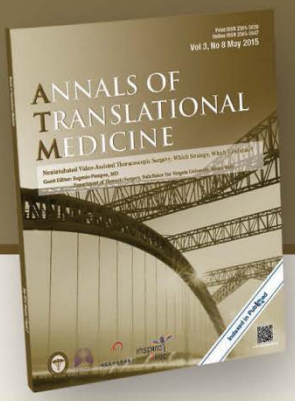
Line 116:
Were instead on was.
Reply: Thanks, modified.
Changes in the text: line 113.

Line 176:
Please complete the sentence.
Reply: The sentence has been completed.
Changes in the text: line 155-156.

Line 220 - 224:
already mentioned earlier, so not needed. Start with the discussion.
Reply: OK, this part of the content has been deleted.
Changes in the text: line 221-216.

Line 232:
Whether or not
Reply: Thanks, modified.
Changes in the text: line 218.

Line 244 - 246:
Giving some probable mechanism for this result would be better rather than rewriting the results here.



Reply: The probable mechanism for this result is supplied.
Changes in the text: line 226-230.

Line 260 - 262:
Giving some probable mechanism for this result would be better rather than rewriting the results here.
Reply: The probable mechanism for this result is supplied.
Changes in the text: line 243-248.

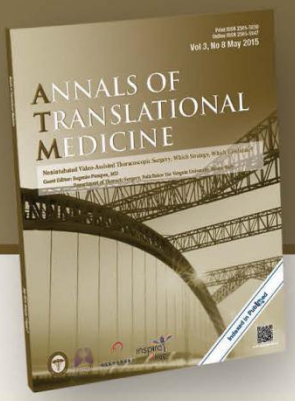
General comments:
The objective in the introduction states “investigating the relationship”.
The objective in the abstract states finding association between pregnancy outcome and presence of ECF.
The results show the comparison of the Mean between the study groups. Nowhere in the results the association can be seen. A figure illustrating the association or relation of ECF with other factors would be appropriate
Reply: Thanks to the reviewers for their valuable comments. The purpose of our research is to explore the relationship between ECF and pregnancy outcome through retrospective research. Therefore, after PSM matching, we compared the pregnancy outcome of the patients with or without ECF. Besides, we calculated the relative risk factor ratios of the pregnancy outcome subgroups, results show that ECF is negatively correlated with biochemical pregnancy, clinical pregnancy, and live birth. The related data can be found in line 182-196.,

**Reviewer C**

Thank you very much for giving me the opportunity to review manuscript ATM-20-3623-R1. The authors investigated relationship between endometrial cavity fluid and clinical outcome of IVF/ICSI. It was interesting to read your findings. The article will be of interest to people in the field. On the other hand, I think that manuscript still have some problems as indicated below.

Major issue
1. You should explain definition of good-quality embryos.
Reply: The definition of good-quality embryos is provided.
Changes in the text: line 148-149.

2. You should mention the detail of embryo transferred-embryos. For example, ratio of IVF/ICSI in ECF and control groups.



Reply: Thanks to the reviewer for the comments, we have added the detailed steps of IVF/ICSI to the manuscript.
Changes in the text: line 157-168.

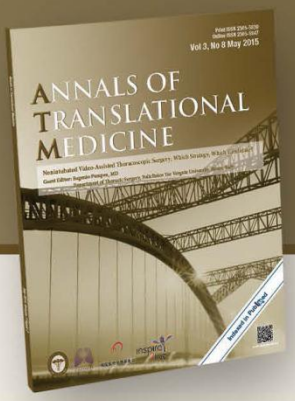
3. Which did you perform fresh ET or frozen-warmed ET? You should explain it. If fresh ET was performed, why did not you select that all embryo/blastocyst were cryopreserved? Because mean number of follicles in both ovaries was approximately 15 (in Table 1) and number of retrieved oocytes was approximately 12.5, I concern about OHSS. Did OHSS occur?
Reply: Our research objects are all fresh embryo transplant patients, which we have added in the article. The proportion of frozen embryo transplants in our hospital during the same cycle is relatively small, so we selected fresh embryo transplant patients for comparison when we conducted the original data collection. Regarding the question of whether the patient has OHSS, after our inspection, we found that some patients in the two groups of patients did have OHSS. Since we chose a 1:1 pairing, we did not pay too much attention to this issue. We can provide relevant data if requested by the author.
Changes in the text: line 158-159.

4. If you performed frozen-warmed ET, you should insert the result of the endometrial thickness on ET-day.
Reply: Since all the patients in our study were transplanted with fresh embryos, there is no need to answer this question, thanks.

5. You should mention luteal support in ECF and control groups.
Reply: We have added the program for luteal support to the draft.
Changes in the text: line 159-160.

6. Would it be possible for you to show the rates of fertilization and embryo development in both groups in Table 2?
Reply: According to the reviewers' comments, we have made statistics on the fertilization rate and embryo development data. The relevant data has been added to Table 2. We found that the fertilization rate of ICSI patients in the ECF group was slightly higher than that in the control group ($P<0.05$), and the number of high-quality embryos There is no difference between the two groups.
Changes in the text: line 354.

7. You should revise following sentence, because it is not clear. Was the incidence of adverse pregnancy outcomes significantly different between ECF and control groups? The incidences of adverse pregnancy outcomes in ECF group significantly higher than that in control groups? Or,



the incidences of abortion during a biochemical pregnancy, ectopic pregnancy, and abortion during clinical pregnancy in ECF significantly higher than in control group, respectively? Which did you mean the former or the latter? To better understand the result of adverse pregnancy outcome, I suggest addition of adverse pregnancy outcome into Table 3.

"Compared with the control group, the incidences of adverse pregnancy outcomes (including abortion during a biochemical pregnancy, ectopic pregnancy, and abortion during clinical pregnancy) (Figure 1)." (line 205-207)
Reply: We have added the adverse pregnancy outcome data to Table 3. We found that the biochemical pregnancy miscarriage rate (p=0.085) and clinical pregnancy miscarriage rate (p=0.087) were higher in the ECF group than in the control group, but there was no significant difference. The statement in the article has been revised.
Changes in the text: line 188-191.

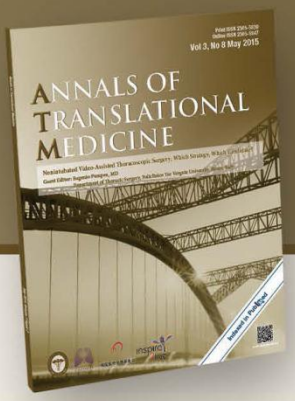
8. Regarding following sentence, I suggest revise it, because the rate of gestational diabetes in ECF group was significantly higher than that in control group.

However, the obstetric outcomes were not affected significantly by the presence of ECF. (line 223-224)
Reply: According to the reviewer's suggestion, we consider deleting this part and starting our discussion from the second paragraph.
Changes in the text: line 221.

9. Regarding following sentence, is this sentence citation? or is this sentence the result of this study? In case of citation, you should insert reference number. In case of the results in this study, you should explain the results in Result section, because the results that showed in Result section only should be mentioned in Discussion section.

In all patients with hydrosalpinx, a comparison between the ECF and control groups showed no significant difference in the clinical pregnancy rate (63/1862, 3.4% vs. 50/1862, 2.7%). (line 244-246)
Reply: Thank you for the reminder, we have modified this.
Changes in the text: line 234-235.

10. Please confirm to the manuscript Akman et al. (line 226-228, reference 8). I think that the citation of this manuscript is not appropriate because this manuscript mentioned that "the drainage of the fluid from the peritoneal surface to the endometrial surface in PCOS could be



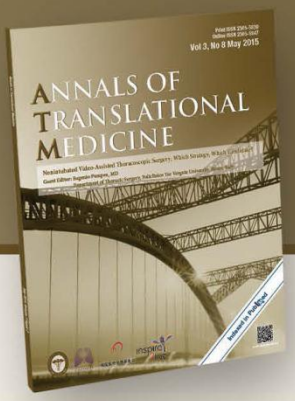
envisaged as being more physiological, whereas in tubal factor cases it raises the suspicions of infectious origin of a previous tubo-ovarian disease."
Reply: Thanks, after careful inspection, we found that there was indeed a misunderstanding of the content of the original article when we cited the literature, and we have corrected this.
Changes in the text: line 211-214.

11. Please confirm to the manuscript of Sharara et al. (line 244, reference 15). I think that the citation of this manuscript is not appropriate because this manuscript showed that "There was a trend for a higher implantation rate and ongoing pregnancy rate in the no hydrosalpinx group compared to the hydrosalpinx group (12.6 versus 9.8%, and 33.7 versus 24.8% respectively); however, this did not reach statistical significance."
Reply: Thanks to the reviewers for pointing out the inappropriateness of our citing literature so carefully, we have modified this.
Changes in the text: line 226-230.

12. Please confirm to the manuscript of Lu et al. (line 248, reference 16). I think that the citation of this manuscript is not appropriate because this manuscript do not mention that the presence of ECF dilutes putative beneficial factors in the uterus following ET.
Reply: Thanks for the reviewer's comments. At the end of the fifth paragraph of the discussion section of this cited article, the author mentioned: In addition to the changed uterine receptivity, the excessive luminal fluid might also dilute important soluble factors (cytokines, chemokines et.al) within the uterine lumen, which might decrease the efficiency of implantation. We cited the view point in the article, please check that.

13. Please confirm to the manuscript of Strandell. (line 252, reference 18). I think that the citation of this manuscript is not appropriate this manuscript did not show positive effect of growth factor from hydrosalpinx fluid on implantation.
Reply: In the *Theory of impaired implantation ability* section of this cited article, the author concluded that the hydrosalpinx fluid has the effect of promoting the secretion of TUN, hCG and progesterone, so it has a certain growth-promoting effect rather than embryo toxicity. We cited this theory of it, please check that.

14. In introduction (in previous studies, line 92-95), "Moreover, ECF disappears by the day of ET if the fluid amount is < 3.5 mm in the anterior–posterior dimension. The presence of this kind of ECF had no influence on embryo implantation and the clinical pregnancy rate was reported to be no worse than patients without ECF."
In discussion (in this study, line 254-256), "All patients with ECF included in this study were identified during COH, and the amount of fluid did not reach the routine treatment standard of



our hospital (< 3.5 mm).” You should discuss the difference between your study and previous study enough. Why did ECF negative effect in your study? Did ECF not disappear by the day of ET? Moreover, if the routine treatment was performed in cases of ≥3.5 mm ECF (ECF was aspirated before ET [exclusion criteria; line 122]), you should mention ECF size (inclusion/exclusion criteria) in Study design section.
Reply: Yes, our research object is the final pregnancy outcome of these patients whose ECF volume is less than 3.5mm but still has ECF at the time of ET. We believe that this persistent ECF is a pathological existence and may affect the process of embryo implantation. According to the reviewer’s opinion, we add the ECF size in study design section.
Changes in the text: line 119.

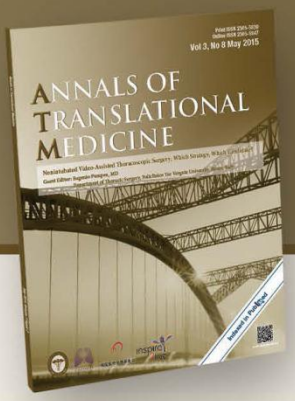
15. You should discuss the result of relationship between Gestational diabetes and ECF enough in Discussion.
Reply: Thanks to the reviewer's comments, we have further discussed the possible relationship between uterine effusion and gestational diabetes.
Changes in the text: line 248-254.

Miner issue
1. You should insert IRB number in Ethical approval section.
Reply: the IRB number was provided.
Changes in the text: line 106.
2. You should provide references for following sentences (line 89-91, line 228-230, line 277-278, line 278-230).
It has been hypothesized that embryonal apposition may not occur when a fluid layer covers the endometrium and that the presence of ECF before ET in IVF cycles is associated with implantation failure.

Following COH, ovarian hyperstimulation syndrome (OHSS) can be linked with extravasation of secretions and obstruction of the cervix leading to ECF, which can occur when there are cervical canal obstructions and pelvic adhesions.

Besides the use of transvaginal sonographic ECF aspiration, moderate curettage of the uterus also works.

Some Chinese traditional herbal medicines such as Leonurus cardiaca might also help with the absorption of effusions in some cases.
Reply: Thanks to the reviewer for the comments. We have adjusted the corresponding references and made accurate explanations of some of our own conclusions.




Please check that.
Changes in the text: line 85-87, line 214-216, line 267-269,line 269-270.

3. I suggest change from the word of “study group” to “ECF group” on line 168, 199, 212 and 216, in Table 1, 2, 3, and 4 because you use the word of “ECF group” in Abstract and Result sections. The lack of uniformity creates confusion.
Reply: According to the reviewer’s comments, all study groups have been replaced by ECF group.
Changes in the text: line 122,154,183,187,189.

4. Does Laboratory tests mean “baseline FSH, LH, and estrogen (line 145)” in Table 1? You should make information in Table 1 clearer.
Reply: Thanks for the reviewer's comments, it has been revised.
Changes in the text: line 352

5. Does “cycle” in Table 2 mean “number of previous cycles of IVF/ICSI (line 148)”? You should make information in Table 2 clearer.
Reply: No, it means the current cycle. We have accurately described it.
Changes in the text: line 138, line 354.

6. Regarding “Duration of Gn – d” in table 2, what do you mean “d”? You should revise it.
Reply: we have revised that.
Changes in the text: line 354.

7. You should insert word of “Table 3” into following location like line 217 (…P = 0.014; Table 4).
(… 69.2%, P < 0.001; Table 3) line 201
(… 63.5%, P < 0001; Table 3) line 204
(… 55.5%, P < 0.001; Table 3) line 209
Reply: Thanks, we have revised that.
Changes in the text: line 183,191,195.

8. You should insert word of “Table 4” into following sentence
Pregnancy-induced hypertension had the highest incidence in both groups (Table 4). (line 215-216)
Reply: OK, we have revised that.
Changes in the text: line 206.

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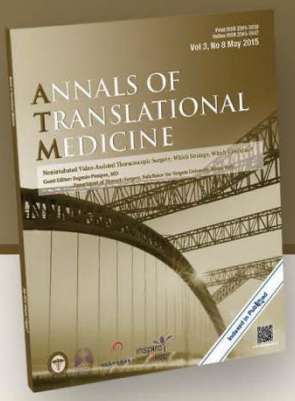

9. You should insert full spell following words.
line 55. ET
line 96. COH
line 124. PSM
Reply: thanks, we have revised that.
Changes in the text: line 61, 82, 92.

Mistyping
You should revise the word of “p” to “P” in abstract section (line 69-76), because you use capital letter (P) in manuscript.

line 54. endometrial cavity fluid(ECF) –> endometrial cavity fluid (ECF) (please insert a space before parenthesis)
line 54. in vitro fertilization(IVF) –> in vitro fertilization (IVF) (please insert a space before parenthesis)
line 55. intracytoplasmic sperm injection(ICSI) –> intracytoplasmic sperm injection (ICSI) (please insert a space before parenthesis)
line 59. in vitro fertilization/intracytoplasmic sperm injection –> IVF/ICSI
line 130. embryo transfer –> ET
line 141. Body mass index (BMI) –> Body mass index (please unify the writing in Table 1, please remove the word of “BMI”, because the word is used this location only)
line 191. hadhave –> had
line 192. baseline variables. (Table 1). –> baseline variables (Table 1). (please remove comma before (Table 1))
line 250. (e.g. integrin...) –> (e.g. integrin…). (please insert a space before integrin)
line 259. (including gestational age, …) –> (including gestational age, …) (please remove a space before including)
line 260. fetuses) –> fetuses) (please remove a space before parenthesis)
Table 1, 2,4 and Figure 1: You should insert a space before parenthesis and before/after equal.
Table 1, Indications for IVF –> Indications for IVF/ICSI (please refer to line 143)
Table 1, and 2. Regarding the unit of liter, please unify the capital letter or lower-case letter.
Reply: According to the problems pointed out by the reviewers, we have made revisions one by one, please refer to the original text for specific revisions.

I hope these comments will be helpful.
Reply: Thanks to the reviewers for their careful comments and suggestions. We have benefited a lot and it will be a big improvement for our article.

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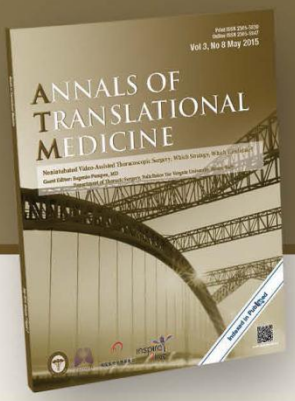


**Reviewer D**

This is a retrospective cohort study with a large sample size, where authors tried to examine the clinical significance of endometrial fluid during IVF/ICSI on outcomes. They found that ECF detected by ultrasonography during COH had a negative association with IVF/ICSI pregnancy outcomes but had no significant association with adverse obstetric outcomes, except from gestational diabetes.

The study should be revisited in terms of structure, accuracy of findings, synchronization of sections and reporting, probably through the help of a native speaker and an IVF expert.

I believe that if improved it could be eligible for publication in the journal.

Reply: Thanks to the reviewers for their comments, we will further improve the quality of the article to ensure that it reaches the publication level.