**Dear Jaydev,**

**Thanking you for reviewing our manuscript entitled “Isolation and *In Vitro* Decidualization of Mouse Primary Endometrial Stromal Cells”. We have revised the manuscript to address each of the comments** followed by our responses highlighted in *blue***. Below is a detailed discussion of our amendments:**

**Reviewer #1:**  
1. Whether the author examined the polyploidization of uterine stromal cells by immunofluorescence or flow cytometry after treatment with estrogen and progesterone. Please provide the data in the results if any or at least be mentioned in the discussion.

Response: Thanks. We have added relevant content in the Discussion part.

2. Line 41, "isolation" should be changed to "isolated".

Response: Thanks for the suggestion. It has been corrected.

3. Please double check line 48, "during the early pregnancy" looks like the redundancy.

Response: Thanks for your reminding. It has been deleted.

4. Line 58, "consists" should be changed to "consists of"

Response: Thanks for your reminding. It has been corrected by the editor.

5. Line 249, please substitute "uterine/endometrial decidualization" for "endometrial stromal cells and decidualization".  
Response: Thanks. We have corrected these in Line 244.

**Reviewer #2:**  
None.

**Reviewer #3:**  
*Major Concerns:*  
This is an important technique however more data is required to show these cells undergo a true deciulal reaction. The only true evidence in the morphology (which in not convincing) and the expression of Snail and Dtprp. This manuscript should show the expression of Pgr, Bmp2 and Wnt 4. The cells should be stained for alkaline phosphatase. To be truly valuable resource a more expensive characterization is required.  
Response: This is a very good point, we appreciate the thoughtful and encouraging comments of this reviewer. The stromal cells experience both morphologic and functional changes after embryo implantation *in vivo*. The decidualized stromal cells around embryo which named primary stromal cells process epithelial characteristics with highly expressed E-cadherin and decreased Snail. While the outer stromal cells are mainly composed by polyploidy stromal cells with secreting nature. It’s hard to imitate all kinds of decidualized stromal cells just by the simple combination of estrogen and progesterone *in vitro*. We failure to detect the increase of Pgr, Bmp2 and Wnt4 in current model, which consistent with the complexity of decidualization. Although our current model will improve in vitro studies, more efforts will be taken to optimize this model in our further study.

*Minor Concerns:*  
Finally, the writing style in the manuscript needs improvement.  
Response: Thanks for your suggestion. The manuscript has been modified by a native English speaker.

We hope that our responses are now satisfactory for consideration of publication in **JOVE**

Again, we appreciate the effort and time of the reviewers in reviewing this manuscript and to you and your staff for handling this task.

Sincerely,



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