November 3, 2014

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 13880-review.docx).

Title: Early gestation chorionic villi-derived stromal cells for fetal tissue engineering

Authors: Lee Lankford, Taryn M. Selby, James C. Becker, Volodymyr Ryzhuk, Connor W. Long, Diana L. Farmer and Aijun Wang

Name of Journal: *World Journal of Stem Cells*

ESPS Manuscript NO: 13880

The manuscript has been improved and revised according to the comments and suggestions of the reviewers and editor:

Reviewer 1 Comments:

1. We appreciate the Reviewer 1's comment about the indications for chorionic villus sampling vs. amniocentesis after 15 weeks of gestation. The aim of our study is to describe chorionic villus tissue-derived PMSCs as a promising source for fetal tissue engineering with unique therapeutic potential. Chorionic villus tissue can be obtained from chorionic villus sampling (CVS) or discarded placentas. In the context of this study we are not discussing CVS as a routine diagnostic procedure, but rather as source of autologous cells for therapies. The majority of indications for *in utero* transplantation of PMSCs can be diagnosed by ultrasound. Thus, CVS would be employed solely to obtain autologous tissue for cell isolation and transplantation, not as a diagnostic procedure. We have revised the text in order to clarify this point.
2. We believe any confusion over the Oil Red staining in our multipotency study image is due to the low resolution of the PDF file type required for initial submission. We are resubmitting the manuscript with a higher resolution figure as well as a magnified view of one cell to highlight the intracellular nature of the oil droplets formed in the differentiated PMSCs. We will also submit our original image file, which will retain maximum clarity.
3. We agree that cytokine expression and secretion may vary from donor to donor. The cell line tested for cytokine secretion was representative of all lines characterized in terms of marker expression and multipotency. Based on previous cytokine array assays that we have performed for other studies, we have noticed many similarities in the cytokines and growth factors secreted by PMSCs from a different isolation protocol and PMSCs from later gestation placental tissue. Nevertheless, no two cell lines that we have tested with these array kits are identical. Therefore, the protein array assays performed on one PMSC cell line shown in this study are meant to serve as an example of the breadth of factors these cells are capable of producing and the variety in their paracrine functionality.

Reviewer 2 Comments:

1. We thank Reviewer 2 for the aforementioned comment and are happy to clear up this confusion. Our description of GFP lentiviral infection does refer to viral transduction. We have revised all references to lentiviral transduction of the cell line to state “viral vector” where previously “vector” had been stated. We also have clarified in the text (in methods) that this transduction involves viral integration of the vector in the host cell genome, and will remain after cells divide. We have worked extensively with this viral vector for cell labeling *in vitro* and *in vivo,* and have observed that cells remain GFP+ for at least 5 passages beyond transduction, and that the GFP expression is stable in cryopreserved cells well beyond 6 months.
2. We have revised the manuscript according to the reviewer’s suggestion that all references to “data” be made plural.

Editor Comments:

1. The manuscript is being revised and submitted as a Word document, with figures included within and as separate .psd files in order to allow for manipulation and editing.
2. Formatting has been corrected for the abstract and several requested sections inserted, including: author contributions, core tip, and comments section.
3. References were formatted according to journal standards, and first pages of references without both PMID and DOI are being provided.

Thank you again for your consideration in publishing our manuscript in the *World Journal of Stem Cells*.

Sincerely,

Aijun Wang, PhD
Assistant Professor
Co-Director, Surgical Bioengineering Laboratory
UC Davis Medical Center, Dept. of Surgery
4625 2nd Ave., Research II, Room 3005
Sacramento, CA 95817
Office: (916) 703-0422
Fax: (916) 703-0430
Email: aawang@ucdavis.edu