



São Paulo, 23 de março de 2021

Prof. Dr. Prof. Tong Cao
Editor-in-Chief of
World Journal of Stem Cells

Dear Dr. Tong Cao,

Attached is the manuscript “COMPARATION OF THE CHONDROGENIC POTENTIAL OF MESENCHYMAL STEM CELLS FROM SYNOVIAL FLUID, BONE MARROW, AND ADIPOSE TISSUE USING A MAGNETIC 3D CELL CULTURE SYSTEM” by Joice Fülber, Fernanda Rodrigues Agreste, Sarah Raphaela Torquato Seidel, Eric Danilo Pauls Sotelo, Ângela Perrone Barbosa, Yara Maria Michelacci and Raquel Yvonne Arantes Baccarin that we wish to submit for publication as “Original article” in World Journal of Stem Cells.

Attached are the modifications introduced in the paper in the light of the reviewers’ comments.

We thank the reviewers’ comments that greatly improved our paper, and hope that the modifications introduced will now meet your requirements.

Hoping to hear from you soon, we are sincerely yours,

Joice Fülber
Postdoctoral Research
FMVZ-USP





Reviewer #1

1. Specific Comments to Authors: *Study reporting extracellular matrix synthesis activity of equine MSCs derived from bone marrow, synovial fluid and adipose tissue during chondrogenic differentiation by extraction of proteoglycans in microspheroids after chondrogenic differentiation is well written and the experimental design is well. However, phenotype of MSCs for surface markers is missing. The phenotype of the isolated cells should be characterized by surface markers and included as a figure with the flow cytometry analysis to confirm the MSC population.*

Response: We agree with the reviewer's opinion. Methods and results of MSCs phenotyping were include (see page 2, line; 48; page 3, line 58; page 8; line 192 and page 12; line 296).

2. Passage number of MSCs used for characterization and differentiation should be indicated. the paper is lacing a clear objective aim and hypothesis.

Response: This information was included in the text (see page 8, line; 193 and page 9, line 207).

3. Assesment of Chondrogeic differentiation was only determined by aggrecan. For proper evaluation of the microspheroid system types II, X, I collagens, and glycosaminoglycan (G AG) are also determined.

Response: We thank the reviewer's comments.

Unfortunately, at this moment, it is impossible to assess the expression of collagen I, collagen II, collagen X and glycosaminoglycans in the microspheroids.

We agree that the determination of type II collagen should be useful (as well as the determination of the transcription factor Sox9, which are cartilage specific markers). Nevertheless, the available primary antibodies did not detect the horse antigens.

The same was true for collagens I and X.

Concerning glycosaminoglycans, due to the microspheroid size, they could be detected only by isotopic metabolic labeling with ³⁵S-sulfate. Other methods, such as Toluidine Blue staining, do not have the necessary sensitivity (limit 0.5 ug). The metabolic labeling with radioisotope has been a very common practice in our laboratory at Escola Paulista de Medicina, Unifesp, for many years (for instance, see Martins et al. *Molecular Vision* 13:142-150, 2007; Soriano et al., *Clinica Chimica Acta* 295:41-62, 2000; Hadad et al., *Biochimica Et Biophysica Acta-General Subjects* 1290:18-28, 1996; Michelacci & Dietrich *Journal of Biological Chemistry* 251:1154-1158, 1976). Nevertheless, it is impossible to perform these experiments now, because due to COVID-19, ³⁵S-sulfate is not available.

However, aggrecan is a cartilage specific marker, and we believe that its expression, at the high levels here presented, is evidence of chondrogenic differentiation.





LANGUAGE QUALITY

Please resolve all language issues in the manuscript based on the peer review report. Please be sure to have a native-English speaker edit the manuscript for grammar, sentence structure, word usage, spelling, capitalization, punctuation, format, and general readability, so that the manuscript's language will meet our direct publishing needs.

Response: The manuscript was reviewed by Editage www.editage.com.

EDITORIAL OFFICE'S COMMENTS

Authors must revise the manuscript according to the Editorial Office's comments and suggestions, which are listed below:

(1) *Science editor:* 1 Scientific quality: The manuscript describes a Basic Study of the chondrogenic potential of mesenchymal stem cells from synovial fluid, bone marrow, and adipose tissue. The topic is within the scope of the WJSC.

(1) Classification: Grade C;

(2) Summary of the Peer-Review Report: The manuscript is well written and the experimental design is well. However, phenotype of MSCs for surface markers is missing. The molecular analysis is required indeed. The questions raised by the reviewers should be answered;

Response: It was done.

(3) Format: There are 4 figures;

(4) References: A total of 53 references are cited, including 5 references published in the last 3 years;

(5) Self-cited references: There are 1 self-cited references; and

(6) References recommendations (kindly remind): The authors have the right to refuse to cite improper references recommended by the peer reviewer(s), especially references published by the peer reviewer(s) him/herself (themselves). If the authors find the peer reviewer(s) request for the authors to cite improper references published by him/herself (themselves), please send the peer reviewer's ID number to editorialoffice@wjgnet.com. The Editorial Office will close and remove the peer reviewer from the F6Publishing system immediately.





2 Language evaluation: Classification: Grade B. A language editing certificate issued by Editage was provided.

3 Academic norms and rules: The authors provided the Biostatistics Review Certificate, the Institutional Review Board Approval Form, the Institutional animal care and use committee statement and the ARRIVE guidelines. No academic misconduct was found in the Bing search.

4 Supplementary comments: This is an invited manuscript. The study was supported by Fundação Coordenação de Aperfeiçoamento de Pessoal Superior. The topic has not previously been published in the WJSC.

5 Issues raised: Please provide the manuscript in MS Word format;

(2) The authors did not provide the approved grant application form(s). Please upload the approved grant application form(s) or funding agency copy of any approval document(s);

Response: This document was included.

(3) The authors did not provide original pictures. Please provide the original figure documents. Please prepare and arrange the figures using PowerPoint to ensure that all graphs or arrows or text portions can be reprocessed by the editor;

Response: It was done.

(4) PMID numbers are missing in the reference list. Please provide the PubMed numbers and DOI citation numbers to the reference list and list all authors of the references. For PMID and DOI numbers of references from English-language journals, please ensure there is a space between the PMID and DOI numbers in the square brackets. Please revise throughout;

Response: It was done.

(5) The “Article Highlights” section is missing. Please add the “Article Highlights” section at the end of the main text.

Response: It was done (see pages 19 and 20).

6 Recommendation: Conditional acceptance.

