26 October 2018

Dear editor and reviewers,

We are very pleased with the results of the evaluation conducted by the Scientific Committee of your renowned journal of our manuscript and have carried out the task of responding to the well-targeted and greatly appreciated observations provided by the reviewers.

In this document, please find, in yellow, the changes kindly suggested by the Reviewers, which now appear as changes in the new version of the manuscript.

Reviewer # 1 (Reviewer's code: 00607640)
SPECIFIC COMMENTS TO AUTHORS

Comments#1 The authors examined the effects of muscle fiber orientations on muscle metabolites using proton magnetic resonance spectroscopy. The results showed that the metabolite profiles in each orientation of muscle fibers to the main magnetic field were different and further suggested careful positioning is one important factor to consider in studying proton magnetic resonance spectroscopy metabolites in muscles. These observations are interesting, and overall results are supported by the data in present study. It is suitable to the Journal and could be helpful in clinic study.

Response: Thank you for suggestion.

Reviewer # 2 (Reviewer's code: 02566952)
SPECIFIC COMMENTS TO AUTHORS

Comments#1. Maybe it should be good to introduce the method of investigation within the title the effects of muscle fibber orientation to main magnetic field on muscle metabolite profiles for MRS acquisition or something similar.

Response: Thank you for suggestion. To introduce the ^{1}H-MRS, it has now been edited according to the comment of the reviewer, in the following manner:

The effects of muscle fiber orientation to main magnetic field on muscle metabolite profiles for MRS acquisition

Comments#2. Please verify if word "wildly" (savage, wild) is what needs to be used in the core tip or maybe rather widely (largely) as it appears in the introduction text.

Response: Thank you for suggestion. The word has now been edited according to the comment of the reviewer, in the following manner:

^{1}H MRS is a technique that is widely used for IMCL and EMCL quantification of muscles as evidenced in various studies.

Comments#3. How do the authors thins the patient should be positioned and what specific muscle types would be more relevant to investigate when trying to detect metabolic syndrome using IMCL/EMCL?
Response: Thank you for suggestion. The results showed that metabolic profiles of positioned at 30˚, 60˚, and 90˚ to the main magnetic field were change when compared to 0˚. Therefore, based on the results we suggested that position should be at 0˚. A specific muscle types is lower extremity muscles such as tibialis anterior, soleus, and gastrocnemius.

Comments#4. Do the authors think a practical protocol could be designed at this stage or further studies will be needed?
Response: Thank you for suggestion. The results showed important data on effect of position to metabolite profiles. Thus, we think that the further studies will be needed to study in human subject.

Comments#5. Given the fact MRS is non-invasive how feasible could be a clinical study in this respect?
Response: Thank you for suggestion. ^{1}H-MRS has been usually used to study metabolic changes in various diseases such as brain tumors and muscle. Thus ^{1}H-MRS could be a clinical study.

Reviewer # 3 (Reviewer's code: 02577402)
SPECIFIC COMMENTS TO AUTHORS
Comments#1. The language needs to improve because of some grammar, spelling and punctuation mistakes.
Response: Thank you for suggestion. It has now been edited according to the comment of the reviewer.

Comments#2. In the Statistical analysis section, please indicate the company, city and country of the manufacturer of the Origin version 8.
Response: Thank you for suggestion. It has now been edited according to the comment of the reviewer, in the following manner:
The data statistical analysis was done by Origin 8.0 software (OriginLab, Northampton, MA, USA).

Comments#3. Use of abbreviation: when using abbreviations, the full phrase should be given at the first time of use. Later, you can always use the abbreviation without mentioning the full phrase. For example, magnetic resonance spectroscopy (MRS).

However, the authors did not abide by this rule all the times. For example, the authors used NMR, SD without indicating the full phrase. Please check the whole article and correct all similar problems.
Response: Thank you for suggestion. It has now been edited according to the comment of the reviewer.

Comments#4. RESULTS: In the end of this section, the authors stated that "The comparisons between ------- was found to be significantly different than -----". Please indicate clearly how it was significantly different than that of other ratios and also give the significant P value. In this section, please state in clear and specific data.
Response: Thank you for suggestion. It has now been edited according to the comment of the reviewer, in the following manner:
The comparisons between 0° and at 30°, 60°, and 90° was done with Wilcoxon sign rank test and was found to be significantly different than that of EMCL and IMCL ratios were obtained from orientations at 0° in every muscle orientation (P-value <0.05).

Comments#5. Discussion: Please give some limitations before making a conclusion. At the end of the discussion, please use one or two sentences to make a conclusion which should be directly relevant to your results.
Response: Thank you for suggestion. It has now been edited according to the comment of the reviewer, in the following manner:
There are limitations in this study, such as the small number of samples and the small size of chicken muscles compared to human muscle. In conclusion, the metabolite profile changes are due to the muscle fiber orientation, which demonstrates that positioning potentially causes inaccuracies in 1H-MRS spectrum analysis.

Sincerely yours,

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