Format for ANSWERING REVIEWERS

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 16973-review.doc).

Title: Standardized quantitative measurements of wrist cartilage in healthy humans using 3T magnetic resonance imaging

Author: Jean-Vincent Zink, Philippe Souteyrand, Sandrine Guis, Christophe Chagnaud, Yann Le Fur, Daniela Militianu, Jean-Pierre Mattei, Michael Rozenbaum, Itzhak Rosner, Maxime Guye, Monique Bernard, David Bendahan

The manuscript has been improved according to the suggestions of reviewers:
Reviewed by 00467045

Standardised quantitative measurements of wrist cartilage in healthy humans
The authors present an interesting paper in a topic relevant to the WJO. There is some good discussion included on MRI and the different scanning sequences.
I recommend a minor revision and have some comments and minor changes for the authors listed below. After the manuscript has been revised, the authors may wish to get the paper checked by a native English speaker to refine the wording in some sections.
We would like to thank the reviewer for his constructive comments. The paper has been thoroughly checked by Professor I. Rosner who is a native English speaker, who grew up in the United states, trained and was on the staff at CWRU School of Medicine in Cleveland, Ohio.
Abstract: Can the authors write the acronyms in full in the Abstract? This has been done for CSA, but not for VIBE or DESS. It should be written in full again at the start of the paper.
    The acronyms have been explained in the abstract and detailed again in the text.

1. *Core content, bullet point 1: 'quantified in wrist' should be 'quantified in <u>the</u> wrist.'*
    The change was made in accordance with the reviewer's suggestion.

2. *Introduction: The sentence on lines 6-8 is not clear (it depicts joint anatomy topographically...).*
    The sentence has been slightly modified as follows : …. as it can provide a 3D dataset of joint anatomy…..

3. *Introduction, line 15 (p 6): Please write the acronym 'OMERACT' in full.*

This acronym has been explained.

4. *Introduction, line 17 (p 6): The words 'Thanks to' should be replaced by 'Due to' or similar as 'Thanks to' is too informal.*
The sentence has been changed in accordance with the reviewer's suggestion.

5. *Introduction, Paragraph 2, Lines 7-8 (p 7): Please reword 'before and after about an hour' slightly to make this section clearer. For example, 'requires repeated investigations prior to, and then after about an hour of contrast agent injection.'*
The sentence has been changed in accordance with the reviewer's suggestion.

6. *Subjects and Methods/MRI: Change 'in the supine position' to 'in a supine position.'*
The sentence has been changed in accordance with the reviewer's suggestion.

7. *MRI: It is difficult for the reader to visualise how the patient's wrists were scanned from the description. Are the authors able to include a photograph?*
Please find below a picture which could be added as the supplementary figure.

8. *MRI: 'Mostly used for abdominal investigations' would be better worded as 'predominately used for abdominal investigations.'*
The term "mostly" has been replaced by the term "mainly".

9. *MRI: The authors have stated that DESS was previously used for knee cartilage imaging. Is it not longer used or is the wording just unclear?*
"was previously" has been replaced by "has been" .

10. *MRI: Table 1 should have a capital 'T.' Can the authors please capitalise the first letter of all table and figure numbers?*
The changes have been made in accordance with the reviewer's suggestion.

11. *Cartilage segmentation and measurements: How was the cross-sectional area calculated in the thresholding process?*
Cartilage cross-sectional areas (CSA) between opposite bones in the carpal region were manually selected. Then a thresholding process was applied on the corresponding region so that only the voxels within the proper signal intensity setting were kept and counted. The cartilage CSA was automatically computed considering the image resolution, the slice thickness and the number of voxels.
These details have been added in the revised version of the manuscript.

12. *Cartilage segmentation and measurements: The authors state that the same (one) radiologist repeated the measurements, but in the results they show data for two operators, each performing the measurements twice. Can they please re-word the methods to reflect that there were two rather than one radiologist?*
This paragraph has been modified as follows: This segmentation process and the corresponding measurements were performed twice by the same experienced radiologist (JVZ, 3-month interval) and once by two senior radiologists (JVZ and PS). The measurements' reliability was investigated on the basis of the comparison between the two operators' measurements. The measurements' reproducibility was investigated via repeated measurements performed by the same operator.

13. *Results: The authors state that the average cartilage cross-sectional area from the VIBE and DESS sequences was 'slightly but significantly lower.' What was the p-value? Was it statistically significantly different and if so, I think the word 'slightly' should be omitted.*
In accordance with the reviewer's suggestion, we have added the p value (5.6×10^{-7}) and the word "slightly" eliminated.

14. *Can the authors please check the caption for Figure 3 as the wording is confusing i.e. which one is (A), (B) or (C)? Also, in Figures 3(A) and 3(B), the horizontal axis is 'cartilage bone height' when I think they mean carpal bone length. Can the authors please label the axes of Figure 3(c)? Which axis is VIBE and which axis is DESS?*
The legend of Figure 3 has been changed in accordance with the reviewer's suggestion

15. *Results (p 15): What do the authors mean by saying the VIBE and DESS measurements are significantly related? I think they mean that the correlation coefficient is high, or the correlation is strong? They have mentioned this again on p 17, where I think they mean high correlation (highly significant relationships - second paragraph on p 17).*
We indeed referred to the high correlation coefficient. The sentence has been changed accordingly.

16. *Results (p 15): The last sentence is unclear and should be reworded.*
The sentence has been modified in accordance with the reviewer's suggestion.

17. *Discussion (p 16): Please reword 'repeated measurements performed twice' as repeated means it was performed twice.*
18. **The sentence has been modified in accordance with the reviewer's suggestion.**
19.
20. *Discussion (p 17): In the second paragraph, what do the authors mean by cause-effect relationship? Can you please elaborate?*
The sentence has been modified as follows: The highly significant relationships reported in Figures 3A and B suggested that cartilage CSA would vary in the same way as the carpal bone length and that the corresponding ratio might be used as a normalized index.

21. *Discussion (p 17): Cannot stand alone as a diagnostic criterion for what?*
The sentence has been changed as follows: cannot stand alone as a diagnostic criterion of cartilage loss.

22. *Discussion (p 18): Please rephrase 'in the 3 dimensional space' to 'in three dimensional space.'*
This sentence has been modified in accordance with the reviewer's suggestion.

23. *Discussion (p 20): In Paragraph 2, what do the authors mean by 'readers' (mentioned twice)? Do they mean the researchers interpreting or measuring data from the MRI films? Please reword this.*
We actually meant 'experienced radiologists'. The sentence has been modified accordingly.

24. *Discussion (p 20): The authors state that one limitation is the small age range of subjects. How is this a limitation? Also, the age ranges are from 23-53 years of age, which I would not consider to be a small age range.*

Considering that cartilage loss can occur with age, we hypothesized that it would be of interest to confirm the linear relationship with older subjects i.e. above 55 y/o.

Reviewed by 00711004

General Comments: This manuscript presents a study to quantify cartilage cross-sectional area at the wrist in healthy subjects and the reproducibility of the proposed measurements. The study is in general well designed and clearly presented in the manuscript. However, the reviewer has some technical concerns below:

1) This is a manual measurement instead of a semi-automatic measurement method claimed by the authors. The reviewer did not see any semi-automatic image-processing method in the measurement.

While the slice selection from the 3D dataset and the selection of the cartilage area are manual, we considered the thresholding aspect as automatic. That is why we used the term "semi-automatic".

2) The measurement plane is determined using manual selection. As shown in Figure 1, the coronal and sagittal plane in B and C are calculated using the axis in A. This reconstructed image is a MPR image. Please clarify it.

None of the images have been calculated. In the first steps of the process, we aimed at selecting a given slice from the 3D dataset using a standardized approach in order to eventually quantify the cartilage CSA.

3) In the measurement of area, the authors indicated "a visual threshold" is applied. This threshold tends to be sensitive to the measurement results. In terms of Figure 2C, it might be possible to calculate a threshold using image-processing method instead of "a visual threshold". This may improve the reproducibility of the measurement.

We erred in not describing our approach with an appropriate choice of words. We indeed used an image processing method and we demonstrated that the corresponding reproducibility and reliability were very robust.

4) If the study can add some number of subjects with RA or OA, the comparison between normal subjects and diseased subjects will improve the significance of the study.

The utilization of this standardized method in patients was beyond the scope of this study. We aimed at this stage to describe the details of the standardized process together with the corresponding reproducibility and reliability. The investigation of patients with RA and OA will be the subject of a future study.

Minor comments:

1.   *P8, Subject mean age: 51, range 23 to 53. For 14 subjects, this is incorrect. Please check.*

In accordance with the reviewer's suggestion, we have checked the ages and the proper values are: range [30-58]; mean = 47.4 ± 8.9. This has been added in the revised version of the manuscript.

2.   *Table 2 Caption: CV1 and CV2 were mentioned but no data in the table.*

The legend of Table 2 has been modified.

3.   *The authors used the terms of CV, variation coefficient, coefficient of variation interchangeably through the manuscript. Please revise it.*

A single term has been used throughout the revised version of the manuscript.

Reviewed by 00458932

The manuscript is of interest. The task would be a challenging one particularly in patients with rheumatological conditions. There is some heterogeneity in the Reference list which should be corrected.

The references list has been modified accordingly.