Reviewer’s report

Title: MR Imaging Features of Orbital Langerhans Cell Histiocytosis

Version: 1 Date: 06 May 2019

Reviewer: Anton Lennikov

Reviewer's report:

Manuscript BOPH-D-19-00089R1 "MR Imaging Features of Orbital Langerhans Cell Histiocytosis" by Wu1et al., is a research article on the topic of diagnostic of Langerhans cell histiocytosis in orbit, manuscript present and summarises different localization and appearance of MRI findings during LCH. The findings of scientific interest and clinical relevance. However, the manuscript requires additional editing, proofreading, and addition of specific histological stainings to be considered for publication.

General comments:
1. The manuscript requires significant editing and proofreading by a native speaker or scientific editing company or preferably both as there are many spelling mistakes, several sentences sound disjointed or involve unusual phrasing and word choices in a multitude of instances across the manuscript.
2. Authors should carefully re-read the manuscript paying extra attention to the usage of spaces between words and sentences in many sentences spaces are missing in some places; there are unnecessary spaces introduced.
3. If authors have taken the photographs of the patients eyes during initial visit and have the authorization from the patients and or patients legal guardians it would be good to present images of the eyes overall look to further support MRI findings as it will benefit the manuscript primary goal to provide the reference for clinicians who may encounter orbital LCH patients.
4. As it a retrospective study authors may include the treatment strategy for orbital LCH and it's outcome.

Specific comments:
Page 1, lines 30-32
As the mean age of the patients was 6.3 years old authors should mention that written informed consent was obtained from patients parents or legal guardians where applicable. Which authors do in the declarations at the end of the article, but not in the statement at the beginning of the article.

Page 3, line 54
I do not think using word short in conjunction with the word diameter is correct.

Page 4, lines 16
"bolus injection of 0.2. mg/kg" - of what? The description and the sources of the contrast are missing.
Page 4, Results
Authors suggested reworking the description of patients age, sex, disease duration into the table.

Page 5, Location and morphology of lesions
Authors encouraged to add visual cues such as arrows or dotted margins to indicate the exact position of lesions on the MRI images. It will improve the understanding of the article by non-specialists in the field.

Page 5, Pre-contrast MRI presentation along with the pathological results
Authors describe hemosiderin and macrophages but only present H&E histology to support their statements. See additional experiments.

Page 6, line 54
"0.2-2.0% cases" - is it worldwide or in some locality? Please clarify.

Page 6, line 57 - "High incidence of the orbital involvement." Where is this data is coming from please provide the reference and please indicate percentage if available regarding the incidence of orbital involvement?

Page 7, line 9 - "boys" - male at or pre-adolescent males. "Boys" is not an appropriate word in the scientific text.

Page 7, lines 6-42 - please revise this fragment of text avoiding using vague terms such as "this group."

Page 7, 42-44 - "Regrettably we could not determined the specific type of rest cases because of the lack of whole body systemic details" - Please rework this sentence as it is confusing to the reader of what exactly authors meant. "Whole body systemic details" is even more confusing statement. Please be more specific of what are you referring to also appropriate grammar is required for sentence to be understandable.

Page 7, line 55 - Coincident - I assume intended meaning is "not consistent."

Page 7, line 58 - actually - Please rework this whole sentence.

Page 8, line 8 - were mostly "crumb or triangular" - I do not understand authors terminology here. Was intended meaning triangular? Is the intended meaning for crumb is porous in structure?

Page 9, line 17 - Authors talk about S100 and CD1a in differential diagnostics of LCH but only present general H&E staining in the figures. See additional experiments.

Page 9, line 32 "general conditions of the patients is poorer" - please revise this sentence to "Patients demonstrate symptoms outside the orbital region, consistent with primary tumor localization, along with typical malignancy associated general symptoms."

Page 9, line 36 - "abdominal primary tumors been found can help diagnosis" - Please revise this sentence to "Presence of the primary tumor is the main finding in the differential diagnostic of the condition."

Page 9, line 43 "worm-eaten" - While I understand what authors try to say, try to use a different synonym to this term such as deteriorated.

Page 10, lines 10-13 - "Osteomyelitis… …can be repaired if anti-inflammatory treatment timely" please revise this sentence it is just disjointed set of words especially in the second part of the sentence.

Page 10, lines 19 - Please spell the acronyms on the first use "Diffusion-weighted magnetic resonance imaging," and since authors have introduced the list of abbreviation, please put it in there. At the same time, the authors did not abbreviate dynamic enhanced scanning.

Figures:
Figure 1-3 Normal MRI should be introduced for comparison. Dotted plot or arrows should indicate the abnormal findings on MRI images for ease of the readers. It would significantly improve the ease of following the MRI images if authors could introduce drawn or 3D rendered artwork of the skull with the localization of abnormalities based on the MRI data.

Figure 4 - Specific stainings are required to make statements regarding cell populations, hemosiderin, and macrophages. H&E staining gives only general information as it is not specific staining for these cell types. Please introduce the scale bar into the images rather than staining the magnification.

Additional experiments:
Authors mention in the background of the article that distinctive marker for LCH is CD1a+ cells, CD1a+ indeed along with CD207 CD1a are specific markers for Langerhans cells, however, only present H&E staining to support their statements regarding the presence of Langerhans cells and hemosiderin in their work. Authors strongly encourage to perform the IHC with patients' histological samples using DAPI/CD1a, DAPI/CD207 (triple DAPI/CD1a/CD207 is preferable) and Prussian blue to accurately detect hemosiderin, i.e. using Iron Stain Kit (such as ab150674). This IHC is easy to perform as authors already have histological sections. If authors have no access to fluorescent microscopy of suitable quality DAB staining with visible light microscopy demonstrated by H&E figure is acceptable. If authors want to demonstrate the macrophages in their samples, they need to use CD14 or CD16 to identify the population positively. Moreover, the combination of CD14/Prussian blue or CD16/Prussian blue is required to identify the phagocytosis of hemosiderin by the macrophages. If possible authors also need to present a healthy orbital material biopsy (or post mortem tissue sample) as a control as well as metastatic orbital biopsy (or post-mortem tissue sample) for comparison.
**Are the methods appropriate and well described?**
If not, please specify what is required in your comments to the authors.

Yes

**Does the work include the necessary controls?**
If not, please specify which controls are required in your comments to the authors.

No

**Are the conclusions drawn adequately supported by the data shown?**
If not, please explain in your comments to the authors.

Yes

**Are you able to assess any statistics in the manuscript or would you recommend an additional statistical review?**
If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

Not relevant to this manuscript

**Quality of written English**
Please indicate the quality of language in the manuscript:

Needs some language corrections before being published

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