

August 10, 2014



Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 12025-review.doc).

Title: Early hepatic complications of Lysosomal acid lipase deficiency in Mexican siblings with new mutations in LIPA gene.

Author: Yuritzi Santillán-Hernández, Enory Almanza-Miranda, Winnie W. Xin, Kendrick Goss, Aurea Vera-Loaiza, María Teresa Gorráez-de la Mora, Raul E. Piña-Aguilar.

Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 12025

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

1 Format has been updated

2 Revision has been made according to the suggestions of the reviewers. See below for specific responses.

Reviewer 1 (02860875)

Thank you for asking me to review this manuscript. Santillán-Hernández et al have produced a case report of two siblings who were found to have cholesterol ester storage disorder. They have described the clinical scenario and identified that the two cases have a novel compound heterozygote genotype for the LIPA gene. The cases are described adequately, diagnosed appropriately and the novel genotype adds to the previously published literature.

Response: Thank you for the comments.

I have the following comments: Major 1. The written English needs to be improved. There are numerous spelling and grammatical errors.

Response: Thank you for the comment. The manuscript received an English review by a native speaker.

2. I am not sure that we can describe the clinical course here as unique; Bernstein et al described a spectrum of clinical courses that almost certainly reflects the functionality of the different mutations of the LIPA gene.

Response: Thank you for the comments. The uniqueness of the case is based in patients showed no detectable LAL activity in whole blood and leukocytes. This normally leads to an early fatal presentation, known as Wolman disease. The reported siblings expand the phenotype of LAL deficiency to an early hepatic presentation without a fatal outcome. Other interesting finding is the fact that missense mutations in exon 4 not generate complete absence of LAL activity. Normally the mutations that lead to undetectable activity are deletions, and non-sense mutations (premature stops).

I would tone down the references to early presentation and prolonged clinical course as somehow noteworthy.

Response: Thank you for the comment. We modified the text to present the cases according the suggestions of the reviewer.

3. Please redraft figure 3; I cannot see how a complete series of US images helps the description of the case. Similarly I am not sure that a complete description of the velocity of venous blood flow within the abdomen for both cases helps the flow of the report.

Response: Thank you for the comments. The purpose of presenting Doppler USG is to highlight the presence of a shunt that is uncommon in adult patients with severe hepatic cirrhosis. This shunt can be the explanation of a non-rapidly progressive course of esophageal varices and portal hypertension. The flow velocities were removed according the suggestions of the reviewer.

4. You should emphasise that the blood spot LAL values for the parents are sub-normal for the mother and low normal for the father. Have you performed an US on either parent?

Response: Thank you for the comments. The very low activity in the mother is another finding that contributes to the uniqueness of the cases. We requested several months ago for the parents to return for further evaluations (hepatic ultrasound and laboratory studies), they have refused follow-up care.

Minor 1. The OMIM number in the abstract is wrong; it should be 278000.

Response: Thank you for the comment. A number 2 was missing, it was corrected.

2. Please could we have reference values for your laboratory (Table 1)

Response: The missing references values were aggregated to the Table 1.

3. Did the CT scan of sibling 1 at age 4 demonstrate adrenal calcification?

Response: No adrenal calcifications were noted. This was specified in the CT description.

Reviewer 2 (02861012)

This study is very clear and well written. The authors show two new mutations in exon 4 of the LIPA gene, which encodes for lysosomal acid lipase (LAL). Clinically LAL deficiency is reported as having one of the two principal phenotypic presentations: the early onset, called Wolman disease (WD) and the late onset called cholesteryl ester storage disease (CESD). The new mutations described here cause a total deficiency of LAL activity and lead to a CESD presentation with early symptomatology and complications.

Response: Thank you for the comments.

Comments to authors: The authors should report the frequency of these new mutations in healthy controls, and whether there is difference in the presence of these mutations in individuals of different origin.

Response: Thank you for the comments. The mutations in exon 4 were not found in other LAL patients studied at Neurogenetics DNA Laboratory of Massachusetts General Hospital.

Perform mutation analysis in control subjects is an expensive procedure. At this point we in our Center in Mexico and our co-authors in MGH have not funding to perform this screening. However, those mutations are not present in the 1092 samples used in the Phase 1 of the project 1000 genomes that included populations with European Ancestry (Toscani and Utah residents with Northern and Western European ancestry), Asian Ancestry (Han Chinese, Japanese, Southern Han Chinese), African Ancestry (African Americans, Luhya and Yoruba) and American Ancestry (Colombians, Mexicans, and Puerto Ricans). Neither in the 6500 samples of individuals with European American and African American ethnicity processed in the NHLBI GO Exome Sequencing Project.

We included this information to answer the request by the reviewer in the corrected version.

Reviewer 3 (02860966)

Thank you for the opportunity to review this interesting paper.

Response: Thank you for the comment.

3 References and typesetting were corrected

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,

A handwritten signature in dark ink, appearing to be 'R. Piña-Aguilar', written in a cursive style.

Raul E. PIÑA-AGUILAR, MD

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