Reviewer’s report

Title: Hepcidin gene polymorphisms and iron overload in β-thalassemia major patients refractory to iron chelating therapy

Version: 0 Date: 19 Jun 2019

Reviewer: Marina Kleanthous

Reviewer's report:

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Comments

1. The 'single nucleotide polymorphisms (SNPs)' should be replaced in the text by 'single nucleotide variations (SNVs)'. SNV is used for both pathogenic and non-pathogenic variants.

2. In the introduction line 21: 'This study aimed to analyze the association of c.-582A>G (rs10421768), c.-153C>T (rs142126068), and c.-443C>T (rs117345431) with iron overload in major β thalassemia patients.' There is no justification in the introduction on why the c.-443C&T (rs117345431) SNV was selected in this study. Also replace the phrase 'major β thalassemia patients' with 'β-thalassemia major patients'

3. The way data for patients are presented in Table 1 is not useful. It should be removed or replaced with a. a table generally describing the patient cohort or b. a table with information about the subgroups of patients presented in Figures 1-3.

4. Table 1: No units are used for the parameters presented in the table

5. In Table 2 line 23 replace 9/8 with 9.8

6. Figures 1-3: The specific SNV should be included in the legend.

7. Figures 1-3: It is not described in the paper how the patients were divided into the four groups: mild, moderate, severe and very severe. Also there is no description of the normal individuals.
8. Figures 1-3: The presentation of data in figures 1-3 is very confusing. Dividing the patients in four groups do not allow for any meaningful statistical analysis.

9. Figures 1-3, upper right part of the figures: Based on genotypes, the patients should be divided into one group with the AA genotype and the other group with the AG or GG genotypes and NOT into one group with the AA or AG genotype and the other group with the GG genotype.

10. Discussion: the first and second paragraph can be transferred into the introduction

11. Statistical analysis should be repeated by comparing the clinical/biochemical parameters of the two groups of patients with the AA genotype and the AG or GG genotypes. A second option is to divide the patients into two groups based on the severity of clinical/biochemical parameters and compare the percentage of the different genotypes in each group, in that case the percentage of each one of the three possible genotypes is determined in each group separately.

12. Methods: a section describing the clinical parameters should be included

13. Table 3: the position of the primers on the reference sequence should be included in the table

Are the methods appropriate and well described?
If not, please specify what is required in your comments to the authors.

No

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

Unable to assess

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

No

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.

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