Reviewer's report

Title: The expression of sirtuins, superoxide dismutase, and lipid peroxidation status in peripheral blood from patients with diabetes and hypothyroidism

Version: 2 Date: 15 Nov 2018

Reviewer: Luis Miguel Roman-Pintos

Reviewer's report:

The present manuscript entitled "The expression of sirtuins and superoxide dismutase in peripheral blood from patients with diabetes and hypothyroidism" was submitted as a research article. The authors aimed to elucidate whether alterations on the expression levels of SIRT1, SIRT3, and SOD2 are related to the observed oxidative stress in type 1 and 2 diabetes, as well as hypothyroidism. The authors concluded that the changes in the expression of SIRT1, SIRT3, and SOD2 they found, may explain the failures of the defense mechanisms against oxidative stress in patients with diabetes and hypothyroidism.

Comments for the authors:

- Authors must be encouraged to follow the STROBE guidelines for cross-sectional studies.

- Please undertake some linguistic editing as there are several errors in all sections of the manuscript.

- In the Introduction section authors avoid the inclusion of TBARS as an objective of the study; however, in Methods, Results and Discussion section, the importance of this variable is evident in the text. Please include as part of the title and aim to support the information mentioned thereafter.

- Abbreviations should be defined at their first occurrence and used thereafter, specifically glycated hemoglobin and thiobarbituric acid reactive substances. Please, revise all sections.

- In the Methods section authors do not describe the setting, relevant dates, including periods of recruitment, samples processing, and data collection.

- Page 3 line 20. There’s no need to clarify the clinical meaning of HbA1c, since the paper is addressed for readers who are familiar with the terminology.

- The eligibility criteria are not precise, please rephrase and be more specific, e.g. define "severe disease complications" of diabetes and hypothyroidism.

- Page 4 line 9. Authors mention a threshold of HbA1c above 7% to diagnose diabetes mellitus; however, the criteria established by the American Diabetes Association is >6.5%. Please correct.
- Please explain how the study size was arrived at.

- Authors do not specify if the samples were processed immediately or stored, please be specific. Also how much blood was drawned for the analysis.

- In the data analysis section, authors mention the use of parametric tests (Student’s t test); however, no formal statistical analysis evaluating data distribution is shown as to sustain the use of parametric tests (Shapiro-Wilks, Kolmogorov-Smirnov). Moreover, qualitative variables are not taken into consideration in the writing of the section.

- Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study.

- Page 5 line 2. The first phrase in the Results section is redundant being that obviously HbA1c has to be higher in patients with diabetes compared to healthy subjects.

- Page 5 lines 8,9,10. The statement is already mentioned in the Methods section (RNA extraction).

- The levels for biochemical determinations and molecular markers should be written in the text and figures.

- In the Discussion section authors declare that "sirtruins and antioxidant enzymes might precede the development of diabetes in patients with HT." because the study was not designed to asseverate this statement. At least a prospective or retrospective study is needed to confirm this hypothesis.

- There are several confounding factors that that have to be considered before jumping into conclusions, such as duration of the disease and HbA1c (not established for hypothyroidism in table 1), TSH and cholesterol in all groups. This is an important limitation of the study and has to be mentioned in the manuscript.

- In table 1 qualitative variables must be expressed as n (%). The use of decimals has to be homologated. When p<0.001 there’s no need to report all the following decimals, p<0.001 should be used.

- Page 13 is blank, please comment if no information was missed out.

- Legends for figure 2 and 3 do not mention T1D group but it’s included in the figure.

- In figure 2, levels of SIRT3 mRNA expression are very similar for T1D and HT groups, however, the statistical significance is very different. Please double check your numbers. The same for figure 4 where levels of T2D and HT are very different but p values are the same.

- Whether patients with HT were taking medication (thyroid hormone) is not mentioned, and this fact can alter the results. Please clarify.
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.

No

Are the conclusions drawn adequately supported by the data shown?
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No

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I am able to assess the statistics

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