Author's response to reviews

Title: Protective effects of Harungana madagascariensis on the Red Blood Cells

Authors:

Prosper C N Biapa (prbiapa@yahoo.fr)
Horea Matei (hmatei@umfcluj.ro)
Ştefana Bălici (stefanabalici@yahoo.com)
Julius E Oben (juliusoben@hotmail.com)
Jeanne Y Ngogang (jngogang@yahoo.fr)

Version: 4 Date: 21 November 2012

Author's response to reviews: see over
The research theme I will like to publish “Protective effects of Harungana madagascariensis on the Red Blood Cells “ deals with the development of new drugs using natural product (medicinal plants), traditionally consumed by our population (Cameroonian) against diseases. This research targets the prevention or the treatment of hemolytic anemia from different origins. I can ensure you that this work is original and therefore could be published in the Journal “BMC Complementary and Alternative Medicine” since thereof considers articles on interventions and resources that complement or replace conventional therapies, with a specific emphasis on research that explores the biological mechanisms of action, as well as their efficacy, safety, costs, patterns of use and/or implementation.

A- Reviewer's report Answers.

Title: Protective effects of Harungana madagascariensis on the Red Blood Cells
Reviewer: Ilavarasan Raju

Question 1. The authors mentioned the animals but not mentioned the details of weight of the animals, sex of the animals details of animal purchase, details of animals feed manufacture and temperature and humidity of lab.

Answer: the question was answered in the revised manuscript

The scientific committee of the University of Yaoundé I and the Cameroon National Ethics Committee approved the experimental procedures. Male rats (200 g) were maintained on a 12h light/dark cycle, at room temperature (26 ºC) with a relative humidity of 25 %, were allowed free access to water and food (made-up with maize, fish, salt, vitamins, soybean and oil).

2. The author no ware mentioned the family of the plant used in the study. and the percentage yield of the extract.

Answer: the question was answered in the revised manuscript

The stem bark of Harungana madagascariensis, from the family of Hypericaceae, has been collected in Okola-Yaounde, in January 2010. The plant has been authenticated at the National Herbarium of Cameroon in Yaounde (Voucher specimen Nº 4224 HNC).

3. Experimental part mention the quantity of ccl4 and olive oil used
Answer: the quantities were mentioned in the tables

4. Invivo experiment, mention the detail experimental design in methodology part and mention how many animals used in each group.

Answer: all these has been summarized in table 1

5. Table 3 mention the dose of olive oil in group 1

Answer: is added in table 3

**Quality of written English:** Needs some language corrections before being published

Answer: has been improved by an expert

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**B- Reviewer's report answers**

**Title:** Protective effects of Harungana madagascariensis on the Red Blood Cells  
**Version:** 2  
**Date:** 3 September 2012  
**Reviewer:** Patcharee Boonsiri

**Level of interest:** An article whose findings are important to those with closely related research interests.

**Quality of written English:** Needs some language corrections before being published

Answer: has been improved by an expert

**about the figures’ format:**

Answer: this has been done

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**C- Reviewer's report**

This manuscript is interesting. There are some parts that should be improved to make this manuscript perfect. In my opinion, this manuscript should be accepted after moderate essential revisions.

1. Is the question posed by the authors well defined?
Yes. There are some studies about this herb, for example, nephroprotective effect and also acute and repeated acetaminophen hepatotoxic rats. The authors reported protective effect of this herb on red blood cells.

2. Are the methods appropriate and well described?
The methods are appropriate but the authors wrote some methods with lots of details but some without details.

Please give more details for

red blood cell water permeability method
Answer: the question was answered in the revised manuscript

membrane lipid extraction
Answer: the question was answered in the revised manuscript

malondialdehyde (MDA) measurement
Answer: the question was answered in the revised manuscript

membrane cholesterol evaluation
Answer: the question was answered in the revised manuscript

assay of catalase and superoxide dismutase activities
Answer: the question was answered in the revised manuscript

3. Are the data sound?
The data are sound but the authors must improve the way of presentation the results because there are too many Figures and Tables. For Figures
Figure 1-6 can be put together in only one Figure and give a), b), c), d), e), f) for each of them.
Answer: done

Give more clear figure legend.
Answer: the question was answered in the revised manuscript.

In Figure 1,2,5,6 the authors used 10 m but in Figure 3, 4 you used 20 m. If possible, please select the same size.
Answer: The 20μm scale was used to emphasize to what extent cells were disrupted by CCl4 or protected by the plant extract.

For Tables
Table 1,2,3 may be written as descriptive in the method part.
Answer: in that way the text would have been too long, we preferred to put them together in the same table 1, with an appropriate legend

The results in Figure 10,11,12 are better show in a Table (not Figures) with statistical significant remark.
Answer: they are put together as table 3

Table 4 can be described in the result part.
Answer: the question was answered in the revised manuscript

Table 5. Is it necessary to give the information of total number of picture (NP) = 23?
Answer: the question was answered in the revised manuscript

4. Does the manuscript adhere to the relevant standards for reporting and data deposition?

5. Are the discussion and conclusions well balanced and adequately supported by the data?
The author should discuss about NMR results in discussion part.
Answer: the question was answered in the revised manuscript

6. Are limitations of the work clearly stated?
No.
Answer: the question was answered in the revised manuscript

Due to the high specificity techniques deployed results obtained by us were focused on certain parameters and this might be viewed as a limitation of this study. Future work needs to be conducted for further investigation of the mechanism of Harungana madagascariensis on the RBCs membrane using complementary techniques.

7. Do the authors clearly acknowledge any work upon which they are building, both published and unpublished?
Yes.

8. Do the title and abstract accurately convey what has been found?
To make the title of this manuscript very clear, I suggest to change to “Protective effects of stem bark of Harungana madagascariensis on the red blood cell membrane”.
Answer: done
In abstract part, conclusion is not a real conclusion. It is about the ethics and methods. Please correct this.
Answer: a slight adjustment has been done. We find it brief and comprehensive since it answers to our general objective.

9. Is the writing acceptable?
The authors must correct the gramma and check spelling.
Answer: done by an expert

9.1 spelling mistakes:
Malonodialdehyde and malonodialdehydro must be “malondialdehyde”. Please replace the correct one because there are many wrong spelling in this manuscript.
RMN : Nuclear Magnetic Resonance must be “NMR : Nuclear Magnetic Resonance”. Please find it in List of abbreviations.
Kj should be KJ (kiloJoule)
figure and table must be Figure” and “Table”.
Reference must be “References”
Answer: done (kJ)

9.2 In abstract, please check “The fresh human washed RBCs with carbone tetrachloride (CCl4). What purpose do the authors want to report? The word “carbone” must be “carbon”.
Answer: has been corrected

9.3 List of abbreviation should be run in alphabetical order. No abbreviation for BW (body weight) and KJ in the list.
Answer: done (bw)
kJ – measurement unit

9.4 The authors should check the format of References part. Here, there are variation in Reference format.
Answer: has been corrected