Title: Ethyl acetate extract of germinated brown rice attenuates hydrogen peroxide-induced oxidative stress in human SH-SY5Y neuroblastoma cells: Role of anti-apoptotic, pro-survival and antioxidant genes.

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Version: 3 Date: 19 May 2013

Author's response to reviews: see over
Reviewer's report
Title: Ethyl acetate extract of germinated brown rice attenuates hydrogen peroxide-induced oxidative stress in human SH-SY5Y neuroblastoma cells: Role of anti-apoptotic, pro-survival and antioxidant genes.

Version: 1 Date: 29 March 2013
Reviewer: Hande Sipahi

Reviewer's report:
Following minor revisions should be made:

1- Page 1, line 61, "any damage" is better.
   Ans: The word “any” has been inserted into the text.

2- Page 4, line 134, Na2CO3 should be changed with subscript
   Ans: The numbers have been changed into subscript.

3- Page 4, line 142, after "Bhd." you do not need to use comma
   Ans: The comma has been removed from the text.

3- Page 4, line 143, "Miami, FL" you should use the same font
   Ans: The font has been standardized.

4- Page 5, line 154, preparation of extract: is it same extraction procedure for BR?
   Ans: Yes, the extraction procedure is the same for BR. Changes has been made accordingly throughout the text to address this matter and for clarity of the readers.

5- Page 6, line 198, "5 % CO2 and 95 % air " is better
   Ans: The word “and” has been inserted into the sentence.

6- Page 8, Statistical analysis: Why did you prefer ANOVA. Are the samples drawn from a normal distribution?
   Ans: The results were analysed using ANOVA since the study design involves more than 2 conditions for comparison i.e. untreated control, H2O2-treated control and GBR-treated group which consists of different concentrations (1, 5 and 10 ppm). As for ANOVA to be conducted, the sample populations need to be normally distributed.

7- You did not mention Figure 5 in the article.
   Ans: Figure 5 has been mentioned in the article.

8- Line 278, references should be changed as "10, 11, 12"
   Line 285 and 298, references should be changed as "14, 15" and "3, 4" And you should control entire manuscript for references.
   Ans: The numberings for the references has been corrected.

Level of interest: An article of outstanding merit and interest in its field
Quality of written English: Acceptable

Statistical review: Yes, and I have assessed the statistics in my report.
Reviewer’s report

Title: Ethyl acetate extract of germinated brown rice attenuates hydrogen peroxide-induced oxidative stress in human SH-SY5Y neuroblastoma cells: Role of anti-apoptotic, pro-survival and antioxidant genes.

Version: 1 Date: 2 April 2013
Reviewer: NAGARAJA HALEAGRAHARA

Reviewer’s report:

Manuscript: Ethyl acetate extract of germinated brown rice attenuates hydrogen peroxide-induced oxidative stress in human SH-SY5Y neuroblastoma cells: 3 Role of anti-apoptotic, pro-survival and antioxidant genes.

1. Is the question posed by the authors well defined?
The research question is not well defined. Authors should provide a clear research hypothesis and the aim/objective of the study in the last paragraph in the introduction section.

Ans: A statement outlining the hypothesis and objectives of the study has been added in the introduction section.

2. Are the methods appropriate and well described?
Yes, the research methods are appropriate and described clearly.

3. Are the data sound?
The research data has been analyzed with appropriate statistical analyses. But the authors have to check the comparison between the groups again. The statistical significance between the groups have not been identified clearly in the bar diagrams. # p < 0.05 versus control has not been presented in Figure 1. BR+250M H2O2 at 15ppm has a high SD and has P<0.05 significance to control. Authors should check the comparison again to confirm this. Figure 4 and 5 comparisons are not clearly stated in the Figure legends. Authors should spell out clearly the what comparisons the different letters on the bars [a, b, c] represent. Some of the groups [Fig 5 AKT1 expression] has very high SD but still the figure shows P<0.05 significance with other groups. This part of the comparison needs a relook.

Ans: The statistical analyses have been revised and the relevant descriptions have been updated too. The letters, when different on any 2 bars of a particular gene, indicate that there is a significant different between the 2 groups of the same gene. On Figure 5, it is true that the AKT1 expression due to GBR EA 1 ppm is not different from the controls due to its high SD, but even after revision, we found it to be different from GBR EA 10 ppm.

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

5. Are the discussion and conclusions well balanced and adequately supported by the data?
Yes, the discussion is well written and adequately supported by the data.

6. Are limitations of the work clearly stated?
No, there is no clear statements regarding the limitations of the study.

**Ans:** the major limitation for the current study is that we could not determine the expression of the proteins for each gene studied because protein expression studies fall outside the scope of the project due to limitation of funds.

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

8. Do the title and abstract accurately convey what has been found?
Yes, title clearly depicts the aim and the study conclusion.

9. Is the writing acceptable?
Writing is acceptable.

10. Discretionary Revisions (which are recommendations for improvement but which the author can choose to ignore)
  • Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

The authors have to check the comparison between the groups again. The statistical significance between the groups have not been identified clearly in the bar diagrams. # p < 0.05 versus control has not been presented in Figure 1. BR+250M H2O2 at 15ppm has a high SD and has P<0.05 significance to control. Authors should check the comparison again to confirm this. Figure 4 and 5 comparisons are not clearly stated in the Figure legends. Authors should spell out clearly the what comparisons the different letters on the bars [a, b, c] represent. Some of the groups [Fig 5 AKT1 expression] has very high SD but still the figure shows P<0.05 significance with other groups. This part of the comparison needs a relook.

**Ans:** The statistical analyses have been revised and the relevant descriptions have been updated too. The letters, when different on any 2 bars of a particular gene, indicate that there is a significant different between the 2 groups of the same gene. On Figure 5, it is true that the AKT 1 expression due to GBR EA 1 ppm is not different from the controls due to its high SD, but even after revision, we found it to be different from GBR EA 10 ppm.

Other Comments:
11. Expand BR at its first use in the abstract.

**Ans:** The correction has been made in the abstract.

12. Expand all the abbreviations in the abstract at its first use.

**Ans:** All abbreviations have been expanded in the abstract at its first use.

13. Aim in the abstract says the study explored the potential antioxidant effect of GBR, leading upto Alzhiemers. But the conclusion in the end didn’t mention anything about it.
The conclusion has been reviewed and the implication of GBR’s high antioxidant potentials on neurodegerative diseases like AD has been stated.

14. Authors should provide few introductory points regarding human neuroblastoma cells.
**Ans:** Introductory points on human neuroblastoma cells have been added.

15. Introduction – line 101…. correct this sentence.
**Ans:** This sentence has been revised

16. Introduction – line 113….correct this sentence.
**Ans:** This sentence has been revised

17. Methods – Line 152….correct 50OC
**Ans:** The correction has been made.

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

**Quality of written English:** Acceptable

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:** I declare that I have no competing interests