Reviewer’s report

Title: Antioxidant Activity of Selenium-enriched Chrysomyia megacephala (Fabricius) Larvae Powder and Its Impact on Intestinal Microflora in D-Galactose Induced Aging Mice

Version: 1 Date: 14 Jun 2020

Reviewer's report:

Very interesting manuscript to read showing good analysis. I have the following comments

- What could account for the non-significant change in gene expression of Nrf2 while protein expression is significantly different in most cases compared to the model group

1. Letter e was omitted in megacephala in the title
2. Line 18 change clarify to assess except if there as there a controversy. Then a sentence or 2 should be added in the background showing the controversy is needed. Same thing with line 92
3. Line 19 Should be Selenium-enriched ("ed" is missing in enrich). This error is recurrent in the manuscript. Correct everywhere
4. Line 24 add S to CML
5. Line 29 Remove And at the beginning of the sentence
6. Line 30 "W" in western should be lower case. Correct throughout manuscript
7. Line 31 add respectively after gene
8. Line 41 in vivo should be italicized. Same thing in line 47, 90. Correct everywhere
9. Line 49 remove "and"
10. Line 51 SOD, GSH-Px, & VE should be written in full and acronyms added
11. Line 57 remove space before fullstop
12. Line 63 Write D-gal in full followed the short form
13. Line 74 change associations to associated
14. Line 89. Include reference of the previous work
15. Line 91 Write 1 or 2 sentences to show the link between oxidative stress and gut microbiota with references
16. Line 94 change the to a
17. Line 99, 106, 107 change America to USA. Indicate city
18. Line 115 and throughout manuscript. "et al." should be in italics except if journal expressly doesn't mind
19. Line 118. Why were 5 weeks old mice used? Are they not very young?
20. Line 126. How many times were mice treated with different substances. It should be mentioned & how long were they kept after treatment
21. Line 131 change oxidation to oxidative stress
22. Line 132 sentence will be clearer if changed to Animals in NC and model groups received water, and animals in the other groups previously described received vitamin E or SCML by intragastric gavage.
23. Line 133 correct concluded
24. Line 136 Micro should be properly written. Correct throughout text and figure legends.

Line 614
25. Line 142. Mention precise when mice were sacrificed
26. Line 147 add space between ph and 7
27. Line 161 correct exaction
28. Line 165 one step of PCR is missing
29. Line 177 Is the dilution for all the 3 antibodies, then it should be indicated
30. Line 180 Methodology for quantification of protein expression is not given
31. Line 186 change "then to run PCR" to "a PCR was performed"
32. Line 191 Write OTU in full
33. Line 198 Correct ANOVE
34. Line 206 remove "were" before receded
35. Line 209 Change "as compare with" to "compared to" throughout the text
36. Line 210 p<0.05 is written without space in between unlike in the figure legends. Be consistent how you write it throughout the manuscript
37. Lines 212 - 217 Refer to fig panels (2A, 2B & 2C) in the text for easy reference
38. Line 225 Here you use kg and in the figure legends, you use Kg. Be consistent throughout the manuscript
39. Line 243 Remove "and". Start the sentence with Compared to
40. Line 261 There is no prior introduction to Nrf2, so you should clearly mention that it is an oxidative stress gene
41. Line 262 Refer to fig 5A for easy referencing
42. Line 266, 288 if you say significantly increase or decrease, you must add the group you are compared to
43. Line 278 Give the importance or relationship of Keap-1 & HO-1 to Nrf2
44. Line 288 Including comma in numbers makes it easier to follow. Write 1,231,539 and 34,209
45. Line 296 If you don't present p values you can't talk of significant. Same with line 302
46. Line 305 Style of writing heading is different from previous headings (capitalization of first letters). Same thing with line 333
47. Line 308 Replace ~ with"-". ~ means approximately. Change in subsequent lines
48. Line 309 Refer to fig 8A. Line 311 refer to fig 8B
49. Line 336 add "and" between Clostridium & Helicobacter
50. Line 339 Replace wasn't with "was no"
51. Line 343 use significant only when backed by p values
52. Line347-349 Rewrite this sentence to clearly bring out its purpose. It appears incomplete
53. Line 350 correct weigh
54. Line 352 there should be no hyphen (-) between gal and injection
55. Line 262 "our work" could refer to previous work. Use "this work" to be more specific. Same thing with line 373
56. Line 363 add "and" before stabilize
57. Line 379 L. in L. casei should be written in full and L casei in italics
58. Line 375 Relationship between Keap1 and Nrf should be stated
59. Line 398 M in microbiome should be lower case
60. Line 409 change result in delay to "resulting in delayed aging"
61. Line 409 Replace those with these
62. Line 412-414 Thickening of intestinal wall is associated more with pathological than beneficial conditions, so I don't see how beneficial this could be. To make a strong statement of thickness of cecal wall, you need to analyze wall thickness and show statistical data. Please include all comments for the authors in this box rather than uploading your report as an attachment. Please only upload as attachments annotated versions of manuscripts, graphs, supporting materials or other aspects of your report which cannot be included in a text format.

Please overwrite this text when adding your comments to the authors.

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

Yes

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

Yes

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

Yes

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If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

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