Reviewer's report

Title: Fucoidan present in brown algae induces apoptosis of HT-29 human colon cancer cells

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Reviewer: Suguru Fukahori

Reviewer's report:

This manuscript describes the anti-tumor effect of fucoidan via both the death receptor-mediated and mitochondria-mediated apoptotic pathway in vitro using HT-29 human colon cancer cells. This report is well written in the detail of apoptotic pathway induced by fucoidan treatment and appears to be worthwhile to be published in this journal, but need some corrections at several parts as follows.

Major Compulsory Revisions

1. Materials
   (p8, line3-4) The reagents employed in this study were purchased from the indicated suppliers: fucoidan.
   The author did not explain the details of fucoidan employed in this study, so should explain how fucoidan is prepared from the seaweed and the details of major components of fucoidan such as L-fucose and sulfate content if known as the previous literature showed that the proportion of these components in the fucoidan plays a great role in the anti-tumor effect.

2. Discussion
   (p16, line21-22, p17, line6-9) The author noted that fucoidan induced apoptosis in HT-29 cells at low concentration, so should add descriptions how the mechanism of apoptosis in this study differ from previous reports in terms of the components of fucoidan.

Discretionary Revisions

3. In results, all obtained data is from only HT-29 human colon cancer cells except Cell culture and cell viability assay. To increase a reliability of remaining studies’ data, the author should obtain the data from HFC human normal colon epithelial cells, too if it is physically possible.

Minor Essential Revisions

4. Cell culture and cell viability assay
   (p8, line10) …with various concentrations of fucoidan.........
   Exact concentrations should be described.
5. (p8, line22) …with various concentrations of fucoidan…………
Same comment.

6. Flow cytometry measurement of mitochondrial membrane potential
(p10, line5) …with various concentrations of fucoidan…………
Same comment.

7. Western blot analysis
(p10, line13-14) We cultured the cells in 100mm dishes and administered fucoidan to them, as described above.
Same comment.

**Level of interest:** An article of importance in its field

**Quality of written English:** Acceptable

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