Author's response to reviews

Title: Selenium enrichment of broccoli sprout extract increases chemosensitivity and apoptosis of LNCaP prostate cancer cells

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Author's response to reviews: see over
August 17th, 2009

Dear Doctors,

Please find the revised version of our manuscript titled “Selenium enrichment of broccoli sprout extract increases chemosensitivity and apoptosis of LNCaP prostate cancer cells”. We have revised the manuscript based on the comments, corrections and suggestions from the reviewers (details as attached). The written English of this revised manuscript have been edited by American Journal Expert, a professional copyediting service (certificate attached).

There are no conflicts of interest of the authors in this manuscript. This manuscript has not been published and is not concurrently under consideration elsewhere.

All of the authors of this study have contributed significantly and agreed to submit this manuscript to BMC Cancer. We believe that BMC Cancer is the ideal media for this manuscript. We would be very grateful if this manuscript could be considered for publication in BMC Cancer.

Sincerely,

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Dear Editor/Reviewer,

Thank you very much for your valuable advices for our manuscript. Based on your advices, we revised the manuscript as follows:

The presentation of drug sensitivity assay results on page 12 (1st paragraph of section) was very confusing and should be re-written.
The paragraph has been re-written. The IC$_{50}$ concentrations of Sulforaphane and Se-methylselenocysteine in CSp and SeSp on each cell lines are now summarized in Supplementary Table 3. (revised manuscript page 12 and Supplementary Table 3)

P 17 you make that statement that Reducing PSA secretion may represent a promising approach to prostate cancer prevention. What justification is there for this statement? PSA is a biomarker and a diagnostic. What evidence is there that reducing the secretion of this marker is an approach for prevention? (It may be an indicator that you have successfully intervened -- although cell culture studies are only suggestive, at best).
We agreed that our statement was overestimated. Therefore, in this revised manuscript, the overstatement sentence has been deleted. (revised manuscript page 18)

P 18-19 You say that Finley and co-workers first introduced that rats were protected from chemically induced mammary tumor development by Se-enrichment of broccoli sprouts. This implies something inaccurate. Fahey and co-workers, in 1997 (Proc Nat Acad Sci), had already reported that broccoli sprouts [without Se enrichment] protected rats from chemically induced mammary tumor development. This paper should be referenced.
Thank you very much for your correction, the mentioned paragraph has been modified and the suggested reference has been referred. (revised manuscript page 19)

p. 19 (top) I believe that the closing sentences in the Results section are not supported by your experimental evidence.
Thank you very much for your correction, the sentence is now the opening sentence of “conclusion”, with the supporting references added. (revised manuscript page 20)
Table 2 (supplemental) Is there a significant difference between the level of sulforaphane in the control sprouts (375.87 uM) and that in the selenium enriched sprouts (316.53 uM)?

No, there were no significant difference between the level of sulforaphane in CSp and SeSp, as we mentioned on the result section. (revised manuscript page 12 and Supplementary Table 2)

Many relevant references have been ignored in the discussion of mechanism. To suggest just a few:

Bhamre et al. (2009) Prostate
Traka et al. (2008) PLoS ONE
Myzak et al. (2007) Exper Biol Med
Shankar et al. (2008) Clin Cancer Res

Thank you very much for the suggestions. The suggested references have been added to the discussion.

There is no discussion of the statistical significance of any of your data. While not necessary in many cases (e.g. Figs 1 & 2), isn’t there need to have at least a passing mention made of the significance of differences in fig 3B and 4? No mention of statistical treatment of data is made in the Methods section and apart from error bars on some of your plots, no other signs that there was a method for determining that observed differences are real differences.

In this revised manuscript, the difference of the effects induced by CSp and SeSp to the PARP, p-Akt and p-mTOR protein expression in figure 4, were analyzed by two-way ANOVA, as it is explained in the method section accordingly. For figure 3B, we only added the error bar and did not analyze the significance of difference. Here, our intention is to shows that LNCaP cells tend to return to their normal cycle upon release from CSp treatment.

Figure 3B consists of four sets of distribution data (0h, 24h, 48h and 72 h; in percentage). Each set consist of inter-dependent cell phases (i.e. subG1+G1+G2/M is always approximately 100%), thus, it’s not appropriate to analyze the significant of difference of a phase from a dataset to the same phase of other dataset because the value of one phase is always affected by other phases of the same dataset.

One of your suggested reference (Bhamre et al., Prostate, 2009) indeed calculate the significance of difference of their cell cycle data similar to figure 3B, by comparing the
percentage of each phase in every treatment to the same phase of control group, however, from the statistical point of view it is theoretically not valid.

We hope you will satisfy with this revised manuscript.

Sincerely,
Authors