Author's response to reviews

Title: Methylseleninic acid restricts tumor growth in nude mice model of metastatic breast cancer probably via inhibiting angiopoietin-2

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Author's response to reviews: see over
April 28, 2012

Dr. Christna Chap
Executive Editor
BMC Cancer

RE: MS: 1227270485664493

Dear Dr. Chap,

I am writing to submit the revised manuscript titled "Methylseleninic acid restricts humor growth in nude mice model of metastatic breast cancer probably via inhibiting angiopoietin-2" (MS: 1227270485664493) for your consideration of publication.

We appreciate the constructive comments of both reviewers, which helped to improve the quality of the manuscript. Detailed responses to the reviewer’s comment are in the next page of the letter. We missed the extended date of revision submission, mostly due to an additional experiment conducted. I have to apologize for this delay.

In the revised manuscripts, we followed formatting requirements from your editorial office and added the section of author contributions, as well as competing interests. We also formatted the manuscript according to the instructions given on the web of manuscript submission site. We also added the sequences of 6 siRNAs used for the RNA interference as the supplementary materials (Table S1).

Please let me know if anything else I need to do to ensure the quality of the manuscript.

Sincerely,

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Response to Reviewer’s comments

Reviewer 1: Tao Zhu

Reviewer’s report:
Wu et al. made use of a ER-/metastatic breast cancer model to characterize the functionality of methylseleninic acid (MSeA). The study revealed Ang-2/Tie2 and VEGF pathways contribute to the anti-tumor effect of MSeA. The study was well-designed with proper control and data was revealing for further investigation of MSeA for possible breast cancer treatment or/and prevention. The English writing was appropriate in general as well.

Reviewer 2: Changqing Su

Reviewer’s report:
In this manuscript, the authors investigated the expression levels of Ang-2 and VEGF on mammary cancer cell line MDA-MB-231 and its xenograft tumors of nude mice after treated with MSeA. They found that treatment with MSeA caused a significant reduction of Ang-2 mRNA transcripts and secretion of Ang-2 proteins, as well as VEGF, in cancer cells. They concluded that MSeA exerts anti-tumor effects partly by inhibiting the Ang-2/Tie2 pathway via inhibiting VEGF. Overall, data in the paper are clearly presented and the paper is well-written, however, the following issues need to be addressed.

Major Comment:
1. VEGF expression was examined by Western blot in cells, and by immunohistochemistry in xenografts. But these two methods are not quantitative, and the conclusion of VEGF expression change needs the quantitative data.

Response: The quantitative data for VEGF expression was now added as Fig. 2B.

2. The authors investigated the expression levels of Ang-2 and VEGF on mammary cancer cell line MDA-MB-231 and its xenograft tumors, and found both of them decreased at expression levels, so they concluded MSeA inhibits Ang-2/Tie2 pathway via inhibiting VEGF. The conclusion is not enough demonstrated. The relationship between their superior and subordinate places need to be further investigated, maybe the change of VEGF is only a concomitant phenomenon. Therefore, the manuscript needs major revisions.

Response: In an effort to address the questions raised by the Reviewer 2, we designed and conducted an additional siRNA experiment to elucidate the relationship between VEGF and Ang-2. Gene specific siRNA was designed and synthesized commercially,
and used to suppress the mRNA expression of VEGF and Ang-2, in sequential, and to assess the level of both target genes relative to the non-siRNA control. The data is now included in the manuscript as the Figure 7.

Comments of Reviewer 2 helped to improve the quality of the manuscript.