Author's response to reviews

Title: Flavones inhibit breast cancer proliferation through the Akt/FOXO3a signaling pathway

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Author's response to reviews: see over
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Author’s response to reviews: see over
Reviewer's report
Title: Flavones inhibit breast cancer proliferation through the Akt/FOXO3a signaling pathway
Version: 4 Date: 3 October 2015
Reviewer: Wangzhi Li

Reviewer's report:
1. The revised manuscripts addressed many of my early concerns and improved the interpretation and writing. However, the authors' response to comments on Figure 4 don't really address the questions and concern: FOXO3a increase is well ahead of suppression of pAkt Ser-473 and the authors don't have sufficient data to show FOXO3a nuclear location is induced by flavone treatment (the data given can only indicate an increase of both cytosol and nuclear increase of FOXO3a). However, in the Results part of the Abstract session, the authors assert that "Flavone, apigenin and luteolin induced forkhead box O3 (FOXO3a) expression and its nuclear localization by inhibiting Phosphoinositide 3-kinase (PI3K) and protein kinase B (PKB)/Akt", this is not accurate and unacceptable for publishing. I would open to accept a revised manuscript if the authors either add more data to support the assertion or the authors revise the assertion.

Akt pathway is an essential molecule for cell survival and growth during development and carcinogenesis. It is a serine-threonine kinase that is regulated mainly by the second messenger PI3K. In addition, Akt regulates cell cycle and proliferation by directly targeting CDKIs p21 and p27, and indirectly by modulating levels of cyclin D1 and p53. Akt promotes cell survival, which involves direct inhibition of pro-apoptotic signals such as FOXO3a.

As mentioned by the reviewer, we did not have direct evidence that FOXO3a translocate from cytoplasm to nucleus. We have modified the assertion according to the suggestion by reviewer. The modified words are marked in red words in the manuscript and are listed below:

Abstract, Paragraph 1, line 19-21
Flavone, apigenin and luteolin induced forkhead box O3 (FOXO3a) expression by inhibiting Phosphoinositide 3-kinase (PI3K) and protein kinase B (PKB/Akt).

Result, Paragraph 5, line 5-8
To investigate whether flavone, apigenin and luteolin affect the FOXO3a expression in breast cancer cells, we performed western blot analyses on the nuclear and cytoplasmic fractions of MCF-7 cells treated with the IC_{50} concentrations of flavone, apigenin, and luteolin for 48 h.

Result, Paragraph 6, line 1-4
FOXO3a is downstream target of the Akt. Akt kinase regulates breast cancer proliferation and survival. Inhibiting Akt phosphorylation modulates the activities of FOXO3a and subsequently affects cell proliferation, apoptosis, and differentiation.

We have read though the entire manuscript and made sure there are no typos and errors.

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

**Quality of written English:** Needs some language corrections before being published

**Statistical review:** Yes, but I do not feel adequately qualified to assess the statistics.

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