Reviewer’s report

Title: Cucurbitacin B inhibits human breast cancer cell proliferation through disruption of microtubule polymerization and nucleophosmin/B23 translocation

Version: 1 Date: 29 April 2012

Reviewer: Gail Fraizer

Reviewer’s report:

This clearly written paper is focused on a very important problem: improving breast cancer therapy. The authors have clearly described experiments and methods designed to characterize the response of breast cancer cells to Cucurbitacin B. Overall the manuscript provides compelling evidence for the utility of Cucurbitacin B as an adjuvant therapy. Results presented show very clear evidence of the anti proliferative and apoptotic effect of Cucurbitacin B on breast cancer cells. Most data is consistent and confirmed by multiple approaches.

Discretionary Revisions:
One weakness is the immunofluorescent images in Figure 6, showing the shift in location of Nucleophosmin/B23 from the nucleolus to nucleoplasm. For untreated cells very little NPM staining is observed and it is localized, unlike the staining in treated cells that is strong and diffusely spread through nucleoplasm. The conclusion that treatment is associated with altered trafficking of NPM seems sound, but a better image might show nucleolar staining better. Also since treatment seems to greatly increase expression detected by immunofluorescence, the author may want to discuss in light of the modest decrease in total protein levels shown by western analysis.

Major Revisions:
Another weakness is the lack of in vitro evidence supporting disruption of microtubular polymerization in Figure 8. The author’s conclusion that this indicates a lack of direct interaction seems sound. However the immunofluorescent images showing alpha tubulin staining are difficult to interpret, perhaps a higher magnification would better reveal changes in staining patterns in MDA-MC-231 cells. The authors may want to discuss the relative increase in alpha tubulin staining in the treated MCF-7 cells.

Minor Essential Revisions:
Figure legend for western described results as the average of three experiments, but quantitation is not shown.
Last sentence in results describing Figure 5 B is unclear.
Typos in discussion:
“MDA-MB-231 was choosed for study” should be “chosen”
“Seldom-filamentous normal microtubule structure could be seen” is unclear
“The addition of paclitaxel was shown to increase the polymeric intensity of the reaction which vincristine was shown to decrease this polymerization.” is unclear

Conclusion
Overall these data convincingly demonstrate anti proliferative and apoptotic effects of Cucurbitacin B. There is also evidence to support the authors conclusions that NPM expression is altered, but the mechanism involved is not as clear due to difficult to interpret immunofluorescent images. If speculation regarding polymerization of the microtubule network were better supported, this interesting manuscript would be greatly improved.

Level of interest: An article of importance in its field

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

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

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