Reviewer’s report

Title: A phenolic ester from Aglaia loheri leaves reveals cytotoxicity towards sensitive and multidrug-resistant cancer cells

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Reviewer: Vivian M. Rumjanek

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A phenolic ester from Aglaia loheri leaves reveals cytotoxicity towards sensitive and multidrug-resistant cancer cells by Dapat et al.

The authors have studied the anti cancer effect of isolates from crude extracts of A. loheri. It was already known that a number of other compounds obtained from different Aglaia species were found to be cytotoxic to tumor cells, supporting the possibility that A.loheri should be investigated. The active fractions obtained from the extracts of A.loheri were selected by bioassay-guided isolation and the active principle was characterized by mass spectroscopy and NMR. The authors tested the isolated active component against a sensitive and a multidrug resistant cell lines, suggesting it as a promising new drug for further development to be used for MDR tumors. The problem of multidrug resistance is, presently, the major drawback of chemotherapy and many efforts are being invested in finding new drugs that are not substrates for MDR transporters.

Despite the potential interest of the work the authors must address the criticisms before a decision can be reached. It is fundamental to distinguish an anti-tumor effect from a non-specific general toxicity.

Major compulsory revision-

It is necessary to clarify some points, related to the bioassays, in the Results section:

1- To be able to distinguish between non-specific cytotoxicity and an anti-tumor effect, it is necessary to perform a control using normal cells. It would be important to use normal peripheral blood mononuclear cells activated with a mitogen such as PHA to see if the effect is specific towards tumor cells.

2- Figure 3 tries to establish that exposure to Maldi 531.2[M+H]+ induces cell death via mitochondria membrane depolarization. It is necessary to explain how, using a cell line resistant to doxorubicin, depolarization can be observed in the presence of doxo (used as a positive control!!). Unless an excessive concentration has been used, doxo should not have an effect. The concentration used as a positive control should be stated.

3- Figure 4 states that the positive control for the induction of apoptosis was 5000ng/ml doxorubicin. Again an unrelated chemotherapeutic drug should have been used considering that the cells are resistant to doxorubicin (however, from
the figure nearly 8% are necrotic and 13% apoptotic what is significant for a 24h assay, if these are cells normally maintained in 5000ng/ml). On the other hand, significant apoptosis with Maldi 531.2[M+H]+ was only observed using extremely high concentrations (500nM- 1000nM)

4- Maybe statistical significance should be stated in a clearer way.

Minor essential revisions-

1- Legend of Fig.2. In the legend it should be stated that the measurement was performed at 72h, to allow comparison with the other assays.

2- Legend of Fig.3. What is the concentration of doxo used as a positive control in the assay, considering that the cell line used is resistant to 5000ng of doxorubicin? The letters used to indicate significance are very confusing. In my copy of the manuscript they seemed to be misplaced in the figure.

**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.

**Declaration of competing interests:**

I declare that I have no competing interests