Reviewer's report

Title: In vitro neuroprotective potential of four medicinal plants against rotenone-induced toxicity in SH-SY5Y neuroblastoma cells

Version: 1 Date: 17 October 2013

Reviewer: Melissa Vetten

Reviewer's report:

Major Compulsory Revisions
1. What was added to the untreated control cells? Was there a vehicle control included in the experimental design?

2. The effects of plant extracts on the cytotoxicity of rotenone was tested at 10 nM, 50 nM and 100 nM, with some extracts demonstrating a cytoprotective properties in cells exposed to 10 nM rotenone. These extracts did not show cytoprotective effects at 50 and 100 nM (page 10 states data not shown). In addition, in preliminary experiments, rotenone experiments were conducted for 72 hours. Why was a concentration of 50 nM and an exposure of 24 hours chosen for further experiments?

3. Page 8, section 3.1.1: the calculated LC50 is approximately double that of the experimentally determined LC50. Is there an explanation for this discrepancy? Are there also perhaps differences in the LC50 values obtained for the plant extracts?

4. Cytotoxicity data using the SRB assay at the same time point and concentration as the ROS, glutathione, MMP and caspase assays should be included as it would assist in comparing the results of the assays. For example, on page 10, the authors state that the extracts did not show any cytoprotective effects at rotenone concentrations of 50 nM (I understood this to be at 72 hours based on Figure 1), but then Figure 5 shows that the extracts have anti-apoptotic activity.

Minor Essential Revisions
5. Page 9, section 3.1.3, states that Figure 1 shows the results of 50 nM rotenone-induced cytotoxicity following pre-treatment; however the legend of figure 1 and the figures itself states that the concentration is 10 nM.

Discretionary Revisions
6. The LC50 values of the plant extracts are mentioned in the abstract, but the data used to obtained these values are not shown anywhere in the main text. Perhaps these results should be included?

7. Page 5, section 2.3: states that the final exposure concentrations of the rotenone is 1.28 x 10^-4 – 50 µM. It may be easier to read as 0.128 nM – 50 µM.

8. Figure 1 legend is a bit unclear. It should clearly state that there was a 1hr
pretreatment.

9. Page 5, section 2.3, first paragraph: Consider moving the last two sentences (“Effects of the plant......a rotenone concentration of 50 nM”) to the paragraph on page 6 so that the methods dealing with the plant extract’s pre-treatment cytotoxicity experiments are all in one paragraph.

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

**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