Reviewer’s report

Title: Ethyl acetate extract of Wedelia chinensis inhibits tert-butyl hydroperoxide-induced damage in PC12 cells and D-galactose-induced neuronal cell loss in mice

Version: 2
Date: 15 September 2014
Reviewer: Suh-Hang Juo

Reviewer’s report:

BMC Complementary and Alternative Medicine
MS ID: 2028313444132600
Title: Ethyl acetate extract of Wedelia chinensis inhibits tert-butyl hydroperoxide-induced damage in PC12 cells and D-galactose-induced neuronal cell loss in mice
Authors: Wea-Lung Lin et al.

The authors investigated the neuroprotective effects on a traditional Chinese medicine (Wedelia Chinensis) using both PC12 cells and an animal model. The data suggest that the extract of this herb and its major components can block cytochrome C release and improve BCL2 family. In addition, antioxidant effects were also noticed. In general, the results are interesting and may encourage scientists to further explore this herb for clinical applications. Some issues need to be considered by the authors:

1. The flow cytometry data (figure) need to be presented.
2. The authors may want to tell the readers how many hours the cells or animals need to be treated with the herb to get neuroprotective effect. The current data only said 24 hours pre-treatment. What if 12 or 6 h pre-treatment?
3. In figure 3, tBHP treatment is 30 min to damage the cells, but why the same treatment needs to be 3 hours in figure 4. In addition, is the dose of tBHP reasonable for humans?
4. What is a reasonable dose of this herb for humans? It is not clear whether the experimental dose way beyond the dose for human consumption.

The sample size is extremely small for SNP association studies, which will lead to either false positive or false negative results.

1. Only 42 cases recruited within 2.5 years, which means only 17 cases each year. So, there may be selection bias since most patients were not recruited.
2. The authors said that this disease is more dominant in men, but only 9 men (vs 33 female) patients in the case group. This also indicates a potential selection bias.
3. The genotyping method is not commonly used for SNP and the detailed
information for genotyping was not presented.

4. Because the genotyping method may not be reliable, and the small sample size also make the data on allele frequency unreliable (for example, the minor allele of IL-6 -174 in health Chinese is generally reported ~25%, but it is only 15% in the current study), the results are unlikely to be correct.