Reviewer's report

Title: Evaluation of direct antiviral activity of Deva-5 compound and extracts of five Asian plants against Influenza A Virus H3N8.

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Reviewer: Maja Nowakowski

Reviewer's report:

This study is a useful attempt to quantify specific antiviral effects of a traditional remedy, Deva-5, and its plant-derived components.

1. The authors clearly define the question addressed by their experiments testing the direct antiviral activity of a traditional compound Deva-5 and extracts of Deva-5 components. This does not include the molecular and phylogenetic analysis of the virus strain used, H3N8.

2. The methods are described in detail and are adequate.

3 - 4. Overall, the data appear to support the authors’ interpretation and are discussed critically. The manuscript meets general standards. However, while data in Figure 3 indicate statistically significant differences, the data presented in Figure 2 and Figure 4 lack any indication of statistical analysis. Including statistical validation of results in Figure legends or within the Figures would considerably improve the manuscript.

5 - 7. The results describing antiviral effects are discussed in detail. The limitations of the work are stated, and results are related to earlier published work and the authors’ preliminary results. The only additional limitations to discuss would include the complex nature of each extract and the need for further analysis to identify the active components within each extract.

8. The title and abstract adequately reflect the work on anti-viral effects of Deva-5 and its components. However, no mention is made of the molecular and phylogenetic analysis, the rationale for performing this analysis, or its relevance to the rest of the work.

9. The writing is acceptable, with minor corrections (listed at the end of this review).

Discretionary revisions: None.

Minor Essential Revisions: It is suggested that the authors include statistical evaluation of results in Figure 2 and Figure 4 (either in Figure legends or incorporated into the Figures). It would be helpful to establish the lowest concentrations at which toxic (or enhancing) effects reach statistical significance (Figure 2). Regarding Figure 4, it would be helpful to identify the highest dilutions of extracts that correspond to statistically significant reduction of infectivity.
Major Compulsory Revisions: A major concern is the considerable effort and space devoted in this work to the molecular and phylogenetic analysis of the virus strain because the data obtained are not discussed in relation to the rest of the work and no rationale is given for performing this extensive analysis. This part of the manuscript should be more fully integrated and its relevance to the rest of the work should be clearly explained.

Minor suggested corrections:

Abstract - Conclusions, last sentence: The results suggest that the former two plants contain substances with high antiviral activity and could be promising...

Methods – Plant material and preparation of extracts, line 5: To minimize the effect of preparation procedures on bioactive compounds, …
- Virus maintenance and plaque titration assay, line 10: …the inocula were…
- Nucleotide sequencing and phylogenetic analysis, line 7/8: …for HA and NA gene fragments, respectively.
- Evaluation of toxicity of herb extracts for MDCK cells, line 5: …viability of cells was evaluated daily…
- Plaque reduction neutralization test, line 5: …antibodies to the homologous H3 subtype of influenza A virus were used as controls as described above.
Line10: …and then inocula were discarded…

Results – Virus neutralization by plant extracts, last line: …the extract and infectivity of the virus…

Discussion, line 6: This effect can be explained…

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

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