Author's response to reviews

Title: Mosquito larvicidal and antimicrobial activity of protein of Solanum villosum leaves

Authors:

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Author's response to reviews: see over
Answer to Reviewers
To
The Reviewer #1,
BMC Complementary and Alternative Medicine

Title: Mosquito larvicidal and antimicrobial activity of protein of Solanum villosum leaves”
Authors: Chowdhury et al

Respected Sir/Madam,
Thank you very much for critically reviewing our manuscript. The specific changes made in the MS are mentioned below. Please note that the revised MS consists of modifications as per recommendation of other reviewers too.

1. The word “antilarvicidal” as shown in the abstract is not correct; it should be “larvicidal” actually.
Author’s response: The word “antilarvicidal” is replaced by “larvicidal” in the modified text.

2. In the method part, the authors described that the plant leaves were collected from May 2006 to June 2007, however, the data shown in Fig. 1 is between March and February. There is an obvious error here. The authors have to correct this point. Also, the amount of protein of young and mature leaves on both axes in Fig. 1 should be labeled with the same scale for comparison.

Author’s response: Actually the work was carried out between March 2006 to Feb 2007. We have made a typographical error in the previous manuscript and wrote that the plant leaves were collected from May 2006 to June 2007. However it has been corrected in the revised text. Also, the amounts of protein of young and mature leaves on both axes in Fig. 1 are now labeled with the same scale.

3. The references cited in context of the manuscript from the method part to results and discussion part is not matched with the numbers listed in the references. The authors have to re-check all the references cited in this manuscript.

Author’s response: We have rechecked the manuscript thoroughly and arranged the references accordingly.
4. As for the statistical analysis, the authors proposed to calculate LC50 and LC90, but there is only LC50 data shown in Table 2. The authors should also show LC90 data obtained from this study for complete information.

Author’s response: It is incorporated in the text.

5. The median lethal concentrations (LC50) of the plant extract against the three mosquito species tested in this study are relatively high (644-747 ppm). In my opinion, I think the protein extracted from *Solanum villosum* leaves in this study showed moderately larvicidal activity only, not really good as mentioned in the manuscript by the authors.

Author’s response: The sentence has been modified accordingly.

6. In this study, the number of concentrations (3), replications (3) and larvae used (10) in each replication of the larval bioassay were quite small. This will have affect on the lethal concentrations obtained in this study. In my opinion, if the authors use more concentrations (4-5), more replications (5-6) and more larvae used (20-25), they will get more accurate lethal concentrations of the plant extract against the tested mosquitoes. I suggest these for the authors to apply for their further studies in the future.

Authors response: Actually we can isolate a very little amount of protein during the present study with which we can’t accommodate more replications. However we know that it was necessary and may lead to the false interpretation and result. Thank you for your suggestion and we will follow your instruction during the future study.

7. The protein extracted from *Solanum villosum* leaves exhibited quite good antibacterial activity against the four bacterial strains in this study. However, the extract was tested at a relatively high concentration (60mg/disc) as compared with the standard antibiotic amoxicillin (30 ug/disc). It will be very useful and informative if the authors can provide more information of antibacterial activity of other lower dosages of this extract.

Author’s response: The results of the lower doses of the extract were not included in the text as the results were not impressive and can be treated as resistant according to the manufacturer’s instruction of antibiotic susceptibility.

Answer to Reviewers
To
The Reviewer #2,
BMC Complementary and Alternative Medicine

MS NO.

Title: Mosquito larvicidal and antimicrobial activity of protein of Solanum villosum leaves"
Authors: Chowdhury et al

Respected Sir/ Madam,
Thank you very much for critically reviewing our manuscript. The specific changes made in the MS are mentioned below. Please note that the revised MS consists of modifications as per recommendation of other reviewers too.

1. No positive controls and negative controls were used. And consequentially the results are not valid; - A similar situation was observed with bacterial assays.

Author’s response: Now a control set for both larvicidal and antibacterial activity is added in the modified text.

2. The number of replicates was very low and antibiotic concentrations were not well chosen (see for example for S. aureus and B. subtillis) and also MICs weren’t determinate. The activity of protein extract was non effective. Authors use a concentration (60 mg) approximately 20000 folds than the antibiotic (30 ug).

Author’s response: Actually we can isolate a very little amount of protein during the present study with which we can’t accommodate more replications. However we know that it was necessary and may lead to the false interpretation and result. Thank you for your suggestion and we will follow your instruction during the future study.

We have determined the Minimum Inhibitory Concentration but not include it in our previous communication. However we have included it in the modified manuscript now.

We have used a high concentration of the plant extract because the results of the lower doses of the extract were not impressive and can be treated as resistant according to the manufacturer’s instruction of antibiotic susceptibility
3. I have some reserves concerning to the statistical analysis, in any situation significant differences were observed; probably NOVA presupposes are not observed and the number of replicated are very low. The statistical treatments need to be removed;

Author’s response: The statistical analysis for seasonal variation by ANOVA has been removed from the modified text.

4. Authors only presented the results, but they didn’t discuss them;

Author’s response: Actually we have included the discussion section along with the result. Therefore the discussion in each case was followed by result. Those have been modified accordingly.

5. The conclusion are very speculative, results didn’t support the conclusions.

Author’s response: The conclusion section has been reconstructed according to the reviewer comment.

Minor comments:

Abstract:
1. The name of the mosquitoes needs to be written in the extended form. It is the first time in the text that the name of this species is mentioned

Author’s response: It is now written in the full form in the modified text.

2. The sentence “Chemical tests for tannins, saponin, flavonoids, steroids, terpenoids, carbohydrates and proteins were carried out on the extracted material of both mature and young leaves using standard procedures.” should be deleted. Authors mentioned this fact in the abstract and in the results sections; however any correlations were established with the activities of S. villosum leaves and any values were presented;

Author’s response: The phytochemical screening portion has been removed from the modified text as we are in agreement with you.

3. Keywords, the methodology is not appropriated for keywords. Methodology development is not a objective of the manuscript;
Author’s response: The methodologies are removed from the modified text.