Author's response to reviews

Title: Ethnobotanical and antimicrobial study of some selected medicinal plants used in Khyber Pakhtunkhwa (KPK) as a potential source to cure infectious diseases

Authors:

Nadeem Khan (khan_m_nadeem@yahoo.com)
Arshad M Abbasi (amabbasi@ciit.net.pk)
Ghulam Dastagir (dastagirbotany@yahoo.com)
Abdul Nazir (nazir_malik1@yahoo.com)
Ghulam M Shah (gmshah72@yahoo.com)
Munir H Shah (mhshahg@hotmail.com)

Version: 4 Date: 18 March 2014

Author's response to reviews: see over
Editor-in-Chief

*BMC Complementary and Alternative Medicines*

Subject: Submission of revised version of research article entitled “Ethnobotanical and antimicrobial study of some selected medicinal plants used in Khyber Pakhtunkhwa (KPK) as a potential source to cure infectious diseases”

Please find herewith the revised version of the research article on the above mentioned subject submitted for publication in “BMC Complementary and Alternative Medicines”. This version is an updated and revised version after making the changes in the manuscripts as suggested by the reviewers (Referee 2 and 3) and the editor. The changes are made accordingly in the main text (manuscript) and in the following pages author has addressed the comments from reviewers and the editor.

I shall be grateful for a prompt action as to the fate of the paper.

With best regards

Yours Sincerely,

*Nadeem Khan*
Assistant Professor
Department of Environmental Sciences
COMSATS Institute of Information Technology,
Abbottabad, Pakistan
Author’s response to reviewers and editor

Title: Ethnobotanical and antimicrobial study of some selected medicinal plants used in Khyber Pakhtunkhwa (KPK) as a potential source to cure infectious diseases

Authors:

Nadeem Khan (khan_m_nadeem@yahoo.com; nadeemkhan@ciit.net.pk)

Arshad Mehmood Abbasi (amabbasi@ciit.net.pk)

Ghulam Dastagir (dastagirbotany@yahoo.com)

Abdul Nazir (nazir_malik1@yahoo.com)

Ghulam Mujtaba Shah (gmshah72@yahoo.com)

Munir H Shah (mhshahg@hotmail.com)

Version 4: 18th March 2014

Author’s response to reviewers/editor

Please see the following pages
Referee 2 from (Jean Paul DZOYEM)

- Major Compulsory Revisions
  - Comment 1. The conclusions drawn from the study by the authors are not supported by the data. Results obtained do not fit into the cut-off of the antimicrobial activity classification of plant extracts (Eloff JN.: Quantification the bioactivity of plant extracts during screening and bioassay guided fractionation. Phytomedicine 2004, 11(4):370-371; Kuete V: Potential of Cameroonian plants and derived-products against microbial infections: A review. Planta Medica 2010, 76: 1479 – 1491). As result, it is not possible to quantify the activity of the samples tested. Therefore, the conclusion should be modified according to the results obtained.

  Thanks a lot for your comments and suggestions. As suggested the conclusion has been changed according to results obtained and the changed text has been embedded in the main text of the manuscript in conclusion section (Track changes).

- Minor Essential Revisions

  Abstract
  - Comments 2. The conclusion draw in the abstract is not related to the study. Please provide a conclusion from the results obtained.

  The text of conclusion has been changed according to the results obtained as suggested by the reviewers (Track changes). Author is thankful to reviewer for suggesting the changes.

  Methods
  - Comment 3. “MIC assay by following the method [24] with little modification”. Modify this sentence as: “MIC was determined as described by Sahin et al [24] with little modification”.

  The text has been changed accordingly and embedded in the main text (Track changes)

  - Comment 4. Authors should add statements about how the inocula were standardized and the possible toxic effect of solvent used to dissolve the extracts.

  Preparation of inocula
  The inocula were prepared by inoculating a loop of each bacterial strain from a 24hrs old culture into a sterile nutrient broth aseptically. The culture was allowed to grow for 24hrs in a shaking incubator at 37°C. The overnight culture is taken and checked until the visible turbidity is equal or greater than that of 0.5 McFarland standards (Pro-Lab Diagnostics) at 560 nm using UV-Visible spectrophotometer (IRMECO UV-VIS U2020, Germany). Sterilized nutrient broth is used as blank. If the absorbance is higher, then the culture is diluted with sterilized nutrient broth and absorbance is noted again. The standardized cultures were used for further analysis. The text has been embedded in the material method section with “Preparation of inocula” as subsection.

  Possible Toxic effects of solvents
  To check the possible toxic effect of various solvents used in the present study (aqueous, ethanolic and n-hexane) different volumes of broth medium and respective solvents were mixed in pre-labelled test tubes. The percentage of the solvent ranged from 1-8%. Then 1000µl of each inoculum was added to the respective tubes and then checked the turbidity after incubating for 24hrs at 37°C. A tube only with broth medium was used as
control while the tubes containing tetracycline and streptomycin (100µg/ml) were sued as positive control. After 24hrs the absorbance was measured for each sample suing at 560 nm using UV-Visible spectrophotometer (IRMECO UV-VIS U2020, Germany). Growth in each of the ‘test’ tube was expressed relative to controls. However, this part is not included in the main text of the manuscript.

Referee 3 (Gerald Ngo TEKE) comments

Actually there is improvement the presentation of the manuscript.

- **Comment 1.** -there is still need to correct some language errors. I suggest it should be read by a native of the language. The changes have been made in the main text and it has already been reviewed by a native English speaker.

- **Comment 2.** Table 2. the decimals are not uniform. The corrections are made and the decimals are now uniform in the Table 2.

- **Comment 3.** Figure 1. following the activity axis how can you say No Activity is representing the highest of the bars? Actually a total of 30 plant extracts (including all three solvents) were used in the study and it was found that 53% (16 out of 30) of the extracts were not effective against any of the testes microorganism. This highest bar in Figure 1 represents that 53% without having any activity.

Editor Victor Kuete comments

- **Comment 1.** Please provide References to the traditional uses of the plants. An additional column in Table 1 with “medicinal plant from literature” has bed added. The references are also embedded in the reference list.

- **Comment 2.** Please use Broth dilution to determine the MIC of your samples and delete the all data related to agar diffusion, as the technique is not appropriate for the screenings of plant extract.

Thanks for your suggestions. I mentioned in last submission (submission 3), I have done MIC values for those extracts which showed antimicrobial activity in agar well diffusion method by using Broth Dilution Method (as suggested by the editor). The method has been added in the Materials and Methods section. The results are presented in Table 3 and also discussed in the Results and Discussion section of the manuscript. However, I also retained the data on agar well diffusion method based on which the MIC values are evaluated.