Author's response to reviews

Title: Evaluation of antimotility effect of Lantana camara L. var. acuelata constituents on neostigmine-induced gastrointestinal transit in mice

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Author's response to reviews:

Author's response to Reviewers' Comments
Title: Evaluation of Antimotility effect of Lantana camara L. var. acuelata constituents on Neostigmine-induced Gastrointestinal transit in mice
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Version: 2 Date: 18.6.05
Author's response to reviews:
Editorial Director
BMC Complementary and Alternative Medicine
Dear Editorial Director
Thank you very much for your e-mail message of May 19, 2005 concerning our manuscript. We sincerely appreciate the very constructive suggestions made by you and the reviewers. We have revised our manuscript in accordance with the comments sent to us. We have now incorporated data on the effect of the Lantana camara methanolic extract on castor oil induced diarrhea in mice (Table No.2, page 11) and have made minor/major additions in Abstract, Introduction, Methods, Results and Discussion and Conclusions sections. We trust that the revisions/additions we have made are satisfactory and that our revised manuscript is now suitable for publication in BMC Complementary and Alternative Medicine.
Our point-by-point answers to the reviewers' comments are described.
We are looking forward to hearing from you soon.
With best regards
Sincerely yours
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Thank you so much for your helpful comments, suggestion, and advice. We tried to revise the manuscript according to your comments.

Detailed comments:

1. The Reviewer has raised doubts about the absorbability of lantadenes as these are nonpolar compounds. We have used 0.25% Carboxy methyl cellulose as the vehicle for the test materials. CMC is an established solubility enhancer and has been shown to reduce peritonitis in the rat model (Ref. a, b). Therefore, the use of CMC as the vehicle shall not only help in the absorption but also inhibit the precipitation of the test compounds.


2. To minimise any discomfort to the animals, the injection volume for each treatment has been kept low. The actual volume injected varied from 0.2 to 0.3 ml, so that each animal in its respective group gets the selected dose, depending upon the weight of the animal. (page 5, bottom 3 lines)

3. In our study, we have used a low dose (125mg/kg), a medium dose (250mg/kg) and two high doses (500mg/kg & 1000mg/kg).

The doses used in our study appear to be relatively high or even very high in the first instance. In our preliminary studies using a low dose (250mg/kg) and a high dose (1000mg/kg), no significant alterations were observed in total bilrubin and SGOT in the treated animals, pointing towards their relative safety in the selected experimental animal i.e. mice.

4. Yes, 0.25% CMC was administered as a vehicle to the control animals. (page 5, bottom 2 lines).

5. Revision and additions in the references has been done and incorporated in the revised manuscript.

Response to the Reviewer's comments:
Reviewer: Dr. Samson Amos

Thank you so much for your helpful comments, suggestion, and advice. We tried to revise the manuscript according to your comments.

Major compulsory revisions:

1. Rationale behind the studies:

Reviewer agrees with us that the plant Lantana camara is toxic to most animal species and GIT motility is interfered with, resulting in constipation. Precisely, this very point (constipating effect) has been made the rationale behind these studies i.e. to develop the plant leaf powder/methanolic extract or some of its active constituent as an antimotility and/or antidiarrheal agent. The only hindrance was the "safe dose" as has been pointed out by the Reviewer. We had addressed ourselves to this very important point and had conducted 'safety profile' studies in a very specific manner in mice (see below). Although the plant parts/plant extracts/concotions have been used in folklore medicine in Africa and Asia and may be other countries, for relieving one from GIT related disorders, no scientific studies (charcoal meal test, gastric emptying test/castor oil induced or any other such test) are available in literature proving or disproving the activity of this plant. Based on these lacunae in scientific literature, we used lantana camara leaf powder, LCME and purified LA to gather insights in this direction.

2. In our study, we have used a low dose (125mg/kg), a medium dose (250mg/kg) and two high doses (500mg/kg & 1000mg/kg).

The doses used in our study appear to be relatively high or even very high in the first instance but we decided to continue to use this range as no significant alterations were observed in total bilrubin and SGOT.
in the treated animals at any of the doses used, pointing towards their relative safety in the selected experimental animal i.e. mice. Close scrutiny of literature has revealed that lantana camara or its isolated constituents are reportedly toxic to sheep, cattle, goats, dogs, deer, guinea pigs, man, cats (Ref. given in the manuscript). No published reference about Lantana toxicity in experimental mice could be found. The route (i.p.) of administration of the test compounds/fractions/extract was decided on the basis that oral route may not be an effective route during acute cases when diarrhea and vomiting are accompanied together. Previous studies (Chitme et al 2004 Ref.No.4 in the manuscript) have as well used the intraperitoneal route of administration of the plant extract in the similar settings i.e. antidiarrheal studies. Moreover, CMC used as a vehicle in our experiments, has been shown to reduce peritonitis in an animal model (Ref. a and b). Under these conditions, the possibility of formation of a Precipitate is reduced but instead the absorption is enhanced.


3. As suggested by the Reviewer, the effect of all four doses of LCME on castor oil induced diarrhea in mice have been performed and results incorporated in the revised manuscript. The relevant additions in Introduction, Methods, Results, Discussion and Conclusions have been made in the revised manuscript.

4. The Reviewer has very rightly suggested - for providing a satisfactory explanation and suggesting a working hypothesis and a probable mechanism of action of Lantana constituent(s), in vitro studies using rabbit or guinea pig ileum should be carried out in future work. In vitro experiments had been performed using mice ileum itself and our preliminary results indicated that LCME/lantadene A had an inhibitory effect on mice ileal smooth muscle contraction. Because of the in vitro solubility problems in Kreb’s- Ringer solution, in vivo feeding of the test preparations for 10 days was done and a significant inhibition in smooth muscle contraction noticed (unpublished observations) in the normal animals and also using acetyl choline and atropine, an antagonist. After completion of these in vitro studies, a probable mechanism can be proposed, the present suggestions invoking an anticholinergic activity remaining highly tentative.

Response to the reviewer’s comments:
Reviewer: Dr. Hassa Mousa

Thank you so much for your helpful comments, suggestion, and advice. We tried to revise the manuscript according to your comments.

Minor essential revisions:
The suggested references have been included in the revised manuscript. (Ref. Nos. 27, 29 and 40)

The suggestion that the word ‘motility’ should replace the word ‘kinetic’ in the title has been accepted and the title or the manuscript has been changed accordingly.

Thanks and regards

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