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Authors' response to final

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FINAL EVALUATOR'S comments on revised paper (if any)

article is much more acceptable for publication in this form.

PART 1:

Journal Name:	British Journal of Pharmaceutical Research
Manuscript Number:	2013_BJPR_3698
Title of the Manuscript:	Bacterial endophytes of the medicinal herb Hygrophila spinosa T. Anders and their antimicrobial activity

PART 2:

Think Evilleditions comments on tevisor paper (it any)	riacinors response to initial
	evaluator's comments
The authors have improved the MS with the corrections performed and I appreciate very much their effort to fix everything I suggested	
accordingly. The MS definitely benefited a lot from the separation between Results and Discussion into two independent sections. The	

Nevertheless. I still think that it suffers from a major problem: because no minimal molecular characterization was employed, the probable species to which these isolates belong could not be presented, so that the discussion about the findings and biotechnological potential of them are still fragile, as a consequence... If the journal understands this is not a major issue if compared to the value of the results and information presented, then publish it (after the few extra corrections indicated below). Once the few extra corrections are performed, I do not need to see the MS again.

I understood the argument presented by the authors, who said this is a "preliminary-first-report-type" of work to show the scientific community about the potential of endophytic isolates from this medicinal species. However, despite of the great improvement of the MS with the new Discussion section, I still feel the work and its relevance would undoubtedly benefit from a closer species identification, through a simple 16s rDNA sequencing (not aiming here at a full molecular characterization, as indicated by the authors as something underway, but simply a technique that could allow one to have an idea of the species involved...). My markings further below reflect this point of view.

As mentioned above, some **further corrections** are needed though; please, see below.

Abstract -

At the 'Conclusion:' "(...) study identified eleven-11 bacterial endophytes harbored by the leaves, stem and root of H. spinosa (...)"

Mat & Met -

Line 108: "(...) The Shannon Weaver biodiversity index H was calculated as follows: (...)"

Line 117: remove ":" after the word 'include'.

Line 123: "(...) method using six test organisms: like-Bacillus subtilis, B. cereus, (...)" (see ":" added!)

Results -

Line 139: "(...) Avoiding the repetitive strains, a total of 11 phenotypically (...)" (see "," added!)

Line 142-147: At least for the last three parameters in the Table 1, the results (numbers) are fully repeated in the text, which is a redundancy! Try to use absolute values in the Table and use the corresponding % in the text, and/or remove values from the text when they

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already appear in the Table.

Table 1: the last column is still not OK – label it as "**total / avg**" and add a footnote to explain that the first three variables are the sum of the endophytes found in all tissues, and that for the last three the averages among the tissues are shown.

Table 2: In the column labeled "size, μm ", I suggest to change "dia" for the symbol of diameter " \varnothing ". I also suggest to change the "X" sign for a smaller version ("x").

Lines 167-169: "(...) Out of 11 isolates, seven were Gram-positive (...)" (see "," added!)

Table 3: At the footnote, change "positive response" for "presence" and "negative response" for "absence".

Line 203: "(...) disc-diffusion method against six different antibiotics like (amoxycillin, bacitracin, (...)" \rightarrow Put the antibiotics names between parenthesis.

Line 204: In the sentence "(...) Results as shown in Table 5 depict that, bacterial endophytes (...)", remove the comma after the word "that".

Table 4: I suggest to change "NIL" in the body of the Table for "0" (the number 'zero'). It'll make it clearer.

Discussion -

Line 236: "(...) H. spinosa, although endophytes could also occur harbor in flower, fruit and seeds. (...)"

Lines 238-241: "(...) Such <u>a higher</u> species richness in leaves may be attributed to the <u>ir</u> anatomical peculiarities of the leaves and micro-environmental <u>peculiarities</u>, <u>as specific</u> conditions rich-in essential nutrients which-drives the selective force for survival of tissue specific endophytic taxa (...)" This is an attempt to turn this sentence clearer in meaning and content... Perhaps some information about what's different in the leaves of this species would help the reader to understand better what are the "peculiarities" mentioned...

Lines 248-250: "(...) distinguishable bacterial endophytes harboringed by leaves, stem and root tissues of *H. spinosa* were characterized in details (Tables 2 - 4) and tentatively identified as members belonging to of the bacterial genera (...)".

Line 251: "(...) These isolates were mostly the <u>belong to a class of</u> fast growing endophytes and (...)" \rightarrow I am not sure this was the meaning of this sentence – I added it anyway in order to clarify and give it some sense...

Line 260: "(...) <u>Although there is not a lot of Information regarding production of enzymes by microbes of plant origins, are few although-endophytic bacteria isolated (...)".</u>

Line 265: The expression "supports earlier observations" is lacking a conclusion of the sentence – earlier observations of what?

Line 266-269: "(...) The presence of nitrate reductase and tryptophanase in some of the isolates appears to suggests they play a key role in the nitrogen cycle, thereby and has ving important agricultural, environmental and public health implications (...)".

Line 277-279: "(...) medicinal plants have been presented [30-32], Furthermore, antimicrobial activities of endophytic bacteria are not uncommon [17, 20, 29]. Li et al. [30], however, have explored endophytic actinomycetes (...)".

Line 284-285: "(...) broad spectrum antimicrobial activity indicating possible biotechnological potential applications (...)".

Conclusion -

Line 295: "(...) appear to be a potential source of antimicrobial metabolites, as well as enzymes for potential biotechnological applications in health, agriculture and industry.

Note: Anonymous Reviewer

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