



SDI Review Form 1.6

PART 1:

Journal Name:	British Microbiology Research Journal
Manuscript Number:	MS: 2012/BMRJ/2302
Title of the Manuscript:	Population Variance and Diversity of Endophytic Bacteria in Soybean (Glycine max (L) Merrill) Cultivated in Vertisol.

General guideline for Peer Review process is available in this link:

(<http://www.sciencedomain.org/page.php?id=sdi-general-editorial-policy#Peer-Review-Guideline>)

- This form has total 9 parts. Kindly note that you should use all the parts of this review form.



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PART 2: Review Comments

	Reviewer's comment	Author's comment <i>(if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)</i>
<u>Compulsory</u> REVISION comments	In this manuscript, the authors reported a study on the soybean endophytic bacteria isolated from roots, stems and leaves at different growth stages. The population density of bacteria was calculated and the isolates were identified to the general level based upon morphology, cultural and biochemical characters. Finally, they concluded that the amount and diversity of endophytic bacteria changed according to the growth stage of the soybean plant. After checking the methods and results, I feel that this is an immature work for making publication. In addition, there are some problems in the experimental designation. So, I have to suggest rejecting it based upon the following reasons: 1. The authors used an enrichment procedure (24h at 30 °C in nutrient broth), but they did not give a justification why they did so. For me, this step is not necessary and it made the subsequent estimation of bacterial abundance and diversity not meaningful. With this procedure, 1) the relative abundance of the fast growing bacteria will be enhanced, and the	Endophytic Population is low hence done.



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	slow growing bacteria or those who cannot grow in this medium will be eliminated in the following isolation procedure; 2) the amount of bacteria after the enrichment is no longer directly related to their abundance inside the plant tissues. So, I would like to suggest the authors making a modification by isolating the bacteria directly from the plant extracts (may be 1 g of plant + 9 mL of sterilized water) on different media (rich medium and some kind selective medium for the slow growing bacteria, like actinomyces). 2. In several cases, the methods are not clear in the current manuscript, such as 1) what are the differences or characters of the sampling sites? (Latitude, longitude, altitude and distances between the sampling sites, climate zone, soil characters, etc.); 2) how the authors selected the isolates? 3) How to calculate the means of CFU? (In Table 1, the means are always greater than the data for each plant part, for me it is not reasonable). 3. No any evidence is presented for the bacterial identification, and the authors only give some general names. This is completely	The media was crudely modified to facilitate the growth of bacteria (any special nutritional requirement) as would take place naturally in tissue. Yes the part is now incorporated.
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	unacceptable. Also, for the identification, I would like to suggest the authors to make 16S rRNA sequence analysis for each group, at least several isolates for each group. 4. In Table 1, the abundance of bacteria in the roots, stems and leaves is very similar. I strongly question these data because from my work and from the published papers cited by the authors (Zinniel et al. 2002; Koklinsky-Sobral et al. 2004), the endophytic bacteria present a level of 10^4 to 10^7 per gram of roots, and 10^2 or less per gram of leaves. I do not believe that the bacterial number in root tissues is so low after the enrichment.	Yes I do agree. The data was very wrongly processed during statistically.
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Minor REVISION comments	Some special comments: 1. In many cases, the point or dot is missing after a sentence. 2. In Abstract, the 4th line of Methodology, “were macerated the filtrate” should be “were macerated and the filtrate” 3. In Abstract, the first words of Results, “A 527 (35.50 %) bacterial isolates” should be “A total of 527 (35.50 %) bacterial isolates” 4. In lines 83 and 84, the abbreviation “Sp.” following each genus name should be eliminated. When you say <i>Pseudomonas</i> sp. (not Sp.), the identity is an (single) unidentified group or strain belonging to this genus. 5. The format of reference citation is not right in many cases, such as line 47 “McWilliams et al., 2004” should be “McWilliams et al. (2004)” 6. In Conclusion (Lines 120-124), You cannot make the conclusion “It appears that the endophytic population is influenced by several factors including environmental conditions, development stages and geographic location”, because no any information about the	Yes the corrections are made.
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	environmental conditions and geographic location was reported in your work. Also, the subsequent two sentences are not correlated to your results, because you did not report any characters of the isolates to support these conclusions.	
<u>Optional/General</u> comments		