



SDI Review Form 1.6

Journal Name:	<u>British Microbiology Research Journal</u>
Manuscript Number:	2013_BMRJ_6587
Title of the Manuscript:	Lipolytic Activity and Molecular Characterization of Pseudomonas aeruginosa and Lysinibacillus sphaericus from domestic oil rich wastewater
Type of the Article	

General guideline for Peer Review process:

This journal's peer review policy states that **NO** manuscript should be rejected only on the basis of '**lack of Novelty**', provided the manuscript is scientifically robust and technically sound.

To know the complete guideline for Peer Review process, reviewers are requested to visit this link:

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SDI Review Form 1.6

PART 1: Review Comments

	Reviewer's comment	Author's comment (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)
Compulsory REVISION comments	<p>Title of the manuscript should be modified. The strain were identified using molecular methods, however the authors do not present molecular characterization of the strains. I suggest also to write ".... Pseudomonas aeruginosa and Lysinibacillus sphaericus strains isolated from...."</p> <p>Delete the first paragraph of the Introduction chapter - description of well known PCR and DNA sequencing techniques in this place is not a good idea. This chapter should focus lypolitic bacteria.</p> <p>Section 3.1., 3.2. I would expect determination of a number of colony forming units as a parameter of bacterial growth, instead of increase in weight of bacteria.</p> <p>Chapter 3.3 lines 163-170. As stated - most of the DNA sequencing reactions revealed multiple peaks, which strongly suggests that the authors did</p>	<p>I agree with the reviewer's comment. The word 'characterization' has been changed to 'identification'; 'isolated' is introduced to the title. First paragraph of the introduction is being deleted.</p> <p>Determination of colony forming unit is as a result of the microbial increase or decrease in weight (microbial mass). In fact, microbial weight is more accurate than colony forming unit since some microbial growth might not be visible to be counted as CFU but could be detected by microbial weight.</p> <p>The cultures worked with are pure culture. The multiple peaks not only resulted from purity of the culture but from other factors</p>



SDI Review Form 1.6

	<p>not work with pure cultures. If so, this would discredit the results presented in previous two chapters of the manuscript, i.e. growth of bacteria in wastewater and palm oil. In my opinion identification of the all tested strains (at the genus level) should be an initial point of this study. line 168.</p> <p>The strains were classified to <i>P. aeruginosa</i> and <i>L. sphaericus</i> species based on the 99 % nucleotide sequence identity of the rDNA sequences. However, according to Drancourt et al (2000. J. Clin. Microbiol. 38:3623-3630) a 97 to 99% identity in 16S rRNA gene sequence is the criterion to identify an organism at the genus level and >99% identity - to the species level. lines 166, 168 - should be <i>Pseudomonas</i> sp. , <i>Bacillus</i> sp.</p> <p>Within the manuscript the Results and Discussion were combined into a single chapter. However section 3.5 contain distinguished Discussion section. The authors should decide about composition of the manuscript.</p>	<p>like non-specific binding of primers to template during amplification as a result of the very low annealing temperature (45°C) and the highly reduced ramp (40%) used (Altschul <i>et al.</i>, 1990) both conditions allowed for promiscuous binding which might be responsible for the challenges faced during sequencing. However, these conditions had to be used to achieve amplification as samples could not be consistently amplified at higher temperatures.</p> <p>I agree with the reviewer's comment.</p> <p>The correction has been done. There is no Section 3.5; the Results and Discussion are now together.</p>
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<p><u>Minor</u> REVISION comments</p>	<p>Figures 1, 2, and 3 have not been cited in the text. They should be omitted.</p> <p>line 124. change " organisms" - to " strains" (change this throughout the text)</p> <p>Please combine data presented in Tables 5a and 5b into more concise form (Single Table).</p> <p>Lines 51-52. Although the bacterial isolates used in this study have been previously described, these data are not available (this section refers to PhD thesis).</p>	<p>The figures 1, 2 and 3 are cited and highlighted in yellow in lines 177 and 182 respectively.</p> <p>Corrections have been made highlighted in yellow.</p> <p>Please I prefer the two data to be separately presented for clarity. I believe it is well explanatory than being combined into one Table.</p> <p>The bacterial isolates has been previously described fully in the PhD thesis and also been described in a published Journal of Microbiology Research, 2013, 3(1): 43-52.</p>
<p><u>Optional/General</u> comments</p>		