



SDI Review Form 1.6

PART 1:

Journal Name:	British Journal of Applied Science & Technology
Manuscript Number:	2013_BJAST_3918
Title of the Manuscript:	The molecular succession of bacterial community of crude oil polluted soil and water samples from the Niger Delta, Nigeria

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 7 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 (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)
<u>Compulsory</u> REVISION comments	Introduction : line 67 Each band theoretically represents a different bacterial population present in the community. Denaturing Gradient Gel Electrophoresis (DGGE) has a broad scope of utilities in scientific research, with the breadth of PCR primers available, DGGE can be used to investigate broad phylogenies or specific target organisms such as pathogens or xenobiotics degraders with a quite high limit of detection (around 10^4). It could be used also to link geographical origin to microbial ecology (Montet ref) Line 81 : Denaturing gradient gel electrophoresis (DGGE) is a molecular fingerprinting method that separates polymerase chain reaction (PCR)-generated double strands DNA products. Line 90: This limitation is however, overcome by using DGGE, the principle of which is based on the denaturing of the double-stranded DNA linked by a GC clamp PCR at	This is noted and it has been corrected in the manuscript as appropriate. This is noted and it has been corrected in the manuscript as appropriate. This is noted and it has been effected in the manuscript as appropriate.



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	different concentrations resulting in a pattern of bands on polyacrylamide gel. Material and methods Line 99 : The water samples were collected aseptically into screw-capped containers while the soil samples were collected into sterile cellophane bags. The authors have to precise how the samples were collected and at which depth? And their sizes? Line 141: explain why you use nested PCR Results Line 171: The bacterial populations ($\times 10^4$) ranged from 30.33 ± 3.9 to 54 ± 4.3 Cfu/mL for samples 1w (water) and 2s (soil) on day 7 ... In general, this results chapter is too short and we want to know what are the samples (numbers)? Discussion Same remarks concerning the numeration, we do not know what are these samples and it is difficult to appreciate. Lines 188 to 207 could be added to result chapter. Line 265 : The bacteria represented in the band 3sa, which was dominant throughout the incubation period are potential candidates for degradation of crude oil which could be	The samples were collected into sterilized screw-capped containers for water samples and cellophane bags for soil samples. Top soil was collected. About 10g was collected for each sample. Nested PCR was used because it involves 2 sets of primers, used in two successive runs of polymerase chain reaction. I intended to use the second primer to amplify a secondary target within the first run product. Sample numbers 1w - Agbada-Aluu water sample 2w – Obite water sample 3w – Bonny water sample 1s - Agbada-Aluu soil sample 2s - Obite soil sample 3s - Bonny soil sample 4 – Control
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	excised, cloned into <i>E.coli</i> and sequenced for molecular identification. Remark of the referee: It could be obtained directly by sequencing the amplicon extracted for DGGE gel. Figures: Plate 3 page 16 Explain the colors Figure 3 page 18: the title of the figure is not clear. pH seems to be the result of the experiment! References: add reference of Montet's team on DGGE	Corrected as appropriate Red colors represent initial colors obtained in the first 7 days of incubation. Blue colors represent disappeared bands Yellow colors represent new bands that emerged during incubation. I think the title is ok but you can suggest an appropriate one. This is noted and it has been included in the reference section.
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Minor REVISION comments	Summary, change the unit Hr to h Same remark for all the text Change l to L (for liter) in all the text Line 104 : The BHB (full text)	These have been corrected as appropriate.
Optional/General comments	The idea to use PCR-DGGE to analyze and follow microorganisms in soil is very interesting. The authors have to improve their text. Usually soil is a hard substrate for molecular biology because of natural inhibitors present in the soil (polymers). We do not know why the authors propose nested PCR that seems not very useful in this work. Sequencing of the strains is missing to interpret the hydrolysis of oils.	As earlier mentioned above, nested PCR was to serve as control so that the wrong locus would not be amplified. More so, I was interested in bacteria not all microorganisms. So, the first primer was to amplify 16S rRNA. In this study, sequencing was not done, but it was established that a particular band is responsible for the degradation. The research still continues.