Hydrocarbon degradation potential of heterotrophic bacteria isolated from oil polluted sites in sakpenwa community in rivers state.

ABSTRACT

In this study, hydrocarbon degradation potentials of heterotrophic bacteria isolated from oil-polluted soil were examined; Samples were collected from Sakpenwa, an oil producing community in Tai LGA of Rivers State, and analyzed for physicochemical and microbiological properties using standard techniques. Hydrocarbon utilizing bacteria (HUB) were isolated by vapour phase transfer method using mineral salt medium. They were subjected to constant shaking in a standard laboratory shaker for 30 days in Bushnell -Haas agar supplemented with 5% of crude oil. Fifteen (15) bacterial isolates were screened for hydrocarbon degradation potentials of which five bacterial isolates exhibited high hydrocarbon degradability. The degradation study was determined by monitoring colour change, Optical density (OD), pH, Total Petroleum Hydrocarbon (TPH), Total Cuturable Heterotrophic Bacterial Counts (TCHBC) and Total Hydrocarbon Contents (THC) respectively for each isolate, The mean total culturable heterotrophic bacterial counts ranged from 1.65×10^{7} to 2.27×10^{8}cfu/ml while the mean total culturable hydrocarbon utilizing bacterial counts ranged from 1.09×10^{4} to 3.9×10^{5}. The optical density varied from 0.09±0.02 - 2.57±0.00 and pH ranged from 2.98±0.09 - 6.98±0.09. The elimination of oil spilled in the environment can be achieved by microbial degradation when added up with some physical and chemical methods

KEY WORDS: hydrocarbon, heterotrophic bacteria, polluted soil, degradation potentials

INTRODUCTION

In Nigeria, 80% of the crude oil used is supplied from the South-South region of the country. Therefore, as a result of high oil exploration activities going on in this part of the country over years (Abu and Chikere, 2006), substances like gaseous emissions, oil spills, effluents and solid waste are discharged into the environment, thus, polluting the environment (Nweke and Okpokwasili, 2004). From statistics, the biotic component of the soil occupied not greater than 5% soil space; living microbes including bacteria, archaea and fungi are responsible for 80-90% of soil processes and formation such as recycling of nutrients, transformation of organic matter, and maintenances of soil structure in microbial decomposers (Nannipieri *et al*. 2003). Since microorganisms in the soil are involved in various biogeochemical processes, soil activities largely depend on them (Mikkonen, 2008).

The degradation of hydrocarbons is influenced by many factors (temperature, relative humidity, soil structure, soil moisture, soil pH, soil biota, pollutant's structure, dose, toxicity, and bioavailability). A study on the degradation of hydrocarbon by aerobic heterotrophic bacteria by Ichor *et al.,* (2014), took cognisance of some of these factors.

MATERIALS AND METHODS

2.1 Study area and sample collection.

The study site was located at the oil polluted sites in Sakpenwa community in Ogoni land, Tai Local Government Area, Rivers State. Soil samples were collected 500m and 1000m away from the major spill sites. Fifty grams (50g) of the oil-polluted soil samples were collected from each of the sampling points using a soil sampler. The collected soil samples

were transported in plastic nylon bags from the polluted sites to the Department of Microbiology, University of Port Harcourt laboratory for analysis within 24 hours (Iheanacho *et al*., 2014).

2.2 Samples preparation

The soil samples collected were passed through a mesh sieve (2mm pore size) to remove large particles and were thoroughly mixed. Thereafter, 5g of each soil sample was suspended in 45 ml of distilled water. The suspended samples were mixed properly in a rotary shaker at 100 rpm at room temperature (28± 2^0C) for 1hour, 30 minutes to liberate the organisms into the liquid medium (Iheanacho *et al*., 2014*)*. The pH of the samples was also taken.

2.3 Isolation and enumeration of total heterotrophic bacteria

The total culturable heterotrophic bacterial count for each degradation set-up was enumerated using the streak plate method (Odokuma and Okpokwasili, 1992). Serial dilutions of the samples were made and 0.1ml aliquot of the 10^{-1} to 10^{-4} dilutions of each sample were transferred onto well dried, sterile nutrient agar plates (in triplicate) and incubated at 37°C for 24. After incubation, the bacterial colonies that grew on the plates were counted and sub-cultured onto fresh nutrient agar plates using the streak-plate method in other to obtain pure cultures of each colony. Discrete colonies on the plates were then transferred into nutrient agar slants, properly labelled and stored at 4^0C as a stock culture for preservation and identification (Odokuma and Ibor, 2002).

2.4 Enumeration of total culturable hydrocarbon utilizing bacteria (TCHUB)

The enumeration of Total Culturable Hydrocarbon Utilizing Bacteria (TCHUB) was done by applying the vapour phase method described by Atuanya and Ibeh, (2004). Appropriate diluent of 0.5ml of the samples collected from the two different set up (1000m and 500m away from a particular polluted site) labelled A and B respectively were inoculated into modified Mineral Salt Agar medium (MSA). The medium was made of 0.42g $MgS0_4.7H_2O$; 0.297g KCl; 0.85g KH_2PO_4; 0.424g $NaNO_3$; 1.27g K_2HPO_4; 20.12g NaCl; 250mg Amphotericin B (sold as Fungizone) and 20g agar powder. These were weighed out and hydrated in 1000mL of sterile distilled water in a conical flask. The media was sterilized by autoclaving at 121°C, 15Psi for 15min, before dispensing into sterile Petri dishes. The gelled Mineral Salt Agar (MSA) was inoculated with 0.5ml of serial dilutions of the polluted soil sites A and B sample respectively. Filter paper (Whatman No 1) was saturated with bonny light crude oil, and the crude oil impregnated papers were aseptically placed onto the covers of Petri dishes and inverted. The hydrocarbon saturated filter papers supply hydrocarbon by vapour-phase transfer to the inoculums (Chikere and Chijioke-Osuji, 2006). The plates were incubated at 28°C±2°C for seven (7) days. Colonies were counted from triplicates and mean values were record and colonies formed were stored as stock cultures at a temperature of 4^0C for preservation and identification.

2.5 Gram staining

Isolates were subjected to Gram's reaction to check whether they are Gram-positive or Gram negative. A thin smear was made on a clean grease-free glass slide. The smear was air dried and heat-fixed by rapidly passing it over the flame of a Bunsen burner three times .The fixed smear was covered with crystal violet stain for 30 to 60 seconds and rapidly washed off with clean water and then flooded with Lugol's iodine for one minute. The iodine was washed off with clean water and the smear was decolorized with alcohol and washed immediately with clean water before counter staining with safranin for one minute. The stain was washed off with clean water and the smear air-dried and observed under microscope using oil immersion objective lens. Gram positive bacteria stained purple while gram negative bacteria stained red or pink (Cheeshrough, 2005).

2.6 Motility test

Twenty-four hour (24hr) culture of the isolates in peptone broth was used for motility test. Five drops of each isolate was placed on a cover slip and the concave shaped slide smeared with Vaseline at the edge of the concavity, gently on the cover slip. This slide was carefully

inverted and the drop on the cover slip observed under high power objective lens (40×) (Cheeshrough, 2005).

2.7 Biochemical tests

The bacterial isolates obtained were characterized and identified based on their cultural, morphological and biochemical characteristics using the scheme of Bergey's Manual of Determinative Bacteriology (Chikere and Okpokwasili, 2003; Oboh *et al*., 2006).

2.7.1 Catalase test

Three millilitres (3ml) of 3% hydrogen peroxide solution was poured into different test tubes and sterile glass rod was used to introduce some colonies of the isolates into the test tubes containing the hydrogen peroxide. The contents of the test tubes were observed for gas bubbling. Catalase positive bacteria showed active bubbles while catalase negative did not (Cheeshrough, 2005).

2.7.2 Urease test

Christensen's solid medium was prepared by dissolving 1g peptone; 5g NaCl; 2g K_2HPO_4 and 10g agar in 1000 ml of distilled water. Phenol red (6ml) was also added and the pH adjusted to 7.0. This was sterilized at 121^0C for 20 minutes at 15psi and allowed to cool to 50^0C. 10 ml of 10% solution of glucose and 100 ml of 20% urea solution were sterilized by filtration. The glucose and urea were mixed aseptically with the agar medium and dispensed in 5ml amounts into bijou bottles and allowed to solidify in the slope position. Urease positive bacteria indicated pink colour while urease negative bacteria did not show pink colour (Cheeshrough, 2005).

2.7.3 Sugar fermentation test

The test was carried out to determine the ability of the isolates to ferment various sugars which is indicated by the production of acids/gas. The following sugars were used: maltose, glucose and lactose. From each sugar, 0.5g was dissolved in 50ml of peptone water and sterilized by membrane filtration. A pinch of phenol red was added as indicator and 5ml aliquots were aseptically dispensed into sterile test tube containing sterile Durham tubes which were inverted in the sterile broth. The broth was inoculated with the isolates using sterile wire loop and incubated at 30^0C for 48 hours. The content was observed for change in colour and/or the production of gas (Cheeshrough, 2005).

2.7.4 Citrate utilization test

This test was used to study the ability of organisms to utilize citrate present in Simon's medium as a sole source of carbon for growth. Simon's citrate agar was prepared, dispensed into test tubes, autoclaved and allowed to solidity in a slanting position. The isolates were streaked from freshly prepared cultures and incubated at 37^0C for 48 hours. The content of the test tubes were observed for the development of growth with blue colour as opposed to the original green colour of the medium which signifies citrate utilization (Cheeshrough, 2005).

2.7.5 Voges –proskauer test

The medium used for this test is glucose phosphate medium. After sterilization, the medium was allowed to cool and the test organism was inoculated into the broth and incubated for five days at 37^0C. After incubation, 1.5ml of 5% alcoholic alpha napthtol and 0.5 ml of 40% aqueous KOH were added. The test tubes were shaken vigorously and allowed to stand for 5 minutes. The content was observed for the development of pink or red colour (Cheeshrough, 2005).

2.7.6 Methyl red test

About 5 drops of methyl red solution was added to 2ml of a five- day old culture of the isolates inoculated in glucose-phosphate broth. Red colouration indicated a positive test while yellow colour indicated negative text (Cheeshrough, 2005).

2.8 Statistical analysis

Statistical analysis was carried out using Statistical Package for Social Sciences (SPSS, Version 20.0). Analysis of variance (ANOVA), P- values test of significance, was carried out

at 95% level of confidence, P - values was use to determine the significance levels between various treatments and data obtained during the study.

RESULTS AND DISCUSSION

The prevailing soil bacteria present in the various study sites including the control were ascertained following standard microscopic, cultural and biochemical methods. The various identification techniques used for the characterization of the bacterial isolates were as shown in Table 1.The bacterial diversity present in the Control soil, Site A (500m) and Site B (1000m) of this study was as represented in Table 2 These isolated bacteria were subjected to hydrocarbon degradation test in order to ascertain those that possess the potential to degrade the hydrocarbon component of the crude oil Table 3.

Table 1: Characterization of bacteria isolated from the study sites.

Strain	1	2	3	4	5	6
Gramstaining	–	–	+	–	–	–
Motility	+	+	+	+	+	+
Catalase	+	+	+	+	+	+
Glucose	+	–	–	+	–	+
Lactose	–	–	–	–	–	–
Butt stant	AB	BB	BB	AB	BB	AB
Gas	+	–	–	+	–	–
H_2S	–	–	+	–	–	+
Citrate	+	–	+	+	–	+
Urease	–	+	+	+	+	–
Methly red	+	+	–	–	–	–
V.P	–	–	+	+	+	+
Glucose	AG	A	A	A	A	A
Sucrose	–	–	–	–	–	–
Probable Organism	*Alcaligenes* sp.	*Pseudomonas* sp.	*Bacillus* sp.	*Acinetobacter* sp.	*Serratia* sp.	*Citrobacter* sp.

AG =acid growth, AB =acid and base, BB=base base, A= acid.

Table 2: Culturable bacterial diversity presents in the various study sites

Control	Polluted site (500m) away	Polluted site (1000m) away
Acinetobacter sp.	*Pseudomonas* sp.	*Alcaligenes* sp.
Alcaligenes sp.	*Bacillus* sp.	*Citrobacter* sp.
Pseudomonas sp.	*Acinetobacter* sp.	*Bacillus* sp.
Serratia sp.		*Acinetobacter* sp.
Bacillus sp.		

Table 3 Culturable hydrocarbon utilizing bacterial isolates from the study sites.

Control	Polluted site (500m) away	Polluted site (1000m) away
Acinetobacter sp.	*Pseudomonas* sp.	*Alcaligenes* sp.
Alcaligenes sp.	*Bacillus* sp.	*Citrobacter* sp.
Pseudomonas sp.	*Acinetobacter* sp.	*Bacillus* sp.
Bacillus sp.		*Acinetobacter* sp.

In Table 4, the physical parameter for *Pseudomonas sp* was recorded for 30 days at 5 days interval, the colour change mean ranged from 2536.67±234.79Pcu on day 0 to 6745.00 ±17.30Pcu for 30 days measured in Cobalt per Unit, while Optical Density ranged from 0.88±0.00 from day 0 to 1.77±0.00 for 30 days. The pH was drastically changing to acidic in the experiment as it ranged from 6.18±0.02 to 5.26±0. 05. The Total Culturable Heterotrophic Bacterial Count (TCHBC) measured in Cfu/ml increases from 1646.67±55.08 on the previous day to 42666.67±3055.05 for day 30. (TPH) decreased from 4.58±0.01 Mg/ml to 0.02±0.00Mg/ml, this show that *Pseudomonas sp.* is active in the degradation process. Then the Total Hydrocarbon Content (THC) decrease in percentage to 68.33 ±2.01%.

Similarly the physical parameter for *Bacillus sp*. was recorded for 30 days at 5 days interval, the colour change mean ranged from 2536.67±243.79Pcu on day 0 to 2416.67 ±35.12Pcu for 30 days measured in Cobalt per Unit, while Optical Density ranged from 0.88±0.00 from day 0 to for 30 days 2.39±0.01. There was a slight increase in pH as it ranged from 6.18±0.02 to 6.30±0.08. The Total Culturable Heterotrophic Bacterial Count (TCHBC) measured in Cfu/ml increases from on the first day of 1180.00±20.00 to 34666.6±7577.35 for 30 days. The Total Petroleum Hydrocarbon (TPH) decreased from 4.58±0.00 Mg/ml to 0.11±0.06 Mg/ml, this show that *Bacillus sp.* is really degrading the hydrocarbon but at a slow rate. Then the Total Hydrocarbon Content THC) decrease in percentage to 53.33± 4.04%.

Furthermore, the physical parameter results for *Acinetobacter sp.* were recorded for 30 days at 5 days interval, the colour change mean ranged from 2536.67±243.79Pcu on day 0 to 5406.67±268.58 Pcu for 30 days measured in Cobalt per Unit, while Optical Density ranged from 0.88±0.00 from day 0 to for 30 days 2.50±0.01. There was a drastically increase in pH from 6.18±0.02 to 8.21±0.14. The Total Culturable Heterotrophic Bacterial Count (TCHBC) measured in Cfu/ml increases from on the first day 1230.00±36.06 to 63333.33±28867.51 for 30 days. The Total Petroleum Hydrocarbon (TPH) decreased from 4.58±0.00 Mg/ml to 0.05±0.07 Mg/ml, this show that *Acinetobacter sp.* is really degrading the hydrocarbon but at a slow rate. Then the Total Hydrocarbon Content (THC) decrease in percentage to 52.67±1.53 %.

Similarly, the physical parameter for *Alcaligenes* sp. was recorded for 30 days at 5 days interval, the colour change mean ranged from 2536.67±243.79Pcu on day 0 to 4786.67 ±32.15Pcu for 30 days measured in Cobalt per Unit, while Optical Density ranged from 0.88±0.00 from day 0 to for 30 days 1.33±0.01. There was a slight decrease in pH as it ranged from 6.18±0.02 to 5.30±0.08. The Total Culturable Heterotrophic Bacterial Count (TCHBC) measured in Cfu/ml increases from on the first day 131.00±45.83 to 51000.00 0±165.15 for 30 days. The Total Petroleum Hydrocarbon (TPH) decreased from 4.58±0.00 Mg/ml to 0.04±0.00 Mg/ml, this show that *Alcaligenes sp.* is really degrading the hydrocarbon. Then the Total Hydrocarbon Content (THC) decrease in percentage to 71.67± 0.58%.

However, the strain identify as *Pseudomonas* sp. possess a steady increase in optical density (OD) from 0.88 to 1.29, which raises the pH from 4.59 to 5.26 and increase in colour change

from 2536.67pcu to 6745.00pcu after 30 days of degradation monitoring As shown in Table 4.5. OD increases slowly as seen in the second strain indentified as *Bacillus* sp. from 0.88 to 2.39, pH of 5.57 – 6.30 and shows a decrease in colour change from 4393.67pcu to 2416.67pcu during the degradation study. There was a steady increase in OD as seen in *Acinetobacter* sp. which increases from 0.88 to 2.50 for 30 days which raises the pH from 5.46 to 8.21 and increase in colour change of 2536.67pcu to 5406.67pcu. Fourth Strain identified as *Alciligenes* sp. showed an increase in OD from 0.88 to 1.33, while pH increases from 2.98 to 5.30 and increase in colour change shows a wide range of 2536.67pcu to 4786.67pcu.

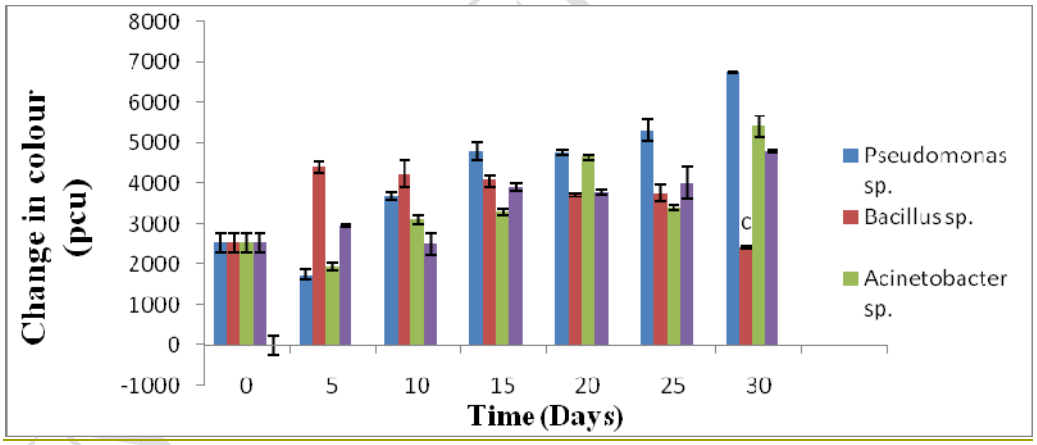
Table 4: Responses of selected hydrocarbon utilizing bacteria as used in biodegradation studies

Day	**Parameters**	***Pseudomonas* sp.**	***Bacillus* sp.**	***Acinetobacter* sp**	***Alcaligenes* sp**
0	Colour (pcu)	2536.67±234.79	2536.67±234.76	2536.67±243.79	2536.67±243.79
	OD	0.88±0.00	0.88±0.00	0.88±0.00	0.88±0.00
	pH	6.18±0.02	6.18±0.02	6.18±0.02	6.18±0.02
	TCHBC(cfu/ml)	1646.67±55.08	1180.00±20.00	1230.00±36.06	1310.00±45.83
	TPH (mg/ml)	4.58±0.00	4.54±0.01	4.58±0.00	4.58±0.00
	THC (%)	100.00±0.00	100.00±0.00	100.00±0.00	100.00±0.00
5	Colour (pcu)	1746.67±122.20	4393.33±125.03	1946.67±83.27	2060.00±30.00
	OD	1.29±0.07	1.44±0.06	1.04±0.03	1.16±0.03
	pH	4.59±0.34	5.57±0.01	5.46±0.08	2.98±0.08
	TCHBC(cfu/ml)	1656.67±65.06	2190.00±101.49	1230.00±20.00	1773.00±61.10
	TPH (mg/ml)	2.84±0.00	2.80±0.02	2.93±0.00	2.86±0.00
	THC (%)	97.67±0.58	94.67±0.58	97.00±1.00	97.33±1.53
10	Colour (pcu)	3690.00±105.83	4240.67±320.78	3103.33±127.41	2496.67±271.54
	OD	1.15±0.00	1.22±0.09	0.91±0.02	1.05±0.03
	pH	4.61±0.45	5.64±0.03	5.51±0.02	4.20±0.02
	TCHBC(cfu/ml)	2030.00±170.00	2273.33±70.24	1600.00±163.71	2346.67±50.33
	TPH (mg/ml)	1.08±0.00	1.08±0.00	1.29±0.00	1.15±0.01
	THC (%)	93.00±4.36	87.33±0.58	94.33±0.59	94.00±1.00
15	Colour (pcu)	4780.00±233.02	4060.67±148.44	3293.33±66.58	3910.00±80.00
	OD	0.25±0.05	1.27±0.00	1.15±0.00	1.23±0.01
	pH	4.99±0.02	5.93±0.04	5.55±0.01	4.53±0.01
	TCHBC(cfu/ml)	35333.33±305.05	15266.67±305.12	18000.00±964.37	13866.67±583.15
	TPH (mg/ml)	0.62±0.00	0.62±0.00	0.68±0.00	0.65±0.01
	THC (%)	82.00±2.31	65.00±1.00	71.33±1.53	90.33±1.53
20	Colour (pcu)	4760.00±60.83	3715.00±37.75	4640.00±72.86	3790.00±69.28
	OD	1.45±0.00	1.32±0.00	1.38±0.00	1.24±0.03
	pH	4.83±0.29	5.95±0.02	5.83±0.05	4.73±0.10
	TCHBC(cfu/ml)	48000.00±458.58	82666.67±541.63	56666.67±527.25	19000.00±590.00
	TPH (mg/ml)	0.12±0.00	0.16±0.00	0.08±0.00	1.16±0.00
	THC (%)	76.33±0.58	63.33±1.15	59.00±1.00	72.33±1.53
25	Colour (pcu)	5300.00±270.14	3760.00±216.56	3396.67±56.86	4003.00±408.57
	OD	1.38±0.00	1.54±0.01	1.44±0.01	1.26±0.05

	pH	4.99±0.03	6.01±0.02	5.83±0.05	4.81±0.04
	TCHBC(cfu/ml)	46000.00±0.00	34666.67±305.05	17366.67±105.87	50000.00±480.89
	TPH (mg/ml)	0.07±0.00	0.14±0.00	0.05±0.00	0.05±0.00
	THC (%)	72.33±1.53	60.33±0.58	57.33±1.53	70.67±2.52
30	Colour (pcu)	6745.00±17.30	2416.67±35.12	5406.67±268.58	4786.67±32.15
	OD	1.77±0.00	2.39±0.01	2.50±0.01	1.33±0.01
	pH	5.26±0.04	6.30±0.08	8.21±0.14	5.30±0.08
	TCHBC(cfu/ml)	42666.67±305.05	34666.60±577.35	63333.33±287.51	51000.00±165.15
	TPH (mg/ml)	0.02±0.00	0.11±0.06	0.05±0.07	0.04±0.00
	THC (%)	68.33±2.01	53.33±4.04	52.67±1.53	70.10±0.58

Mean ± standard deviation of triplicate determination

The information from the degradation studies as shown in the table above (Table 4) were analysed and represented in bar charts. Changes in colour (pcu) against Time(Days) (Figure 1), Changes in total hydrocarbon content(%) against Time(Days) (Figure 2), Changes in Optical Density (OD) against Time(Days) (Figure 3), Changes in total petroluem hydrocarbon in (mg/ml) against Time (Days) (Figure 4) Changes in pH against Time(Days) (Figure 5), Changes in total cuturable heterotrophic bacteria counts (cfu/ml) against Time(Days) caused by some of the isolates during the degradation studies were represented.



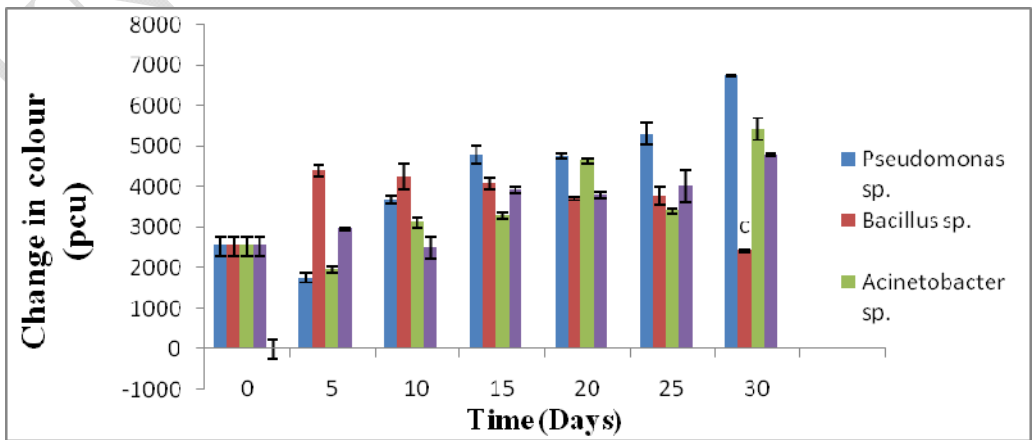
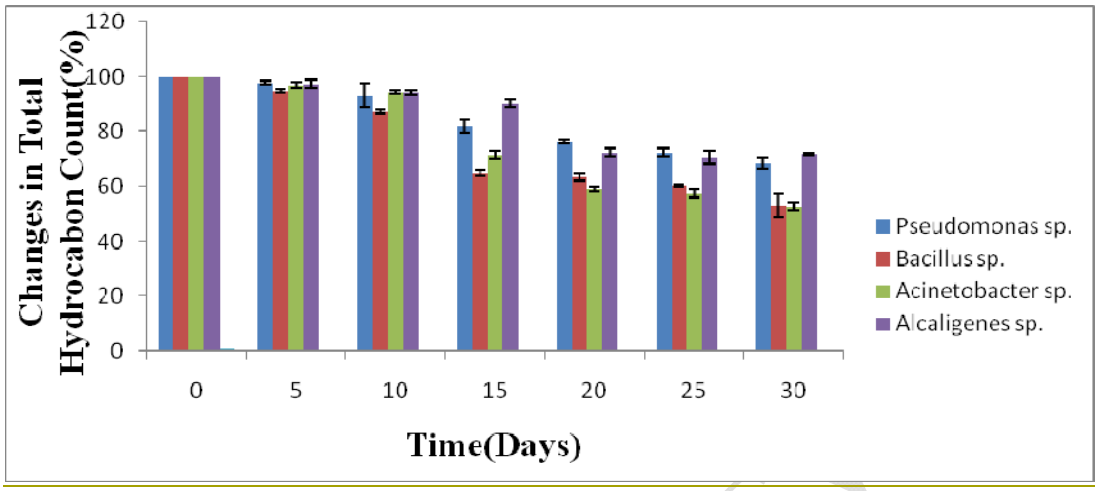


Figure 1: Changes in colour (pcu) against Time(Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations)



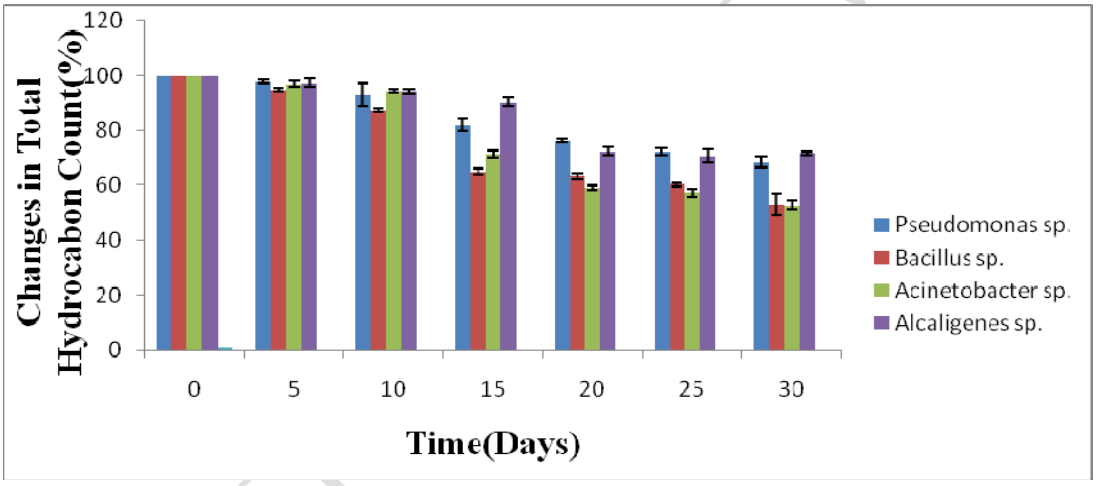
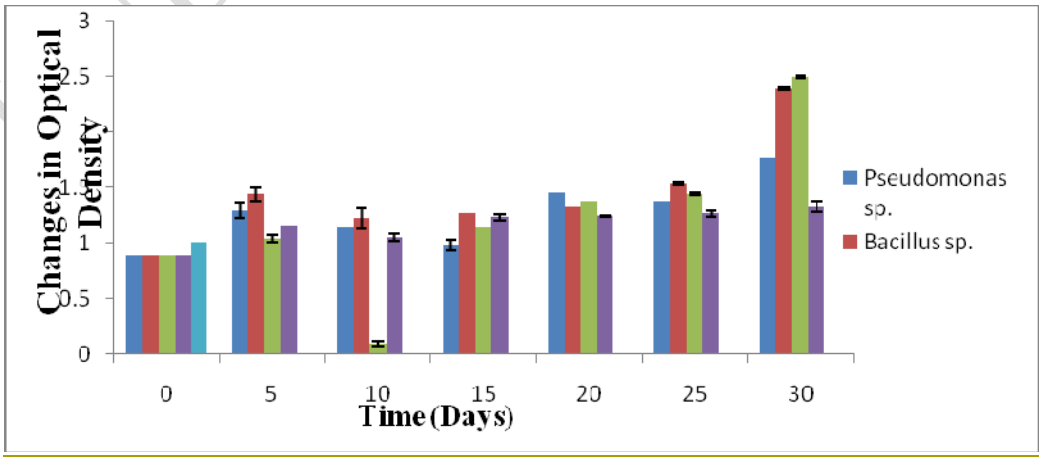


Figure 2: Changes in Total Hydrocarbon Content(%) against Time(Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations)



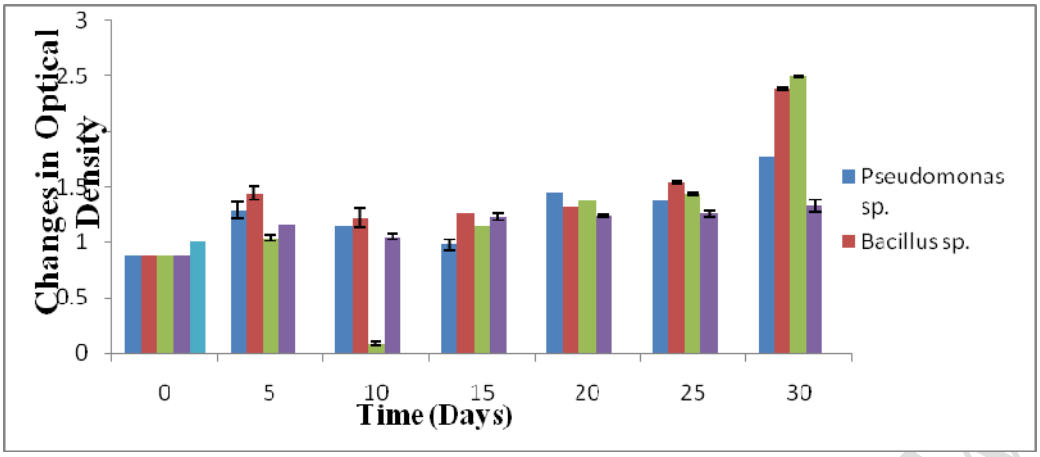


Figure 3: Changes in Optical Density (OD) against Time in (Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations)

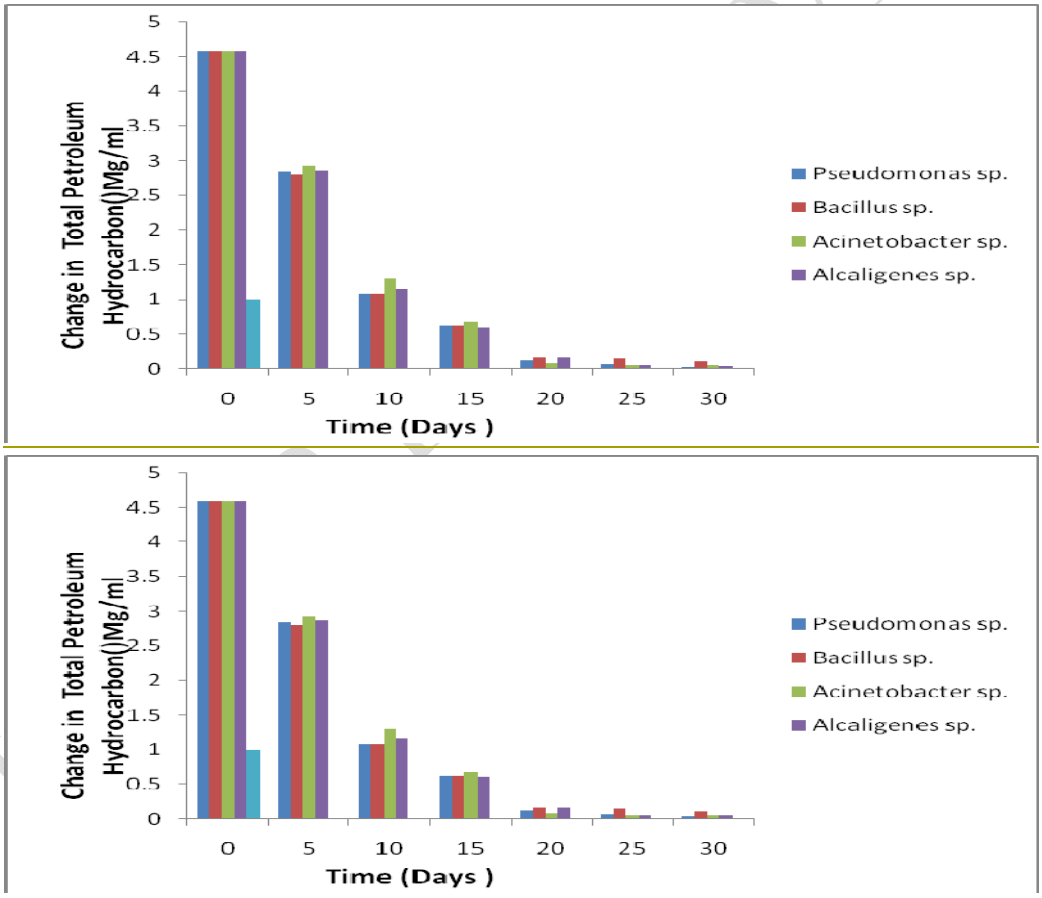


Figure 4: Changes in total petroluem hydrocarbon in (mg/ml) against Time in (Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations)

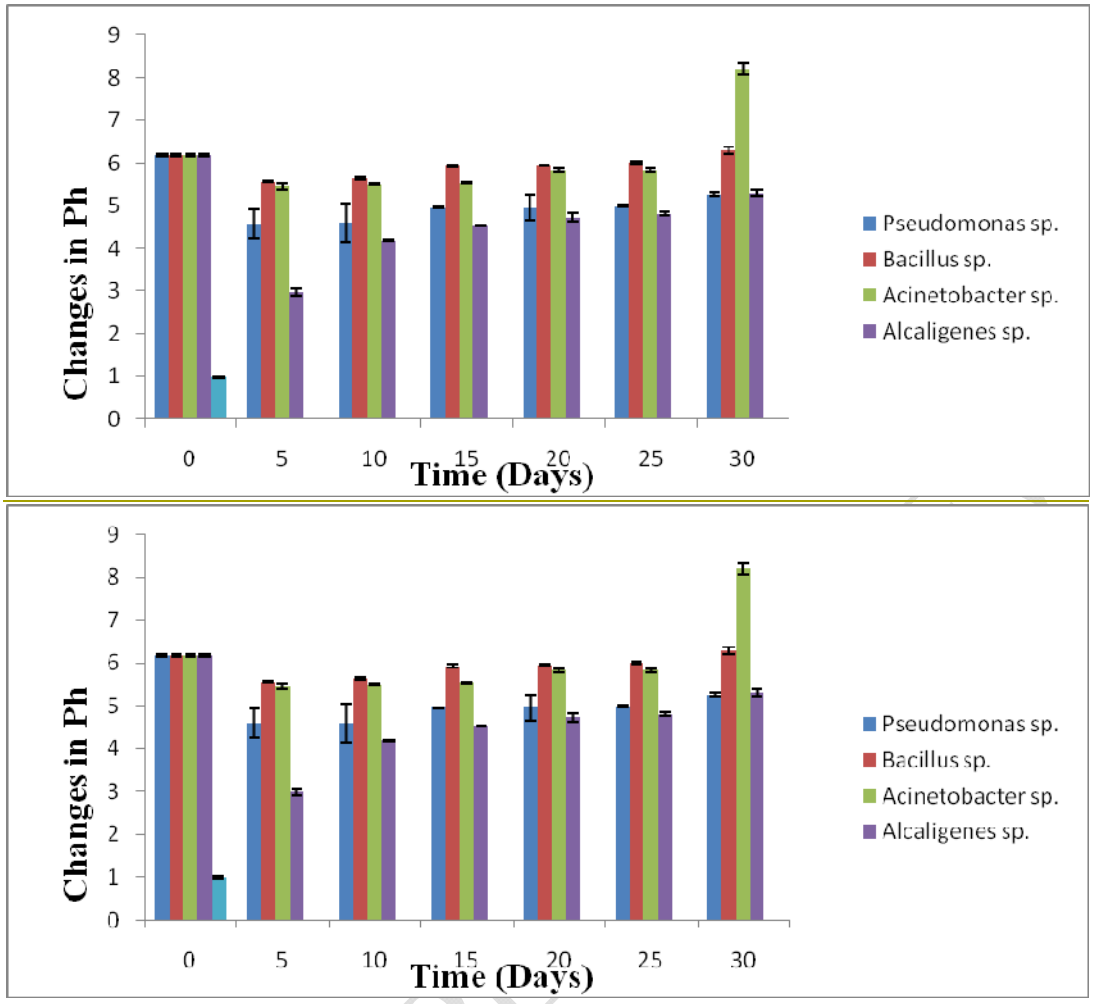
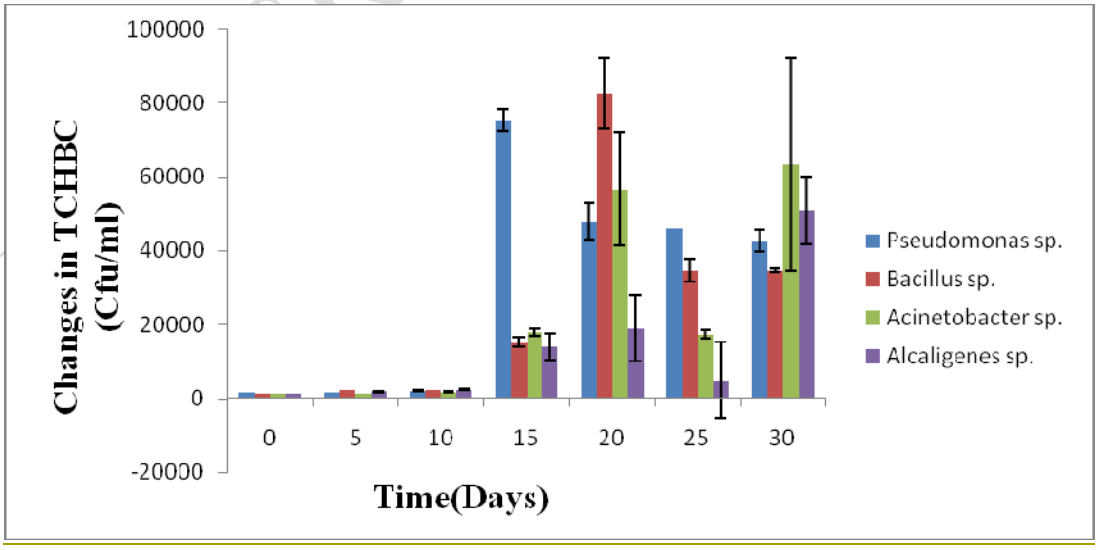


Figure 5: Changes in pH against Time in (Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations.



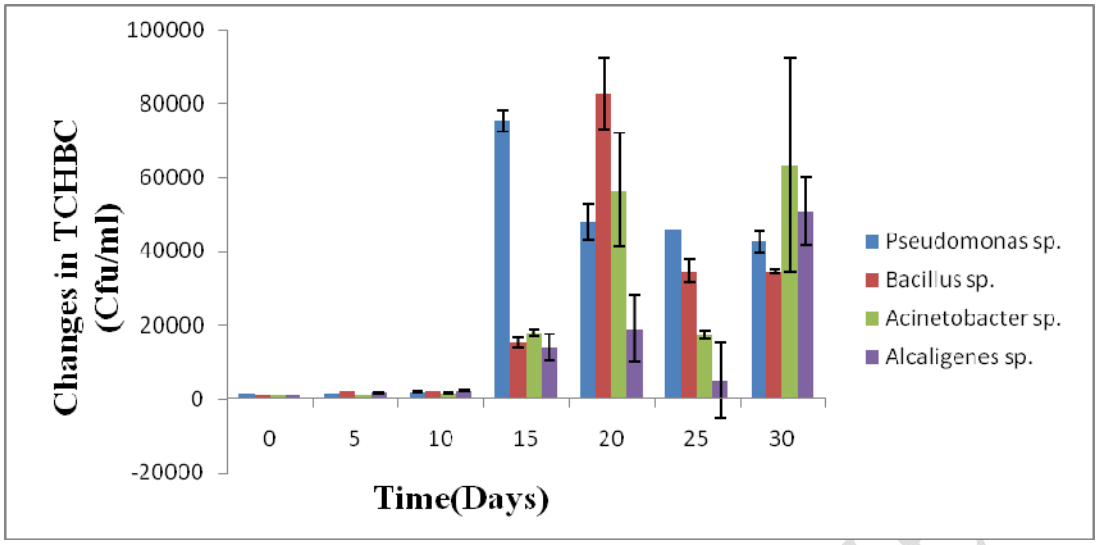


Figure 6: Changes in total cuturable heterotrophic bacteria counts (cfu/ml) against Time in (Days) caused by some of the isolates during the degradation studies (Data are mean ±S.D of triplicate determinations)

The ability of these bacteria to be predominantly Gram- negative is contrary to Austin *et al.* (1997) who isolated predominantly gram-negative hydrocarbon utilizing bacteria from soil and aquatic environment. The isolation of Bacillus species is in agreement with the work of Okpokwasili and Okorie (1990), who found that Bacillus species could also be one of the predominant Gram positive organisms found in oil polluted areas

These findings showed that the biodegradation of complex hydrocarbons in nature usually required the cooperation of more than a single species. The microbial populations consisting of the strains belonging to various genera have been detected in petroleum-contaminated soil (Ilori *et al*,. 2006; Kim and Crowley 2007). This suggested that the strains from various genera have their roles in the hydrocarbon transformation processes (Ghazali *et al*,. 2004; Cunliffe and Kertesz 2006). Individual microorganisms can metabolize only a limited range of hydrocarbon substrates; hence assemblages of the mixed populations with overall broad enzymatic capacities would be required to achieve considerable biodegradation of petroleum hydrocarbons.

Statistical analysis of the result shows that there is significant difference between various heterotrophic bacteria and soil samples.

CONCLUSION

Environment pollution caused by released of a wide range of compound as a consequence of industrial progress has assumed serious proportions. To prevent development of hazardous waste the process of bioremediation has been followed. Our present study follows the isolation of hydrocarbon degrading bacteria from oil polluted sites. Sample was collected from contaminated sites of Sakpenwa Community, Tai L.G.A, Rivers State, These were brought then to the laboratory and isolation was done on the basis of gram staining. Biochemical tests were performed with isolates. The isolates were screened for their oil degrading capacity.

Having the knowledge of the processes and factors involves in the biodegradation experiment is of great ecological significance in the society. The elimination of oil spilled in the environment can be achieved by microbial degradation when added up with some physical and chemical methods

Further scale-up studies as applicable need to be carried out in increasing the degrading ability and stability of the crude oil degrading isolate and its usage as a possible commercial strain

We concluded that the use of consortium species makes biodegradation study more effective.

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