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ULTIMATE BIODEGRADABILITY POTENTIAL OF DRILLING FLUIDS AND OIL SPILL DISPERSANTS IN FRESHWATER ECOSYSTEM

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Abstract

The present study investigates ultimate biodegradability potential of drilling fluids-oil base and water base, and oil spill dispersants - OSD/LT and OSD/Seacare employed in the upstream sector of the Nigerian petroleum industry in fresh water environment. Percentage ultimate biodegradability and petroleum product utilizing bacteria (PPUB-log10cfu/ml) were used as methods for the assessment of biodegradability potentials of the drilling fluids and oil spill dispersants. The results of the percentage (%) ultimate biodegradability of the drilling fluids and oil spill dispersants at day 20 showed the following trend: water base (55.5%) > oil base (54.8%)> OSD/Seacare (54.1%) >OSD/LT (41.1%). The results of petroleum products utilizing bacteria (PPUB-log10cfu/ml) at day 20 followed the trend: Water b base DF (4.89±0.65)>OSD/LT (4.71±0.05)> oil base DF (3.45±0.31)> OSD/Seacare (3.30±0.58). The PPUB-log10cfu/ml decreased from day 1 to 20 with drilling fluid water base contaminated fresh

water sample having the highest cumulative count (4.89±065). The perturbation of biogeochemical cycles by these pollutants could have altered the process of decomposition, mineratization and nutrient regeneration. In conclusion, the study showed that drilling fluids were relatively more biodegradable than oil spill dispersants.

Introduction

The petroleum (oil and gas) industry in Nigeria is composed of two major sectors: the "Upstream Sector" and "Downstream Sector". The "upstream sector" focus on oil exploration and production (E&P) whereas the "Downstream sector" deals with crude oil refining, processing, distribution and marketing (UNEP 2006; NRC, 2005 and NEST 1991).

Petroleum-based products: kerosene, gasoline, diesel fuel, home heating oil etc, are the major source of energy for industries and other domestic activities. Petroleum is also the raw material for many chemical products such as plastics, paints and cosmetics. Environmental pollutions originating from petroleum and petrochemical products has been recognized as a significant and serious safety and health hazard threats (ACGIH, 2003; DPR 2002). Most components of oil are toxic to humans and wildlife as they are easily incorporated into the food chain with the resultant effect of bioaccumulation, bioconcentration and biomagnifications or bioamplification. Expectedly, this has attracted and stimulated scientists' interests worldwide to examine the distribution, fate of oil and its derivatives in the environment (Schlemmer et al., 2002; USA EPA, 2010; NRC, 2005, and NEST, 1991).

Drilling fluids, oil spill dispersants, degreasers and industrial detergents are commonly employed in upstream sector of the Nigeria petroleum (oil and gas) industries (DPR, 2002). In Nigeria, the two commonly used types of drilling fluids are water based and oil based (mineral/synthetic or pesudo) muds. Drilling fluids are mixture of natural and synthetic chemical compounds used to cool and lubricate the drill bit, clean the hole bottom, carry cuttings to the surface, control formation pressure and improve the function of the drill string and tools in the hole (DPR, 2002, Burke, & Veil, 2001).

Dispersants are chemical agents that reduce interfacial tension between oil and water in order to enhance the natural process of dispersion by generating larger numbers of small droplets of oil that are entrained into the water column (USA EPA 2010). They are class of chemical compounds used in the control of oil spillage in marine water, fresh water and brackish water (NRC, 2005; API, 1997 & 2003).

Biodegradability of drilling fluids and oil spill dispersants or the amount of oxygen required to breakdown these chemicals in environment is a major concern to environmentalist when using them. Dispersants in particular, exhibits high demand for oxygen, hence their use in oil spillage in polluted coastal bay or inland waters with limited air circulation could deplete or lower the dissolved oxygen resources, thereby causing damage to biological community in such waters (Hamdam and Fulmer, 2011, ACGIH, 2003).

Biodegradation is an important factor for reducing and removal of organic contaminants from the environments. The evaluation of biodegradability of

anthropogenic organic substrate or substance is an essential parameter for environmental risk assessment and requires appropriate legislations as observed by the National Research Council (NRC, 1989, API, 2003.

A changing pattern of anthropogenic activities have been linked to inconsistencies in the relationship of ecological systems in the environment. The choice of fossil fuel materials used for energy production is directly responsible for increase in carbon dioxide and other gasses resulting in the current trend of global warming. Consequently, new petroleum products used in the upstream sector of the Nigeria petroleum industry whose biodegradability rate are extremely low and ecotoxicological indices are high constitute greater harmful effects in the aquatic environment (Nrior and Odokuma 2015, Nrior and Onwunka, 2017).

The present study is aimed at evaluating and comparing the biodegradability potentials of the drilling fluids and oil spill dispersants and possible discharge dilution in fresh water environment.

Materials and Methods

Source of Sample

Water sample: Fresh water samples were collected with sterile plastic ten (10) litre containers from Asarama stream in Asarama town, Andoni Local Government Area in Rivers state, Nigeria. The containers were rinsed three times with the water samples to be collected at the site before collection was made. The river serves as the major sources of drinking water.

Source of Microorganism for Biodegradability Test

The microorganisms for the biodegradation study were naturally occurring micro flora in the fresh water environment.

Source and Type of Drilling Fluids and Oil Spill Dispersants

Drilling fluids – water and oil base where obtained from Addax Petroleum Limited, Izombe, Owerri, and Imo State Nigeria. Oil spill dispersants (OSD) – OSD/LT and OSD/Seacare were purchased from Offshore Chemicals, Trans –Amadi Industrial layout, Port Harcourt.

Biodegradation Monitoring

Biodegradation monitoring were set up for each drilling fluid (water base and oil base) and oil spill dispersant (OSD/LT and OSD/Seacare). 300ml of the freshwater from Asarama Stream were dispensed into three 500ml Erlenmeyer flask separately. After that, 1% (3ml) of drilling fluid (water base) was dispensed into the first flask and 1% (3ml) of drilling fluid (oil base) was dispensed into the second flask. The third flask was not contaminated with any drilling fluid and was used as a control. Same processes were carried out for oil spill dispersants (OSD/LT and OSD/Seacare).

The flasks were perforatedly plugged to allow aeration, and were kept at ambient temperature (28 ± 2^{0}C) for 20 days. Samples were taken at day 0, 5, 10, 15, and 20 from the Erlenmeyer flasks containing fresh water contaminated with the test chemicals. This was to determine the hydrogen ion concentration (pH), total dissolved solids (TDS), total heterotrophic bacterial counts, total fungal counts, and hydrocarbon utilizing bacteria (Nrior and Odokuma, 2015).

Microbiological Analysis

Isolation and Enumeration of Total Heterotrophic Bacteria

Total heterotrophic bacteria for each biodegradation set up were enumerated by spread plate method. 0.1ml aliquot of the 10^{-1} to 10^{-4} was transferred unto well-dried nutrient agar plates and incubated at 37^0C for 24 to 48h, after incubation the bacterial colonies that grew on the plates were counted and sub-cultured unto fresh nutrient agar plates using the streak plate technique. Discrete colonies on the plates were aseptically transferred into agar slants, properly labeled and stored as stock cultures for preservation and identification (Odokuma and Ibor, 2002; Nrior and Odokuma, 2015).

Isolation and Enumeration of Total Fungal Count

The total fungi population in the biodegradation set up (Habitat water sample and TCE) were enumerated and isolated by inoculating 0.1ml aliquot of the mixture unto well-dried potato dextrose agar containing antibiotics (Tetracycline, Penicillin and Ampicillin) to inhibit bacterial growth. Pure cultures of the fungi isolates were enumerated and transferred unto potato dextrose agar slants as stock cultures for preservation and identification (Odokuma and Okpokwasili, 1992; Nrior and Odokuma, 2015; Nrior *et al.,* 2017).

Isolation and Enumeration of Hydrocarbon Utilizing Bacteria

Enumeration of hydrocarbon utilizing bacteria was performed by inoculating 0.1ml aliquot of the dilutions unto mineral salt agar plates containing the hydrocarbons (Odokuma and Okpokwasili, 1992; Nrior and Onwuka, 2017). Colonies were counted after 48 to 72 h incubation at ambient temperature. The bacterial colonies on the plates after incubation were counted and sub-cultured onto fresh mineral salt agar plate.

Identification of Bacterial and Fungal Isolates

The cultural, morphological and biochemical characteristics of the discrete bacterial isolates were compared with the recommendation in Bergey's manual of determinative bacteriology (1994). The morphological and biochemical test include; gram staining, motility, catalase, oxidase, citrate utilization, hydrogen sulphide production, indole production, methyl red and voges proskauer tests. The presence or absence of septa in the mycelium, type of spore, presence of primary or secondary sterigmata, and other microscopic characteristics as well as cultural characteristics were used in the identification of the fungal isolates of the biodegradation flask set up (Cheesbrough, 2006).

Physico-chemical Analysis

The physico-chemical parameters analyzed were: pH using electrometric pH meter (Jenway 3015 method), dissolved oxygen (DO) and biochemical oxygen demand (BOD) were determined by modified winkler method (APHA, 2010; Nrior and Onwuka, 2017), chemical oxygen demand (COD) was determined by permanganate oxidation method from the biodegradation set-up on days 0, 5, 10, 15 and 20.

Ultimate Biodegradation Monitoring Using the Percentage Ratio of BOD to COD Ultimate biodegradation was monitored using the percentage ratio of BOD to CO2. Adopting standard method of APHA (2010), the biochemical oxygen demand (BOD) of

each biodegradation set up was monitored at 0, 5, 10, 15 and 20 days, whereas chemical oxygen demand was determined at day 0. Ultimate biodegradation referable to also as the percentage of carbon in the material that is potentially mineralizable was calculated from the percentage of the ratio of BOD for day 0, 5, 15, and 20 to COD at day 0 (API, 1997; API, 2003; and, Nrior and Odokuma, 2015). The percentage of mineralizable carbon in the test compounds that was actually mineralized was derived from the formular (API, 1997; API, 2003, and, Nrior and Odokuma, 2015):

$$\frac{P}{I} \times 100$$

$$100 - M = N$$

Where

P = percentage of potentially mineralizable carbon in the test compound
I = Percentage of potentially mineralizable carbon in test compound at day O
N = Percentage of potentially mineralizable carbon in test compound that was actually mineralized.

Results and Discussion

The result of physico-chemical properties of the fresh water environment is presented in Table 1. The general appearance of the water was clear, odour have unobjectionable sensory evaluation. Colour was 20.0, Hazen; pH (7.67), Temperature (28.04) Electrical conductivity (EC) (20.0 µS/cm), Turbidity (1.00 NTU), Total hardness (30.70 mg/L), Total alkalinity (22.0 mg/L), Chloride (28.0 mg/L).

Total Dissolved Solids (10.0 mg/L), Total Solids (18.5 mg/L), and Total Suspended Solids (8.5 mg/L). The values of other parameters were: Nitrate (2.30 mg/L), Sulphate (9.80 mg/L), Calcium (7.75 mg/L), Magnesium (1.40 mg/L). Biochemical Oxygen Demand (BOD: 21.90 mg/L) and Chemical Oxygen Demand (COD: 52.60 mg/L).

Table 1: Physico-chemical Characteristics of Fresh Water

S/N	Physico-chemical characteristics	Unit	Fresh water
1	General Appearance	-	Clear
2	Odour	-	unobjectionable
3	Colour	Hazen	20.00
4	pH	-	7.67
5	Electrical Conductivity (EC)	µS/cm	20.00
6	Temperature	^{0}C	28.04
7	Turbidity	NTU	1.00
8	Total Hardness	mg/L	30.70
9	Total Alkalinity	mg/L	22.00
10	Chloride	mg/L	28.00
11	Total Suspended Solids (TSS)	mg/L	8.50
12	Total Dissolved Solids (TDS)	mg/L	10.00
13	Total Solids	mg/L	18.50
14	Nitrate	mg/L	2.30
15	Sulphate	mg/L	9.80
16	Calcium	mg/L	7.75
17	Magnesium	mg/L	1.40

18	Biochemical Oxygen Demand (BOD)	mg/L	21.90
19	Chemical Oxygen Demand (COD)	mg/L	52.60
20	Total Iron (Fe)	mg/L	0.65
21	Lead (Pb)	mg/L	<0.001
22	Copper (Cu)	mg/L	0.001

The results of physical and chemical properties of the biodegradation set-up showed that the alkaline pH range as well as hemophilic temperature range favored the acclimatization process for the native hydrocarbon utilizing microbial population. These physic-chemical factors were particularly important for the survival of hydrocarbon utilizing microbial consortium in the aquatic ecosystems. These findings corroborated the findings of Nrior *et al (*2017). With respect to sensory evaluation the odour of the fresh water was unobjectionable.

Eight bacterial isolates obtained from the mixture of Drilling fluid - oil base and water base, Oil spill dispersant - OSD/LT and OSD/Seacare with freshwater sample were: Micrococus, Corynebacterium, Bacillus, Pseudomonas, Staphylococcus, Proteus, Escherichia, Citrobacter (Table 2). Total heterotrophic viable bacteria (THVB) count decreases from day 1 to day 20; with oil spill dispersant OSD/LT contaminated freshwater sample having the highest cumulative count (6.78±0.36 log10cfu/ml-Table 3).

Statistical evaluation of the growth of THVB result during the biodegradation of Drilling fluids and Oil Spill Dispersants revealed the following order: Total Heterotrophic Bacteria (THB - log10 cfu/ml) in fresh water: OSD/LT (6.78±0.36) > OSD/Seacare (6.67±0.29) > Control (6.37±0.19) > Water base (6.32±0.23) > Oil base (6.21±0.64). Generally, this meant that the intermediates produced from the degradation of Drilling fluids and Oil Spill Dispersants in these aquatic systems favoured the growth of a larger population of heterotrophic bacteria. Odokuma and Ibor, 2002 observed a similar trend.

Six genera of fungal isolates identified were – Aspergillus, Alternaria, Fusarium, Penicilium, Rhizopus and Mucor (Tables 4&5). The Macroscopic morphology was based on best growth temperature, growth rate, colour on SDA, colour on reverse side, texture and special feature while the microscopic morphologies and identities of the different species of the fungal isolates was based on characteristic features of conidiophores, phialides, vesicle, sclerotia, hulle cells, sporangiophore, apophysis, columella, sporangium and rhizoids. The fungal species identified were: Aspergillus flavus, Aspergillus fumigates, Aspergillus versicolor, Penicilium marneffei, Penicilium chrysogenum, Rhizopus microspores, Rhizopus arrhizus, Mucor racemosus, Mucor amphibiorum (Tables 4 and 5).

The trend observed in THVB was same with total fungal population. Statistical evaluation of the growth of total fungi during the biodegradation of drilling fluids and oil spill dispersants revealed the following: Total viable fungi count (TVFC - log10 cfu/ml) in freshwater: OSD/Seacare (4.26±0.36)>control (3.67±0.19)>OSD/LT (3.61±0.22)>Water base (3.24±0.64)>Oil base (2.60±0.15; Table 6).

These test drilling fluids and oil spill dispersants showed mild increases and decreases in the total microbial (fungal) population in the freshwater used as inoculums. This observation is in agreement with the report of Nrior and Odokuma,

(2015) that, oil spill dispersants support mild increases (stimulation) and decrease (inhibition) in the growth of specific heterotrophic fresh water bacteria. This response also applies to fungal population in this study.

Responding to changes in the environment is a fundamental property of a living cell and chemotaxis is the best studied bacterial behavioural response that navigates the bacteria to niches that are optimum for their growth and survival (Nrior et al., 2017). However, Odokuma and Okpokwasili (1992); Nrior and Obire (2014) have observed that biodegradation of oil spill dispersants in artificial media of varying NaCl concentrations (0mg/l, 20,000mg/l and 40,000mg/l) decreases with increase NaCl concentration. Acclimatization of the microbial population with petroleum product components enhances the biodegradation efficiency of the microorganisms. Although bacterial population was more than fungal petroleum products degraders in the fresh water ecosystem, which agrees with previous findings of Odokuma and Okpokwasili (1992) and Nrior et al., 2017.

The adaptability of native microbial population in the fresh water to petroleum products components would be the reason for their success at mineralizing the petroleum products in the experimental set-up where the physicochemical properties of the ecosystem were supportive to the survival of these microorganisms (Nrior and Odokuma, 2015).

Evaluation of Hydrocarbon utilizing bacteria (HUB) in freshwater ecosystem revealed their population as follows: (Table 7) Water base (4.89±0.65) > OSD/LT (4. 71±0.05) > Oil base (3.45±0.31) > OSD/Seacare (3.30±0.68) > Control (2.34±0.23). The study revealed that Pseudomonas spp, Proteus spp, Micrococcus spp, Bacillus spp, Rhizopus spp, Aspergillus spp and Penicilium spp. genera isolated from the fresh water ecosystems were capable of utilizing petroleum products as their carbon source. Similar trend in the ability of natural microbiota to degrade novel or synthetic compounds has been reported (Ogbulie et. al., 2008 and Nrior and Onwuka, 2017).

Such similarity in the utilization of novel compounds by natural microflora is expected, since such breakdown depends on the possession of plasmids that are not naturally present in all microorganisms (Ogbuilie et. al., 2008). Hence, the ability to utilize xenobiotics must be dependent on the possession of the requisite enzymes necessary for such degradation.

Evaluation of percentage (%) ultimate biodegradation at day 20 (Table 8) showed: water base (55.5) > oil base (54.8) > OSD/Seacare (54.1) > OSD/LT (41.1). The difference in biodegradation potential of individual types of drilling fluids and oil spill dispersants could be due to differences in composition of each test chemical and difference in genetic makeup of indigenous microorganisms carrying out the degradation process (Patrick et al., 1991; Nrior et al, 2017), prolonged or previous exposure to the effluent (Obire and Nrior, 2014), mutation (Zelibor et al., 2000) and relative utilization of the effluent for metabolism.

Isolate	Shape	Colour	Texture	Elevation	Translucent	Gram	MR	Indole	Citrate	Coagulase	Oxidase	Catalase	Motility	Glucose	Lactose	Sucrose	Matrose	Probable Isolate
a)	Bacilli	Cream	Moist	Raised	Opaque	+	-	+	+	-	+	+	-	A	AG	-	-	*Micrococcus sp*
b)	Cocci	Cream	Dry	Flat	Flat	+	+	+	-	-	+	+	+	AG	-	A	A	*Corynebacterium*
c)	Rod	Cream	Mucoid	Raised	Raised	-	+	-	+	-	+	+	+	A	-	-	-	*Bacillus sp*
d)	Rod	Bluish gray	Moist	Flat	Flat	+	-	+	-	-	+	+	+	+	+	+	-	*Pseudomonas sp*
e)	Cocci	Yellow	Moist	Smooth	Transchisent	+	-	+	-	+	+	+	-	-	-	-	-	*Staphylococcus spp*
f)	Rod	Cream	Moist	Raised	Opaque	-	-	+	-	-	-	-	+	A	A	-	AG	*E.coli*
g)	Rod	Cream	Moist	Raised	Opaque	-	-	-	-	+	-	+	+	A	-	-	-	*Citrobacter sp*
h)	Rod	White	Sticky	Raised	Transchisent	-	-	-	-	+	+	+	+	AG	-	-	-	*Proteus sp*

Table 3: Total Heterotrophic Viable Bacterial Count During the Study

Day	Fresh Water + Control Log10cfu/ml	Fresh Water + Waterbase DF Log10cfu/ml	Fresh Water + Oilbase DF Log10cfu/ml	Fresh Water + OSD/LT Log10cfu/ml	Fresh Water + OSD/Seacare Log10cfu/ml
0	6.48	6.41	6.92	7.00	7.00
5	6.46	6.38	6.70	7.08	7.76
10	6.57	6.63	5.74	7.04	6.86
15	6.18	6.08	6.30	6.38	4.45
20	6.15	6.11	5.40	6.40	6.30
	6.37±0.19	6.32±0.34	6.21±0.64	6.78±0.36	6.67±0.25

Table 4: Macroscopic Morphologies of the Fungal Isolates and Their Identities

Species	Best growth temp.	Growth rate	Colour on SDA	Colour on reverse side	Texture	Special feature
Aspergillus flavus	25^0C	Rapid	Yellow to green	Goldish to red brown	Woolly cottony to somewhat granular	Sclerotia is dark brown
Aspergillus fumigates	25^0C	Rapid	Blue-green to gray	White to tan	Woolly cottony to somewhat granular	Display lavender diffusible pigment
Aspergillus versicolor	25^0C	Slow to moderate	Viariously coloured (versicolored), white at the beginning, turns	White to yellow or purplish red	Woolly to cottony	Exudates is pink to reddish brown

			to yellow, tan, pale green or pink			
Penicilium marneffei	35^0C	Slow	Creamy to slightly pink	Red	Glabrous to convoluted	Filamentous, flat, radially sulcate colonies
Penicilium chrysogenum	25^0C	Rapid	White to blue-green	Pale to yellowship	Velvety, woolly	The colonies initially white and become blue- green, pinkish
Rhizopus microspores	46^0C	Rapid	Initially white, quickly becoming pale gray and then developing small black dots in the mycelium	Pale	Cottony candy	Develop small black dots in mycelium which are mature sporangia
Rhizopus arrhizus	46^0C	Very rapid	Initially white, quickly becoming pale gray and then developing small black dots in the mycelium	White	Cottony candy	Develop small black dots in mycelium which are mature sporangia
Mucor racemosus	25^0C	Rapid	White	White	Cottony candy	Fluffy appearance
Mucor amphibiorum	25^0C	Slow	Grayish brown	White	Conttony	Fluffy appearance with height of several centimeters

Table 5: Microscopic Morphologies of the Fungal Isolates and Their Identities

Species	**Conidiopore**	**Phialides**	**Vesicle**	**Sclerotia**	**Hulle cells**	**Sporangio-phore**	**Apophysis**	**Columella**	**Sporangium**	**Rhizoid**
Aspergillus flavus	Colourless rough	Uni-/biseriate	Round, radiate head	+	-	-	-	-	-	-
Aspergillus fumigates	Short (<3300), smooth, colourless	Uniseriate	Round columnar head	-	-	-	-	-	-	-
Aspergillus versicolo	Long, smooth, colourles	Biseriate	Round, loosely radiate	-	+	-	-	-	-	-

r	s		head							
Peniclillum marneffei	Elongated sausage-shaped cells	Form brush-like clusters called "penicillin"	-	-	-	-	-	-	-	-
Penicilum chrysogenum	Branched	Flask-shaped on metulae, form brush-like cluster called "penicillin"	-	-	-	-	-	-	-	-
Rhizopus microsporus	-	-	-	-	-	Unbranched and brown	Not prominent	Spherical	Spherical	Simple, hyaline
Rhizopus arrhizus	-	-	-	-	-	Unbranched, longitudinally seriate	Prominent	Ellipsoidal	Globose	Simple, hyaline
Mucor racemosus	-	-	-	-	-	Branched	-	+	Spherical	-
Mucor amphibiorum	-	-	-	-	-		-	+	Spherical	-

Key: - Absent + Present

Table 6: Total Heterotrophic Viable Fungal Count During the Study

	Fresh water	Fresh water +	Fresh water +	Fresh water +	Fresh water +
Day	**Control**	**Waterbase DF**	**Oilbase DF**	**OSD/LT**	**OSD/Seacare**
	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml

0	4.45	4.38	2.85	3.86	4.32
5	2.95	2.90	2.48	3.83	3.71
10	3.70	3.00	2.48	3.48	4.68
15	3.60	3.00	2.60	3.40	4.4
20	3.65	2.90	2.60	2.48	4.18
	3.67±0.53	3.24±0.64	2.60±0.15	3.61±0.22	4.26±0.36

Table 7: Petroleum Product Utilizing Bacterial Count During the Study

Day	Fresh water Control	Fresh water + Waterbase DF	Fresh water + Oilbase DF	Fresh water + OSD/LT	Fresh water + OSD/Seacare
	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml	Log10cfu/ml
0	2.6	4.48	3.90	4.23	4.08
5	2.3	5.70	3.60	4.20	3.90
10	2.43	5.48	3.43	4.15	3.20
15	2.3	4.48	3.23	4.15	2.70
20	2.0	4.30	3.11	4.11	2.6
	2.34± 0.23	4.89 ± 0.65	3.45 ± 0.31	4.71 ± 0.05	3.30 ± 0.58

Table 8: Percentage (%) Ultimate Biodegradation of the Drilling fluids and Oil Spill Dispersants During the Study

Day	**Fresh water Control**	**Fresh water + Waterbase DF**	**Fresh water + Oilbase DF**	**Fresh water + OSD/LT**	**Fresh water + OSD/Seacare**
	%	**%**	**%**	**%**	**%**
0	0	0	0	0	0
5	15.7	15.8	17.8	4.8	2.9
10	31.6	26.3	22.1	12.0	14.4
15	3.62	44.5	42.8	16.7	32.1
20	37.4	55.5	54.8	41.1	54.1
	24.18± 16.05	**28.42 ± 22.15**	**27.50 ± 21.56**	**14.92 ± 15.99**	**20.70 ± 22.53**

Conclusion

This study reveals that the freshwater from Asarama Stream is heavily impacted with toxic substances from oil companies constantly using these petroleum products in Rivers State and Nigeria in general. Furthermore, the study

showed that the same species of bacteria from different aquatic ecosystems could respond differently to the same level of a toxicant from time to time even when all other variables are held constant.

This study showed that drilling fluids were relatively more degradable than oil spill dispersants in fresh water ecosystem.

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