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Successional changes of algae as toxicity indices in an induced semi-natural crude oil/dispersant contaminated aquatic ecosystem

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ABSTRACT

Changes in the composition and diversity of chlorophyceae and cyanophyceae algae during biodegradation of 2% forcados blend crude in a semi-natural contaminated aquatic ecosystem was studied. FOOD 3 bacteria inocula served as oil degrader with changes in nutrient levels and an oil dispersant – "ESSO". Filamentous algal forms dominated species encountered with coelosphaerium and volvox as the colonial forms found. Changes in algal counts over a two week period is reported. Growth medium made up of crude oil and dispersant was more deleterious to algae growth than that containing oil alone. Green algae were more susceptible to toxic effect of crude and dispersant than the blue green algae. Anabaena and Oscillatoria were least susceptible to toxic effect of the oil/dispersant. Spirogyra and Ulothrix are being proposed as test organisms for the dispersant toxicity test based on their susceptibility within 4 days of exposure to dispersant concentrations between 1.5% and 2.5%.

Key words: Algae, crude oil, dispersant, toxicity index, aquatic *ecosystem*.

INTRODUCTION

Oil spill is the result of accidental or intentional release of petroleum products into the environment as a result of human activity, which include drilling, manufacturing, storing, transporting and waste management. (Sharma, 2008). Nigeria like other oil producing countries has to contend with oil spills in her course of prospecting for oil (Fafioye and Owa, 2000).

Oil pollution is very damaging to aquatic ecosystem Aquatic plants responds in various different ways to oil and many species may be killed off during oil pollution (Thomas, 2011).

Bioremediation may be used to accelerate the process of biodegradation of the oil after a spill. In this process dispersant cause disintegration of oil accumulation creating smaller droplet which are more accessible and easier to degrade by Microorganism (Prince;, 1997; NRC, 2005.) Microbial degradation is much quicker, if the oil is in very small droplet. (Biello, 2010).

However, relatively little is known about the environmental effects, toxicity and the fate of dispersant applied to marine oil spills. (Place *et. al.*, 2000, Rio – Martinez *et. al.*, 2012). The objective of the present study is to determine the concentration at which dispersant could be applied in an oil pollution without having serious consequences on the fresh water microflora.

MATERIALS AND METHODS

Sample Collection:- Water samples were collected in clean 5 litre kegs, covered and brought to the laboratory form Awba Dam, University of Ibadan. Each sample lasted for 14 days after which it was discarded.

Sterilization of glass wares:- All glass wares used for the test were washed with detergent, rinsed in several changes of tap water and dried before sterilization at 180^0c for 3 hours in an oven.

Inocula used for Biodegradation studies
A bacterium culture designated 'FOOD 3' was used. The organism was supplied by the Department of Microbiology University of Ibadan. Each inoculum was culured in Nutrient broth for 96 hours using the shaker incubator.

Determination of Dispersant Concentration that Enhance Biodegradation
Different concentration of dispersant (ESSO Chemicals) chemicals were added to the troughs containing the oil degradation mixture i.e. tanks with oil polluted water, mineral salt and inocula.

Table 1: Experimental Set-up Showing the Varied Dispersant Concentration Used

Tank	Dispersant Concentration %
i.	-
ii.	0.5
iii.	1
iv.	1.5
v.	2
vi.	2.5

Crude oil – 12ml
Water volume – 60ml
Inoculum – 20ml
& N & P for cop 103 – 2% N and 3% P

RESULTS

The algae taxa present in FOOD 3 seeded tank with varied dispersant concentration and nutrient enrichment (2% Nitrogen, 3% Phosphorus is shown in Table 2. The Blue algae genera present at the beginning of the experiment include *Coelosphaerium, Oscillatoria, Nostoc, Arabaena* and *Spirullina*. The green algae include *Spirogyra Oedogonium, Chlamydomonas Ulothrix* and *Volvox*. At the end of the experiment Green algae *Spirogyra, Chlamydomonas* and *Volvox* and blue green algae *Nostoc* were absent in all dispersant concentration.

Table 3 shows the surviving population count of algae in varied dispersant concentratins while the statistical difference between Blue Green Algae and Green Algae in the tanks are shown in table 4.

FOOD 3

TABLE 2: Algae taxa present in seeded tank with varied dispersant concentration and 2%N, 3%P

Algae General	Day O						Day 4						Day 7						Day 10							Day 14				
	0%	**0.5%**	**1.0%**	**1.5%**	**0%**	**0.5%**	**1.0%**	**1.5%**	**2.0%**	**1.5%**	**2.0%**	**2.5%**	**0%**	**0.5%**	**1.0%**	**1.5%**	**2.0%**	**2.5%**	**0%**	**0%**	**0.5%**	**1.0%**	**1.5%**	**2.0%**	**2.5%**	**0%**	**0.5%**	**1.0%**	**1.5%**	**2.0%**
Blue green algae																														
Coelosphaerium	+	+	+	-	+	+	+	-	+	+	+	+	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	-	+
Oscillatoria	+	+	+	+	+	-	+	+	+	-	-	-	+	+	+	+	+	+	+	+	+	-	+	-	-	+	-	+	+	+
Nostoc	+	+	+	+	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Anabaena	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+
Spirullina	+	+	+	+	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
Green algae																														
Spirogyra	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Oedogonium	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+
Chlamydomonas	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-	-	-	-	-	-
Ulothrix	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Volvox	+	+	+	+	-	-	+	+	-	-	-	-	+	+	+	-	+	+	+	-	-	-	+	+	-	-	-	+	+	-

\+ - *Positive*

\- - *Negative*

Table 3: Surviving Population Count (X 10^5) of algae in varied dispersant concentration at 2%N, 3%P

Age of culture in days	Algae taxa	0%	0.5%	1%	1.5%	2%	2.5%	Mean count
0	BGA	48	50	40	40	48	45	45.2
	GA	86	41	80	85	86	45	37.2
4	BGA	50	44	47	50	52	50	48.8
	GA	80	34	38	30	23	20	29.3
7	BGA	45	39	44	40	45	46	48.8
	GA	25	26	26	15	30	25	24.5
10	BGA	45	30	32	30	42	45	37.3
	GA	30	15	16	15	18	20	19.0
14	BGA	40	48	45	30	54	44	43.5
	GA	12	15	14	13	12	15	13.5

BGA = Blue green algae
GA = Green algae

Table 4: Student t-test showing statistical difference between blue-green and Green algae in FOOD-3 seeded tanks at varied dispersant concentration with days of incubation

Days of incubation	Df	t-value	t-tab	Sig at 0.05
0	5	4.18	2.571	S
4	5	5.38	2.571	S
7	5	10.58	2.571	S
10	5	9.35	2.571	S
14	5	9.07	2.571	S

DISCUSSION

The blue green algae present at the beginning of the experiment included *Coelosphaerium, Oscillatoria, Nostoc, Anabaena* and *Spirullina* while the green algae were Spirogyra *Oedogonium, Ulothrix, Volvox* and *Chlamydomonas* as seen in Table 2.

On day 4, a decrease in the surviving general of algae was observed especially among the green algae. It was seen that higher concentration of the dispersant affected flamentous *spirogyra* sp and *Ulothrix* sp which were absent in 1 and 1.5% dispersant concentrations. This may be due to high level of crude oil and toxic chemical dispersant killing marine life and oxygen producing algae. Grattan et al., (2011) found increased mortality in planktonic copepods exposed to dispersant with stronger effects on small-sized species.

On day 7, *Anabaena* and *Oscillatoria* were the surviving genera of the blue green algae at all dispersant concentration. *Spirogyra* and *Ulothrix* filaments which were found at the start of the experiment in the various concentrations disappeared from day 7 and was not seen till the end of the experiment. This may serve as test organism in studies to determine dispersant toxicity.

The algae genera in each of the dispersant concentration on day 10 and 14 were similar consisting of *Coeloosphaerrium, Oscillatoria* and *Anabaena* for the blue green algae and the green algae *Oedogonium*. This correlates with the result of Bott and Rogenmuser (1978) where algae species composition changed with the loss of some filamentous algae particularly with fuel oil treatment. Table 3 shows the surviving population counts of algae in the dispersant concentrations. There was a gradual decline in the green algae count from the 1st day to the 14th day. Maglir, (2010) pointed out that oil slick as well as the chemical dispersant will kill off the phytoplankton and algae species for which oil is a toxin and will block off sunlight needed for photosynthesizing algae.

Statistical analysis of the data shown in table 4 shows that there was a significant difference in blue green and green algae count throughout the period of the experiment.

CONCLUSION

The algae studies show that treatment with dispersant showed a depression in algae diversity. The blue green algae *Oscillatoria* and *Anabaena* survived treatment with the various dispersant concentrations. The green algae *Spirogyra* and *Ulothrix* also disappeared at all dispersant concentration from day 7 till the end of the experiment. This work revealed that the effect of microbial clean up exercise on the natural algae communities of the ecosystem must be investigated before a large scale eradication exercise is embarked upon on a large water body. The green algae *Spirogyra* and *Ulothrix* will be good as test organism to determine the toxicity index of dispersant.

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