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The Challenges of a Changing World :
Perspective of Nigerian Women Scientists in

Chemical, Environmental and Pharmaceutical Research

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Assessment of the Effect of Water Soluble Fractions of Toluene and Hexane by Two Microalgae

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ABSTRACT
The assessment of the effects of water soluble hydrocarbons of crude oil by two microalgae namely *Scenedesmus quadricauda* and *Oscillatoria borneii* were carried out in the laboratory. The microalgae were grown in a batch culture in the presence of various concentrations (0%, 25%, 50%, 75% and 100%) of the water soluble fractions of Toluene and Hexane. Their growth pattern was assessed in a 14 days experiment. Algal growth was measured using HACH DR 2000 spectrophotometer at absorbance of 745nm. Growth stimulation was recorded by both microalgae in control, 25% and 50% concentrations of the water soluble fractions of both hydrocarbons while 75% and 100% concentrations of the WSFs of both hydrocarbons inhibited growth. Percentage inhibition of WSF of the aromatic hydrocarbon (Toluene) was more than that of the aliphatic (hexane). Comparatively, the Cyanobacteria was more inhibited than the chlorophyte by the WSF of both hydrocarbons.
Key words : Assessment, WSF (water soluble fractions), OD (Optical density), Microalgae.

INTRODUCTION

Crude oil is a collection of hundreds of different substances, including compounds of suphur, oxygen, nitrogen as well as the metal Nickel and vanadium, the highest proportion being hydrocarbons[10]. It is a complex mixture of hydrocarbons with 4- 26 or more carbon atoms per molecule [4]. The constituents may be straight, branched or cyclic chains including polycyclic aromatic hydrocarbons (PAHS) which are known to be potent carcinogenic[4].

The rapid increase in the demand for and utilization of petroleum has resulted in a steady increased level of petroleum exploration and hence pollution of both fresh water and marine environment arising from products leakage and spill during production, processing, storage, transportation, distribution, damage to pipeline, tankers, bunkery activities, sabotage to well head and accidental spillages into the aquatic environment. These activities coupled with increasing demand for petroleum products has made more complex the problem of oil pollution of marine and fresh water ecosystems. When oil spill occurs, there is a gradual mixing and stirring of the oil water mixture by physical factor[1]. This stirring gives rise to higher percentage of water soluble fractions of crude oil. Water soluble components of crude oil in addition to Toluene and Hexane include a variety of compounds that are toxic to a wide spectrum of marine

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plants and animals[8]. Attempt has been made by various authors to assess the rate of dissolution and solubility of different refined products and crude oil but work on effects of water soluble fractions of hydrocarbons on microalgae are rare. Related works include those of [2, 3, 4, 6, and 9].

MATERIALS AND METHODS

Test algae

The experimental microalgae used in this study were, *Scenedesmus quadricauda* and *Oscillatoria borneii*. They were isolated and maintained in chu's growth medium in the phycology laboratory in Unversity of Benin.

Isolation of pure culture of experimental microalgae

Pure cultures of experimental microalgae were obtained by series of sub culturing in Chu 10 artificial growth mediums.

Culture media

The growth medium used is an artificial medium formulated in accordance with Chu's modified medium No 10 solution of 1942 [7].

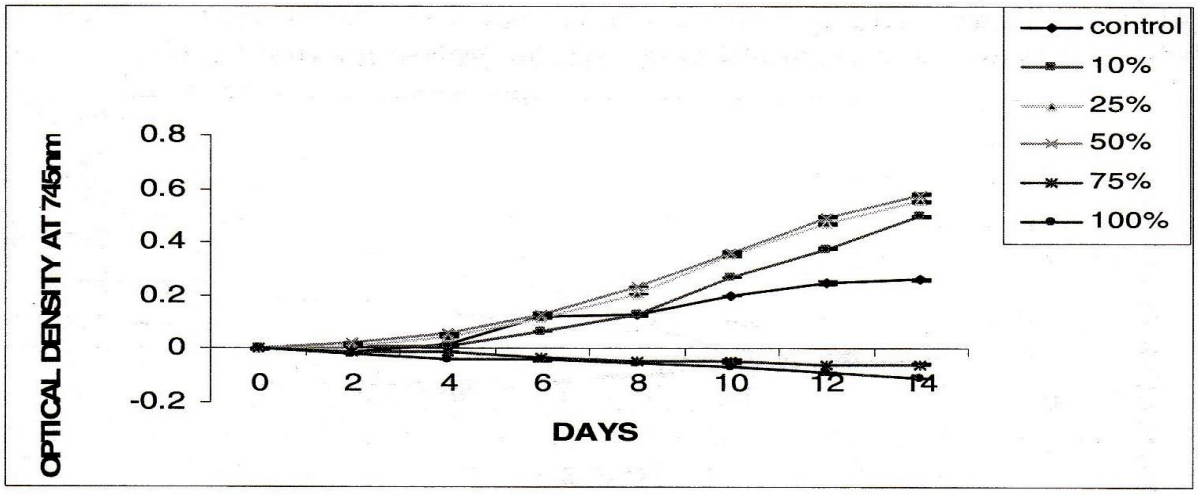

Fig.1: Growth Response of *Scenedesmus quadricaudal* in WSF of Hexane.

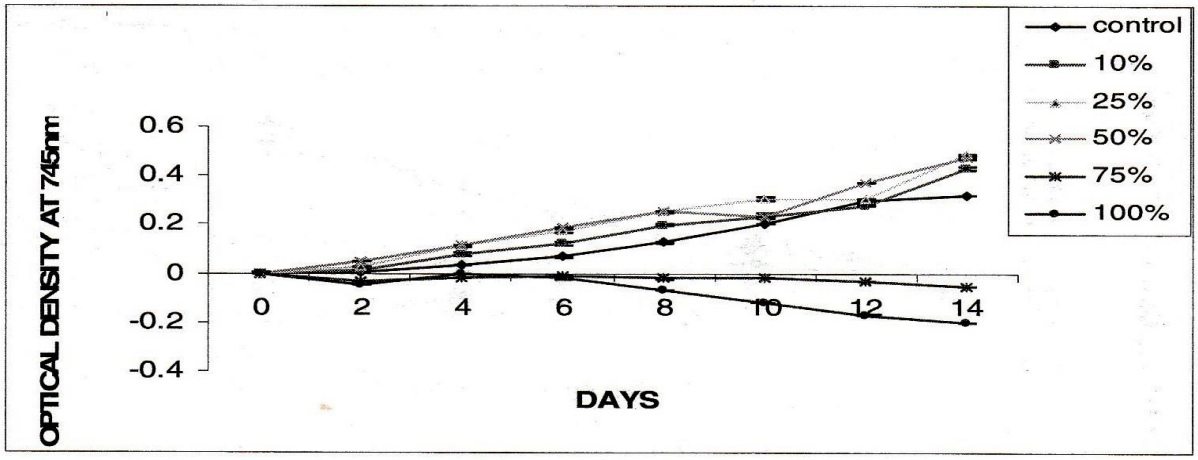

Fig 2: Growth response of *Scenedesmus quadricauda* in WSFof Toluene.

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Culture vessels

Experimental vessels were 950ml round bottom transparent bottle. They were washed thoroughly with detergent and further rinsed with 70% sulphuric acid solution to remove any trace of algal spore present. The bottles were further rinsed with distil water and were oven-dried at 35^0C for 72hours.

Inoculation

Inoculation was done using 2ml of a unialgal inoculum of each of the experimental microalgae (*Scenedesmus quadricauda* and *Oscillatoria borneii*).

Preparation of water soluble fraction

The WSF was prepared according to the method of [11]. A sample of crude oil was slowly mixed separately in an equal volume of distilled water in ratio 1:9 in a 2L screw-cap conical flask. This was placed on Gallen-kamp table top magnetic stirrer and stirred with 7/cm magnetic rod for 24hrs at room temperature (27^0C ± 2^0C.). After mixing, the oil-water mixture was allowed to stand ove night in a separating funnel. The filtrate which is the water soluble fraction was separated from the supernatant and referred to as stock or 100% WSF.

Growth measurement

Algal growth measurements were done using absorbance at 745nm from 25mls of aliquot collected every 2 days from the experimental set up using the methods prescribed by [5].

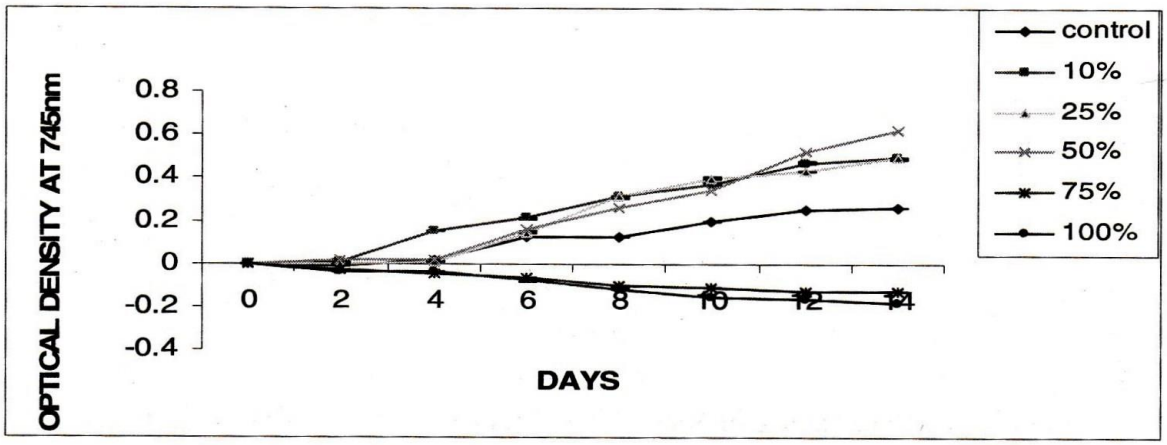

Fig 3: Growth response of *Oscillatoria borneii* in WSF of Hexane.

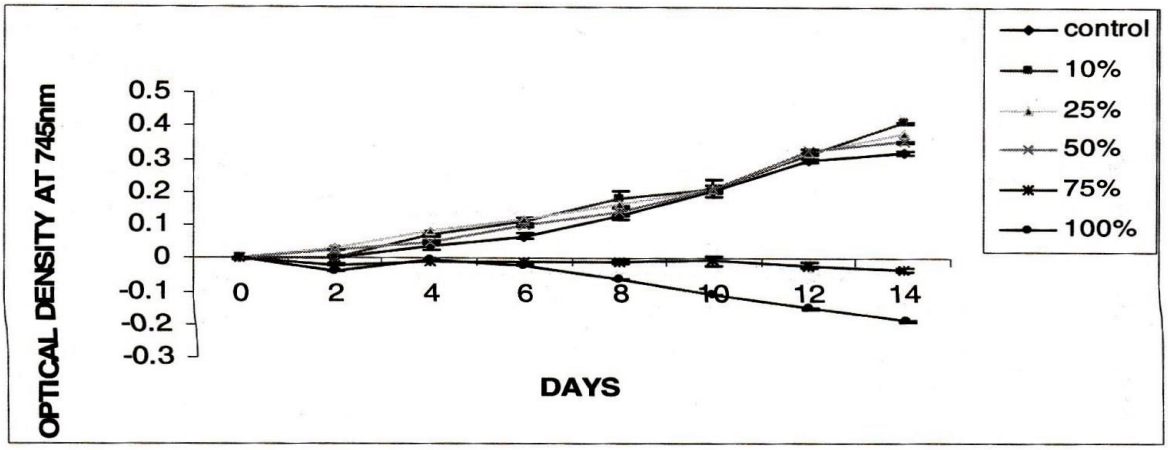

Fig 4: Growth response of *Oscillatoria borneii* in WSF of Toluene.

RESULTS AND DISCUSSION

Figures 1-4 show comparative growth response of *Scenedesmus quadricaudal* and *Oscillatoria borneii* in different concentrations of WSFs of Hexane and Toluene. The figures show a lag phase of growth in all concentrations of the treatments between the start of the experiment and day 4. A gradual increase in growth was then observed in lower and mid concentrations till day 14 of the experiment. Generally, Irrespective of the WSFs, the two microalgae showed growth stimulation at lower and mid concentrations and complete growth inhibition at higher concentrations. The lag phase of growth observed at the onset of the experiment by both microalgae could be due to any of the follow-

1. Algal cells were adjusting to the new environment.
2. Algal cells could also be experiencing a resting period during which they adapt to changing environment stress factors.
3. Again, the algal cells could have altered their needs, thus taking some time to start their normal physiological activities.

The growth stimulation observed at lower and mid concentrations could be due to the fact that algal cells are growing and dividing at a maximal rate, given their different genetic strength, the nature of the medium and the optimum conditions under which they are growing. Paramount among these conditions could be utilization of some hydrocarbons in the presence of nutrient and light as their carbon source for photosynthesis. The inhibition observed at higher concentrations could be attributed to nutrient deficiency, death of some algal cell and possibly anoxic condition created by the presence of the WSFs.

CONCLUSION

In view of the results obtained, growth and utilization of water soluble fractions of both hydrocarbons occurs only at lower and mid concentrations in the presence of nutrient ions while inhibition was recorded in higher concentrations.

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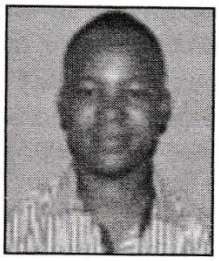

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