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HYDROCARBONS DEGRADING YEASTS FROM COCHIN BACKWATER*

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ABSTRACT

Yeasts isolated from Cochin Backwaters were screened for their ability to degrade petroleum products. Out of 35 isolates of yeasts tested, 77% showed assimilation of diesel, 74% kerosene and 23% crude oil. These yeasts belonged to the genera *Debaryomyces, Hansenula, kluyveromyces, Pichia, Saccharomyces, Candida, Geotrichum, Rhodotorula* and *Trichosporon.* The growth rate in oil medium varied for each isolate. Among these yeasts *Candida tropicalis* was the most efficient in oil degradation. The potential role of marine yeasts in mediating oil degradation in marine environment is discussed.

INTRODUCTION

SPILLAGE of oil is a pollution threat to the marine environment and is of much concern to marine ecologists. Sen Gupta and Kureishy (1981) have shown that the trade and tanker routes across the Arabian Sea are more contaminated by petroleum hydrocarbons than southern Bay of Bengal. With the advent of increased water transport, indifferent washing of tanks of large oil carriers just outside barmouth, expose backwaters and estuaries for heavy oil pollution. Cochin Backwater is no exception. Therefore, the situation warrants a study of different aspects of petroleum degradation and also the means to control the environmental damage. So far, only a few studies have been made in this line. Recent studies have shown the role of filamentous fungi, yeasts and bacteria in biodegradation of oil in marine environment (Ahearn and Crow, 1986; Sorkhoh *et al.*, 1990).

The aim of this study is to conduct a survey of the Cochin Backwater for marine yeasts capable of oil degradation on the presumption that the study will lead to the isolation of potent oil degraders.

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MATERIALS AND METHODS

A total of 35 species of yeasts isolated from water and sediment samples at different locations of Cochin Backwater (Prabhakaran, 1991) were screened for hydrocarbon assimilation.

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The screening of hydrocarbon assimilation was conducted in the liquid nutrient medium (Chatterjee *et al.*, 1978), prepared with 50% aged seawater maintaining the pH at 7.0. The medium was dispensed in 16 mm tubes (5 ml each) and sterilized. Hydrocarbons to be tested were sterilized separately and 0.5 ml was added in each tube. Medium devoid of carbon source served as control. A loopful of organisms from an actively growing slant was inoculated into each tube. The inoculated tubes were shaken occasionally and incubated at 28°C for about a week. The hydrocarbon utilization was assessed by the proliferation and growth of yeast in the oil-medium interface.

Results and Discussion

The studies on bio-degradation of organic compounds by yeasts collected from different stations in Cochin Backwater clearly show that they have a useful role in the degradation of petroleum products. The ability of selected yeasts to assimilate petroleum products is indicated in Table 1. From Table 1 it can be seen that out of 35 isolates 27 (77%) were able to assimilate diesel and 26 (74%) were able to assimilate kerosene. About 8 (23%) isolates showed clear growth and 16 (46%) isolates showed poor growth in crude oil. The growth rate varied for each isolate and was observed in water-oil interface. The heaviest and most rapid growth was obtained in 3-4 days with *Candida tropicalis* (Fig. 1).

Microscopic examinations of the yeast/hydrocarbon system revealed development of the yeast cells over the periphery of the oil droplet. This affected the surface tension of the globules (Fig. 2, 3).

TABLE 1. *Hydrocarbon assimilation of yeasts isolated from Cochin Backwater*

Organism	Hydrocarbon assimilation		
	Crude Oil	Diesel	Kerosene
Ascosporogenous yeasts			
Debaryomyces hansenii	–	–	–
D. marma	+	+	+
D. vanriji	+	+	+
Hansenula anomala	±	+	+
Kluyveromyces marxianus	–	+	+
Pichia bovis	±	+	+
P. guilliermondii	+	+	+
Saccharomyces cerevisiae	–	–	–
S. exiguus	±	+	+
S. kluyveri	–	–	–
Saccharomyces sp.	–	–	–
Asporogenous Yeasts			
Candida albicans	±	+	+
C. atmospherica	+	+	+
C halophila	±	+	+
C. intermedia	±	+	+
C. krusei	±	+	+
C. membranaefaciens	+	+	+
C. parapsilosis	±	+	+
C. pseudointermedia	±	+	+
C. sake	±	+	+
C. solani	±	+	+
C. tropicalis	+	+	+
Candida sp.	–	–	–
Cryptococcus laurentii	–	–	–
Geotrichum candidum	±	+	+
Rhodotorula aurantica	±	+	+
R. qlutinis	±	+	+
R. graminis	–	+	+
R. lactosa	–	+	+
R. minuta	±	+	+
R. rubra	+	+	+
Trichosporon aquatile	–	–	–
T. cutaneum	±	+	–
T. penicillatum	+	+	+
Sporobolomyces roseus	–	–	–

+ Full growth, - No growth and ± Poor growth

Bacteria have already been proposed as a means of bio-degradation of petroleum effluents in the marine environment. The present investigation suggests yeasts as possible bio-degraders of oil pollutants. From the results it can be seen that majority of the yeasts have potential capability of biodegradation of petroleum compounds. Most of the isolates have the capability of degrading more than one hydrocarbon tested (Table 1). Meyers and Ahearn (1972) have discussed the application of yeasts ot mediate oil decomposition in the marine ecosystem. They have further stated that yeasts are more resistant than bacteria to UV rays and to fluctuations in osmotic pressure and salinity and hence may have a potential role in the degradation of effluents in shallow areas such as estuaries where biological treatement methods are preferred. However, the hydrocarbonoclastic yeasts and moulds have not been examined intensively as probable *in situ* biodegradation agents in spite of the wealth of the experimental laboratory data (Meyers and Ahearn, 1972).

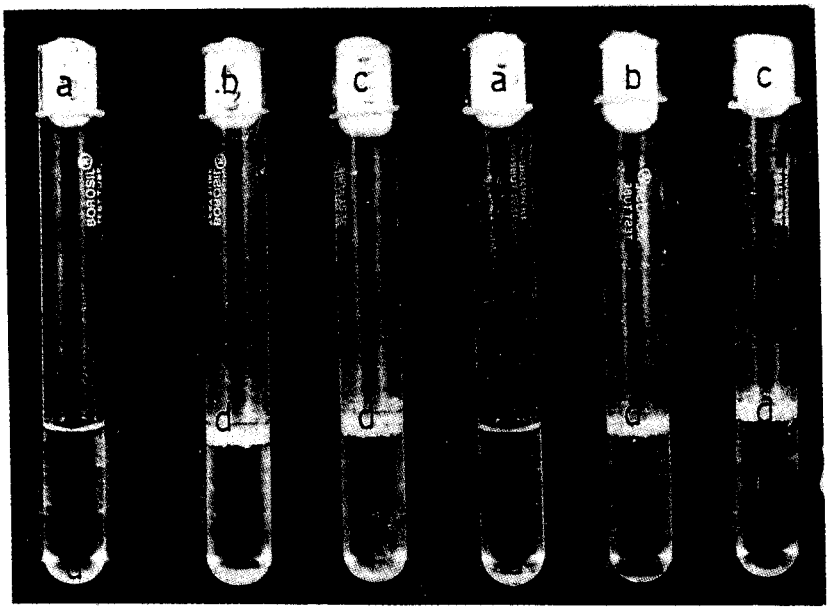

Fig. 1. **Composite plate showing hydrocarbon assimilation by *Candida tropicalis* : a. Control, b. Kerosene,** c. Dieseal and d. Organism after 4 days and 7 days respectively.

The high frequency distribution of versatile hydrocarbonoclastic yeasts such as *Candida tropicalis, C. membranaefaciens* and *Rhodotorula rubra*, etc. is recorded in high densities in the Backwaters (Prabhakaran, 1991) which signifies to the presence of petroleum effluents in the ecosystem. Ahearn and Meyers (1976) have also reported widespread occurrence of hydrocarbonoclastic yeasts in the neritic environment. Considerable studies have to be made for the selection of yeast species which can enhance the degradation of petroleum effluents in the marine and marine influenced ecosystems.

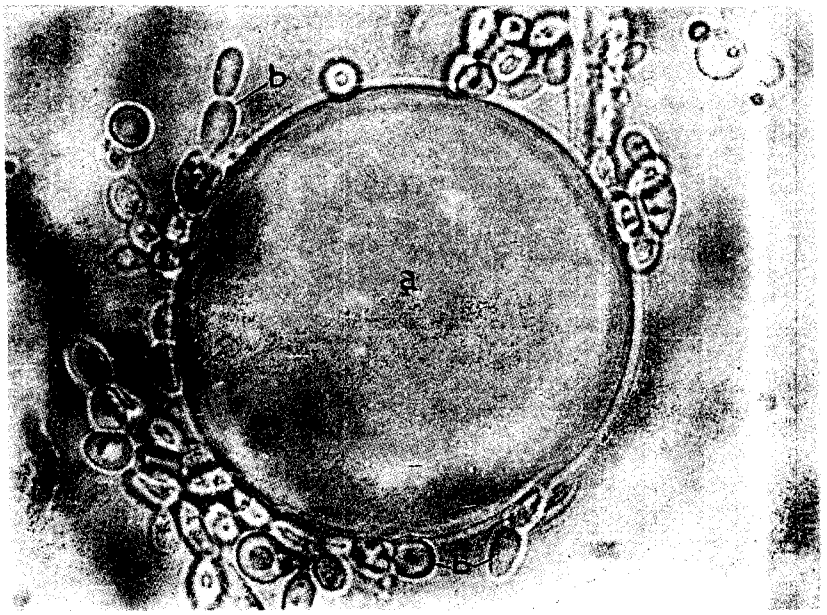

Fig. 2. Photomicrograph showing 3 days old growth of *Candida tropicalis* cells on diesel globule — under high magnification : a. oil droplet and b. yeast cells.

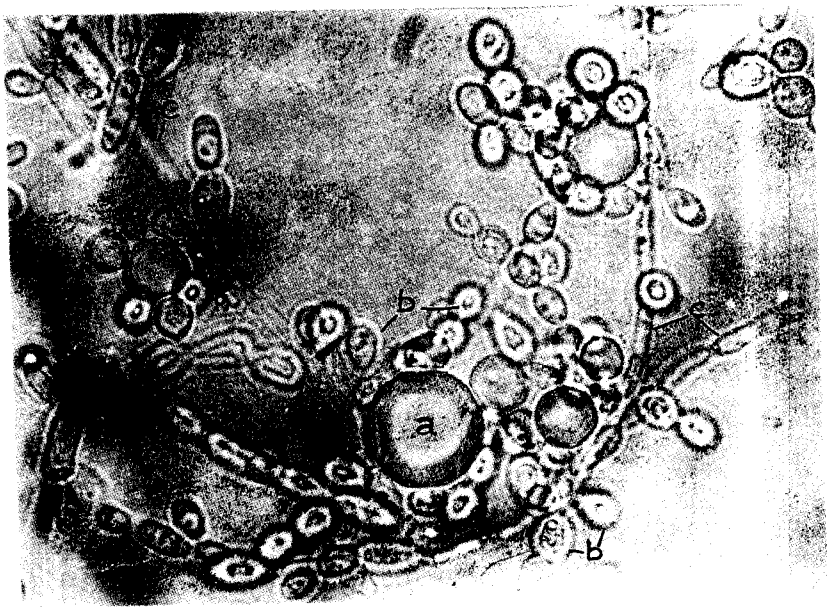

Fig. 3. Photomicrograph showing 7 days old growth of *Candida tropicalis* showing complete entrapment of diesel oil droplets : a. oil droplet, b. yeast cells and c. pseudohyphae.

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