

**BIODEGRADATION OF ALIPHATIC HYDROCARBONS
BY OIL - DEGRADING BACTERIA
ISOLATED FROM COASTAL WATERS**

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**BIODEGRADATION OF ALIPHATIC HYDROCARBONS BY
OIL-DEGRADING BACTERIA ISOLATED FROM COASTAL
WATERS**

by

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ABSTRACT

By employing a marine oil-degrading bacteria isolated from Port Dickson coastal waters; the degradation rates of several selected aliphatic hydrocarbons as well as PETRONAS Tapis A crude oil's aliphatic fraction were determined quantitively by using the Gravimetric Method. Since no identification was conducted in this study, therefore the bacteria was known as Nap C. The selected aliphatic hydrocarbons used for this study were n-octadecane ($C_{18}H_{38}$), n-tetracosane ($C_{24}H_{50}$), n-octacosane ($C_{28}H_{58}$) and n-dotriacontane ($C_{32}H_{66}$).

Degradation of aliphatic hydrocarbons started only after a two to four day lag period, for the induced and non-induced Nap C cells, and reached the maximum rate within two weeks. The optimal physico-chemical conditions for the cells' growth were found at pH 8, salinity of 30 ppt and temperature of 30^0C . In addition, the incorporation of phosphate nutrient (2.5 $\mu g\text{-at P/l}$, 25 $\mu g\text{-at P/l}$ and 50 $\mu g\text{-at P/l}$) caused only little improvement of biodegradation but an addition of ammonium nutrient (10 $\mu g\text{-at N/l}$, 50 $\mu g\text{-at N/l}$ and 100 $\mu g\text{-at N/l}$) contributed to a greater biodegradation rate in the ascending trend. However, the combination of 100 $\mu g\text{-at N/l}$ and 50 $\mu g\text{-at P/l}$ did not support a better growth of the cells. Owing to this, the nutrients concentration for this study was set at 100 $\mu g\text{-at N/l}$ with 2.5 $\mu g\text{-at P/l}$.

Under those optimum parameters, the biodegradation rates of C18, C24, C28 and C32, each added at an amount of 1ml (20 mg/ml) in 250 ml of synthetic marine medium;

were 0.2330 $\mu\text{g/l/day/cell}$, 0.1826 $\mu\text{g/l/day/cell}$, 0.1385 $\mu\text{g/l/day/cell}$ and 0.1133 $\mu\text{g/l/day/cell}$ respectively for the induced cells while for the non-induced cells the values were 0.04242 $\mu\text{g/l/day/cell}$, 0.02015 $\mu\text{g/l/day/cell}$, 0.01802 $\mu\text{g/l/day/cell}$ and 0.00848 $\mu\text{g/l/day/cell}$ which is nine fold lower than that of induced cells.

Liquid chromatography analyses demonstrated that the PETRONAS Tapis A crude oil composed of 48.48 % of aliphatic fraction. The experiment conducted on this compound showed that the biodegradation rates by both induced and non-induced cells were 0.05198 $\mu\text{g/l/day/cell}$ and 0.01009 $\mu\text{g/l/day/cell}$ respectively. This results revealed that the hydrocarbons induced Nap C was more capable to dergade the aliphatic hydrocarbons 5 times faster than the non-induced cells.

ABSTRAK

Dengan menggunakan sejenis bacteria pengurai hidrokarbon yang diperolehi dari perairan persisiran laut port Dickson, kadar degrasi beberapa hidrokarbon alifatik dan juga minyak mentah PETRONAS telah ditentukan secara kuantitatif melalui Analisis gravimetrik. Oleh kerana jenis bakteria yang diguna untuk mendegradasikan hidrokarbon alifatik tersebut tidak dikenalpasti, maka ia hanya dikenali sebagai Nap C bagi kajian ini. Justeru, jenis komponen alifatik yang dipilih dalam kajian ini meliputi n-octadecane ($C_{18}H_{38}$), n-tetracosane ($C_{24}H_{50}$), n-octacosane ($C_{28}H_{58}$) dan n-dotriacontane ($C_{32}H_{66}$).

Keputusan kajian ini menunjukkan bahawa proses degradasi hanya bermula selepas suatu fasa lamban selama 2-4 hari dan mencapai kadar maksimumnya dalam jangkamasa 2 minggu. Dalam konteks ini, keadaan fiziko-kimia yang optimum bagi pertumbuhan sel Nap C adalah pada tahap pH 8, dengan saliniti 30 ppt dan pada suhu 30°C . Sehubungan itu, percampuran nutrien fosfat (pada tahap 2.5 $\mu\text{g-at P/l}$, 25 $\mu\text{g-at P/l}$ dan 50 $\mu\text{g-at P/l}$) hanya menyebabkan peningkatan yang rendah bagi proses degrasi tetapi pertambahan nutrien ammonium pula (pada tahap 10 $\mu\text{g-at N/l}$, 50 $\mu\text{g-at N/l}$ dan 100 $\mu\text{g-at N/l}$) menyumbangkan kadar yang lebih tinggi terhadap proses degradasi ini selaras dengan pertambahan kepekatan nutrien tersebut. Namun, satu kombinasi 100 $\mu\text{g-at N/l}$ dan 50 $\mu\text{g-at P/l}$ tidak dapat menyokong pertumbuhan sel Nap C yang lebih baik.

Dalam keadaan optimum ini, kadar biodegradasi bagi C18, C24, C28 dan C32, di mana ia dimasukkan sebanyak ke dalam setiap kelalang kultur pada kepekatan 20mg/ml

ke dalam 250 ml media sintetik marin; adalah sebanyak 0.2330 $\mu\text{g/l/hari/sel}$, 0.1826 $\mu\text{g/l/hari/sel}$, 0.1385 $\mu\text{g/l/hari/sel}$ dan 0.1133 $\mu\text{g/l/hari/sel}$ masing-masing bagi sel yang terdedah terlebih dahulu kepada alifatik hidrokarbon. Manakala bagi sel yang tidak terdedah kepada alifatik hidrokarbon, kadar degradasinya pula adalah 0.04242 $\mu\text{g/l/hari/sel}$, 0.02015 $\mu\text{g/l/hari/sel}$, 0.01802 $\mu\text{g/l/hari/sel}$ dan 0.00848 $\mu\text{g/l/hari/sel}$, di mana nilai ini adalah lebih rendah sebanyak 9 kali (secara purata) ganda jika dibandingkan dengan kadar tersebut di atas.

Analisis chromatografi cecair pula menentukan bahawa terdapat 48.48 % kompaun alifatik di dalam jumlah komposisi minyak mentah PETRONAS. Sehubungan itu, eksperimen degradasi kompaun ini menjelaskan bahawa sel tersebut dapat mendegradasikan hidrokarbon ini dengan kadar 0.05198 $\mu\text{g/l/hari/sel}$ dan 0.01009 $\mu\text{g/l/hari/sel}$ masing-masing bagi sel yang terdedah serta tidak terdedah kepada komponen alifatik. Keputusan ini sekali lagi membuktikan bahawa sel yang terdedah sebelumnya mempunyai keupayaan yang lebih tinggi pada kadar 5 kali lebih tinggi untuk mendegradasikan komponen alifatik sekiranya dibandingkan dengan sel yang tidak terdedah sebelumnya.