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HYDROCARBONOCLASTIC BACTERIA FROM JAKARTA BAY AND SERIBU ISLANDS

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ABSTRACT

Jakarta Bay has been known as one of the most polluted marine environment in Indonesia, with no exception by oil. Seribu Islands waters, located in the north of Jakarta Bay may have been impacted by this polluted condition. It's sometimes also hit by oil spillage from pipe leakage. The purpose of this study is to isolate and identify hydrocarbonoclastic bacteria (oil and Polyaromatic Hydrocarbon degrading bacteria) from Jakarta Bay and Seribu Island waters. The bacteria were isolated from water and sediment/sand. Isolation was prepared by enriched samples in SWP medium with Arabian Light Crude Oil (ALCO). Screening for PAH degrading bacteria has been completed by using sublimation plate method in ONR7a medium and screening for oil degrading bacteria were conducted by using oil plated method with the same medium. Bacteria identifications were done based on 16sRNA gene. The results were analyzed using BLAST and showed that 131 potential hydrocarbonoclastic bacteria have been isolated from Jakarta Bay and Seribu Island waters. Most of them were oil degrading bacteria (41.98%) and the rest were PAH degrading bacteria. Oil pollution level may impact the number of strain of hydrocarbonoclastic bacteria isolated. Among the hydrocarbonoclastic bacteria isolated from Jakarta Bay and Seribu Islands, *Alcanivorax, Marinobacter, Achromobacter* and *Bacillus* were common hydrocarbonoclastic genera in Jakarta Bay and its surrounding waters. *Alcanivorax* spp. is important oil and PAH-degrader found not only in temperate waters, but in tropical waters as well.

Keywords: Hydrocarbonoclastic bacteria, Oil pollution, PAH, Jakarta Bay, Seribu Island.

INTRODUCTION

Jakarta Bay is located north of Jakarta. It is a rather shallow bay with the average depth of 15 m, covering an area of about 514 km². Seribu Islands is a group of islands located in front of Jakarta Bay stretching along 80 km in a north-westerly direction. The environmental conditions of Jakarta Bay and Seribu Islands have been declining due to the influence of increasing anthropogenic activities.

Oil concentration in Jakarta Bay has exceeded Indonesian Marine Water Quality Standard for marine life (1 ppm) (Environment Minister Decree, No.51, 2004). Razak (2005) mentioned that oil concentrations ranged between 14.23 and 78.00 ppm, with the highest concentration found in the western part of Jakarta Bay. Jakarta Bay receives water from 13 rivers which carry waste disposal from developing urban area, such as Jakarta, Bogor, Depok, Tangerang and Bekasi. Moreover, there are International Harbour (Tanjung Priok) and fishery ports located here, which cause high oil concentrations in this waters.

Seribu Islands has been used for marine transportation and oil/gas exploration. Oil spills regularly hit this area almost twice a year. Even, there were 7 times oil spill accidents occurred in period of December 2003–November 2004 which contaminated about 78 islands (Suara

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Pembaharuan Daily, 2004). Therefore, an action should be done to minimize the impact of oil pollution in Jakarta Bay and surrounding waters.

Oil is a complex mixture of thousands of compounds which are classified into four groups, namely: saturates, aromatics, resins, and asphaltenes (Leahy and Colwell, 1990). Polycyclic aromatic hydrocarbons (PAHs) are the pollutants of major concern, because of their carcinogenic and mutagenic properties (White, 1986). Physical and mechanical methods are fairly well established to deal with oil pollution both at sea and on shorelines (Swannell *et al.,* 1996). Recently, the bioremediation (biological methods) have received the most attention due to the environmentally friendly and cost effective. However, this study is still very rarely conducted in tropical waters. Bioremediation is the act of adding materials to contaminate environments, such as oil spill sites, to cause an acceleration of the natural biodegradation process. (U.S. Congress, 1991).

In terms of oil pollution in Jakarta Bay or other Indonesian marine waters, bioremediation method seems to be promising. To apply this method in Indonesian waters, the first step should be done is to understand the ecological processes of oil biodegradation in Indonesian seawater. Therefore, the hydrocarbonoclastic bacteria, biodegrading bacteria particularly oil and PAHs as the indigenous bacteria of Jakarta Bay and Seribu Islands should be first evaluated. This study was a preliminary study aimed to isolate and identify the hydrocarbonoclastic bacteria from Jakarta Bay and Seribu Islands.

MATERIALS AND METHODS

Study Site and Abiotic Parameters

The study site was established from Jakarta Bay to Seribu Islands represented by 5 sampling points (Fig. 1).

In terms of oil pollution these 5 sampling points could be divided into 2 categories, i.e. oil polluted areas and control areas (Table 1). Sea water was collected using bucket from the surface water (0–30 cm) and sediment samples were taken using Van Veen grab (the thickness of sediment was 5 cm from the surface). Salinity, temperature and dissolved oxygen (DO) were measured by refractometer, thermometer and DO meter respectively.

Enrichment and Isolation of Hidrocarbonoclastic Degrading Microorganisms

The enrichment of seawater samples were conducted by inoculating each of 190 ml of seawater sample in an Erlenmeyer flask and supplemented with 10 ml of 20 x stock SWP medium (contains NH_4NO_3, K_2HPO_4, ferric

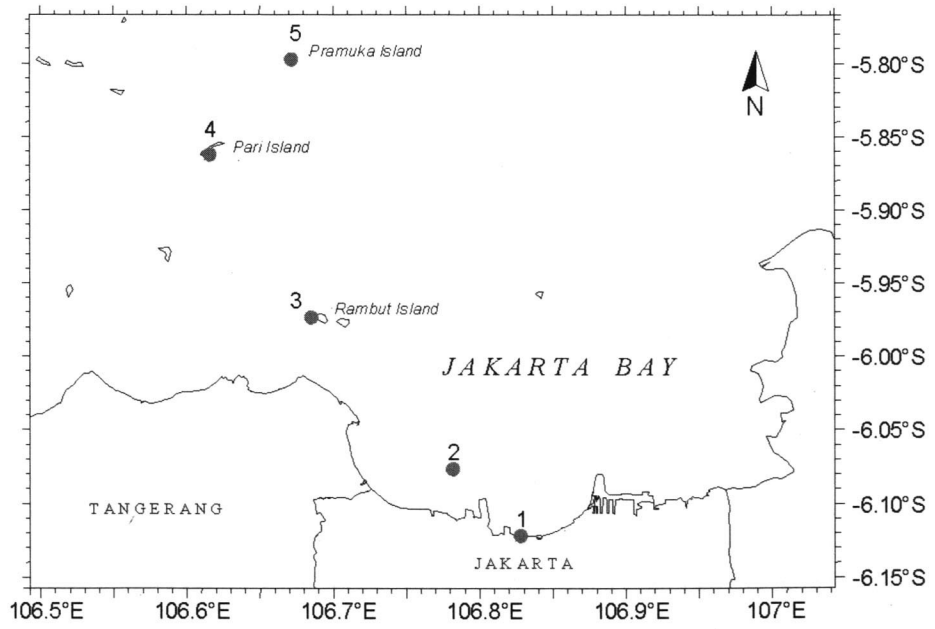

Figure1. Sampling points for isolating oil degrading bacteria in Jakarta Bay and Seribu Islands

Table1. Five sampling points of the study site based on the two categories

No.	Oil polluted areas	Utilization
1.	Marina Port	Personal yacht harbour
2.	Muara Baru offshore	Transfering place of oil from tanker to the oil bottom sea line for Jakarta Power Plant
3.	Pramuka Island	Experienced hit by heavy oil spill
	Control areas	
4.	Rambut Island	Bird sanctuary (non inhabited island)
5.	Pari Islands	Field station of Marine Science Research Center and seaweed farming

citrate and yeast extract) and 1ml of ALCO. The enrichment of sediment samples were carried out in the same way as seawater samples. The cultures were incubated at 30°C in orbital shaker (100 rpm). During the incubation period, the loss of water of the enriched culture was supplied by distilled water and maintained until crude-oil disappeared.

The procedure of hydrocarbonoclastic bacteria isolation are as follows, one ml of supernatant was taken out aseptically from culture and diluted with filtered-autoclave sterilized seawater after different periods of times (3, 14, 28, 42, and 56 days). A series of 10-fold dilutions of supernatants spread out on ONR7a-1.5% agar medium (Dyksterhouse *et al.*, 1995).

To isolate oil degrading bacteria, the agar medium was supplemented with ALCO. PAH were used to supplement the isolation of PAHs degrading bacteria. ALCO and PAH compounds are the only sources of carbon and energy. ALCO was added by layering the surface of the agar and PAH compound were added by sublimation method (Alley and Brown, 2000). The temperature and time needed for sublimating PAHs are listed in Table 2.

After sublimation, the plate was incubated in room temperature for 1 to 2 weeks. The colony was surrounded by a clear zone and the interested colony (change media color) was further purified by streaking on the same type of plates for confirmation test. The purified isolates was collected by re-streaking on rich medium (Marine agar medium) for checking out.

16S rRNA Gene Sequence Determination

Total genomic DNA for 16S ribosomal DNA (rDNA) amplification was isolated directly from the colonies grown on marine agar (DIFCO) by using InstaGene (Bio-Rad). PCR amplification of the 16S rRNA genes was obtained using the forward primer 16F27 (AGAGTTTGATCMTGGCTCAG) and reverse primer 16R1492 (TACGGYTACCTTGTTACGACTT). PCR was performed with an initial denaturation step consisting of 95°C for 4 min, 30 cycles of 1 min at 95°C, 1 min at 55°C, 2 min at 72°C and 72°C for 7 min (final extension step). Primers and free nucleotides were removed from PCR mixtures by using a QIAquick PCR purification kit (QIAGEN). Direct sequence determination of the PCR-ampliRed DNA was carried out using an automated DNA sequencer and Taq cycle-sequencing reactions according to the protocols of the manufacturer (Perkin-Elmer Applied Biosystems).

RESULTS AND DISCUSSION

Abiotic Parameters

The abiotic parameters at each sampling points are listed in Table 3.

Table 2. Temperature and time used for sublimating PAHs

Poly Aromatic Hydrocarbon	Sublimation Temperature (°C)	Time (minutes)
Dibenzothiophene	100	5
Phenanthrene	100	5
Fluorene	100	5
Fluoranthene	95	10
Naphtalene	75	1

The water depth, salinity, temperature and dissolved oxygen varied 1.2–18 m, 6–32‰, 29.2–29.9°C and not detected – 7.9 mg/l respectively (Table 3). The results showed that Marina Port and the offshore area of Muara Baru waters were estuarine ecosystem whereas Rambut, Pari and Pramuka Islands waters were marine environment.

Although both Marina Port and Muara Baru were estuarine ecosystem, the DO values were different at both sites. It might be due to the pollution levels were different. The environmental condition of Marina Port was anaerobic indicated by the black sediment which was reflected in the water with strong sulphuric gas odor. The DO values of the other sampling sites were still good, ranged between 7.7 and 7.9. Four years ago Pramuka Island waters was hit by heavy oil pollution but seemed to be recovering in the present study, although some tar balls were stranded on dead corals.

Diversity of Hydrocarbonoclastic Bacteria

The result showed that the fastest oil disappearance in enriched culture was found in sediment and seawater samples collected from Marina Port. It might indicate that the fastest oil degradation occurred in Marina Port. Its occurrence was in anaerobic condition due to the lowest dissolved oxygen and low salinity was observed here (Table 2). Brakstad and Lodeng (2003) reported that hydrocarbon degradation is usually oxygen dependent and catalyzed by di- or mono-oxygenases. Biodegradation rates of alkanes in seawater at different distances from the pollution sources did not differ significantly. It can occur when nitrate, ferrous iron or sulfate used as an electron acceptor, or under conditions of methanogenesis. Several classes of petroleum hydrocarbons, including alkanes (Widdel and Rabus, 2001) and mono- and PAH (So *et al.*, 2003) degrade in this condition. However, Harayama *et al.* (2004) mentioned that anaerobic biodegradation processes were slower than that the aerobic ones. They might be a significant component of natural attenuation owing to the abundance of anaerobic electron acceptors relative to dissolved oxygen. Unfortunately, no confirmation at which site the highest degradation rate in this study occurred due to the measurements of oil concentrations and degradations rates were not conducted.

By using conventional method namely culture-based technique, 131 different hydrocarbonoclastic bacterial strains had been obtained from enriched culture of Jakarta Bay and Seribu Islands. Jakarta Bay and Seribu Islands contributed 52% and 48% respectively of all isolates collected (Fig. 2). It seems different from bacterial community analysis using culture-independent molecular technique. Okazaki (person.com) mentioned that the number of clones from Pari seawater (171 clones) was higher than those from Jakarta Bay seawater (139 clones). Harayama *et al.* (2004) suggested that bacteria in seawater cannot be readily cultured and bacteria isolated by culture-dependent method represent a minor population than that in natural bacterial communities. There are some reasons for low success of culturing marine bacteria, such as: some microbes can not grow in pure culture due to chemical component for their growth provided by other microbes, lack of communication between cell, lack of growth factor, disruption of oxidative stress, unbalanced growth caused by in excess or too low of substrate and lysogenic phages induction when famished.

Zhu *et al.* (2001) reviewed that some representative bacteria have the capabilities to degrade petroleum hydrocarbons i.e. *Achromobacter, Acinetobacter, Alcaligenes, Arthrobacter, Bacillus, Brevibacterium, Corynebacterium, Flavobacterium, Nocardia, Pseudomonas* and *Vibrio*. Darmayati (2003) isolated some petroleum degrading bacteria, i.e. *Pseudomonas*

Table 3. The abiotic parameters at each sampling points

No.	Sampling points	Depth (m)	Temperature (°C)	Salinity (‰)	DO mg/L
1.	Marina Port	2	29.2	6	nd
2.	Muara Baru offshore	12	29.9	16	7.3
3.	Rambut Island	18	29.7	30	7.7
4.	Pari Island	1.2	29.8	32	7.9
5.	Pramuka Island	2	29.9	32	7.9

nd: not detected

cepacia and *P. gladioli* from oil polluted marine sediment of East Kalimantan. In addition, *Achromobacter putrefaciens, Acinetobacter haemolyticus* and *Vibrio alginolyticus* were also obtained from marine sediment of Java sea (Darmayati, unpublished data). Feliatra (1998) isolated *Acinetobacter, Arhtrobacter, Micrococcus* and *Bacillus* from Dumai waters and Malaka straits. The genus of *Alcanivorax, Marinobacter, Bacillus* and *Achromobacter* found at the study site were common genera.

The common genus found at almost all sampling points was *Alcanivorax* consisted of *A. dieselolei*, *Alcanivorax* sp. TE-9, *Alcanivorax* sp. EPR 6, and *Alcanivorax* sp. B-1084 (Table 6). The members of *Alcanivorax/Fundibacter*

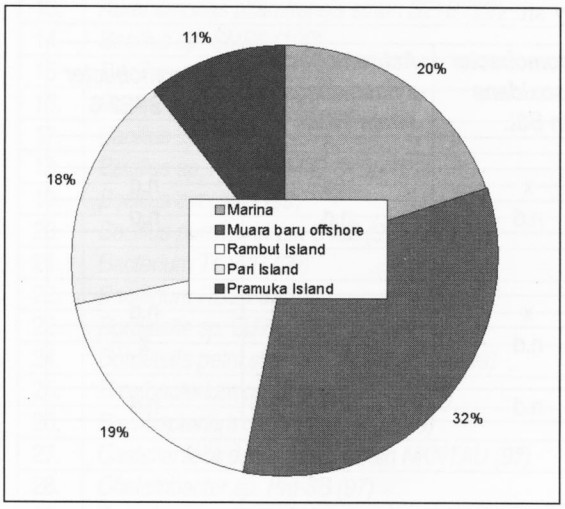

Figure 2. The Percentage of hydrocabono clastic bacteria strains in Jakarta Bay and Seribu Islands

group have cosmopolitan distribution. Their occurrences have been documented in several areas such as: Indonesia waters (present study), United Kingdom (Roling *et al.*, 2002), Italy (Yakimov *et al.*, 2005), Japan (Kasai *et al.*, 2002) and Germany (Bruns and Berthe-Corti, 1999). More than 250 *Alcanivorax*-affiliated bacteria have been isolated or detected as 16s rRNA gene sequences in all types of marine environment, retrieved from microbial communities inhabiting cold polar areas and the organism itself has been isolated so far from temperate lower latitudes (Yakimov *et al.*, 2007). Harayama *et al.*, (2004) mentioned that these bacteria used limited carbon sources with a preference for petroleum hydrocarbon and thus 'professional hydrocarbonoclastic' bacteria. *Alcanivorax* strains grow on n-alkanes and branched alkanes, could not use any sugar or amino acids as carbon sources. This study emphasized that the *Alcanivorax* strains was also found in tropical marine waters. It might indicate that this genus played a significant role in global marine hydrocarbon degradation.

The genus of *Marinobacter* could be found in oil polluted and unpolluted waters. Two species of *Marinobacter*, i.e. *Marinobacter vinifirmus strain FB1* and *Marinobacter sp. strain HS225* were isolated from the offshore area of Muara Baru and Pramuka Island. The species of *Marinobacter aquaeolei isolate OC-9* was found in Pari and Rambut Islands.

The present study found that the genus of *Bacillus* mostly detected in clean water, i.e. Rambut and Pari Island (Table 4). *Bacillus circulans, B. pumilus, Bacillus* sp. FPI/2002 and *Bacillus* sp. SMB8 were isolated from Rambut Island and *B. pumilus, Bacillus* sp. CO64, *Bacillus* sp. JZDN 42, *Bacillus* sp. PD-E-(s)-I-D-8(3) and *Bacillus* sp. SMB8 were found in Pari Island. *Bacillus* sp. SMB8 belonge to the genus of *Bacillus* was the only species that could be isolated from the offshore area of Muara Baru (polluted area). Doddamani and Ninnekar (2000) mention that *Bacillus* species are known to be versatile in degrading a wide variety of aromatic compounds in the natural environment. They isolated the *Bacillus* sp. that was able to degrade phenanthrene and utilize naphthalene, anthracene, and biphenyl.

In contrast, the genus of *Achromobacter* were mostly isolated from polluted marine waters (Marina Port). It also occurred in soil polluted by engine oil in Nigeria (Adelowo *et al.*, 2006). There were 4 different strains of *Achromobacter xylosoxidans* isolated from this area. From the offshore area of Muara Baru only one strain of *Achromobacter* could be isolated i.e. *Achromobacter* sp. SPE2-6. The species of *Achromobacter xylosoxidans* strain B8L was isolated from Marina Port and Rambut Island (Table 5).

The number of strains at each sampling site varied and the oil pollutant might affect the cultured strain variability. The Result of partial 16SRNA sequencing showed that 131 isolates consisted of 65 identified different strains (Table 6). The highest portion of strains (32%) was obtained from the offshore area of Muara Baru (Jakarta Bay) and low portions were found in Seribu Island (Pramuka,

Table 4. The genus of *Bacillus* detected at the study sites

Location	*Bacillus circulans*	*Bacillus pumilus*	*Bacillus sp.* FPI/2002	*Bacillus sp.* SMB8	*Bacillus sp* CO64	*Bacillus sp* JZDN 42	*Bacillus sp* PD-E-(s)-1-D-8(3)
Clean Waters							
Rambut Island	x	x	x	x	n.d	n.d	n.d
Pari Island	n.d	x	n.d	x	x	x	x
Oil Polluted waters							
Marina Port	n.d	n.d	n.d	n.d	n.d	n.d	n.d
The offshore area of Muara Baru	n.d	n.d	n.d	x	n.d	n.d	n.d
Pramuka Island	n.d	n.d	n.d	n.d	n.d	n.d	n.d

nd: not detected; x: presence

Table 5. The genus of *Achromobacter* detected at the study sites

Location	*Achromobacter xylosoxidans*	*Achromobacter xylosoxidans strain AU1011*	*Achromobacter xylosoxidans strain B8L*	*Achromobacter xylosoxidans strain TKW4*	*Achromobacter sp.* SPE2-6
Clean Waters					
Rambut Island	n.d	n.d	x	n.d	n.d
Pari Island	n.d	n.d	n.d	n.d	n.d
Oil Polluted waters					
Marina Port	x	x	x	x	n.d
The offshore area Muara Baru	n.d	n.d	n.d	n.d	x
Pramuka Island	n.d	n.d	n.d	n.d	n.d

nd: not detected; x: presence

Rambut and Pari Island: 11–19%). Prince and Atlas (2005) reviewed that oil degrading microorganism may be ubiquitous but only a small fraction of the biota of an uncontaminated sites, probably because they are substrate limited. An oil spill removes this limitation, and there is generally a bloom of hydrocarbon organisms so that they become a major fraction of the microbial population.

The number of oil degrading bacteria strains in Marina Port, the offshore area of Muara Baru and Pramuka waters considered as oil polluted area varied. It might be caused by the oil pollution levels of these sites were different. Marina Port and the offshore area of Muara Baru have received oil continuously from run off. It is considered that Pramuka Island waters might be in recovering condition due to no high oil spill has occurred since 2005. According to visual observation, oil concentration in Marina Port might be in the high level because this port is a crowded yacht harbor. Besides refueling has occurred in this area. This condition is not favorable for microorganism, except for anaerobic microorganism.

There were 11 isolates which indicatively represent new strains in the present study, but further intensive study is needed to obtain more data because in this study only partial sequencing was done. Their 16S rRNA gene sequences showed 92–97 percentage homology (Table 7).

Pollutant in Concern

Based on the number of strains, the composition of hydrocarbonoclastic bacteria at 5 different sampling points were different (Fig.3). According to Colwell and Walker (1977), microbial community structure depends on oil composition, climate condition and seasonal variation. Therefore, although we understand that only small portion of bacteria can be cultured but

Table 6. Diversity of the indigenous hydrocarbonoclastic bacteria in Jakarta Bay and Seribu Islands waters

No.	Strains (% homology)	Marina Port	Muara Baru offshore	Rambut Island	Pari Island	Pramuka Island
1.	*Alcanivorax dieselolei strai B-5 clone 1 (98)*	+	-	-	-	-
2.	*Alcanivorax sp. TE-9 (99-100)*	+	+	-	-	+
3.	*Alcanivorax sp. B-1084 (100)*	-	+	-	-	-
4.	*Alcanivorax sp. EPR 6 (99-100)*	-	+	+	+	-
5.	*Alcaligenes sp. mp-2 (99)*	-	-	-	-	+
6.	*Agricultural soil bacterium isolate S1-15 (96)*	-	+	-	-	-
7.	*Fulvimarina sp. K416 (99)*	-	-	-	+	-
8.	*Achromobacter sp. SPE2-6 (99)*	-	+	-	-	-
9.	*Achromobacter xylosoxidans strain AU1011 (99)*	+	-	-	-	-
10.	*A. xylosoxidans strain B8L(98-99)*	+	-	+	-	-
11.	*A. xylosoxidans strain TKW4 (99)*	+	-	-	-	-
12.	*A. xylosoxidans (100)*	+	-	-	-	-
13.	*Aurantimonas altamirensis strain S21B (99)*	-	-	-	+	-
14.	*Bacillus sp. SMB8 (100)*	-	+	+	+	-
15.	*Bacillus sp. FPI/2002 (99)*	-	-	+	+	-
16.	*Bacillus sp. CO64 (100)*	-	-	-	+	-
17.	*Bacillus sp. JZDN42 (97)*	-	-	-	+	-
18.	*Bacillus sp. Pd-E-(S)-I-D-8 (3) (100)*	-	-	-	+	-
19.	*Bacillus circulans (99)*	-	-	+	-	-
20.	*Bacillus pumilus strain B402 (99)*	-	-	+	+	-
21.	*Bacterium Te 27R (99)*	-	-	+	-	-
22.	*Bacterium RBS4-92(98)*	-	-	-	+	-
23.	*Bordetella sp. QJ7-8 (99)*	+	-	-	-	-
24.	*Bordetella petrii strain GDH030510 (95-99)*	+	-	-	-	-
25.	*Brevibacterium casei strain 3S (a) (99)*	-	+	-	-	-
26.	*Brevibacterium casei strain 3Tg (99)*	-	+	+	-	-
27.	*Castellaniella denitrificans strain NKNTAU (97)*	+	-	-	-	-
28.	*Chelatobacter sp. Pht-3B (97)*	-	-	+	-	-
29.	*Cytophaga sp.TUT1213 (100)*	+	+	+	-	-
30.	*Gamma proteobacterium PIGHI. 1-A2 (92)*	-	+	-	-	-
31.	*Halomonas sp.HS 207 (98)*	-	-	-	-	+
32.	*Halomonas sp. SBJ 85 (98)*	-	+	+	-	-
33.	*Kartchnercaverns bacterium HI-11 (98)*	-	+	-	-	-
34.	*Kordiimonas givangyangensisi strain GW14-5 (99)*	-	-	-	-	+
35.	*Leucobacter aridicollis (99)*	-	-	-	+	-
36.	*Marine bacterium MBIC 1357 (100)*	-	+	-	-	-
37.	*Marinibacillus campisalis (98)*	+	-	-	-	-
38.	*Marinobacter sp. HS 225 strain HS 225 (99)*	-	-	-	-	+
39.	*Marinobacter vinifirmus strain FBI (99)*	-	+	-	-	-
40.	*Marinobacter aquaeolei isolate OC-9 (99)*	-	-	+	+	-
41.	*Mesorhizobium sp. LZXC 36 (99)*	-	-	+	+	-
42.	*Mesorhizobium sp. TUT 1018 (99)*	-	-	-	-	+
43.	*Mesorhizobium sp. W6-20 (99)*	-	+	-	-	-
44.	*Oceanobacillus iheyensis HTE 831 (100)*	-	+	-	-	-
45.	*Ochrobactrum anthrop (99)i*	+	-	-	-	-
46.	*Phenanthrene-degrading bacterium 19. M20 (98)*	-	+	-	-	-
47.	*Phenanthrene-degrading bacterium M20 (98)*	-	+	-	-	-
48.	*Phyllobactericeae bacterium NL-21 (99)*	-	-	-	+	-
49.	*Porphyrobacter donghaensis strain SW-158 (97)*	+	-	-	-	-
50.	*Porphyrobacter donghaensis strain SW-158 (100)*	-	+	-	-	-
51.	*Porphyrobacter tepidarius strain OKSAPO (97)*	-	+	-	-	-

Table 6. (Continued)

No.	Strains (% homology)	Marina Port	Muara Baru offshore	Rambut Island	Pari Island	Pramuka Island
52.	*Pseudomonas aeruginosa PAOI (96-100)*	+	-	-	-	-
53.	*Pseudomonas aeruginosa strain XJU-2 (100)*	+	-	-	-	-
54.	*Pseudomonas stutzeri ATTC 17594 (100)*	-	+	+	-	-
55.	*Pseudaminobacter sp. W 11-4 (99)*	-	-	-	+	-
56.	*Pseudidiomarina taiwnensis strain PITI (97)*	-	-	-	-	+
57.	*Rhodocista sp. JZHS 37 (100)*	-	-	+	-	-
58.	*Rhodospirillaceae bacterium No. 71 (99)*	-	-	+	-	-
59	*Roseobacter sp. 812 (99-100)*	-	+	-	-	+
60.	*Stenotrophomonas acidiminiphila (100)*	+	-	-	-	-
61.	*Sphingomonas sp. MBIC 3990 (99)*	+	-	-	-	-
62	*Spingopyxis sp. 0-1A (99)*	-	+	+	-	-
63.	*Thalassospira sp. DBT-2 (95)*	-	+	-	-	-
64.	*Thalassospira lucentensis (99)*	-	-	-	-	+
65.	*Tristella sp. Zp5 (100)*	-	+	-	-	-
66.	*Tristella mobilis (99)*	-	+	-	+	-

Tabel 7. 16S rRNA sequences are most closely related to GeneBank Database

No.	Name Code	Most closely related sequence (accession no.)	% homology
1.	ID06-002O	*Bordetella petrii,* strain GDH03510 (AJ870969)	95
2.	ID06-004O	*Castellaniella denitrificans strain NKNTAU (U82826)*	*97*
3.	ID06-008O	*Agricultural soil bacterium isolate SI-15 (AJ252582)*	*96*
4.	ID06-077O	*Gamma proteobacterium PI_GH1.1.A2 (AY162032)*	*92*
5.	ID06-097O	*Pseudomonas aeruginosa PAO1 (AE004091)*	96
6.	ID06-105O	*Bacillus sp. JZDN42 (DQ659021)*	97
7.	ID06-108O	*Pseudodiomarinna taiwanensis strain PITI*	97
8.	ID06-113O	*Porphyrobacter donghaensis strain SW-158 (AY559429)*	97
9.	ID06-147O	*Chelatobacter sp.Pht-38 (DQ659453)*	97
10	ID06-155O	*Thalassospira sp. DBT-2 (DQ659435)*	95
11.	ID06-162O	*Porphyirobacter tepidarius strain OK5PO*	97

we still believe that we can estimate oil pollution based on the number of isolated cultured bacteria. Unfortunately the population number of bacteria was not measured in the present study. It was only the number of hydrocarbonoclastic bacteria strain that could be informed. Hydrocarbonoclastic bacteria collected from sediment and seawater were dominated by oil degrading bacteria that occurred in Marina Port and Pari Island. The domination of PAH-degrading bacteria, especially Phenanthrene degrading bacteria was detected in the offshore area of Muara Baru and Pramuka Island. In Rambut island, the number of oil-degrading bacteria and PAH-degrading bacteria were comparable. It might indicate that the offshore area of Muara Baru and Pramuka Island were contaminated by PAH, while, Marina Port and Pari Island were contaminated by crude oil.

CONCLUSION

Oil pollution level might affect the number of strains of isolated hydrocarbonoclastic bacteria. *Alcanivorax* spp. (one of the important hydrocarbonoclastic bacteria) was also found in tropical waters. Four genera of hydrocarbonoclastic bacteria, i.e. *Alcanivorax, Marinobacter, Achromobacter* and *Bacillus* are common in Jakarta Bay and its surrounding waters.

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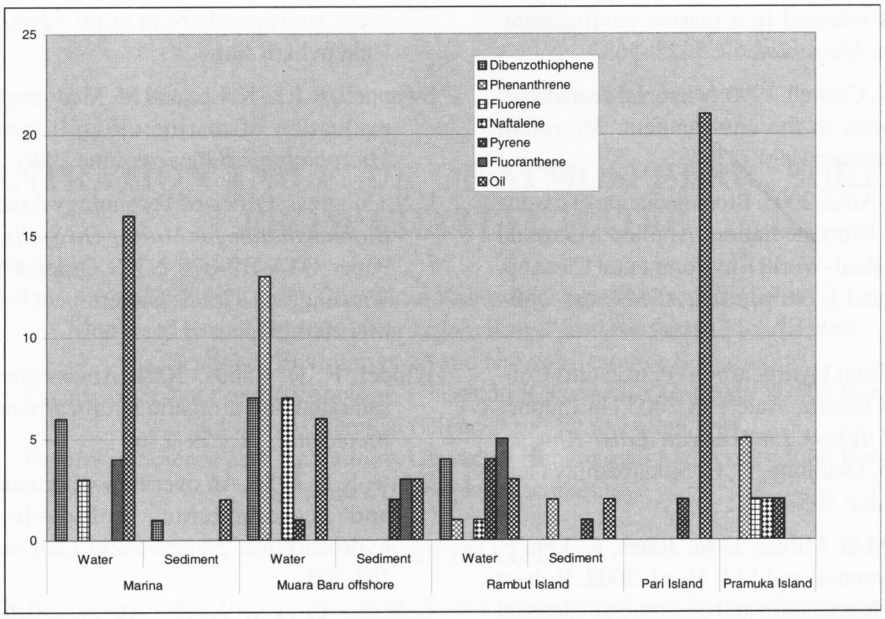

Figure 3. Distribution of oil and PAH degrading bacteria in Jakarta Bay and Seribu Islands

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