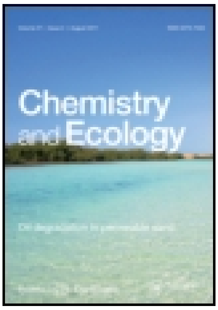

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Experimental degradation of oil in permeable sand from the Gulf of Aqaba, Red Sea

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Degradation rates of light and heavy oil in permeable carbonate sands from the Gulf of Aqaba were investigated to evaluate the ability of sediments to degrade oil compounds. Silicate sands that are less permeable and different properties from carbonate sands were used for comparison. Estimates of oil degradation rates were based on oxygen consumption rates, calculated by incubating natural carbonate sands with oil. The degradation rates of light oil were twofold higher than those of heavy oil, which may be attributed to the presence of a higher carbon number in heavy oil compared with light oil. Degradation rates of light oil in carbonate sands were twofold higher than in silicate sands. Oil degradation rates calculated using the bottle incubation technique were three- to fourfold higher than rates from chamber incubations, indicating the importance of adequate mixing between oil particles and sediments during degradation processes. This study suggests that permeable sands, through their chemical and physical properties, increase oil biodegradation rates by enhancing flow through sediment particles and positively impacting bioturbation processes.

Keywords: oil degradation rate; permeable sand; Gulf of Aqaba; total hydrocarbons

1. Introduction

Oil introduced into the marine environment by accidental spills can spread rapidly by physical, chemical and biological processes, including current dilution, evaporation, dispersion and emulsification [1]. Oil may be attached to sand particles because of gravity during these processes, and adsorption onto suspended matter may occur, followed by sinking, sedimentation and tar ball formation [2,3]. In coastal regions characterised by high concentrations of suspended particles, ∼20–30% of total spill volume has been reported to reach sediments [4].

Benthic microorganisms can play an important role in the biodegradation of oil compounds in sediments. Over 200 species of bacteria, yeasts and fungi have been shown to degrade hydrocarbons in the environment [5]. Bacteria are considered to be the predominant hydrocarbon-degraders in marine environments [6–8] and the higher degradation rates of oil in sediments compared

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with water have been linked to the presence of oil-contaminant-degrading bacteria in sediments. Moreover, nearly 90% of the oil contaminants and bacteria are usually attached to sediments [9].

Oil degradation is highly dependent on environmental conditions, chemical structure [10,11] and the concentrations of oil compounds [12]. In addition, the physical and chemical properties of the sediment also have an impact on the degradation process of any organic material [13–17]. Because of the porous structure, carbonate sands found in tropical or subtropical regions within coral reef ecosystems [18] are able to accumulate both oil compounds and bacteria by chemical and physical adsorption. These sands may play an important role in the degradation of organic compounds because of their high permeability, porosity and buffer efficiency [16–21]. Rusch et al. [22] found that highly permeable sands can act as efficient biocatalytical filters that enhance the mineralisation of organic matter in the sands. Therefore, oil compounds can be degraded aerobically and anaerobically in sediments using microbenthic communities existing in the sediments [8,23–27].

An increase in shipping traffic on the Jordanian side of the Gulf of Aqaba has also increased the frequency of oil spills ranging from small- to medium-scale spills. Small spills include the accidental or intentional release of oil-contaminated bilge or ballast water from freighters; whereas, spills from oily ballast water, bunker oil releases, defective equipment and mishandling at oil terminals/pipeline facilities are examples of medium oil spills [28]. Deliberately leaked or accidentally spilt oil can spread on the sea surface and subsequently evaporate, disperse and/or deposit on the sediments, contributing significantly to sources of water pollution affecting sediments, water quality and biota. According to Wahbeh [29], oil pollution in the Gulf of Aqaba may damage the reproductive system of corals, interfere with the production of larvae and inhibit normal settling. Moreover, continuous thick films of oil may interrupt the photosynthetic cycle of specific coral reefs.

The aim of this study was to investigate the degradation rates of raw oil (light and heavy fraction) in natural permeable carbonate sands along the Jordanian coast bordering the Gulf of Aqaba in order to understand the ability of the permeable carbonate sands to degrade oil compounds that may be accidentally introduced into the sea. A review of the existing literature shows that research on oil spills and pollution in the Gulf of Aqaba is limited. This study will help develop a better understanding of the dynamics of oil degradation in the Aqaba waters and contribute to the development of effective techniques for managing oil pollution.

2. Methods

2.1. *Incubations*

Experiments were designed to evaluate the degradation rates of light and heavy oil in permeable sands using chamber and bottle incubations as described below.

2.1.1. *Chamber incubations*

Carbonate sands that consist mainly of calcareous biogenic materials were collected from a coral reef location near the Marine Science Station (MSS) (29°27′30.52″N, 34°58′34.03″E) by a SCUBA diver. In order to demonstrate the importance of permeability and sand properties in the oil degradation rate, a chamber incubation experiment was conducted using less-permeable silicate sands. These sands, originating from terrigenous sources, were collected from a site near that of the carbonate sands ($\sim$150 m distance). Some properties of the carbonate and silicate sands are shown in Table 1. The permeability of the sands was measured using a constant head permeameter

Table 1. Properties of carbonate and silicate sands.

Character	Carbonate	Silicate
$CaCO_3$ content (%)	92 ± 4	11 ± 4
Organic content (%)	0.32 ± 0.05	0.28 ± 0.05
Mean grain size (μm)	458 ± 10	275 ± 10
Permeability ($m^2 \times 10^{12}$)	114.5 ± 10	43.2 ± 10
Porosity (%)	44 ± 4	34 ± 4
Bacterial cell number ($cm^3 \times 10^7$)	7 ± 1	4 ± 1
Shells number (m^2)	452 ± 20	243 ± 20
Snails number (m^2)	765 ± 40	324 ± 40
Polychaetes	45 ± 6	22 ± 6

as described by Klute and Dirksen [30]. Porosity was calculated from weight loss of wet sediment after drying at 60 °C for 24 h. A bacteria count was conducted using an epifluorescence microscope following the method described by Rusch et al. [31]. The concentration of calcium carbonate in the sediment was determined by complexometric titration of calcium carbonate with 0.1 N of HCl, according to Muller [32].

For chamber incubations, the sand was mixed with 50 mL oil and placed in various acrylic flux chambers, each with an inner diameter of 9.5 cm and a height of 40 cm. The sediment column length was 25 cm. Some chambers were used as controls and remained without any added material. The chambers were then filled with seawater (~15 cm depth) and covered with gastight plastic lids with two sampling ports. To avoid stratification, the water above the sediment in each chamber was stirred using a rotating disk placed above the sediment surface [18]. Three experiments were conducted: (1) six chambers were incubated using natural sands mixed with light oil and four chambers were used as controls without mixed oil, (2) four chambers were incubated using the sands mixed with heavy oil and four chambers were used as controls, and (3) four chambers were incubated using silicate sands mixed with light oil and two chambers were used as controls. Water samples were collected above the sediment level to monitor the concentration of dissolved oxygen as a function of incubation time. The oxygen concentration was determined using the Winkler method according to Grasshoff et al. [33].

2.1.2. *Bottle incubations*

This experiment was performed to estimate oxygen consumption and oil degradation under good mixing conditions. For this purpose, 100 mL of natural carbonate sands and 5 mL of light and heavy oil were incubated separately in a gas tight bottle (250 mL) with 145 mL filtered (0.45 μmfilter) seawater. Twelve bottles were used for each oil type and 12 others were used as controls without oil. The bottles were closed tightly and shaken continuously using a mechanical shaker for 1, 48, 96 and 218 h. At the end of each incubation period, a water sample was taken from each bottle for determination of oxygen by Winkler titration.

2.2. ***Rate calculation***

Degradation rates for oil of both incubation types were calculated from oxygen consumption rates, which were evaluated using the linear regressions of oxygen concentrations over time after subtraction of the rates recorded in the control chambers. The rate was calculated using a ratio of 1 for CO_2 (production): 1 for O_2 (consumption) as used previously [18,34,35].

3. Results

3.1. *Chamber incubations*

Oxygen concentrations decreased in all chambers at different rates (Figure 1). In the first incubation, oxygen decreased gradually for controls in the first 192 h at a rate of $\sim$0.28 μM·hr^{1}, and then decreased at a slower rate to the end of incubation. For chambers with light oil, oxygen concentrations gradually decreased in the first 192 h at a faster rate ($\sim$0.57 μM·h^{-1}) compared with controls. In the second incubation for heavy oil, the trend in oxygen concentration in the control was almost the same as for the first incubation (Figure 2). Oxygen concentrations for

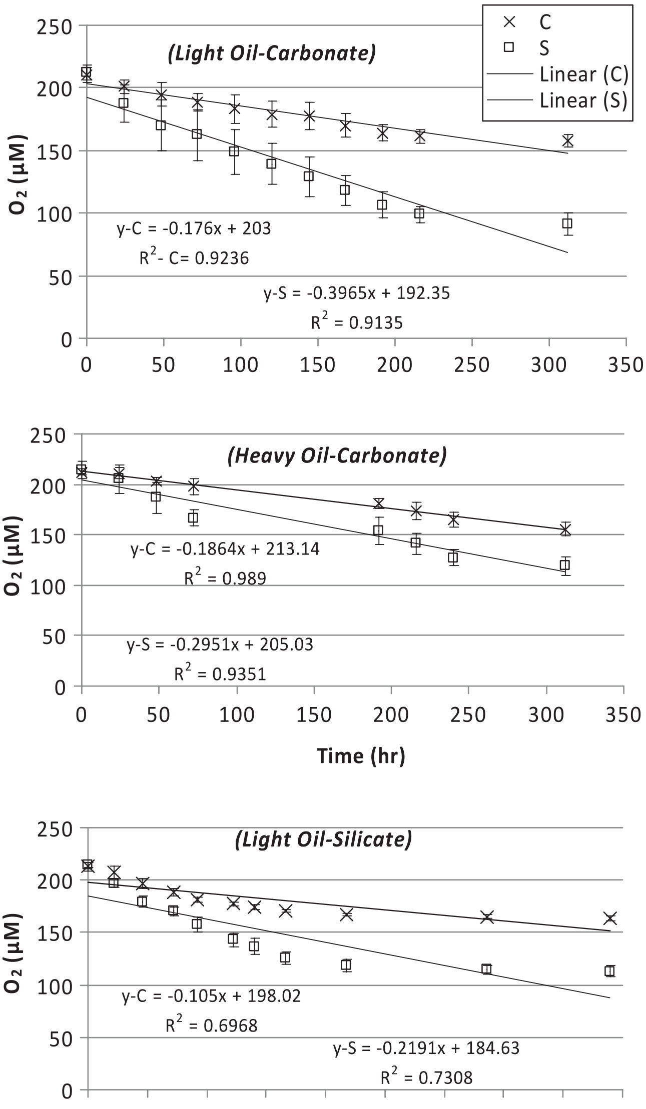

Figure 1. Time course of dissolved oxygen concentrations (μM) during the incubation of control (C) and spiked (S) sands of light and heavy oil. Error bars represent the standard deviation of the concentrations from six chambers for light oil and four chambers for heavy oil and control.

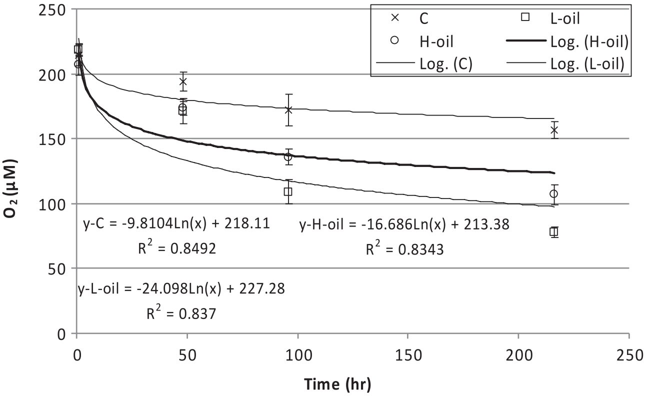

Figure 2. Time course of dissolved oxygen concentrations (μM) during the incubation of control (C), light oil (L-Oil), and heavy oil (H-Oil). Error bars represent the standard deviation of the concentrations from three bottles for each incubation period.

heavy oil also decreased gradually in the first 240 h at faster rate than in the control (0.35 and 0.19 μM·h^{-1} respectively). This was followed by a slower rate of decrease in oxygen. In the chambers with silicate sands, the dearease in oxygen in the light oil followed the same trend as for the carbonate sand, but at a slower rate. In the first 218 h, the rate was 0.44 μM·h^{1}, which decreased to the end of incubation.

3.2. *Bottle incubation*

Oxygen concentrations decreased gradually in the first 96 h for all bottles (Figure 2). In the bottles with light oil, oxygen decreased at faster rates compared with heavy oil and controls (1.15, 0.74 and 0.43 μM·h^{1}, respectively). The final oxygen concentrations in the bottle incubations were 156, 78 and 107 μM for control, light oil and heavy oil respectively.

4. Discussion

4.1. *Oil degradation rates in carbonate sands*

Rates of oil degradation in permeable carbonate sands were calculated for light and heavy oil using the oxygen time course in the two incubation experiments. The degradation of light and heavy oil occurred rapidly in the first few hours of incubation and then started to slow (Figures 1 and 2), which may be due to a decline in the availability of nutrients required for microbial degradation [11]. The degradation rates for light oil were approximately twofold higher than the degradation rates for heavy oil in both chamber and bottle incubations (Table 2). This may be attributed to the fact that heavy oil contains a higher carbon number than light oil and the biodegradation of oil usually increases with decreasing carbon number [25,36]. Branched and cyclic oil compounds that are denser than linear compounds are known to be more resistant to degradation [10,37,38]. Furthermore, resin and asphalts, the heaviest oil compounds, are considered to have low degradation rates [39] or to be refractory to degradation [1]. Degradation rates estimated from bottle incubations were higher than rates estimated using the chamber incubation technique. In the bottle incubations, continuous shaking allowed the oil particles to mix well with the sand particles and maximise nutrient utilisation for bacterial biodegradation.

Table 2. Average degradation rates and standard deviations of light and or heavy oil (mg C·m^{-3}·days^{-1}) in carbonate and silicate sands.

	Chamber incubation Carbonate	Chamber incubation Silicate	Bottle incubation Carbonate
Light oil	34.9 ± 7	18.1 ± 3	155.8 ± 7
Heavy oil	17.2 ± 7		79.2 ± 15

4.2. *Role of permeable sands in oil degradation*

The degradation of light oil in carbonate sands was approximately twofold higher than the rate in silicate sands (Table 2). Carbonate sands had higher permeability and porosity than silicate sands (Table 1). Rasheed et al. [18], used sieved carbonate and silicate sands (250–500 μM) to compared the degradation rate of some natural algae in the sands. They measured different permeabilities for both sands and found a higher degradation rate for carbonate sieved sand compared with sieved silicate sand (1.5-fold). Because of their porous structure, carbonate permeable sands can transport particulate and dissolved matter through the upper layer of the seafloor [40] more rapidly than the less-permeable silicate sand. Advective flow in permeable sand, in addition to diffusion, assists in increased circulation of water through the sediment and introduces more oxygen and other electron acceptors deeper into sediments, leading to an enhanced degradation rate of organic matter [15,17,19,41]. An increased oxygen content drives the aerobic processes that form major pathways for oil degradation in marine sediments [42–44]. In addition, the increased numbers of bacteria found in permeable sands (Table 1) also help in aerobic biodegradation of oil [19,35].

Numbers of macrofauna (shells, snails and polychaetes) were higher in the carbonate sands than the silicate sands (Table 1). This may increase the translocation of oil in the different sand layers [45], which may enhance the degradation of oil in the sand. Polychaetes would transport organic matter oil in the sediment layer below 1 cm [46]. Bioturbation is known to have a significant influence in oil degradation in marine coastal sediments [38]. Because of their porous structure and nutrient availability, permeable coral sands host high numbers of micro and macro benthic fauna that enhance bioturbation in these sands (Table 1) [19,47,48]. Bioturbation processes increase the mechanical mixing and oxygenation of sand, which in turn enhance the degradation of organic matter in sand [47,49]. Gilbert et al. [50] found that bioturbation processes play an important role in the burial and degradation of hydrocarbons, and in the absence of macrofauna, oil compounds were not buried. Biosurfactants present in large amounts in the digestive tract of benthic macrofauna [51] are able to produce digestive solubilisers that enhance hydrocarbon bioavailability and possibly increase transport into the water column [38,52,53]. The burial of oil by bioturbation reduces its chance of being released into the water column, enhances the possibility of its preservation and increases the time available for degradation [38].

A comparison of the results of this study with the published literature is not feasible because most previous studies usually deal with the degradation of specific oil constituents such as alkanes, alkenes, polyaromatic hydrocarbons, etc. using specific microbial treatment. The main concern of our article is the degradation of raw oil in permeable sands; this is a new area of study and to the best of our knowledge there is hardly anything in the literature that can be used for direct comparison. The closest example, perhaps, is a study by Del Arco and Franca [12] focusing on oil degradation in sandy permeable sands and experimental determination of the degradation of grease, *n*-alkanes and pristine using different microbial type and biomass treatments.

5. Conclusion

Degradation of light and heavy oil occurred rapidly in the first few hours of incubation and then tapered off, which may due to the decarese in the concentration of oxygen and nutrients required for microbial degradation. The rates of degradation estimated from bottle incubations, where continuous shaking allowed the oil particles to mix well with sand particles and maximise nutrient utilisation for bacterial degradation, were higher than those estimated from chamber incubations. The degradation rates of oil in permeable carbonate sands were higher than the rates for silicate sands. The results and discussion show that permeable sands play an important role in oil degradation due to their physical and chemical properties by enhancing flow of oil through sediment particles and positively impacting bioturbation processes.

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