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Microbial Functional Group activities of an Oil Processing Facility and How They relate to Corrosion and Souring.

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ABSTRACT

Microbial functional group activities such as the ability to reduce sulfate and generate sulfide by sulfate reducing bacteria (SRB), oxidize sulfide and reduce nitrate by sulfide oxidizing, nitrate reducing bacteria (soNRB), and nitrate reduction by heterotrophic nitrate reducing bacteria (hNRB) were determined in samples collected from an oil producing and processing facility in Coleville synthetic brine medium (CSB-K). Corrosion rates of the initial raw samples were measured with a linear polarization resistance (LPR) probe. Skimmer pit water samples (ES_SP) recorded the highest SRB activity in both VFA and lactate media (120 and 167 units/day). Same sample also recorded relatively high hNRB activity (127 units/day) and soNRB activity (116 units/day) with a relatively high corrosion rate (0.23±0.042 mm/yr). Other samples that recorded relatively high corrosion rates with corresponding high microbial activities include; PG_S (0.2±0.07), ES_SW (0.14±0.028) ES_MX (0.1±0.028) and ES_PW(0.08±0.042). Sample AB_PW recorded relatively low corrosion rate (0.05±0.042) with a corresponding low microbial activity. The study have clearly demonstrated how the knowledge of the presence and activities of the three microbial functional groups studied can be used to mitigate corrosion and souring episodes.

Keywords: SRB, soNRB, hNRB, Souring, Corrosion

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INTRODUCTION

Microorganisms thrive in oil reservoirs under strict anaerobic conditions and the major metabolic processes in oil reservoirs are sulfate reduction, methanogenesis, fermentation and to some extent nitrate reduction [1,2]. The anaerobic food chain of oil field microorganisms is therefore based on the use of organic compounds by the fermentative bacteria and SRB that oxidizes organic matter under anaerobic conditions and methanogenesis through carbon dioxide reduction and hydrogen scavenging may be the dominant terminal metabolic process [2]. The potential electron donors for fermentation and sulfate reduction include numerous organic molecules such as acetate, formate, propionate, butyrate and benzoate [1,3].

Sulfate reducing bacteria and Archaea constitute a large and heterogeneous physiological group of strictly anaerobic prokaryotes that are commonly found in oil fields. They share the ability of anaerobic respiration using sulfate as a terminal electron acceptor and organic compounds or hydrogen as electron donors in a process known as dissimilatory sulfate reduction as opposed to assimilatory sulfate reduction [4]. SRB are widespread in nature and they play an important role in global sulfur cycle and in marine sediments, they can account for up to 50% of the total carbon mineralization [4]. SRB are also known for their ability to reduce sulfate thereby producing toxic hydrogen sulfide gas which results to souring and also for their corroding activities [1,3,5]. Various nitrate reducing microorganisms with autotrophic [6], heterotrophic [7] and Chemolitotrophic [8] abilities have been isolated. Methanogens have also been frequently isolated from oil reservoirs [5,9]. Methanogens are group of microorganisms that metabolize hydrogen, carbon dioxide, acetate, methyl amines and dimethyl sulfides with the concurrent production of methane and they are distributed among five orders namely; *Methanomicrobiales, Methanobacteriales, Methanosarcina, Methanococcales and Metahnopyrales* [9]. Another important group of oilfield microorganisms are the fermentative group which has been identified in 4 out of the 6 currently recognized genera of the phylum *Thermotoga, Thermosipho, Geotoga and Petrotoga* [5].

Three main microbial functional group activities that play significant roles in corrosion and souring episodes are, i. The ability of SRB to reduce sulfate and generate sulfide, ii. The ability of heterotrophic nitrogen reducing bacteria (hNRB) to reduce nitrate to nitrite and the ability of the sulfide oxidizing nitrate reducing bacteria (soNRB) to oxidize sulfide and reduce nitrate [10]. SRB for instance can initiate an incomplete oxidation of oil organics to acetate and carbon dioxide or complete oxidation of acetate to carbon dioxide and the reduction of sulfate to sulfide. hNRB can initiate the incomplete oxidation of oil organics to acetate or carbon dioxide and reduction of nitrate to nitrite and then to either nitrogen or ammonia while so-NRB oxidizes sulfide to sulfate or sulfure with nitrate being reduced to nitrite and then to either nitrogen (with NO and N_2O) as intermediates or ammonia without intermediates as shown in Figure 1 [11]. The implication of this is that while SRB is problem causing, hNRB and so-NRB are beneficial to the environment.

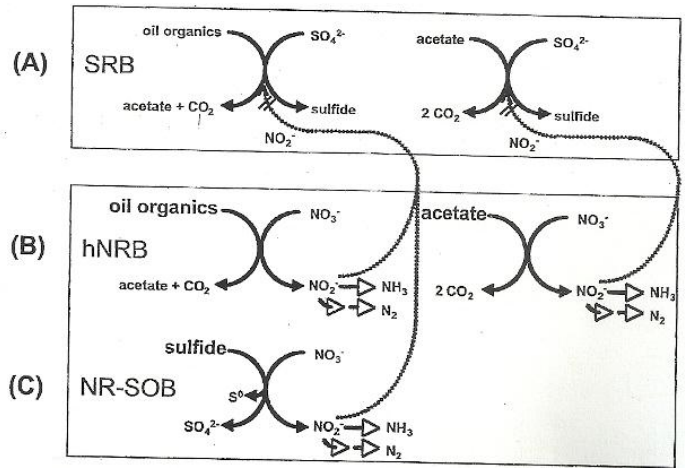

Figure 1: Functional group microbial activities. (A). SRB couple incomplete oxidation of oil organics to acetate and CO_2 or complete oxidation of acetate to CO_2 to the reduction of sulfate to sulfide.(B). hNRB couple incomplete oxidation of oil organics or complete oxidation of acetate to CO_2 to reduction of nitrate to nitrite and then to either nitrogen or ammonia. (C). NR-SOB (so-NRB) oxidize sulfide to sulfur or sulfate with nitrate being reduced to nitrite and then to either nitrogen or ammonia. (adapted from Voordouw, 2008),

Nitrite which is a product of nitrate reduction by hNRB and the so-NRB is a powerful SRB inhibitor and Nitrite has worked efficiently with some biocides to inhibit SRB [11]. Nitrate can also inhibit SRB activities by stimulation of hNRB (competitive exclusion) [12]. Recently, it has been discovered that so-NRB can be used to control souring by its ability to oxidize sulfide(lowering sulfide levels) and reduce nitrate to nitrite which further inhibit SRB [11,13]. Naturally, so-NRB and hNRB are very useful for the control of souring and corrosion and need not be inhibited by biocides.

The main goal of the present investigation therefore is to determine how microbial functional group activities influence souring and corrosion episodes in the oil facility under investigation. It is believed that a clear understanding of these functional group activities can give an insight on the how best to mitigate souring and corrosion related problems in the affected oil fields.

MATERIALS AND METHODS

Sample collection

Samples ES_SP (Escravos skimmer pit water), ES_SW (Escravos seawater), AB_PW (Abiteye Produced water), ES_PW (Escravos produced water), PG_S (Pigging solids) and ES_MX (Escravos seawater and produced water mixture) were collected in sterile 500 ml Nalgene sample bottles which were filled to the brim to exclude air and analyzed within 48hrs of collection.

Chemical Analysis

The samples were analyzed for pH, SO_4^{2-}, HS^- , NH_4^+, NO_3^-, NO_2^- and organic acids such as acetate, propionate and butyrate. The pH was analyzed using Orion pH meter. SO_4^{2-} was analyzed in two ways, through High Performance Liquid Chromatography (HPLC) and through turbidimetry using $BaCl_2$ [14]. HS^-, a dissolved sulfide was analyzed using diamine method [15]. NH_4^+ was analyzed using the indol-phenol method. NO_3^-, NO_2^- and organic acids such as acetate, propionate and butyrate were analyzed using HPLC. SO_4^{2-}, NO_3^- and NO_2^- were analyzed using 100 µL of the samples with 400 µL HPLC anion buffer while organic acid analysis used 300 µL of the samples and 20 µL 1 M phosphoric acid as described elsewhere [16].

Corrosion rate measurement

The electrochemical corrosion rate of the initial samples was carried out with a linear polarization resistance (LPR) probe using a portable meter (AquaMate® Portable CORRATER® LPR Corrosion Rate Instrument, Cosasco, USA). The probe was put into each sample bottle (15ml volume) and nitrogen gas was continuously flowed into the headspace of the bottle during the corrosion rate measurement.

Microbiological assay

The medium that was used for the microbiological assay was Coleville synthetic brine (CSB-K) with composition (g/L) as previously described [16]; NaCl(1.50), $CaCl_2$ $2H_2O$ (0.21), $MgCl_2$ $5H_2O$ (0.54), NH_4Cl (0.30), KCl (0.10), KH_2PO_4 (0.05) Resazurin, (1%) 2-3 drops. These chemicals were mixed and dissolved in MQ water in an Erlenmeyer flask and were transferred to a widdel flask for autoclaving. After autoclaving, more components were added: Trace elements (1 ml), Selenate-tungstate (1 ml), $NaHCO_3$ (1 M) 30 ml, Na_2S (1 M)1 ml, HCl (2 M) 2 ml, pH adjusted to 7.4. The Widdel flask was connected to a gas stream of 90% N and 10% CO_2. About 70 ml of the medium was then aseptically and anaerobically dispensed to 125 ml serum bottles with a gas phase of 90% N and 10% CO_2 and closed with a sterile butyl rubber stopper.

Components added to CSB-K for specific microbiological tests

The following electron donors and acceptors were added to the CSB-K medium in serum bottles to determine the functional group activity of major bacterial groups:

a. Sulfate-reducing bacteria (SRB) – 40 mM lactate and 20 mM sulfate; 3 mM VFA and 20 mM sulphate
b. Heterotrophic nitrate reducing bacteria (hNRB) – 3 mM VFA and 10 mM nitrate
c. Sulfide-oxidizing, nitrate-reducing bacteria (so-NRB) – 5 mM sulfide and 10 mM nitrate

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3.5 ml of the samples (5%) were added to the prepared media bottles and incubated at 37°C in a shaker for about 30 days. Using a sterile syringe needle, 1 ml of the sample was taken periodically for every 2 days within the first one week and subsequently for every 7 days and analyzed for sulfide, sulfate, nitrate and nitrite using HPLC. Microbial activities were calculated as $100/t_{1/2}$, where $t_{1/2}$ is the time (days) needed to reduce half of the sulfate (SRB activity), nitrate (hNRB and so-NRB activities) and sulfide concentrations (so-NRB).

Most Probable Number (MPN) Measurement

To quantify presence of SRB in the samples, API RP-38 broth media were used. Serial dilution of the samples in API RP-38 broth media of up to 10 fold was made. With the use of a sterile syringe needle, 1 ml of each sample was inoculated serially to the 9 ml medium up to the 10^{th} tube making a ten-fold dilution. Samples were then incubated at 37°C for up to 30 days. Formation of black precipitates of iron sulfide was used as a diagnostic tool to confirm the presence of SRB. For acid producing bacteria, prepared ZPRA-5 acid produced media (Phenol red-dextrose reagent) with a salinity of 5000ppm was used. Change in color from orange to yellow shows presence of acid producers (Fermentation of dextrose).

RESULTS

Chemical characterization of samples

pH of samples ranged between 6.9 and 7.4. Considerable amount of sulfate was found in Escravos sea water (ES_SW), Escravos produced water (ES_PW), Abiteye Produced water (AB_PW) and the point of discharge where sea water mixes with produced water (ES_MX). Traces of hydrogen sulfide was found in Escravos skimmer pit water (ES_SP) and point of produced water discharge (ES_MX) while considerable sulfide concentrations was found in the pigging solid samples (PG_S). NH_4^+ was present in all samples. Nitrate was also present in all samples except sample AB_PW. No nitrite was found in all samples. The VFA, acetate and propionate were present at considerable concentrations in samples ES_SP, ES_SW, PG_S and ES_MX but butyrate was absent in all the samples. Detailed results are shown in Table 1.

Table 1: Chemical analysis of samples (Values in millimolar)

S/N	Sample Code	pH	HS^- Chemical	SO_4^{2-} Chemical	SO_4^{2-} HPLC	NH_4^+ Chemical	NO_3^- HPLC	NO_2^- HPLC	Acetate HPLC	Propionate HPLC	Butyrate HPLC
1	ES_SP	6.9	0.16	2.45	1.56	2.14	0.04	0	20.40	2.80	0
2	ES_SW	7.1	0	24.6	22.80	0.18	0.02	0	8.8	1.6	0
3	AB_PW	7.4	0	10.25	8.65	1.50	0	0	0.65	0.4	0
4	ES_PW	7.2	0	16.45	13.20	0.16	0.05	0	0.90	2.2	0
5	PG_S	6.9	2.2	1.16	2.16	2.71	0.54	0	8.80	3.4	0
6	ES_MX	7.1	0.12	14.30	11.80	2.40	0.03	0	14.4	1.5	0

Microbiological counts using MPN technique

The results of microbiological analysis carried out on samples using MPN technique showed relatively high concentrations of sulfate reducing bacteria (SRB) in samples ES_SP, PG_S and ES_MX. Heterotrophic nitrate reducing bacteria (hNRB) were present in all samples but the soNRB were only present in samples ES_SP, ES_SW, AB_PW and ES_MX. Detailed results are shown in table 2.

Table 2: Most Probable Number (MPN) results of samples.

S/N	Sample code	Sample description	# of SRBs per ml	SRB	hNRB	so-NRB
1	ES_SP	Escravos Skimmer pit water	10^6	+	+	+
2	ES_SW	Escravos Seawater	10^2	+	+	+
3	AB_PW	Abiteye Produced Water	BD	-	+	+
4	ES_PW	Escravos produced water	10^2	+	+	-
5	PG_S	Pig run solids	10^6	+	+	-
6	ES_MX	Escravos/ produced water Mixture	10^3	+	+	+

BD - below detection - absence of bacteria + presence of bacteria

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Functional group microbial activities of oil field samples

Escravos skimmer pit water (ES_SP)

Microbiological activity results revealed that sulfates were readily utilized and reduced by the resident SRBs leading to the appreciable production of hydrogen sulfide as shown in Figure 2. Sulfate reduction was observed both in lactate and VFA media as shown in Figure 3a. SRB activity test in both VFA and lactate media recorded (120 and 167 units/day). Nitrate was considerably reduced by the hNRB within the 28 days of exposure and hNRB activity was 127units per day. The soNRB also reduced nitrate considerably as shown in Figure 3b but sulfide oxidation was not properly measured due to time lag and its high volatility. soNRB activity recorded 116 units/day.

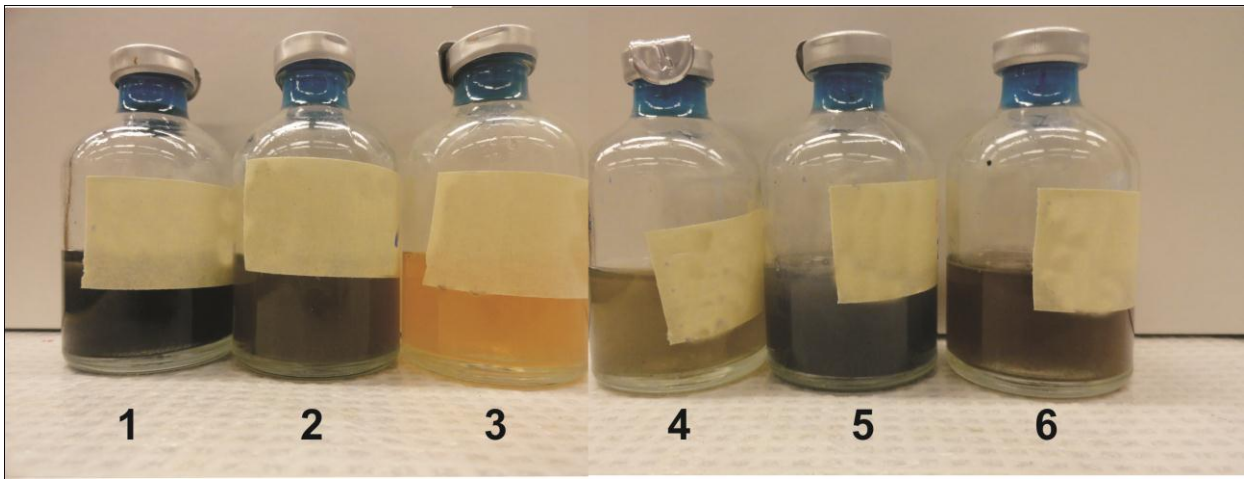
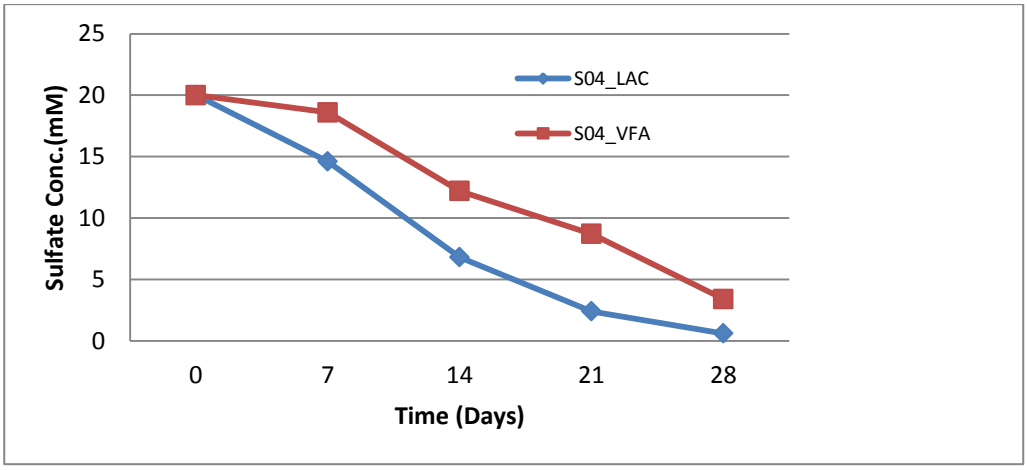


Figure 2: Functional group activity tests of samples 1(ES_SP), 2(ES_SW), 3(AB_PW), 4(ES_PW), 5(PG_S), and 6(ES_MX). Black precipitates are as a result of hydrogen sulfide production. Photographs were taken following 4 weeks of incubation.

Escravos seawater (ES_SW)

Escravos seawater samples (ES_SW) also showed appreciable reduction of sulfate and hydrogen sulfide production as shown in Figure 2. Sulfate was also considerably reduced in both lactate and VFA media with an SRB activity that ranged between 86-97 units/day in VFA and lactate media (Fig. 4a). hNRB and soNRB activities also showed considerable reduction of nitrate as shown in Figure 4b. hNRB activity was 148 units/day while soNRB activity was 132 units/day.



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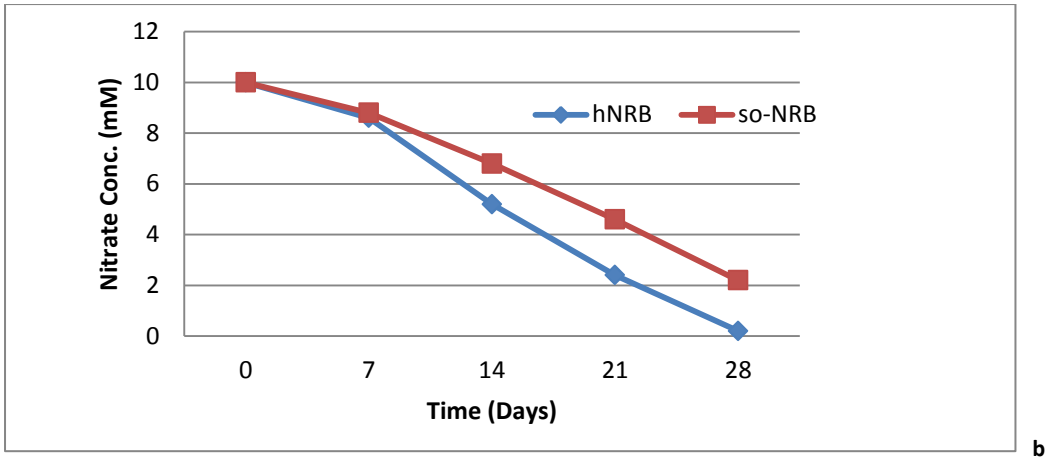

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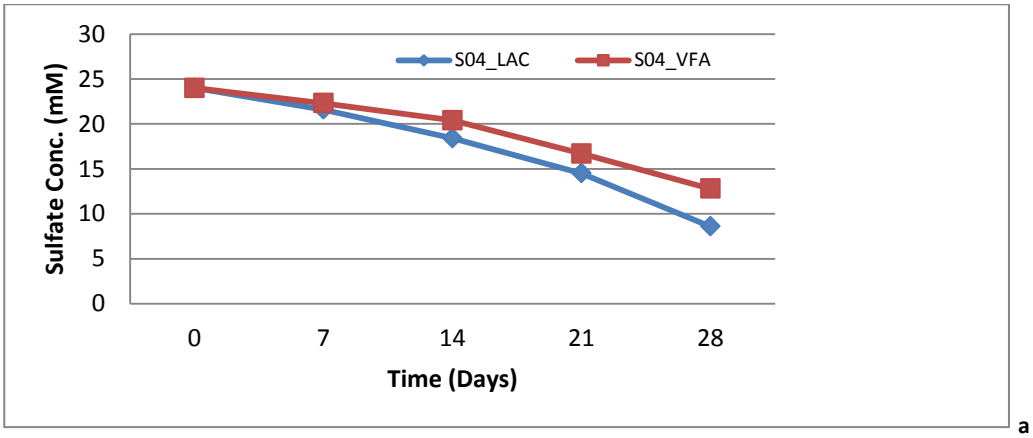
Figure 3: Microbial activities in sample ES_SP showing a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

Abiteye Produced Water (AB_PW)

Abiteye produced water (AB_PW) showed relatively low microbial activity and consequently, hydrogen sulfide presence was not observed as shown in Figure 2. As expected the rate of sulfate reduction in both lactate and VFA media were relatively low (Fig. 5a). SRB activity in both VFA and lactate media ranged between 20 – 36 units/day. On the contrary, the activities of hNRB and soNRB were relatively high as seen in the drastic reduction of nitrate (Fig. 5b). hNRB activity was 165 units per day while soNRB activity was 120 units/day.



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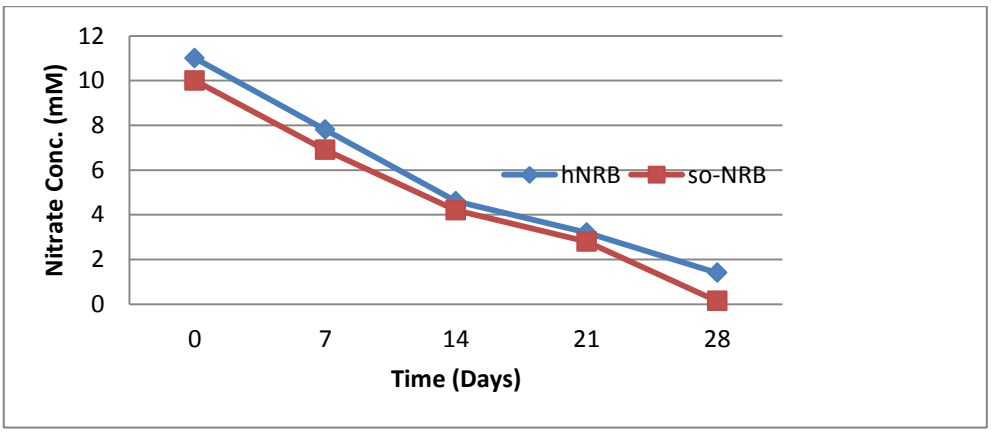

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Figure 4: Microbial activities in sample ES_SW showing a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

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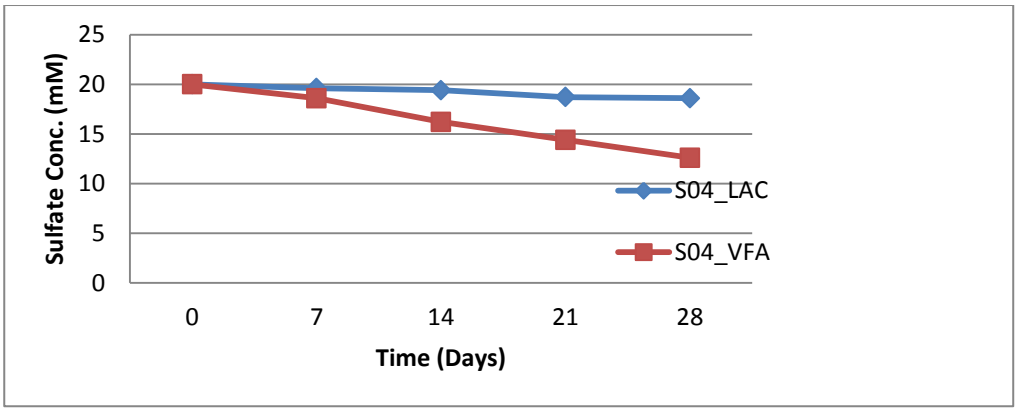
Escravos Produced Water (ES_PW)

Escravos produced water showed relatively low SRB activity as observed in low sulfide concentration (Fig.2). Rate of sulfate reduction in both lactate and VFA media were relatively low (Fig.6a).SRB activity in VFA and lactate media ranged between 50-85 units/day. Heterotrophic NRB activities were considerably high (136 units/day) but those of soNRB were relatively low (36 units per day) (Fig.6b).



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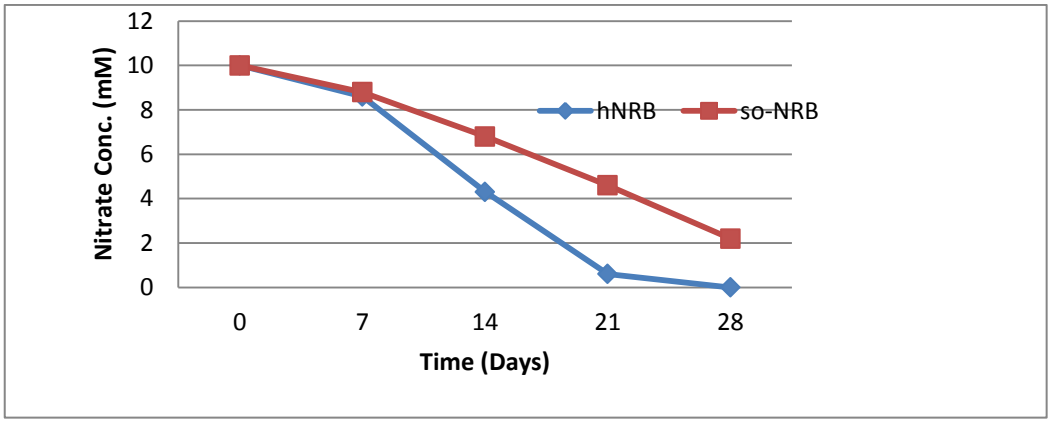

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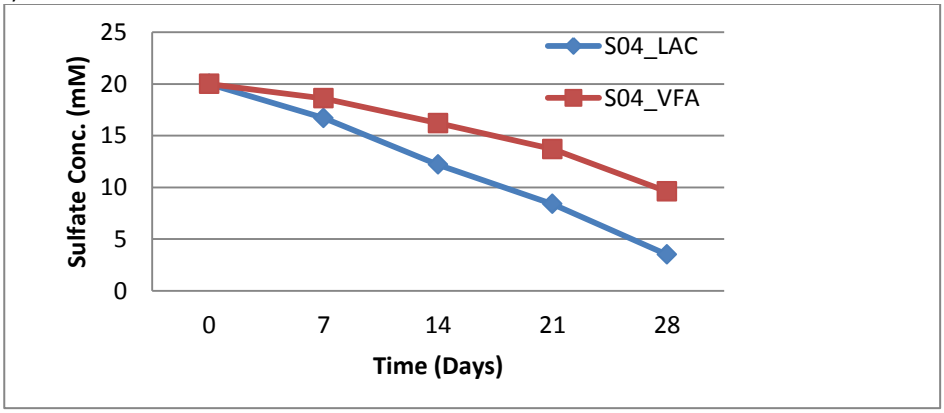
Figure 5: Microbial activities in sample AB_PW showing a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

Pigrun Solid Samples (PG_S)

Pigrun solid samples (PG_S) showed high sulfide concentrations (Fig.2) and the rate of sulfate reduction in both lactate and VFA media was drastic (Fig.7a), an indication of high SRB concentrations. SRB activity in VFA and lactate media ranged between 87-110 units per day. The activities of hNRB was high (115 units/day) as shown in the drastic reduction of nitrate but those of soNRB were relatively low (16 units per day) (Fig. 7b).



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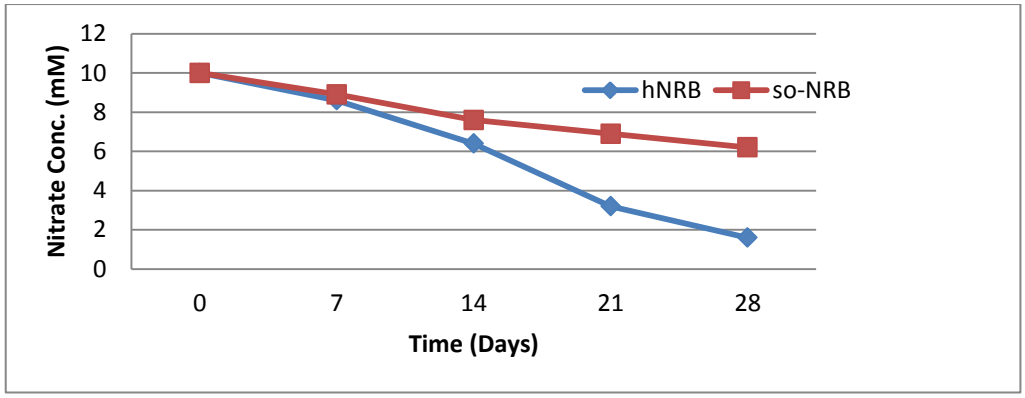

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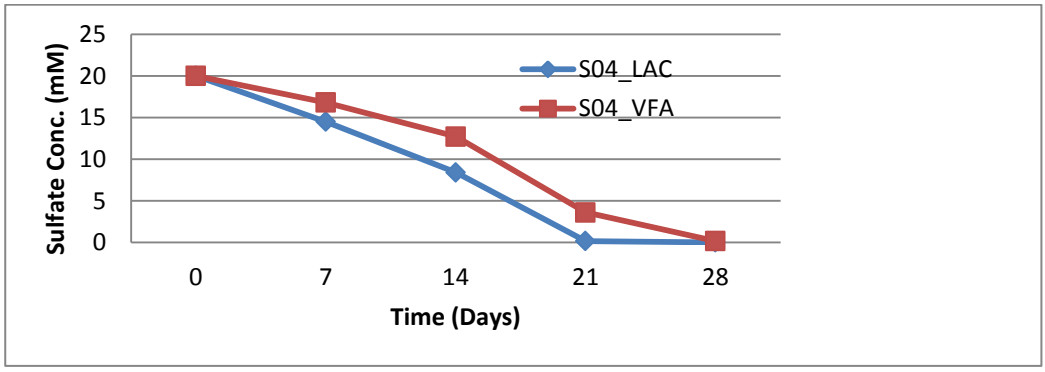
Figure 6: Microbial activities in sample ES_PW showing a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

Escravos/Produced water mixture (ES_MX)

This sample was collected from the produced water discharge area at Escravos facility where seawater mixes with produced water. The sample recorded considerable high microbial activity and relatively high sulfide concentration (Fig.2). Rate of sulfate reduction in both lactate and VFA media was drastic (Fig. 8a), same with the activities of hNRB and soNRB (Fig.8b). SRB activity in VFA and lactate media ranged between 110-138 units/day. hNRB activity recorded 140 units/day while soNRB activity recorded 186 units/day.



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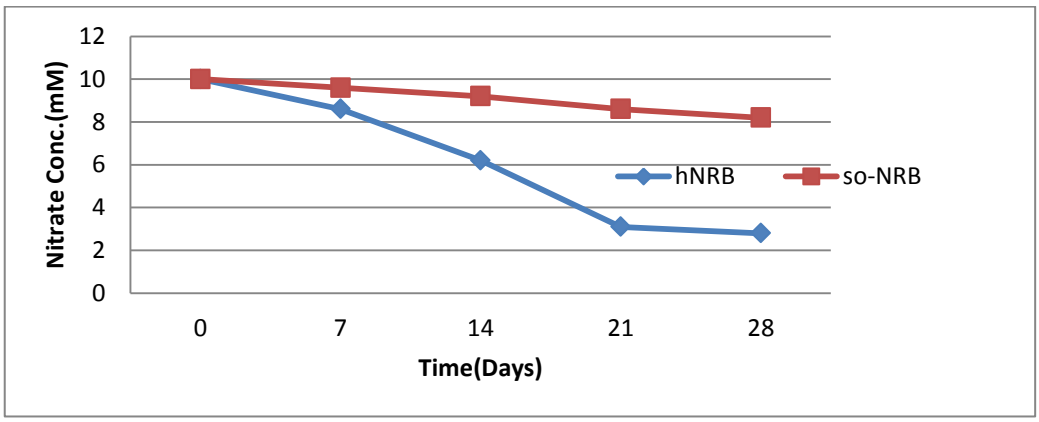

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Figure 7: Microbial activities in sample PG_S showing a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

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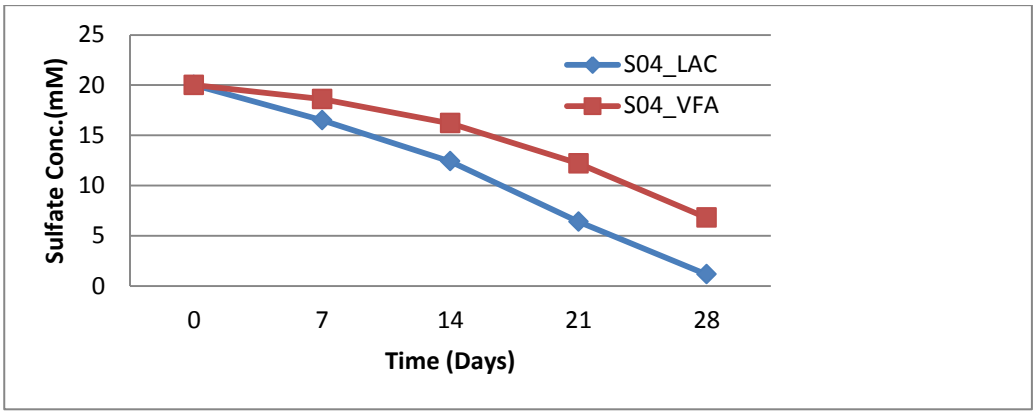
Corrosion rates of initial raw samples

Corrosion rate measurements of initial raw samples indicated that Escravos skimmer pit samples (ES_SP) recorded the highest corrosion rate (0.23 ± 0.042 mm/yr) and this correlated with high microbial activity in CBS-K medium. Other samples that recorded relatively high corrosion rates in (mm/yr.) that also correlated with high microbial activity in CBS-K medium are; PG_S (0.2 ± 0.07), ES_SW (0.14 ± 0.028), ES_MX (0.1 ± 0.028), and ES_PW (0.08 ± 0.042). AB_PW recorded relatively lower corrosion rates (0.05 ± 0.042) which also correlated with low microbial activity. Detailed corrosion rate results are shown in Figure 9.



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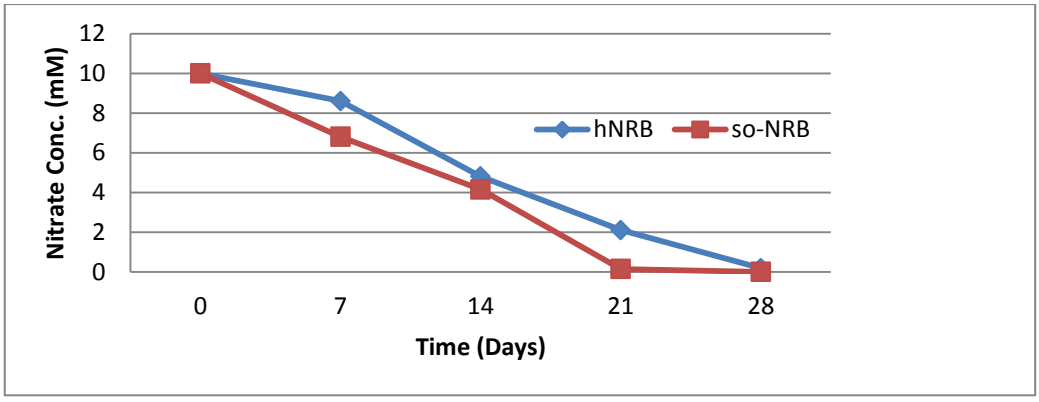

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Figure 8: Microbial activities in sample ES_MX showing; a. SRB activities in both lactate and VFA media and b. Activities of hNRB and soNRB

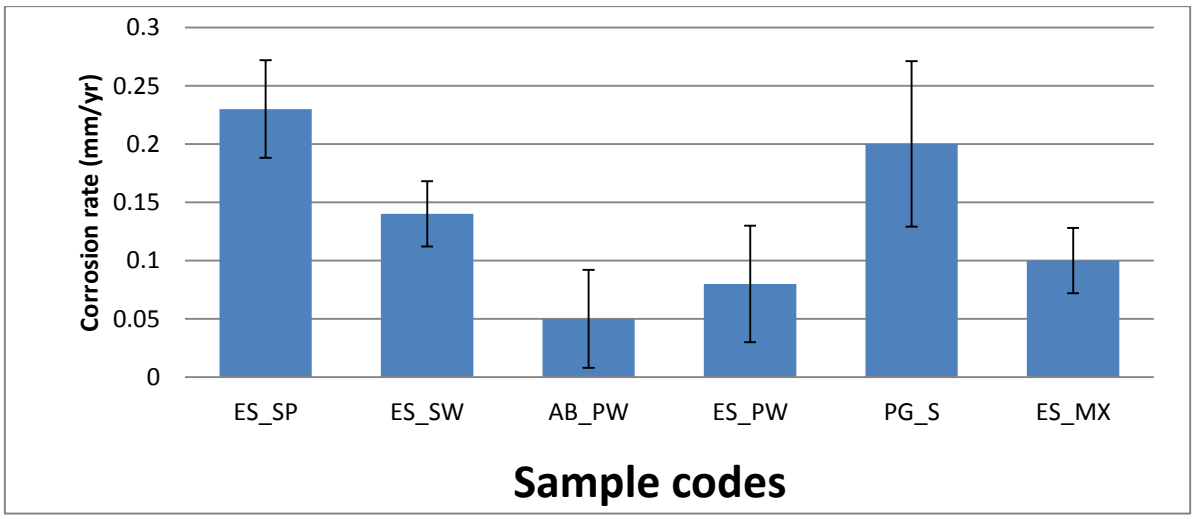

Figure 9: Corrosion rate measurements of samples

DISCUSSIONS

Results of chemical analysis indicate that skimmer pit samples (ES-SP) and pig run samples (PG_S) recorded relatively low sulfate concentrations (1.56 and 2.16 mM) respectively but with substantial concentrations of organic nutrients and SRBs. This is an indication that the organic nutrient present encouraged the growth and proliferation of SRBs which led to rapid utilization of sulfate and generation of hydrogen sulfide. This opinion has been advanced by some other investigators [10,17]. Samples collected from the area where Escravos produced water discharge mixes with sea water (ES_MX) also recorded relatively high concentration of organic nutrients, sulfate and SRBs. This is expected because both the sea water and produced water have relatively high sulfate content and low dilution effect at the point of discharge ensures that considerable concentrations of organic nutrients, sulfate and SRBs are maintained at any point in time. The rest of the samples comprising of ES_SW, AB_PW and ES_PW recorded relatively high sulfate concentration but with lower concentrations of organic nutrients when compared with the other samples. These samples also showed lower concentrations of SRBs probably due to lower organic nutrient content. hNRBs were found to be present in all samples while soNRBs were also present in all samples except samples ES_PW and PG_S. Previous studies in Nigerian oil fields have also shown the presence of SRBs, hNRBs and soNRBs [16].

On how functional group activities relate to souring and corrosion, we observed that the sample that recorded the highest SRB activity (ES_SP, 120-167 units/day), also recorded high hNRB and soNRB activities (127 and 116 units/day respectively) and the highest corrosion rates (0.23±0.042 mm/yr) with clear evidence of sulfide. On the other hand, sample AB_PW that recorded the lowest SRB activity (20-36 units/day) but with considerable hNRB and soNRB activity (165 and 120 units/day respectively), recorded the lowest corrosion rates (0.05±0.042) with no evidence of sulfide production. The activities of SRB, hNRB and soNRB as it relates to corrosion and souring have also been advanced by some other investigators [12,16,18,19] and it has been clearly established that while SRB activities induces souring and corrosion, those of hNRB and soNRB seem to limit the rate of SRB activity by competitive exclusion. It has also been established that both sulfide oxidation and nitrate reduction by the soNRB and nitrate reduction by the hNRB generates nitrite that also inhibits SRB activities [13,20]. Pig run samples from Nigerian oil rich Niger Delta have in the past recorded high microbial activity and corrosion rates (16,21) and the results of sample PG-S confirmed this assertion.

Some investigators [3,10] have further confirmed that the hNRBs and the so-NRBs are the two main physiological types of NRBs that are involved in the control of SRB activity, the implication of this is that wherever they are present, the prediction is that they can inhibit the growth of SRB with the injection of nitrate that stimulates their growth and activities [10,18,22]. With the presence of the indigenous hNRBs and the so-NRBs in the skimmer pit and the produced water discharge point at sea which were already saturated with SRBs, controlling the SRBs with nitrate injection may be the most appropriate solution to the potential menace of the SRBs in these fields.

CONCLUSIONS

The present study have clearly established that knowledge of the presence and activities of SRB, hNRB and soNRB in oil fields can help the operators map out clear strategies that can be used to mitigate incidence of corrosion and souring in the affected oil fields.

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