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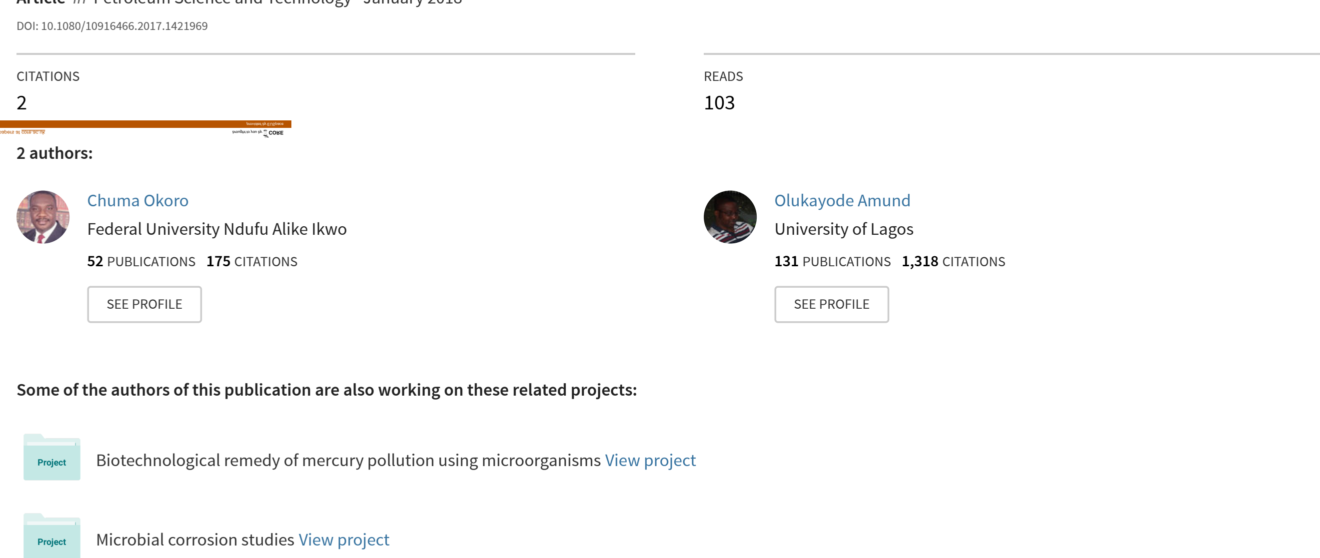

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Microbial community structure of a low sulfate oil producing facility indicate dominance of oil degrading/nitrate reducing bacteria and Methanogens

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ABSTRACT
Analysis of microbial community structure of a low sulfate oil producing facility in Nigeria using 16S rRNA gene sequencing technique revealed dominance of oil degrading and nitrate reducing bacteria and methanogenic archaea in produced waters and oil samples namely, *Marinobacter* (37%), *Azovibrio* (21%), *Thauera* (10–28%), and *Methanolobus* (22%). On the contrary, the associated oil pipeline samples revealed massive dominance of potentially corrosive *Methanolobus* (60%) and *Methanobacterium* (25-27%). Further experimentation shows that the methanogens implicated in oil pipelines are corrosive moderate halophile that utilizes H_2/CO_2 and methanol as substrates. More emphasis should therefore be on methanogenic archaea as opposed to sulfate reducing bacteria (SRBs) during mitigation plans for microbially induced corrosion (MIC) in a low sulfate oil producing facility.

KEYWORDS
low sulfate oil facility; oil degrading bacteria; nitrate reducing bacteria; methanogens; MIC

1. Introduction

Petroleum reservoirs harbor a rich and diverse community of microorganisms including, fermentative, sulfure and sulfate reducing, nitrate reducing, metal reducing and methanogenic microorganisms (Magot et al. 2000). Among these microorganisms, the heterotrophic and hydrogenotrophic species found mostly in methanogenic archaea, sulfate reducing and oil degrading bacteria are considered common inhabitants of oil field environments (Fardeau et al. 2000; Magot et al. 2000). Presently, an assessment of microbial diversity and habitat conditions within a petroleum reservoir is now recognized as an important component of reservoir management (Li et al. 2012). A comprehensive assessment of the diversity, metabolic processes and habitat conditions for petroleum microorganisms is therefore of practical importance in assessing the economic potentials of oil fields.

In the past, a great deal of attention has been paid by researchers to microbial related problems and microbial community structures in sulfate rich oil environments (Dahle et al. 2008; Grawbowski et al. 2005; Grigoryan et al. 2009) with little information on microbial community structures and associated problems in low sulfate oil environments. Persistent corrosion of pipelines transporting crude oil and water in a low sulfate oil producing facility in Nigeria (Okoro et al. 2017), provided strong incentives for a comprehensive assessment of its microbial community structures and the corrosive potential of the dominant methanogens.

The study is therefore expected to provide an insight on why corrosion persists in low sulfate environment with little or no sulfate reducing bacterial activity.

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2. Materials and methods

2.1. Sample collection

Oil and water samples were collected from Obigbo North facility in sterile Nalgene sample bottles and filled to the brim to exclude air. Solid deposit samples from crude oil transport pipelines (OBPG-1 and OBPG-2) were collected the same period from two different locations in sterile zip lock nylon bags.

2.2. Chemical analysis

The pH of the samples was measured using an Orion pH meter. Aqueous sulfide was analyzed using the diamine method (Truper and Schlegel 1964) and NH_4^+ with the indophenol method (Apha 1980). Sulfate, NO_3^-, NO_2^- and the volatile fatty acids (VFA) such as acetate, propionate and butyrate were analyzed by high-performance liquid chromatography (HPLC), as described by Grigoryan et al. (2009). Measurement of dissolved oxygen concentration in samples was determined using the method of Eaton et al. (1995). Fe^{2+} assay was conducted as described in Okoro et al. (2013) while NaCl and conductivity measurements were carried out as described in Eaton et al. (1995).

2.3. Measurement of microbial activities and most probable numbers

The activities of SRB, as well as of heterotrophic nitrate reducing (hNRB) and of sulfide-oxidizing nitrate-reducing bacteria (soNRB) were measured in Coleville synthetic brine (CSB-K) medium as previously described (Okoro et al. 2014). The most probable number (MPN) of lactate-utilizing SRB and acid producing bacteria (APB) were also determined as described in (Okoro et al. 2015).

2.4. Carbon steel coupon corrosion testing

Carbon steel ASTM A366 (ASTM international designation A 1008/A) coupons ($2 \times 1 \times 0.1$ cm) containing 0.08% (w/w) carbon were cleaned according to a standard protocol (NACE 2013). The corrosion rate (CR; millimeter yr^{-1}) was determined as described in Okoro et al. (2014) from the metal weight loss (ΔW in g) as:

$$CR = \frac{87600 \ \Delta W}{A \ D \ T}$$

where A, D and T represent the coupon area (6.1 cm^2), the density of the steel (7.85 g/cm^3) and the incubation time (h), respectively.

2.5. Methane production test (Methanogen activity)

The ability of viable organisms in the samples to produce methane was determined using CSB-K medium amended with H_2/CO_2 (80:20) v/v head space and 20 mM methanol. 2.5 ml of sample (or 1g of solid sample) was inoculated onto 30 ml of CSB-K medium in an 80 ml serum bottle with and without two carbon steel coupons ($5 \times 0.5 \times 0.1$ cm). The head space of each bottle was filled with 80% H_2 and 20% CO_2 gas. The setup without inoculating the sample was served as a control. Each sample was incubated at 32°C and 100 rpm for a period of 6 weeks. Aliquote (0.2 ml) of culture headspace was removed periodically and the methane content was determined using HP 5890 gas chromatograph equipped with the Porapak R 80/100 column (0.049 cm × 5.49 m) with helium as carrier gas (flow rate of 54 ml/min) and a detector (5975C Inert XLMSD series, Agilent). After culturing, corrosion rates of coupons were determined by the weight loss method as described in Section 2.4.

2.6. DNA extraction, amplification, sequencing and bioinformatic analysis

DNA extraction and amplification was carried out as described in Okoro et al. (2014). PCR products (typically 100 ng) were sent to the Genome Quebec and McGill University Innovation Centre for pyrosequencing. The entire set of the raw reads is available from the Sequence Read Archive at the National Center for Biotechnology Information (NCBI) under **accession numbers; SRR1508449, SRR1508450, SRR1508452, SRR1508453 and SRR1508454.** Injection water samples (OBIW) was not sequenced due to low concentrations of DNA.

3. Results

3.1. Chemical characterization of samples

physico-chemical properties of samples are shown in Table 1. pH values of all samples ranged between 6.85 and 8.32 while site temperature ranged between 25 and 45°C. Dissolved oxygen concentration in samples ranged between 0.1 and 0.8 mg/L. Sodium chloride level was higher in produced water (OBPW-T, OBPW-NT) and crude oil samples (OBCR) and the value ranged between 157 and 179 mM. On the contrary, the underground injection water sample (OBIW) and pig run solid samples (OBPIG-1 and OBPIG-2) recorded lower salinity (NaCl) levels (1.6–32 mM). Sulfate level was zero in the underground injection water (OBIW) and very low in the rest of samples except pig run samples (OBPIG-1 and OBPIG-2) that also recorded significant sulfide concentration and traces of ferrous ion. The level of organic nutrients (VFA), acetate, butyrate and propionate were zero in all samples except OBPG-1 and OBPG-2 that recorded low concentrations of acetate and propionate as shown in Table 1.

3.2. Microbial counts and activities

Acid producing bacteria (APB) and Sulfate reducing bacterial (SRB) populations were found to be moderate in all samples (10^1–10^4 cells/ml), as shown in Table 2. However, all the samples possess high hNRB activity (43–168 units/day) and low SRB_VS and soNRB activity (>2 units/day). SRB activity with lactate was moderate in all samples (20–43 units/day) except OBIW that recorded less than 2 units/day of activity (Table 2). Overall, the organisms found in samples preferred lactate as an electron acceptor than VFA (acetate, butyrate and propionate). Very low concentrations of VFAs in the original samples seem to support this.

3.3. Corrosion rate measurements under methanogenic conditions

Pig runs solid samples, OBPG-1 and OBPG-2 showed the highest methane production (1.65 ± 0.042 and 1.58 ± 0.028 mmol) and corrosion rates (0.054 ± 0.0042 and 0.061 ± 0.0028 mm/yr.) respectively followed by Obigbo crude (OBCR) with a respective methane production and corrosion rates of 1.48 ± 0.028 mmol and 0.038 ± 0.0028 mm/yr. The respective methane production and corrosion rates of produced waters, OBPW-T and OBPW-NT (1.26 ± 0.039mmol; 0.036 ± 0.0042 mm/yr. and 1.28 ± 0.028 mmol; 0.03 ± 0.0028 mm/yr.) were equally high. Expectedly, the zero sulfate and low saline underground injection water recorded lower methane production (0.65 ± 0.042 mmol) and corrosion rates (0.0018 ± 0.00042 mm/yr.) as shown in Figure 1.

3.4. Assessment of microbial communities in samples

Bacterial taxa dominated in samples OBPW-T, OBPW-NT and OBCR while Archaeal taxa dominated in samples OBPG-1 and OBPG-2. In produced water samples (OBPW-T and OBPW-NT), Betaproteobacteria dominated the microbial community in both samples (33–42%). Other dominant members of microbial community in samples OBPW-T apart from Betaproteobacteria (33%) are *Azovibrio* (21%), *Thauera* (10%), *Sprochaeta* (10%), *Methanolobus* (7%) and *Azospira* (7%). In OBPW-NT, apart from

Table 1. Chemical composition of samples from Obigbo North (unit; mM).

S/N	Sample Code	Sample Description	pH	Site Temp. (O^C)	DO	NaCl	Conduct-ivity (mS/cm)	So_4^{2+}	HS^-	NH_4^+	Fe^{2+}	Ace.	But.	Prop.
1	OBPW-T	Mechanically treated produced water from Obigbo North with oil content below 100 ppm	8.13	40–45	0.80	174	17.94	0.09	**0.02**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
2	OBPW-NT	Untreated produced water from Obigbo North with oil content of about 300 ppm	8.01	35-40	0.20	157	16.23	**0.00**	0.03	0.01	0.00	0.45	**0.00**	**0.00**
3	OBIW	Zero sulfate underground water for injection at Obigbo North facility	7.66	25	0.60	1.60	0.15	0.04	0.02	0.08	0.00	0.00	**0.00**	**0.00**
4	OBCR	Obigbo crude oil for export	8.32	35–40	0.40	179	20.59	0.01	0.02	0.03	0.00	**0.00**	**0.00**	**0.00**
5	OBPIG-1	Pig runs samples from Obigbo pipeline collected from the surface	6.93	35–40	0.10	28	2.86	8.87	3.14	0.00	0.16	0.03	**0.00**	**0.12**
6	OBPIG-2	Pig runs samples from Obigbo pipelines collected at 1 cm from the surface	6.85	35–40	0.10	32	3.38	11.57	2.71	0.00	0.69	0.04	**0.00**	**0.20**

Ace = Acetate; But = Butyrate; Prop = Propionate. DO = Dissolved oxygen (mg/L)

Table 2. Viable bacterial counts (10 x/ml) and activities of Bacteria and Methanogens in samples.

Sample Code	Bacterial cell number (10^x/ml)		Bacterial activity (units/day) with substrate amendment				Methanogen activity* (mmol)
	SRB	APB	SRB_LS	SRB_VS	hNRB	soNRB	
OBPW-T	3	3	45	3.50	110	1.50	0.65
OBPW-NT	2	2	20	3.60	64	1.60	0.70
OBIW	1	3	2.5	2.50	45	1.20	0.40
OBCR	3	3	44	2.50	125	1.30	0.80
OBPG-1	4	4	43	2.40	165	1.80	0.85
OBPG-2	4	4	44	2.60	156	1.60	0.80

SRB = Sulfate reducing bacteria; **APB** = Acid producing bacteria; **SRB_LS** = SRB in lactate media+Sulfate; SRB_VS = SRB in Volatile Fatty Acid media+Sulfate; **hNRB** = Heterotrophic nitrate reducing bacteria; soNRB = Sulfide oxidizing nitrate reducing bacteria
*Methanogen activity is maximum methane produced after 2 weeks of incubation

Betaproteobacteria (42%), *Marinobacter* was also dominant (34%), followed by *Bacteroidetes* (7%) and *Methanolobus* (6%). In Obigbo crude samples (OBCR), microbial community was dominated by *Thauera* (28%), *Methanolobus* (22%), *Bacteroidetes* (18%), *Rhodobactereceae* (7%) and *Pseudomonas* (6%). Surprisingly but interestingly, the microbial communities of pig run samples from crude oil transport pipelines (OBPG-1 and OBPG-2) differed significantly from produced water and crude samples. Detailed phylogenetic classification of pyrosequencing reads of samples are shown in Table 3.

4. Discussion

A recent study by Okoro et al. (2017) showed that Methanogens probably dominated pipeline corrosions in a low sulfate oil environment and that provided some incentives for the present study. It was observed that methanogenic activities in samples from the present study were relatively high when compared with the activities of the SRBs. Several authors have advanced that in the absence of sulfate or nitrate, water mediated fermentation of oil organics to methane and carbon dioxide becomes a dominant metabolic process (Grabowski et al. 2005; Youssef et al. 2009). Pyrosequenced data revealed dominance of betaproteobacteria (33–42%) in the produced water samples (OBPW-T and OBPW-NT) (Table 3 and Figure S1). Specific genera of Betaproteobacteria implicated include *Azovibrio* (21%), *Thauera* (10%) and *Azospira*

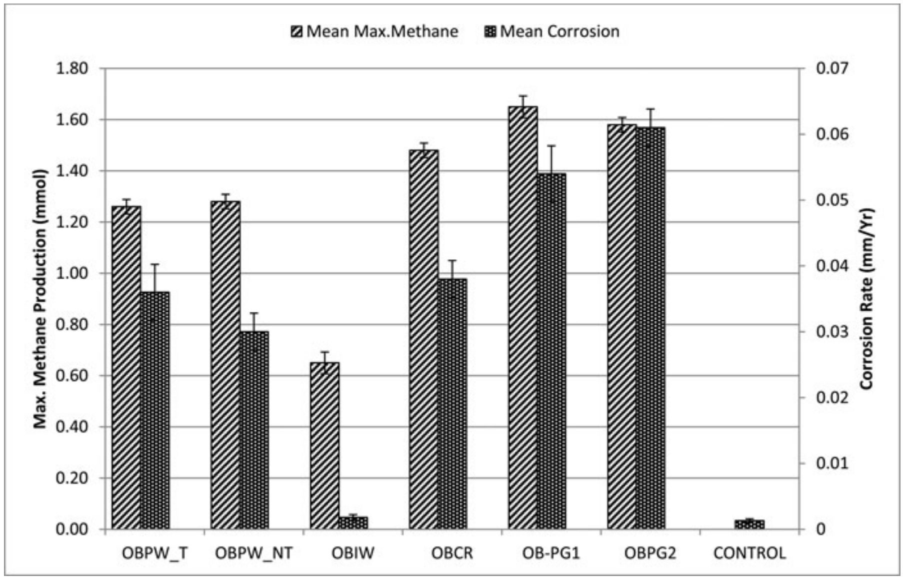

Figure 1. Methane production and corrosion rate of samples after 4 weeks of exposure with metal coupons.

Table 3. Detailed Phylogenetic classification of pyrosequencing reads of samples used in the study.

Serial No of Sample on Tree		**1**	**2**	**3**	**4**	**5**
Type of Sample		**OBPW-T**	**OBPW-NT**	**OBCR**	**OBPG-1**	**OBPG-2**
Sequence Code		**V30_1349**	**V30_1350**	**V30_1352**	**V30_1353**	**V30_1354**
Total Reads		**1579**	**1732**	**946**	**1851**	**1906**
Number of OTU'S		**117**	**86**	**99**	**129**	**115**
Number of Taxa		**72**	**39**	**62**	**73**	**69**
Estimated OTU's (Chao)		**223**	**121**	**237**	**293**	**189**
Normalized Shanon Index		**3.26**	**2.72**	**3.21**	**2.35**	**2.32**
Accession Numbers		**SRR1508449**	**SRR1508450**	**SRR1508452**	**SRR1508453**	**SRR1508454**
Bacterial taxa (%)		**93.4**	**94.5**	**86**	**22**	**22.3**
Archaeal taxa (%)		**6.60**	**5.50**	**14**	**78**	**77.70**
Entries: Taxon: Phylum; Class; Order; Family; Genus						
1	Archaea; Euryarchaeota; Methanobacteria; Methanobacteriales; Methanobacteriaceae; Methanobacterium	**0.26**	**0.00**	**0.00**	**22.70**	**22.40**
2	Archaea; Euryarchaeota; Methanomicrobia; Methanomicrobiales; Methanocalculus	**0.00**	**0.00**	**0.00**	0.49	0.74
3	Archaea; Euryarchaeota; Methanomicrobia; Methanosarcinales	**0.56**	**0.00**	**0.00**	**2.00**	**1.63**
4	Archaea; Euryarchaeota; Methanomicrobia; Methanosarcinales; Methanosarcinaceae; Methanolobus	**5.38**	**5.49**	**13.95**	**51.70**	**52.00**
5	Bacteria	0.95	0.40	**1.06**	0.22	0.16
6	Bacteria; Bacteroidetes	0.13	**6.35**	0.95	0.50	**0.00**
7	Bacteria; Bacteroidetes; Bacteroidia; Bacteroidales; Marinilabiaceae; Anaerophaga	0.18	**2.54**	0.00	0.50	**0.50**
8	Bacteria; Bacteroidetes; Sphingobacteria; Sphingobacteriales; WCHB1-69	0.95	**0.00**	**2.11**	**0.22**	**0.00**
9	Bacteria; Bacteroidetes; VC2.1	**1.46**	**0.00**	**11.10**	**0.00**	**0.00**
10	Bacteria; Candidate_division_WS6	0.41	**0.26**	0.21	0.54	**1.63**
11	Bacteria; Chloroflexi; Anaerolineae; Anaerolineales; Anaerolineaceae; uncultured	0.13	**0.00**	0.12	4.32	**2.20**
12	Bacteria; Deferribacteres; Deferribacteres; Deferribacterales; Deferribacteraceae; Calditerrivibrio	0.06	**3.23**	0.11	0.00	**0.05**
13	Bacteria; Firmicutes	**3.48**	**0.12**	0.32	0.00	**0.00**
14	Bacteria; Firmicutes; Clostridia; Clostridiales; Eubacteriaceae; Acetobacterium	0.83	**0.00**	**1.37**	0.76	**1.01**
15	Bacteria; Firmicutes; Clostridia; Clostridiales; Family_XI_Incertae_Sedis; Sedimentibacter	0.63	**0.23**	**1.27**	0.16	0.37
16	Bacteria; Firmicutes; Clostridia; Clostridiales; Family_XI_Incertae_Sedis; Tissierella	**1.58**	**0.16**	**1.37**	0.32	**0.42**
17	Bacteria; Firmicutes; Clostridia; Clostridiales; Lachnospiraceae; uncultured	0.38	**0.00**	**1.16**	**0.00**	**0.00**
18	Bacteria; Firmicutes; Mollicutes; Acholeplasmatales; Acholeplasmataceae; Acholeplasma	**1.90**	**0.00**	0.74	**0.00**	**0.00**
19	Bacteria; Proteobacteria	0.51	**1.32**	**8.03**	0.54	0.42

20	Bacteria; Proteobacteria; Alphaproteobacteria; Rhizobiales; Hyphomicrobiaceae; Xanthobacter	**0.00**	**0.00**	0.00	0.97	**2.62**
21	Bacteria; Proteobacteria; Alphaproteobacteria; Rhodobacterales; Rhodobacteraceae	**0.00**	**0.00**	**4.44**	0.05	0.26
22	Bacteria; Proteobacteria; Alphaproteobacteria; Rhodospirillales; Oleomonas	**0.00**	**0.00**	**0.00**	0.65	**1.36**
23	Bacteria; Proteobacteria; Alphaproteobacteria; Rhodospirillales; Rhodospirillaceae; Magnetospirillum	**0.23**	**0.00**	**1.16**	**0.00**	0.50
24	Bacteria; Proteobacteria; Alphaproteobacteria; Rhodospirillales; Rhodospirillaceae; Rhodospirillum	0.38	0.28	**1.80**	0.00	**0.00**
25	Bacteria; Proteobacteria; Betaproteobacteria	**25.97**	**38.45**	**8.56**	0.05	**0.10**
26	Bacteria; Proteobacteria; Betaproteobacteria; Burkholderiales; Alcaligenaceae	**1.77**	0.00	**0.00**	**0.70**	0.73
27	Bacteria; Proteobacteria; Betaproteobacteria; Burkholderiales; Comamonadaceae	**1.14**	0.32	**0.00**	**0.00**	0.50
28	Bacteria; Proteobacteria; Betaproteobacteria; Burkholderiales; Comamonadaceae; Brachymonas	**2.98**	0.00	**0.00**	**0.50**	**0.05**
29	Bacteria; Proteobacteria; Betaproteobacteria; Hydrogenophilales; Hydrogenophilaceae	0.463	**1.039**	**0.11**	**0.00**	**0.00**
30	Bacteria; Proteobacteria; Betaproteobacteria; Hydrogenophilales; Hydrogenophilaceae; Petrobacter	0.53	**1.386**	**0.50**	**0.50**	**0.00**
31	Bacteria; Proteobacteria; Betaproteobacteria; Rhodocyclales; Rhodocyclaceae; Azospira	**5.82**	0.00	**1.59**	**0.00**	**0.00**
32	Bacteria; Proteobacteria; Betaproteobacteria; Rhodocyclales; Rhodocyclaceae; Azovibrio	**16.78**	0.33	0.63	0.05	**0.16**
33	Bacteria; Proteobacteria; Betaproteobacteria; Rhodocyclales; Rhodocyclaceae; Thauera	**8.23**	0.92	**17.34**	**3.01**	**3.67**
34	Bacteria; Proteobacteria; Deltaproteobacteria	0.00	**2.14**	0.00	**0.00**	**0.00**
35	Bacteria; Proteobacteria; Gammaproteobacteria	**1.33**	0.12	0.95	**0.12**	**0.11**
36	Bacteria; Proteobacteria; Gammaproteobacteria; Alteromonadales; Alteromonadaceae; Marinobacter	0.00	**31.01**	0.50	**0.16**	**0.21**
37	Bacteria; Proteobacteria; Gammaproteobacteria; Oceanospirillales; Oceanospirillaceae; Pseudospirillum; OM182	0.25	**1.16**	**0.00**	**0.50**	**0.00**
38	Bacteria; Proteobacteria; Gammaproteobacteria; Pseudomonadales; Pseudomonadaceae; Pseudomonas	0.32	**0.00**	**3.70**	0.38	0.37
39	Bacteria; Spirochaetes; Spirochaetes	**1.20**	**0.23**	**1.90**	0.11	**0.05**
40	Bacteria; Spirochaetes; Spirochaetes; Spirochaetales; Spirochaetaceae; Spirochaeta	**6.20**	**0.21**	**1.01**	0.05	**0.00**
41	Bacteria; Thermotogae; Thermotogae; Thermotogales; Thermotogaceae; Kosmotoga	0.25	**0.00**	0.50	**2.60**	**0.53**
42	Classified trace bacteria	3.76	2.22	2.58	4.39	4.78
	TOTAL (%)	**96.88**	**99.66**	**96.75**	**97.82**	**97.90**

The number of good sequence reads for each sample (Total reads), the total number of OTU's (95% sequence identity) and the number of taxa that these represent are indicated. The estimated maximum number of OTU's (Chao), The Shannon index (normalized for the same number of reads) and the accession numbers of samples are also listed. The fraction of reads (%) are indicated in bold for all taxa which have at least one fraction in excess of 1%.

(7%). *Azovibrio* and *Azospira* which belong to the family Rhodobactereceae and are both nitrate reducers and also fix nitrogen. *Azospira* posseses nitrogenase activity and have been implicated in perchlorate reduction in contaminated soils and surface waters (Hutchinson 2013). *Thauera* on the other hand is a known oil degrading bacteria and has been implicated in anaerobic degradation of aromatic hydrocarbons (Widdel and Rabin 2001; Harayama et al. 1999). Another dominant microbial community in produced water samples is *Marinobacter* (37%). Though an aerobic organism, *Marinobacter* is suspected to be indigenous to oil bearing systems (Liebensteiner et al. 2014) and has frequently been implicated with petroleum hydrocarbon degradation in hypersaline environments (Mcgenity et al. 2012). *Marinobacter* has also been implicated in nitrate reduction (Gao et al. 2015). Sufficient concentration of dissolved oxygen in produced water samples (1.2 mg/L) may have been responsible for the survival and activity of *Marinobacter* in an anaerobic environment. Other less dominant microorganisms found in produced water such as *Calditerrivibrio* (4%) and *Petrobacter* (2%) are active nitrate reducers (Youssef et al. 2009) *Methanolobus* (6-7%) is the only methanogenic archeae in produced waters. *Methanolobus* is potentially corrosive and its metabolization of methyl amines and dimethyl sulfides leads to concurrent production of methane which increases reservoir pressure and decreases oil viscosity (Gao et al. 2015). *Pseudomonas* and *Bacteroidetes* are common with oil environments and they are potential hydrocarbon degraders (Luo et al. 2014; Peng et al. 2015). *Acetobacterium* also present in crude oil sample are known as acetogens because they produce acetic acid as a bi-product of anaerobic metabolism. They also oxidize hydrogen and reduce carbon dioxide to acetic acid (Balch et al. 1977). It was observed that the microbial community composition of pipeline samples (OBPG-1 and OBPG-2) differed considerably from those of produced waters and oil (OBPW-T, OBPW-NT and OBCR), while bacteria taxa dominated microbial community composition of produced water and oil samples, archaea taxa dominated those of pipeline samples. The dominant archaeal taxa were *Mathanolobus* (60%) and *Methanobacterium* (25–27%). While the methylotrophic *Methanolobus* were found in all samples, hydrogenotrophic *Methanobacterium* were found only in the pipeline samples where they are suspected to play active role in metal corrosion via cathodic depolarization of metals. Dominance of *Methanolobus* and *Methanobacterium* in the oil pipeline samples is of interest to us because both organisms are potentially corrosive. *Methanolobus* is methylotrophic because they utilize methyl compounds as substrates while *Methanobacterium* is hydrogenotrophic and can use hydrogen on steel surfaces for their metabolism thereby accelerating steel corrosion (Dinh et al. 2004).

5. Conclusion

The results obtained from the present study indicate that oil degrading and nitrate reducing bacteria such as *Marinobacter* (37%), *Azovibrio* (21%), *Thauera* (10-28%) and *Methanolobus* (22%) clearly dominate the microbial community of oil and water samples while *Methanolobus* (60%) and *Methanobacterium* (25-27%) dominate the pipeline pig-run samples. This suggests that corrosive methanogens may have a significant role to play in a low sulfate oil producing facility where the activities of the SRBs are negligible.

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