



Evaluation of the Bacteriological Diversity and the Physicochemical Properties of Kolo-Creek Oil Field Wastewater

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Authors' contributions

This work was carried out in collaboration between both authors. Author WSA designed the study and wrote the protocol and first draft of the manuscript. Author ON performed the statistical analysis managed the analyses of the study and the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Evaluation of bacteriological diversity and physicochemical parameters as well as the heavy metals present in the water samples collected from Kolo creek flow station located in Bayelsa State in the Niger Delta, Nigeria was carried out for the period of eight months (March to October). Oilfield wastewater samples were obtained in four different locations of the Kolo creek in sterile bottles and were transported to the Microbiology Laboratory of the Rivers State University for analysis. 10-fold serial dilutions were carried out and aliquots of 0.1 ml were inoculated and spread plated onto sterile nutrient agar, MacConkey agar and mineral salt agar plates. The water samples were analyzed for the total heterotrophic bacteria, coliform bacteria and hydrocarbon utilizing bacteria. The total heterotrophic and coliform bacteria load in log₁₀CFU/ml ranged from 6.25 to 13.62 and 4.04 to 5.75 respectively, while the hydrocarbon utilizing bacteria ranged from 0 to 4.81. Higher Mean monthly counts of the total heterotrophic bacteria, coliform bacteria and hydrocarbon utilizing bacteria were 13.48 in March, 5.44 in April and 4.51 log₁₀ CFU/ml in March, respectively. While lower mean monthly counts recorded were 6.72 in August, 4.29 in August and 0 log₁₀CFU/ml in April,

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respectively. Fifteen bacteria belonging to *Micrococcus*, *Chromobacterium*, *Listeria*, *Actinomyces*, *Staphylococcus*, *Bacillus*, *Enterobacter*, *Serratia*, *Escherichia coli*, *Vibrio cholera*, *Vibrio parahaemolyticus*, *Proteus*, *Shigella*, *Pseudomonas* and *Acinetobacter* species were identified. While *Bacillus*, *Pseudomonas*, *Serratia*, *Micrococcus*, *Proteus*, and *Acinetobacter* species were the hydrocarbon utilizing bacteria identified. The bacteria populations varied across the months. The physicochemical parameters as well as the heavy metals were within the permissible limits set by regulatory bodies. The hydrocarbon utilizing bacteria in this study could be used for bioremediation study.

Keywords: Oilfield wastewater; bacteriological diversity; physicochemical parameters; heavy metals.

1. INTRODUCTION

The Petroleum industry in Nigeria is a complex combination of independent operations including exploration and production operations, the processing of crude oil into consumer products, transportation and marketing activities. At each stage of operation, gaseous, liquid and solid waste materials are produced and discharged. These can adversely affect the air, water, and soil environment if not properly discharged and controlled [1].

Oilfield wastewater or produced water is usually very salty and may contain suspended and dissolved solids, residual hydrocarbons, numerous organic species, heavy metals, naturally occurring radioactive and chemicals used in hydrocarbon extraction [2,3]. Several studies investigated the characteristics of produced water and its impact on the surrounding environment. Neff et al. [4] described produced water for ocean discharge as containing up to 48 ppm of oil. Besides, most produced waters are more saline than seawater [4,5]. Produced waters may contain concentrations of chloride 150 to 180 g/L (sea water contains an average of 35 g/L) [6]. With these levels of salts the water becomes toxic for many forms of life [6]. Produced waters may also contain chemical additives used in drilling and producing operations [7] and in the oil/water separation process. These chemicals can affect the oil/water partition coefficient, toxicity, bioavailability, and biodegradability [5]. The negative effect of produced waters on the Indonesian environment was confirmed by Smith et al. [8]. According to Oboh et al. [9] the discharged of oilfield produced water had high metal ions and total hydrocarbon concentrations.

Oil field wastes water is discharged into the sea after it has been separated from oil drawn from the reservoir [10,11].

In recent years studies have been carried out on the effect of oilfield wastewater on aquatic environments [12,13]. Due to marked increase in offshore oil operations, the studies in these areas have not been widespread especially studies from onshore field locations (Land Rigs). Therefore there is need to assess the constituents of both physicochemical and microbiology oil field wastewater from Land Rigs as well as Swamps or Offshore Rigs.

2. MATERIALS AND METHODS

2.1 Collection of Oilfield Wastewater Samples

Oilfield wastewater were collected from the point of discharge at Kolo creek flow station an onshore oil production platform located in Bayelsa State, Nigeria (Fig. 1). The oilfield wastewater samples were collected using 4 Litre capacity plastic bottles. Prior to the collection of the oilfield wastewater the interior of the nozzle of the outlet biofilter was flushed for few minutes before collecting directly into the 4 litre plastic bottles. The plastic bottles were appropriately labeled and stored in an ice packed cooler. The stored samples were immediately transported to the laboratory within 24 hours for processing and analyses. Samples were collected monthly for a period of eight months (March to October, 2018).

2.2 Microbiological Analysis of the Oilfield Wastewater

Samples for Microbiological analysis were transported to the laboratory immediately after collection in Ice packed coolers. Total heterotrophic bacterial (THB) count, coliform bacteria, hydrocarbon utilizing bacteria (HUB) of oilfield wastewater were analyzed using standard laboratory procedure and methods described by Wemedo et al. [14].

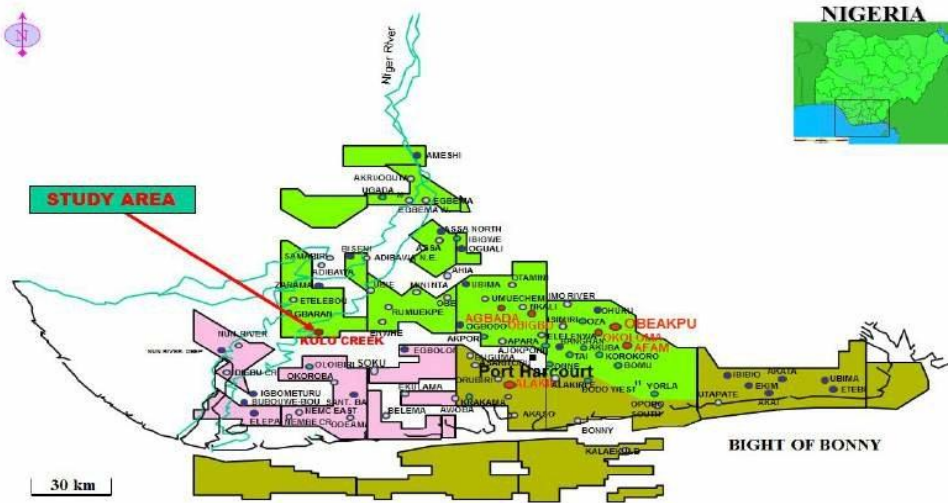


Fig. 1. Map showing the study area

2.3 Physicochemical Analysis of Oilfield Wastewater Samples

Physicochemical analyses of the oilfield wastewater samples were conducted according to standard procedures of APHA [15] and ASTM [16]. The physicochemical parameters determined include pH, temperature, turbidity, total dissolved solids (TDS), total suspended solids (TSS), salinity, conductivity, biological oxygen demand (BOD₅), chemical oxygen demand (COD), total hydrocarbon content, odour and heavy metals such as lead, zinc, total iron, chromium, mercury, arsenic, copper, and cadmium.

3. RESULTS

The monthly counts and means of the total bacteria count of the oilfield wastewater are presented in Table 1. The monthly count in all the sources ranged from 6.25 – 13.62 (log₁₀CFU/ml); mean monthly counts ranged between 6.72 and 13.48 (log₁₀CFU/ml), being the

lowest counts in August and the highest counts recorded in March, respectively.

The total coliform bacteria count as shown in Table 2 ranged from 4.04 to 5.75 (log₁₀CFU/ml). Mean monthly counts ranged between 4.29 and 5.44 (log₁₀CFU/ml), being the lowest counts in August and highest counts recorded in April respectively.

Table 3 shows the total hydrocarbon utilizing bacteria count which ranged from 0 to 4.81 (log₁₀CFU/ml). While the mean monthly count ranged between 0 and 4.51(log₁₀CFU/ml) been the lowest counts in April and the highest counts recorded in March respectively.

The result of the mean values of physicochemical constituents of the oilfield wastewater in the various pits (1A to 1D) is as shown in Table 4.

The result of the mean values of the heavy metals of the oilfield wastewater in the various pits (1A to 1D) is as shown in Table 5.

Table 1. The total heterotrophic bacteria count (log₁₀CFU/ml) of the Kolo creek flow station for the eight months duration

Sampling points	Months							
	Mar	April	May	Jun	July	Aug	Sep	Oct
A	13.62	7.27	6.60	6.54	6.83	6.56	6.76	6.72
B	13.60	7.34	7.34	6.79	6.76	6.25	6.76	6.78
C	13.38	7.30	6.94	6.96	6.78	7.09	6.65	6.85
D	13.31	6.99	6.91	7.29	6.95	6.97	6.83	6.68
Mean	13.48	7.23	6.95	6.90	6.83	6.72	6.75	6.76

Table 2. The total coliform bacteria population (\log_{10} CFU/ml) of the Kolo creek flow station for the eight months duration

Sampling points	Months							
	Mar	April	May	Jun	July	Aug	Sep	Oct
A	5.31	5.46	4.95	5.05	4.79	4.04	4.75	4.76
B	5.39	5.42	5.47	4.96	4.80	4.26	4.60	4.52
C	5.45	5.44	5.08	4.30	5.75	4.46	4.65	4.81
D	5.41	5.45	4.88	4.60	4.89	4.40	4.49	4.58
Mean	5.39	5.44	5.10	4.73	5.06	4.29	4.62	4.67

Table 3. The total hydrocarbon utilizing bacteria population (\log_{10} CFU/ml) of the Kolo creek flow station for the eight months duration

Sampling points	Months							
	Mar	April	May	Jun	July	Aug	Sep	Oct
A	4.81	0	3.48	4.20	4.20	4.23	3.85	4
B	4.15	0	4.15	3.90	3.95	4.15	3.60	4.08
C	4.73	0	3.60	3.60	3.90	4.53	3.60	3.78
D	4.36	0	3.95	4	4.04	4.60	3.95	3.78
Mean	4.51		3.80	3.93	4.02	4.38	3.75	3.91

4. DISCUSSION

The bacteriological diversity and the physicochemical properties of Kolo-creek flow station oil field wastewater were evaluated. The bacterial densities were very high and fluctuated in the various months. The total heterotrophic bacterial load obtained in this study across the samples was generally high in all the samples. Mean counts of viable heterotrophic bacteria were highest in the month of March and very low in the month of August. Also, the total coliform bacteria were highest in the month of April for all the samples and lowest in the month of August. The hydrocarbon-utilizing bacteria load fluctuated between samples as well as in the different months. It was observed that the hydrocarbon utilizing bacteria were high in some samples and low or even zero in other samples, showing a wide range between the lowest value and the highest value. The high microbial load observed in this study could be influenced by physicochemical parameters of the environment as well as other activities taking place. Statistically, there was significant difference between the monthly counts of hydrocarbon-utilizing bacteria. Wemedo et al. [14] reported that there was no significant difference between the hydrocarbon utilizing bacteria observed between the months of study. The presence of hydrocarbon utilizing bacteria suggests that the Kolo creek oil field contains some level of hydrocarbon residues which could have

supported their growth and multiplication. Thus, the presence of hydrocarbon utilizers in the Kolo creek could enhance the removal of hydrocarbon pollutant from the water. This agreed with findings from previous studies [13,14].

The bacteria genera identified in this study include *Micrococcus*, *Chromobacterium*, *Listeria*, *Actinomyces*, *Staphylococcus*, *Bacillus*, *Enterobacter*, *Serratia*, *Escherichia coli*, *Vibrio cholera*, *Vibrio parahaemolyticus*, *Proteus*, *Shigella*, *Pseudomonas* and *Acinetobacter* species. The bacteria genera were greatly influenced by the months as well as the sample locations as instability of isolates were observed across the months. Findings in this study agreed with that of Wemedo et al. [14] who reported that the bacteria densities fluctuated on monthly basis. Also, these microorganisms identified in this study could suggest that they are normal flora of the water or may have been introduced into the water body. For instance, *Escherichia coli* has been reported to be an indicator of contamination arising from faecal materials as it is a normal flora of the gut [17]. The hydrocarbon utilizing bacteria identified in this study were *Bacillus*, *Pseudomonas*, *Serratia*, *Micrococcus*, *Proteus*, and *Acinetobacter* species. Similar organisms have been isolated from previous studies [18,19] who isolated *Pseudomonas*, *Bacillus*, *Micrococcus* and other bacterial strains from wastewater and sediments.

Table 4. Mean values physiochemistry – Kolo creek from march to october for pits 1a to 1d

Parameters	March to October								Dpr limits	FMEnv LIMITS
	March	April	May	June	July	August	September	October		
pH	6.78	6.8	7.05	7.23	7.4	7.3	7.1	7.3	6.5-8.5	6.0- 9.0
Temperature, °C	24.18	24.6	24	23.98	24	23.8	23.8	24.1	25	20-33
Turbidity, NTU	2	3	6.5	5.3	5.3	5.8	7	3.8	10	-
Odour	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable	Unobjectionable
TDS, mg/l	131.75	194.3	83.5	90.25	137.75	644.25	690.75	30.5	2,000	-
TSS, mg/l	18.25	10.5	19	18.8	15.5	16.25	17.5	19	30	NS
BOD, mg/l	1.25	2	1.5	2	2	2	2.25	1.75	10	4
COD, mg/l	5.75	3.3	5.25	6	5	5.25	5.5	5.5	10	-
THC, mg/l	0.78	1.2	1.5	1.25	1.5	1.8	1.3	1.8	10	NS
Salinity, mg/l	100.5	165.8	137.75	177.5	204	162	147.8	4.75	600	-
Conductivity, µS/cm	148.5	310.8	561	513.8	199	334.75	303.75	65	-	-

Table 5. Heavy metals - mean values - Kolo creek flow station

Parameters	March to October								DPR limits	FMEnv LIMITS
	March	April	May	June	July	August	September	October		
Lead, mg/l	0.01	0.02	0.03	0.02	0.02	0.01	0.02	0.03	0.05	0.017 (1.7µg/l)
Zinc, mg/l	0.02	0.03	0.03	0.02	0.03	0.03	0.03	0.02	1	0.03
Copper, mg/l	0.03	0.02	0.01	0.01	0.01	0.01	0.36	0.33	1.5	0.02-0.04 (2-4µg/l)
Total Iron, mg/l	0.02	0.02	0.11	0.15	0.18	0.42	0.9	1.9	1	1
Chromium, mg/l	0.02	0.01	0.02	0.02	0.01	0.01	0.01	0.13	0.03	0.02-2.0
Cadmium, mg/l	0.02	0.02	0.02	0.02	0.01	0.01	0.02	0.04	-	-
Mercury, mg/l	0.02	0.04	0.06	0.01	0.001	0.01	0.18	0.14	-	0.01 (1.0µg/l)
Arsenic, mg/l	0.04	0.01	0.06	0.04	0.13	0.01	0.03	0.11	-	0.5

The result of the physicochemical parameters of the Kolo creek flow station oilfield wastewater showed that all the physicochemical parameters are within the admissible limits set by DPR despite the fluctuations across the months. The pH varied between slightly acidic to slightly alkaline. The pH values recorded in this study is tolerable for the proliferation of bacteria. Thus, this could be the reason why there are diversities of bacteria genera including the hydrocarbon utilizing bacteria. Bartha and Atlas [20] reported that an alkaline pH enhances biodegradation of crude oil by bacteria utilizers. Also, the pH of water is important because many biological activities can occur only within a narrow range, thus any variations beyond an acceptable limit could be fatal to a particular organism. Similarly, the temperature varied slightly across the months and supports the growth of varieties of bacteria including the hydrocarbon utilizing bacteria. This is in agreement with previous studies which have reported that the favourable temperature for the growth of bacteria as well as hydrocarbon utilizing bacteria in marine environments is between the range of 15-30°C [21]. Temperature is one of the most important ecological and physical factors which has a profound influence on both the living and non-living components of the environment, thereby affecting organisms and the functioning of an ecosystem.

Turbidity is defined as the measure of the clarity or cloudiness of water and the values are attained by measuring the scattering and absorbing effect that suspended particles have on light [22]. Turbidity values of the Kolo creek from March to October ranged from 2.0 NTU to 7.0, whereas the limit set by DPR is 10 NTU. Thus, the turbidity is within admissible limits and may suggest that the Kolo creek is not very contaminated and was not to a greater degree influenced by run off during the rainy period.

Similarly, the result for the heavy metal analysis showed that they were all within the permissible limits and at such do not pose serious danger to the public.

5. CONCLUSION

In this study, the viable bacteria as well as the coliform load is high. Though the physic-chemical parameters as well as the heavy metal analysis are within the permissible limits for both DPR (department of petroleum resources) and Federal ministry of environment in the Kolo-creek, the viable bacteria identified in this study could

contain pathogenic strains which could result to infections especially of the gastrointestinal tract. Furthermore, the presence of hydrocarbon utilizing bacteria in the Kolo creek could enhance the elimination or complete degradation of notable oil in the water.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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