

Produced Water Treatment Using Bacteria and Microalgae Consortia

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Abstract- Due to the many processes involved in oil and gas production, Produced Water is usually given off as a major by-product. This wastewater constitutes of contaminants like dissolved salts, hydrocarbons, organic matter and other toxins which pose significant risks to the environment if discharged without the necessary treatments. This study analyzed the treatment potential of hydrocarbon-degrading bacteria and microalgae species, utilizing Produced water gotten from the Obaji Oil field, Rivers State, Nigeria. A total of five microalgae species -*Chlorella vulgaris*, *Scenedesmus obliquus*, *Arthrospira platensis*, *Monoraphidium* sp. and *Neochloris oleoabundans* -were screened and acclimatized, while bacterial isolates were obtained from rhizosphere soil and identified as *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Acinetobacter calcoaceticus*, *Rhodococcus erythropolis* and *Sphingomonas paucimobilis*. The treatments were carried out in batch reactor systems at 50%, 60%, 75% and 100% produced water concentrations for seven days. Notable changes in temperature, pH, Total Dissolved Solids (TDS), Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Hydrocarbon Content (THC) and Dissolved Oxygen (DO) were observed. Microalgae treatment reduced the major pollutants and produced biomass, while bacterial treatment gave noticeable reductions in TDS, BOD and THC. At 100% produced water concentration, bacterial treatment achieved 12.7%, 16.8%, 23.7% and 14.6% reductions in TDS, COD, BOD and THC respectively. At the same concentration, microalgal treatment reduced TDS by 11.9%, COD by 24.7%, BOD by 31.4% and THC by 22.8%. Comparing the given results with Nigerian Upstream Petroleum Regulatory Commission (NUPRC) Onshore Discharge standards showed that the treated water still surpassed the limits for major parameters. The study indicated that biological treatment alone, of the produced water sample, was inadequate for direct discharge into the environment. The findings from this study demonstrated the viability of microbial treatment and aids further development of combined bacteria-microalgae systems for produced water remediation.

Keywords- Produced water, bioremediation, hydrocarbons, wastewater.

I. INTRODUCTION

Produced water, a by-product of crude oil extraction, has three primary sources in the oil and gas industry, namely; Formation water, Injection water and Condensed water. The constituents of produced water vary with reservoir characteristics, production practices and the age of the producing field. The wastewater may contain dissolved and dissipated hydrocarbons, salts, phenolic compounds, organic acids, benzene, toluene, ethylbenzene and xylene (BTEX), heavy metals and production chemicals. The salinity is usually high because of the complex contaminants and this limits the efficiency of biological treatment systems. High salinity imposes osmotic shock on microorganisms and reduces metabolic and photosynthetic activity (Khosravi & Alamdari, 2009; Lusnier et al., 2019). Discharge of untreated wastewater

into the environment is of significant concern (Beyer et al., 2020; Cozzarelli et al., 2017)

Traditionally, produced water treatment involves physical separation, filtration, floatation, adsorption, chemical precipitation, coagulation, oxidation and membrane processes. These methods target specific contaminants and their efficiency depends on the level of contaminants in the produced water. Physical and Chemical methods can be utilized but may involve large capital or operating costs, energy requirements, chemical consumption and secondary waste generation, especially when the salinity is high (Ahmadun et al., 2009; Al-Ghouti et al., 2019; Fakhru'l-Razi et al., 2009; Igunnu & Chen, 2014; Lusnier et al., 2019).

Biological treatment offers a lower-energy alternative because microorganisms can effectively assimilate organic compounds and nutrients. Microalgae are particularly harnessed because they can assimilate nutrients, biosorb some metals, contribute to organic-pollutant removal and produce biomass. Bacteria directly degrade a wide range of hydrocarbons and other organic compounds. However, the efficacy of such treatment systems depends on the composition of the produced water and the ability of the selected microorganisms to acclimatize to its salinity and contaminant load (Chan et al., 2022).

The present study analyzed the performance of bacterial and microalgal treatment systems as components of a microbial-enhanced produced water strategy.

The study aimed to identify suitable bacterial isolates and microalgal species, assess treatment of produced water at different concentrations, estimate microalgal biomass production and compare the performance of the two biological systems at the same produced water concentrations.

II. MATERIALS AND METHODS

A. Produced Water Sources and Experimental Design

The sample of produced water used for the experiment was collected from the Obaji Flow station in ONELGA, Rivers State, Nigeria. The experiments were carried out in batch reactor systems. Treatment concentrations were 50%, 60%, 75%, and 100% produced water. Table I shows the treatment variations where a constant 10ml of microbial inoculum was combined with different volumes of Produced Water and Milli-Q water to obtain a total volume of 100ml.

Table I: Treatment Variation

Treatment	Inoculum (mL)	Milli-Q water (mL)	Produced water (mL)	Total (mL)
50%	10	45	45	100
60%	10	36	54	100
75%	10	22.5	67.5	100
100%	10	0	90	100

B. Screening and Cultivation of Microalgae

Microalgae gotten from freshwater sources were screened in two stages. Initial screening was based on growth performance; final screening under produced-water conditions. Five microalgal species were selected: *Chlorella vulgaris*,

Scenedesmus obliquus, *Arthrospira platensis*, *Monoraphidium* sp. and *Neochloris oleoabundans*. Cultures were maintained using BG-11/F/2-based media. The cultivation conditions employed were 25°C and a 16 h light/8h dark cycle. The growth of the species was monitored through biomass concentration and optical density observations. Table II indicates the capabilities of the selected species.

Table II. Identified Microalgae species

Species	Key abilities
<i>Chlorella vulgaris</i>	Nutrient uptake; high growth rate
<i>Scenedesmus obliquus</i>	Heavy-metal adsorption; tolerance to varying conditions
<i>Arthrospira platensis</i>	Biomass production; high lipid potential
<i>Monoraphidium</i> sp.	CO ₂ absorption and nutrient uptake
<i>Neochloris oleoabundans</i>	Organic-pollutant and nutrient uptake

C. Bacteria Isolation and Identification

One gram of rhizosphere soil was suspended in 9 mL sterile 0.85% NaCl solution and diluted stepwise. A 0.1 mL sample was smeared on nutrient agar and incubated at 37°C for 24 hours. Isolated colonies were subcultured to obtain pure isolates. Gram staining and biochemical tests, including catalase and oxidase reactions, were used for initial identification. The isolates were identified to be: *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Acinetobacter calcoaceticus*, *Rhodococcus erythropolis* and *Sphingomonas paucimobilis*. The calculated Total Heterotrophic Bacterial abundance was 7.4×10^7 CFU/g.

For bacterial treatments, isolates were incubated at 30°C for seven days. The extensive experimental system used an Innova 44 Incubator Shaker at 120rpm to grow cultures. Several water-quality monitoring parameters were measured during the treatment period. Temperature, TDS, and pH were determined using portable meters; COD and BOD were measured with standard procedures; DO was calculated electrometrically (Winkler procedures) and heavy metals were measured by Atomic Absorption Spectrophotometry after acid digestion.

The major performance parameters were changes in TDS, COD, BOD and THC. Removal percentage was calculated as:

$$\text{Removal \%} = \frac{C_o - C_t}{C_o} \times 100 \quad (1)$$

C_0
where: C_0 is the initial concentration and C_t is the concentration after treatment.

III. RESULTS AND DISCUSSION

A. Microalgal Growth

The biomass densities of *Chlorella vulgaris*, *Scenedesmus obliquus*, *Arthrospira platensis*, *Monoraphidium sp.* and *Neochloris oleoabundans* in a standard BG-11 medium was recorded to be: 0.97, 0.76, 0.60, 0.60 and 0.42 g/L, respectively. At 100% produced water concentration, microalgal species showed different growth capabilities; *Chlorella vulgaris* stabilized at 0.40 g/L, *Scenedesmus obliquus* reached 0.12 g/L, *Neochloris oleoabundans* climaxed at approximately 0.12 g/L and declined afterward. *Arthrospira platensis* achieved approximately 0.64 g/L while *Monoraphidium sp.* showed no calculable growth as shown in Figure I. The results showed that the species expressed differences in resistance to the salinity and contaminant conditions of the produced water.

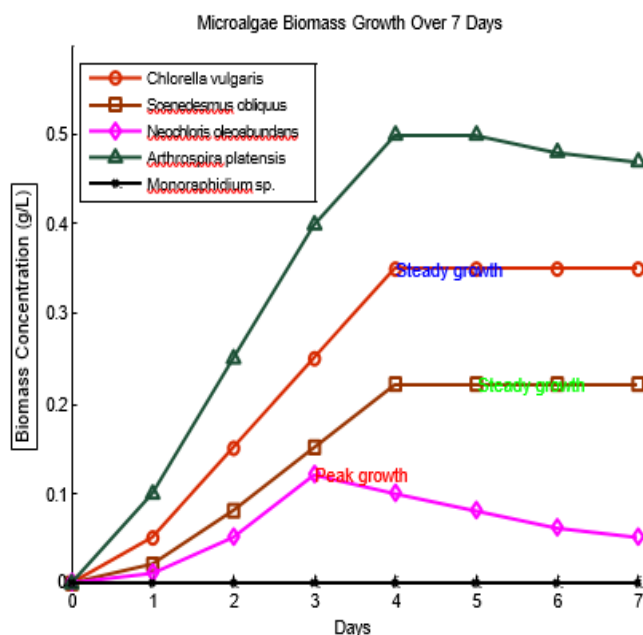


Fig I. Growth rate in 100% produced water

B. Bacterial Treatment Performance

Table 2 displayed that there were constant reductions across all tested concentrations after bacterial treatment. At 50% produced water, there was a 17.1% reduction in TDS, 16.8% reduction in COD and a 25.0% reduction in BOD. After

treatment at 100% concentration, TDS showed a 12.7% reduction, COD reduced by 16.8% BOD by 23.7% and THC by 14.6%. DO increased considerably at 100% concentration from 3.5 to 4.5mg/L due to a lesser demand for oxygen as biodegradable organic matter was consumed. The final value was below the discharge standard.

Table III: Results after bacterial treatment

Produced-water concentration	Parameter	Day 0	Day 7	Removal (%)
50%	TDS (g/L)	36.8	30.5	17.1
50%	BOD (mg/L)	32	24	25.0
50%	COD (mg/L)	950	790	16.8
50%	THC (mg/L)	9.2	7.2	21.7
60%	TDS (g/L)	40.0	34.0	15.0
60%	BOD (mg/L)	34	25	26.5
60%	COD (mg/L)	1050	860	18.1
60%	THC (mg/L)	10.5	8.8	16.2
75%	TDS (g/L)	45.8	39.0	14.8
75%	BOD (mg/L)	36	27	25.0
75%	COD (mg/L)	1150	940	18.3
75%	THC (mg/L)	11.8	10.0	15.3
100%	TDS (g/L)	50.2	43.8	12.7
100%	BOD (mg/L)	38	29	23.7
100%	COD (mg/L)	1250	1040	16.8
100%	THC (mg/L)	13.0	11.1	14.6

C. Microalgal Treatment Performance

Microalgal treatment produced increasing improvements in water quality. BOD and COD reduced by 33.3% and 26.0% respectively at 50% produced water concentration. At 100% concentration, TDS decreased by 11.95, BOD by 31.4%, COD by 24.7% and THC by 22.8%. Table 3 indicated a 14.7% decrease in DO which can be attributed to the balance between oxygen production by photosynthetic activity, microbial oxygen consumption and changing light or culture activity.

Table IV: Results after microalgal treatment

Produced-water concentration	Parameter	Day 0	Day 7	Removal (%)
50%	TDS (g/L)	37.5	33.0	12.0
50%	BOD (mg/L)	30	20	33.3
50%	COD (mg/L)	1000	740	26.0
50%	THC (mg/L)	54.3	42.5	21.7
60%	TDS (g/L)	42.0	38.0	9.5
60%	BOD (mg/L)	32	22	31.3
60%	COD (mg/L)	1200	900	25.0
60%	THC (mg/L)	65.2	51.0	21.8
75%	TDS (g/L)	48.0	43.0	10.4
75%	BOD (mg/L)	34	24	29.4
75%	COD (mg/L)	1300	950	26.9
100%	TDS (g/L)	52.2	46.0	11.9
100%	BOD (mg/L)	35	24	31.4
100%	COD (mg/L)	1540	1160	24.7
100%	THC (mg/L)	84.7	65.4	22.8

D. Heavy Metal Reduction

Both systems showed reductions in heavy metals, although the extent of removal varied by metal and treatment. The variation as shown in Table V is valued with differences in Biosorption, Bioaccumulation and microbial transformation mechanisms.

Table V: Analysis of heavy metals

Metal	Raw (mg/L)	Algae-treated (mg/L)	Bacteria-treated (mg/L)
Arsenic	3.435	0.016	0.004
Silver	1.287	0.002	0.197
Strontium	5.831	0.934	2.451
Manganese	5.000	0.976	1.098
Molybdenum	4.987	3.100	1.002
Cobalt	6.123	2.132	2.109
Antimony	9.543	1.095	3.856
Zinc	3.498	0.867	0.902
Chromium	5.000	0.978	0.470
Boron	21.345	3.243	5.213
Nickel	7.230	1.100	0.076

E. Comparative Performance

Both treatments demonstrated matching strengths. Bacterial treatment generally produced a robust reduction in TDS and BOD and a stronger overall THC reduction in the combined comparison. Algae showed a driven COD reduction and production of biomass as extra benefit.

The data also showed that an increase in the concentration of the produced water comprehensively reduced the pollutant removal efficiency, conforming with the restrictive effects of salinity and contaminants loading. These observations further agree with the need for integrating bacterial and microalgae procedures rather than relying solely on either system.

F. Regulatory Assessment

Table VI reports that biological treatments improved key water-quality monitoring parameters but failed to meet the criteria for discharge into the environment based on NUPRC standards. TDS, COD and THC were above the desired limits, while DO after bacterial treatment remained below the reported minimum.

Table VI: Comparison of both treatments

Parameter	Pre-treatment	Day 7 after microalgae	Day 7 after bacteria
pH	6.5	7.3	7.0
Temperature (°C)	24	25.8	20
TDS (mg/L)	52,200	46,000	43,800
BOD (mg/L)	35	24	29
COD (mg/L)	1,540	1160	1,040

THC (mg/L)	84.7	65.4	11.1
DO (mg/L)	3.7	6.7	4.5

Both biological systems were observed over the treatment period of seven days for notable changes in key parameters. Based on reported results after treatment, the biological systems can be considered to be a treatment stage requiring further treatment technologies rather than as a complete stand-alone discharge treatment.

Table VII: Comparison with discharge standards

Parameter	Reported NUPRC limit	Status after bacteria	Status after microalgae
pH	6.5–8.5	Compliant	Compliant
Temperature	≤35	Compliant	Compliant
TDS (mg/L)	2,000	Non-compliant	Non-compliant
BOD (mg/L)	50	Compliant	Compliant
COD (mg/L)	125	Non-compliant	Non-compliant
THC (mg/L)	10	Non-compliant	Non-compliant
DO (mg/L)	≥5.0	Non-compliant	Compliant

IV. CONCLUSION

The research demonstrated the pollutant-removal capabilities of combining different strains of locally-isolated microalgae and bacteria isolates. The study offers novel insights into the interactions between microalgae species and bacterial strains in reducing pollutants in produced water under varying salinity and contaminant loads.

REFERENCES

- Ahmadun, F.-R., Pendashteh, A., Abdullah, L. C., Biak, D. R. A., Madaeni, S. S., & Abidin, Z. Z. (2009). Review of technologies for oil and gas produced water treatment. *Journal of Hazardous Materials*, 170(2–3), 530–551.
- Al-Ghouti, M. A., Al-Kaabi, M. A., Ashfaq, M. Y., & Da'na, D. A. (2019). Produced water characteristics, treatment and reuse: A review. *Journal of Water Process Engineering*, 28, 222–239.
- Arbib, Z., Ruiz, J., Álvarez-Díaz, P., Garrido-Pérez, C., Barragan, J., & Perales, J. A. (2014). Photobiological removal of nutrients from wastewater by microalgae. *Environmental Science and Pollution Research*, 21, 2560–2569.
- Babu, A. J., Kumaresan, S., & others. (2020). Algal-based wastewater treatment and resource recovery studies cited in the dissertation.
- Bwapwa, J. K., Anandraj, A., & Trois, C. (2017). Microalgae-based treatment of petroleum hydrocarbons in wastewater: Review of potential applications.
- Campos, J. C., Borges, R. M. H., Oliveira Filho, A. M., Nobrega, R., & Sant'Anna, G. L. Jr. (2002). Oilfield wastewater treatment by combined microfiltration and biological processes.
- Chan, C. X., et al. (2022). Synergistic effects of microalgae and bacterial consortia for wastewater treatment.
- Christenson, L., & Sims, R. (2011). Production and harvesting of microalgae for wastewater treatment, biofuels, and bioproducts. *Biotechnology Advances*, 29(6), 686–702.
- Clark, C. E., & Veil, J. A. (2009). Produced water volumes and management practices in the United States. Argonne National Laboratory.
- Fakhru'l-Razi, A., Pendashteh, A., Abdullah, L. C., Biak, D. R. A., Madaeni, S. S., & Abidin, Z. (2009). Review of technologies for oil and gas produced water treatment. *Journal of Hazardous Materials*, 170, 530–551.
- Gerde, J. A., et al. (2014). Evaluation of microalgal biomass for nutraceutical and biofuel applications.
- Igunnu, E. T., & Chen, G. Z. (2014). Produced water treatment technologies. *International Journal of Low-Carbon Technologies*, 9, 157–177.
- Jiménez, M. M., Micó, M., Arnaldos, M., Medina, F., & Contreras, S. (2018). Desalination and fouling of NF/low pressure RO membrane for shale gas frac-produced water treatment. *Chemosphere*, 192, 186–208.
- Kondash, A. J., et al. (2020). Studies on produced-water volumes and characteristics cited in the dissertation.
- Lusinier, A., et al. (2019). Biological treatment of produced water under saline conditions.
- Pendashteh, A., Abdullah, L. C., Awang Biak, D. R., Madaeni, S. S., & Zainal Abidin, Z. (2012). Review of technologies for oil and gas produced water treatment. *Journal of Hazardous Materials*, 170(2–3), 530–551.

18. Priyadarshani, I., Rath, B., & Singh, Y. (2011). Heavy metal tolerance of freshwater microalga *Scenedesmus* sp. *Journal of Microbiology and Biotechnology Research*, 1(4), 90–98.
19. Rawat, I., Ranjith Kumar, R., Mutanda, T., & Bux, F. (2011). Dual role of microalgae: Phycoremediation of domestic wastewater and biomass production for sustainable biofuels. *Applied Energy*, 88(10), 3411–3424.
20. Renuka, N., Guldhe, A., Prasanna, R., Singh, P., & Bux, F. (2013). Performance of an integrated microalgae-based production system for the treatment of wastewater and biomass production. *Water Research*, 47(13), 4945–4953.
21. Veil, J. A., Puder, M. G., Elcock, D., & Redweik, R. J. Jr. (2004). A white paper describing produced water from production of crude oil, natural gas, and coal bed methane.