

Physicochemical and microbiological characterisation of effluent from a faecal-sludge treatment plant in Adjagbo, Benin, West Africa.

Abstract:

Rapid population growth, intensive urbanisation and the expansion of human and industrial activities have led to a substantial increase in faecal-sludge production in urban areas of developing countries. This study assessed the physicochemical and microbiological quality of effluent discharged from the Adjagbo faecal-sludge treatment plant in the municipality of Abomey-Calavi, Benin. Twenty-four samples were collected seasonally from four strategically selected locations within the treatment plant. Physicochemical parameters were determined in accordance with French Association for Standardisation (AFNOR) methods, while microbiological parameters were assessed using membrane-filtration and colony-enumeration methods. One-way analysis of variance was used to evaluate the influence of season on the measured parameters. Principal component analysis was performed to examine correlations among the variables.

At the outlet of the polishing lagoon, the mean pH and nitrate concentrations were 7.35 and 45.80 mg L⁻¹, respectively, during the rainy season, and 7.54 and 51.87 mg L⁻¹, respectively, during the dry season. These values were generally within the recommended range for irrigation water. However, the mean concentrations of total suspended solids were 2,873.87 mg L⁻¹ during the rainy season and 2,167.85 mg L⁻¹ during the dry season. The corresponding mean biochemical oxygen demand over five days (BOD₅) values were 2,805.75 and 2,407.63 mg O₂ L⁻¹, while the chemical oxygen demand (COD) values were 5,195.99 and 3,887.50 mg O₂ L⁻¹, respectively. *Escherichia coli* concentrations reached 347,377.81 and 1,357,412.50 CFU per 100 mL during the rainy and dry seasons, respectively.

These results indicate substantial organic and microbiological contamination of the effluent discharged from the polishing lagoon. Additional treatment is therefore required before discharge into the environment or any form of reuse.

Keywords: faecal sludge; treatment plant; physicochemical quality; microbiological quality; Abomey-Calavi.

Introduction

In the context of rapid population growth and industrial development, effective waste management has become an increasingly important concern for the international community .

Among the different types of waste generated, faecal sludge requires particular attention because of the environmental and public-health risks associated with its inadequate management . Although many urban residents in developing countries rely on on-site sanitation systems, appropriate faecal-sludge management chains are often lacking . Consequently, emptied faecal sludge is frequently discharged near residential areas, on roadsides, in drainage channels and in vacant spaces. Such practices expose communities to waterborne diseases, including diarrhoeal illnesses, cholera, dysentery, typhoid fever and parasitic infections, while also disrupting ecosystem functioning . To address these challenges, the United Nations 2030 Agenda for Sustainable Development, adopted in 2015, recognised the central role of sanitation in sustainable development and committed to achieving universal access to safely managed water and sanitation services by 2030 .

In the major cities of Benin, particularly in the municipality of Abomey-Calavi, the most common improved sanitation facilities include watertight pit latrines, soakaway pits and septic tanks . Inadequate use and maintenance of these facilities may lead to the contamination of soils, groundwater and surface water . To address this situation, the Government of Benin included the construction of three faecal-sludge treatment plants in its Government Action Programme for 2021-2026. These facilities are located, or planned, in the municipalities of Sèmè-Podji, Abomey-Calavi and Parakou, and use different treatment technologies .

The plant in Sèmè-Podji uses unplanted drying beds with a treatment capacity of 500 m³ day⁻¹. In Abomey-Calavi, the treatment process consists of planted drying beds containing *Echinochloa pyramidalis*, with a capacity of 600 m³ day⁻¹, combined with a lagoon system intended to improve treatment efficiency . Following appropriate treatment, faecal sludge and its liquid fraction may potentially be converted into resources suitable for agricultural use, including irrigation water. Nevertheless, treated effluent may remain a source of pollution because of its organic, mineral and microbiological load.

This raises an important question: what is the actual physicochemical and microbiological quality of effluent discharged from the plant, and what environmental and public-health risks may be associated with its release? Water plays a central role in human, animal and plant life and in the development of societies. It also provides the medium in which numerous metabolic reactions occur in terrestrial and aquatic organisms . The present study therefore aimed to characterise the physicochemical and microbiological quality of effluent from the Abomey-Calavi faecal-sludge treatment plant and to compare the observed values with relevant World Health Organization (WHO) and Beninese standards.

Materials and methods

Study site

This study was conducted in Abomey-Calavi, Benin, at the faecal-sludge treatment plant located in the Kanssounkpa district, Atadjè neighbourhood. The plant uses planted drying beds containing reeds for faecal-sludge treatment (Figure 1). The municipality is bordered to the north by Zè, to the south by the Atlantic Ocean, to the east by Sô-Ava and Cotonou, and to the west by Tori-Bossito and Ouidah. It covers an area of approximately 539 km².

The study area has a sub-equatorial climate characterised by a major rainy season from mid-March to mid-July, a minor rainy season from mid-September to mid-November, a major dry season from mid-November to mid-March and a minor dry season from mid-July to mid-September. Mean annual rainfall is approximately 1,200 mm, of which 700–800 mm occurs during the first rainy season and 400–500 mm during the second. Mean monthly temperatures vary relatively little, ranging from 27 to 31°C .

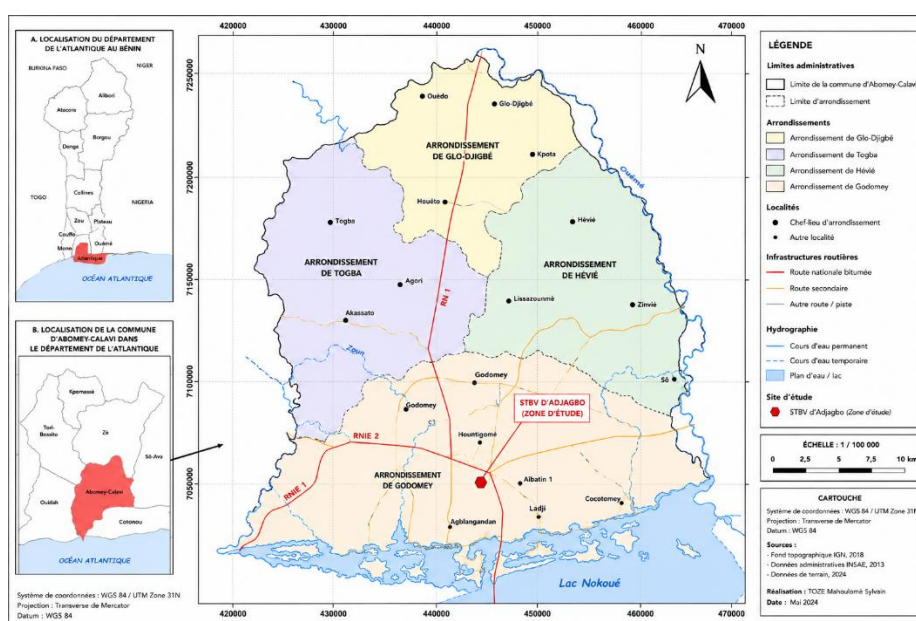


Figure 1. Map showing the location of Abomey-Calavi municipality and the Adjagbo faecal-sludge treatment plan

Sampling

Three samples were collected at each of four strategically selected points. Raw sludge (B0) was collected during discharge from emptying trucks. Pre-treated sludge (B1) was collected from the plant's buffer basin during the pre-treatment stage. Additional water samples were collected at the inlet of the maturation lagoon (B2), downstream of the planted drying beds, and at the plant outlet (B3), before final discharge (Table1). This sampling strategy was designed to assess treatment performance across the planted drying-bed and lagoon systems.

All sampling bottles were labelled appropriately and transported immediately to the laboratory in an insulated cooler at 4°C. Analyses were conducted within a maximum of 8 hours after collection.

Four sampling campaigns were conducted in May, September and December 2023 and February 2024. These periods were selected to cover the different seasons occurring in Abomey-Calavi and to assess seasonal variation in physicochemical and microbiological parameters.

Table 1. Geographical coordinates of the sampling points at the faecal-sludge treatment plant.

Code	Sampling point	Longitude	Latitude
B0	Raw sludge	N 6°31'17,5''	E002°18'49,3''
B1	Buffer basin	N 6°31'19,3''	E002°18'49,8''
B2	Inlet of the maturation lagoon	N 6°31'14,6''	E002°18'47,4''
B3	Outlet of the polishing lagoon	N 6°31'59,9''	E002°18'48,0''

Physicochemical analyses

pH was measured in situ using a Hach HQ40d multimeter in diluted sludge and water samples. Nitrogenous and phosphorus pollutants were quantified colorimetrically using a QW324A UV2700 spectrophotometer, in accordance with AFNOR procedures.

COD was determined by oxidation in an acidic medium using an excess of potassium dichromate at 150°C. Total suspended solids were determined by filtering a known volume of sludge through a 0.45-µm cellulose membrane, following AFNOR procedures. BOD₅ was measured using Oxytop probes maintained at a constant incubation temperature of 20°C in a thermostatically controlled cabinet, in accordance with NF EN 1899-2. The method is based on the change in oxygen pressure during incubation.

Microbiological analyses

Faecal coliforms were enumerated using the surface-spreading method, in accordance with AFNOR procedures. Violet Red Bile Lactose agar was used for faecal-coliform enumeration. Total coliforms and *E. coli* were enumerated using the multiple-tube method in accordance with NF T90-413.

The presumptive test was performed using Brilliant Green Lactose Bile Broth, while confirmation was conducted using Brilliant Green Lactose Bile Broth and simple peptone water for *E. coli*. Samples were incubated at 37°C for 24 hours.

Statistical analysis

Data from the physicochemical and microbiological analyses were entered and processed using Microsoft Excel. Arithmetic means and standard deviations were calculated. Analysis of variance was performed to assess the effect of season on the measured parameters. Principal component analysis and correlation analysis were conducted using R software, version 4.2.2.

Results

Physicochemical characterisation

Table 2 presents the physicochemical parameters measured during the rainy and dry seasons. pH (figure A) values ranged from 6.56 to 8.10 during the rainy season, with a mean of 7.35, and from 6.84 to 7.85 during the dry season, with a mean of 7.54. The highest pH values were recorded in the discharged effluent samples from B3, whereas the lowest values were observed at B2. The pH values were generally consistent with the recommended range for irrigation water under both Beninese and WHO guidelines.

Total suspended solids (figure B) ranged from 64.17 to 8,480 mg L⁻¹ during the rainy season, with a mean of 2,873.87 mg L⁻¹, and from 50 to 5,273.75 mg L⁻¹ during the dry season, with a mean of 2,167.85 mg L⁻¹. The lowest concentrations were recorded at B3, whereas the highest concentrations were observed in the raw sludge samples at B0. These values did not comply with the applicable discharge standards .

Phosphate concentrations (figure I) ranged from 11.32 to 227.55 mg L⁻¹ during the rainy season and from 20.86 to 180.51 mg L⁻¹ during the dry season. The lowest values were recorded in the discharged effluent at B3, while the highest values were observed at B2. The measured concentrations exceeded the permissible value of 2 mg L⁻¹ specified by WHO and the Beninese Agency for Standardisation.

Ammonium concentrations (figure G) ranged from 441.5 to 2,283.5 mg L⁻¹ during the rainy season and from 32.75 to 2,516 mg L⁻¹ during the dry season. Although a marked reduction to below 200 mg L⁻¹ was observed at the plant outlet, the concentrations remained above the applicable discharge standards. Nitrite concentrations during the rainy season ranged from 8.52 to 25.73 mg L⁻¹ and exceeded the limit of 3 mg L⁻¹ recommended by WHO and the Beninese Agency for Standardisation.

Nitrate concentrations (figure E) ranged from 11.24 to 89.60 mg L⁻¹ during the rainy season and from 3.93 to 129.30 mg L⁻¹ during the dry season. At the outlet of the polishing lagoon, nitrate concentrations were generally close to, or within, the recommended limit for irrigation water. However, the maximum values recorded in the dataset exceeded 50 mg L⁻¹ and should therefore be interpreted cautiously.

COD values (figure C) ranged from 201.5 to 14,660 mg L⁻¹ during the rainy season and from 339 to 8,960 mg L⁻¹ during the dry season. A substantial reduction in COD was observed along the treatment train, with the highest values at the inlet and the lowest values at the outlet. This reduction may reflect the degradation of organic matter by microorganisms and the retention of particulate material during treatment. Nevertheless, the COD concentrations remained above the WHO and Beninese discharge limit of 90 mg L⁻¹.

BOD₅ values (figure D) ranged from 36.75 to 5,521 mg O₂ L⁻¹ during the rainy season and from 48.5 to 4,604 mg O₂ L⁻¹ during the dry season. At B3, mean BOD₅ values were 36.75 and 48.5 mg O₂ L⁻¹ during the rainy and dry seasons, respectively. These values indicate that the effluent still contained a substantial biodegradable organic load.

Table 2. Physicochemical characteristics of samples collected during the rainy and dry seasons.

Season	Parameter	B0	B1	B2	B3	Mean	Minimum	Maximum	SD
Wet season	pH	7.31	7.42	6.56	8.1	7.35	6.56	8.1	0.63
	TSS	8480	1286.2	1665.0	64.1	2873.8	64.17	8480	3799.3
			5	7	7	7			4
	COD	14660	5393.7	528.7	201.	5195.9	201.5	14660	6741.2
			5		5	9			9
	BOD ₅	5521	5521	144.25	36.7	2805.7	36.75	5521	3135.6
					5	5			1
	NO ₃ ⁻	41.17	41.17	89.6	11.2	45.8	11.24	89.6	32.43
Dry season					4				
	NO ₂ ⁻	25.73	25.73	11.84	8.52	17.96	8.52	25.73	9.08
	NH ₄ ⁺	2283.5	2283.5	2093.5	441.	1775.5	441.5	2283.5	893.83
					5				
	TN	1312	1312	193.6	42.9	715.14	42.95	1312	691.94
					5				
	TP	227.55	227.55	49.8	11.3	129.06	11.32	227.55	114.81
					2				
Dry season	pH	7.77	7.71	6.84	7.85	7.54	6.84	7.85	0.47
	TSS	5273.7	2772.5	575.15	50	2167.8	50	5273.75	2382.8

n	5			5			7		
COD	8960	5055	1196	339	3887.5	339	8960	3955.1	4
BOD₅	4604	4604	374	48.5	2407.6	48.5	4604	2539.6	3
NO₃⁻	37.13	37.13	129.3	3.93	51.87	3.93	129.3	53.94	—
NO₂⁻	—	—	—	—	—	—	—	—	—
NH₄⁺	2516	2516	900	32.7	1491.1	32.75	2516	1235.1	8
TN	1933.5	1933.5	363.4	85.0	1078.8	85.05	1933.5	993.37	6
TP	180.51	180.51	91.1	20.8	118.25	20.86	180.51	77.4	6

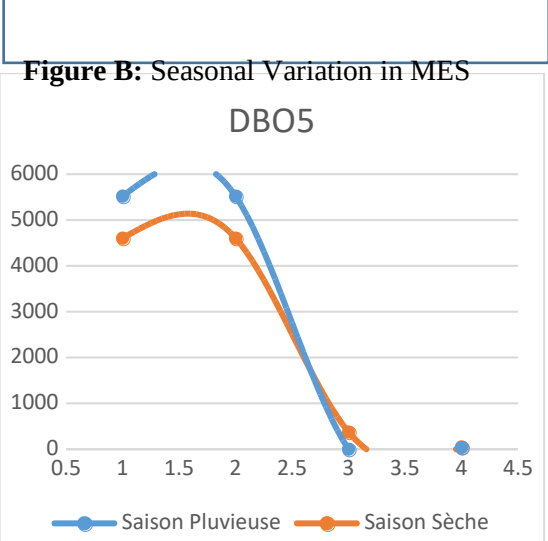
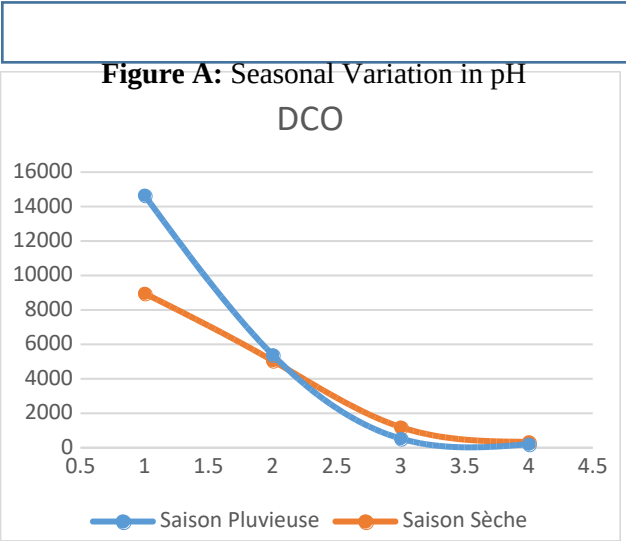
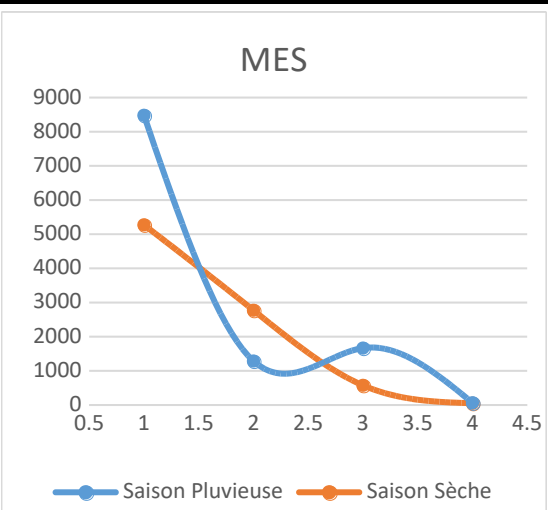
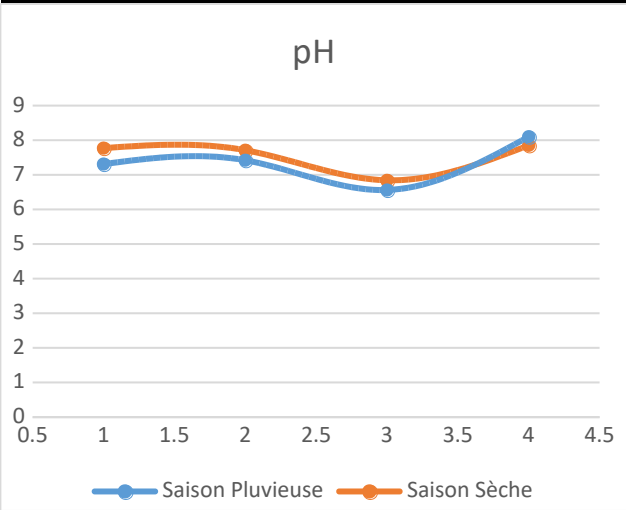
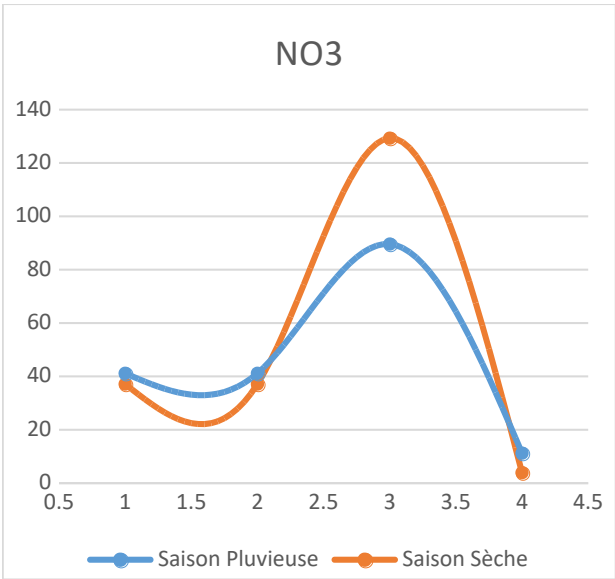


Figure C: Seasonal Variation in COD

Figure D: Seasonal Variation in BOD5



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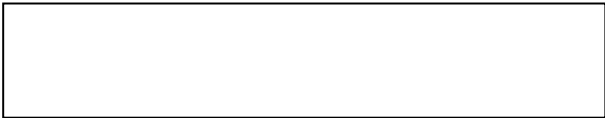


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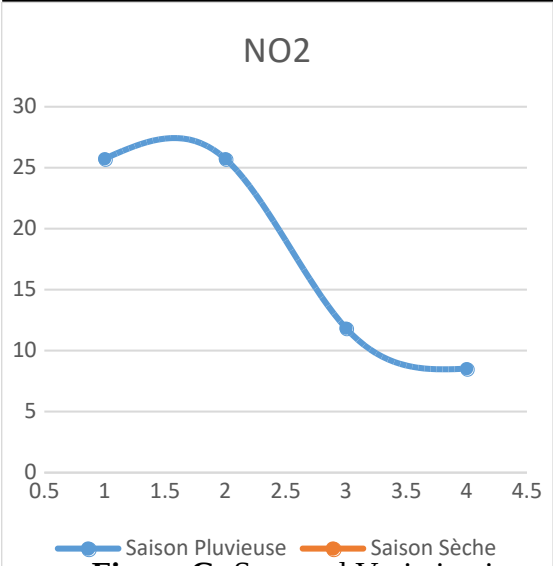
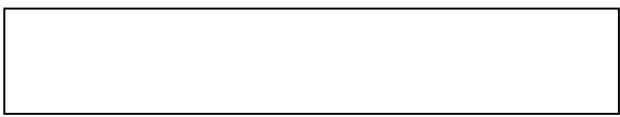
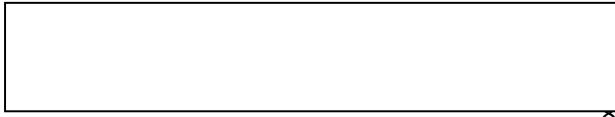
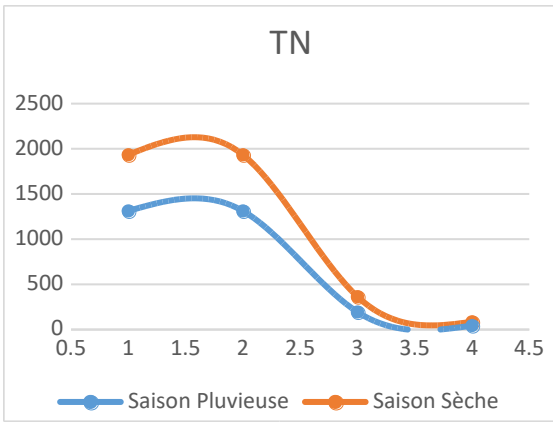
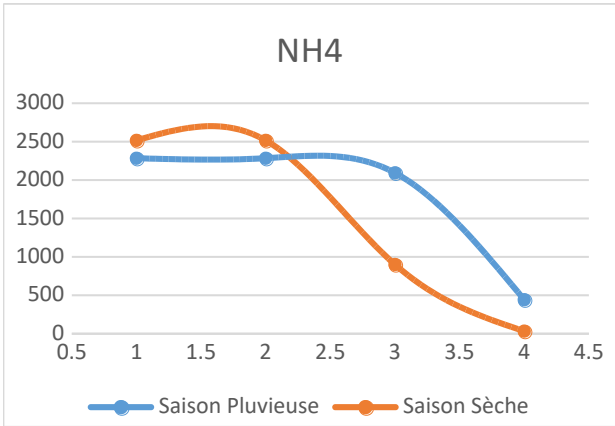


Figure G: Seasonal Variation in Ammoniacal Nitrogen

Figure H: Seasonal Variation in Total Nitrogen



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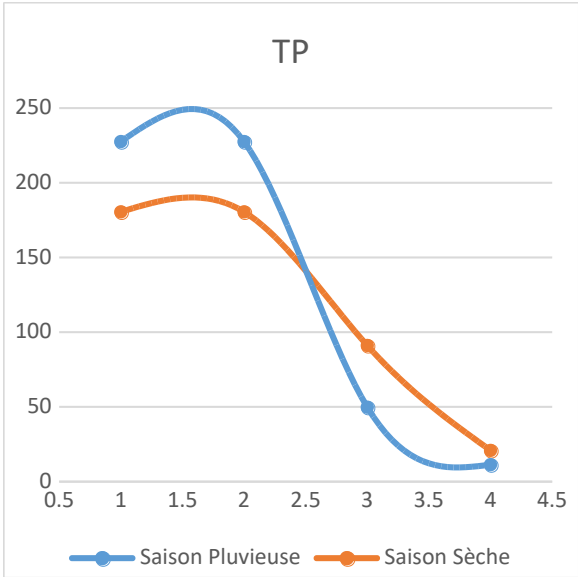


Figure I: Seasonal Variation in Total Phosphate

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180 **Microbiological characterisation**

181 Table 3 presents the microbiological results. During the rainy season, microbial densities
182 ranged from 10.25 to 693,550 CFU per 100 mL for *E. coli*, from 1,830 to 23,000 CFU per
183 100 mL for total coliforms and from 7,086.6 to 1,463,200 CFU per 100 mL for faecal
184 coliforms. During the dry season, *E. coli* concentrations ranged from 50 to 2,710,000 CFU
185 per 100 mL, while faecal-coliform concentrations ranged from 7,740 to 4,270,000 CFU per
186 100 mL. The highest bacterial concentrations were generally observed during the dry season.

187 **Table 3.** Seasonal variation in the microbiological parameters measured in the samples,
188 expressed as CFU per 100 mL

Season	Wet season			Dry season		
Microbiologic al parameter	<i>Escherichi a coli</i>	<i>Total coliform s</i>	<i>Faecal coliforms</i>	<i>Escherichia coli</i>	<i>Total coliform s</i>	<i>Faecal coliforms</i>
B0	693,55	23	1,463.20	2,710,000	—	4,270,000
B1	693,55	23	1,463.20	2,710,000	—	4,270,000
B2	2,401	5,777.33	122,813.3 3	9,6	—	160,9
B3	10.25	1,83	7,086.67	50	—	7,74

Mean	347,377.8 1	13,401.8 3	764,075.0 0	1,357,412.5 0	—	2,177,160.0 0
Min	10.25	1,83	7,086.67	50	—	7,74
Max	693,55	23	1,463,200	2,710,000	—	4,270,000
SD	399,726.4 0	11,199.5 5	808,661.3 2	1,561,838.3 8	—	2,417,412.2 6
WHO guideline value	≤1,000	≤1,000	≤1,000	≤1,000	≤1,000	≤1,000

The concentrations of indicator organisms substantially exceeded the limits reported in the applicable WHO and Beninese guidelines. For water intended for human consumption, microbiological quality criteria generally require the absence, or very low concentrations, of faecal indicator organisms. However, because the present study concerns treated effluent and potential irrigation reuse, the manuscript should clearly distinguish between standards for drinking water, irrigation water and wastewater discharge.

Total coliform concentrations varied markedly among sampling points. During the rainy season, concentrations were reported to range from 8,320 to 36,000 CFU per 100 mL. These values exceeded the threshold of 1,000 CFU per 100 mL cited in the manuscript and indicate that the discharged effluent was not microbiologically suitable for unrestricted reuse.

Faecal-coliform concentrations also varied substantially between seasons. During the rainy season, values ranged from 240 to 4.27×10^5 CFU per 100 mL, while during the dry season they ranged from 136 to 4.27×10^5 CFU per 100 mL. The highest concentrations greatly exceeded the Beninese discharge standard cited in the manuscript. These results indicate persistent faecal contamination and suggest that the effluent may contain other enteric pathogens.

The detection of high concentrations of *E. coli* and faecal coliforms indicates recent faecal contamination. Consequently, the effluent may pose a health risk, particularly where it is used for irrigation of crops eaten raw or where communities are exposed to contaminated surface water. The presence of indicator organisms does not, by itself, demonstrate the presence of specific pathogens such as enterotoxigenic *E. coli* or *Shigella dysenteriae*; pathogen-specific conclusions would require targeted microbiological testing.

Seasonal effects

The ANOVA results are presented in Table 4. Season did not significantly affect the parameters measured in the raw sludge mixture at B0. At B1, seasonal effects were observed for ammonium at the 10% significance level and for *E. coli* at the 5% level. Seasonal variation also affected total phosphorus and *E. coli* at B2. At the inlet of the maturation lagoon, pH and BOD₅ were influenced by season. The effect of season on COD became significant from the outlet of the first lagoon and remained evident at the outlet of the polishing lagoon. At B3, seasonal effects were also observed for BOD₅ and total phosphorus. These findings suggest that seasonality influenced the performance of specific treatment stages rather than affecting all measured parameters uniformly. Rainfall, dilution, hydraulic loading, temperature and microbial activity may all contribute to seasonal changes in effluent quality.

Principal component analysis

Principal component analysis was used to summarise the relationships among the physicochemical and microbiological variables. The first two principal components explained 60.51% of the total variance. Dimension 1 accounted for 41.50% of the variance, whereas Dimension 2 accounted for 19.01%.

Dimension 1 was positively associated with pH, dry matter, total suspended solids, ammonium, total nitrogen, *E. coli* and total coliforms. It was also positively correlated with COD, BOD₅, nitrate, nitrite and total phosphorus. Dimension 2 showed positive associations with pH, COD, BOD₅, nitrate, nitrite, *E. coli* and coliforms.

The raw sludge samples from the emptying-truck mixture were positively associated with Dimension 2 and showed variation in pH, *E. coli* and faecal coliforms. They were negatively associated with Dimension 1, with BOD₅ and total phosphorus contributing to their position in the factorial space. COD, nitrate and nitrite displayed similar patterns of variation within the truck-mixture samples.

Overall, the PCA indicates that suspended solids, dry matter, COD, BOD₅, ammonium, nitrite and total nitrogen were important factors characterising the quality of the faecal-sludge-derived effluent. However, the final manuscript should include the PCA loading table and the percentage contribution of each variable to the components so that these interpretations can be independently assessed.

Analysis of variance

Table 4 presents the influence of season on the physicochemical and microbiological parameters measured in the investigated samples. The ANOVA results showed that season did not significantly affect the parameters measured in the truck-mixture samples at B0. At B1,

seasonal effects were observed for ammonium at the 10% significance level and for *E. coli* at the 5% significance level. Seasonal variation also affected total phosphorus and *E. coli* at the inlet of the maturation lagoon (B2).

Table 4. Assessment of the effect of season on the physicochemical and microbiological parameters of the investigated water samples.

Parameter	B0	B1	B2	B3
pH	0.178	0.151	0.042 (**)	0.241
TSS	0.157	0.737	-	-
TDS	0.974	0.244	0.973	0.783
COD	0.404	0.836	0.272	0.013 (***)
BOD ₅	0.479	0.760	0.003 (***)	0.078 (*)
NO ₃ ⁻	0.687	0.814	0.625	0.803
NO ₂ ⁻	-	-	-	-
NH ₄ ⁺	0.463	0.087 (*)	0.348	0.153
TN	0.261	0.913	0.188	0.232
TP	0.819	0.092 (*)	0.033 (**)	0.003 (***)
<i>E. coli</i>	0.186	0.017 (**)	0.038 (**)	0.298
Total coliforms	0.990	-	0.995	-
Faecal coliforms	0.466	0.494	0.719	0.141

Note: *: Significant effect at the 10% level; **: significant effect at the 5% level; ***: significant effect at the 1% level.

At the same sampling point, pH and BOD₅ were also significantly influenced by season. The effect of season on COD became apparent from the outlet of the first lagoon and persisted through to the outlet of the polishing lagoon, which represented the final discharge point. At the polishing-lagoon outlet, BOD₅ and total phosphorus were also significantly affected by season.

The results indicate that seasonal variation affected selected parameters at specific stages of the treatment process rather than exerting a uniform effect throughout the plant. This pattern may reflect seasonal changes in rainfall, hydraulic loading, dilution, temperature and microbial activity.

Principal component analysis

Principal component analysis was used to assess the correlation among physicochemical and microbiological.

The first two principal components explained 60.51% of the total variability in the physicochemical and microbiological characteristics of the faecal sludge collected at the Abomey-Calavi treatment plant. As shown in the accompanying table, the first principal component, Dimension 1, explained 41.50% of the variance, whereas the second component, Dimension 2, explained 19.01%.

Dimension 1 was positively correlated with pH, dry matter, total suspended solids, ammonium, total nitrogen, *E. coli* and total coliforms. It was also positively associated with COD, BOD₅, nitrate, nitrite and total phosphorus. Dimension 2 showed positive correlations with pH, COD, BOD₅, nitrate, nitrite, *E. coli* and total coliforms.

The manuscript also reports a negative correlation between faecal coliforms and key parameters, including dry matter, total suspended solids, ammonium, total nitrogen and total phosphorus. This interpretation should be checked against the PCA loading matrix because the abbreviation “FEC” is not defined and appears to refer to faecal coliforms.

The projection of the physicochemical and microbiological variables onto the first two principal components showed that the raw sludge from the truck mixture was positively associated with Dimension 2. The raw sludge samples were particularly associated with variation in pH, *E. coli* and faecal coliforms.

The truck-mixture samples showed a negative association with Dimension 1, with BOD₅ and total phosphorus contributing to their position in the factorial space. COD, nitrate and nitrite also displayed similar patterns of variation in the truck-mixture samples.

Overall, the PCA indicates that several groups of variables were important in characterising the samples. These included physical parameters, such as total suspended solids and dry matter; organic pollution parameters, particularly COD and BOD₅; and mineral or nutrient parameters, including nitrite, ammonium and total nitrogen. These variables should therefore be considered when evaluating the treatment performance and environmental quality of effluent from the faecal-sludge treatment plant.

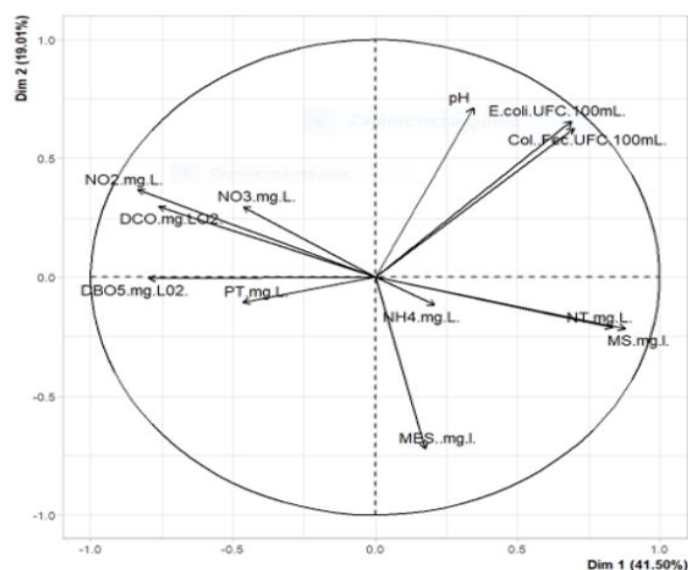


Figure 3.18. Projection of the physicochemical and microbiological parameters of raw sludge onto the first two factorial axes.

Discussion

Physicochemical characterisation

The results of the water analyses showed that the effluent was slightly alkaline during both seasons. The recorded pH values 7.35 during the rainy season and 7.54 during the dry season were within the applicable discharge standards. This alkalinity is consistent with findings reported for Lake Toho in southern Benin, groundwater from basement aquifers in Côte d'Ivoire, and water bodies in the Brussels region .

The pH values recorded in the present study were higher than those measured in 2017 in the same watercourse (7.16) and in the coastal lagoon (7.32), but lower than those reported in 2014 for the Ahémé-Guézin lagoon complex (7.76 and 9.15) . These differences may be explained by variations in acid rainfall, biological activity and the nature of wastewater discharges.¹⁵

In the chemical assessment of aquatic environments, total suspended solids (TSS) should be understood as the mineral and organic particles present in water. According to Rodier, TSS concentrations are influenced by the nature of the geological formations crossed by the water, season, rainfall, flow conditions and the characteristics of the discharged waste. The substantial reduction in TSS between the raw sludge and the buffer basin indicates the effectiveness of physical sedimentation. This reduction may also be explained by the treatment mechanism of planted drying beds, which promotes TSS retention through the combined action of plant roots and the filter substrate.

However, TSS concentrations at the outlet of the polishing lagoon (B3) 64.17 mg L⁻¹ during the rainy season and 50 mg L⁻¹ during the dry season were at, or exceeded, the Beninese standard of 50 mg L⁻¹ for direct discharge into the environment . Elevated TSS concentrations may impair the quality of the receiving water by increasing turbidity, reducing light penetration and decreasing dissolved oxygen concentrations. These effects may limit the development of aquatic life in Lake Toho, where the effluent is discharged. Increased suspended-solid concentrations may also contribute to water warming, thereby reducing habitat quality for cold-water organisms .

Nitrogen and nitrogen-derived compounds in the effluent

Ammonium concentrations ranged from 441.5 to 2,283.5 mg L⁻¹ during the rainy season and from 32.75 to 2,516 mg L⁻¹ during the dry season. These concentrations did not comply with the discharge standards established by the Beninese Agency for Standardisation or with the WHO recommendation of less than 1 mg L⁻¹ . Consequently, the effluent discharged into Lake Toho represents a potential source of ammoniacal-nitrogen pollution and may contribute to eutrophication of the receiving environment.

Nitrite concentrations, which reached 17.96 mg L⁻¹ during the rainy season, were described as relatively low across the sampling points. This may be related to the rapid oxidation of nitrite to nitrate . These findings are consistent with those reported by who observed concentrations well below 3 mg L⁻¹ at some sites and only trace concentrations at several stations along the Ouémé River. and similar nitrite concentrations have also been reported in the Ubu River in the Niger Delta and domestic waste water in Kombolcha (northeast of Ethiopia). Although nitrite toxicity in fish is species-dependent, concentrations close to 0.015 mg L⁻¹ have been associated with fish mortality through hypoxia in some circumstances . The concentrations recorded in the present study, which reached 25.73 mg L⁻¹, could therefore pose a risk to fish populations in Lake Toho. They may also contribute to unexplained fish mortality, as reported in the lake in 2020 .

However, this potential effect should be confirmed through targeted ecological and toxicological investigations .

In studies of nitrogen transformation in aquatic systems, nitrate is generally regarded as the terminal product of the oxidation of organic nitrogen compounds. In the present study, nitrate concentrations ranged from 11.24 to 89.60 mg L⁻¹ during the rainy season and from 3.93 to 129.30 mg L⁻¹ during the dry season. The manuscript states that these values were below the WHO guideline value of 50 mg L⁻¹ and the corresponding Beninese standard . However,

because some individual concentrations exceeded 50 mg L⁻¹, this interpretation should be revised to distinguish between mean concentrations and individual observations.

Given the slightly alkaline conditions observed during the study, the relatively low nitrate concentrations in the discharged effluent may partly reflect the volatilisation of ammonium as ammonia .

The chemical notation and the specific nitrogen species involved should be verified in the final version. Although no universally applicable toxicity threshold has been established for total nitrogen, concentrations above 1.0 mg L⁻¹ in surface waters are commonly considered indicative of a potential eutrophication problem . The concentrations observed, particularly at the outlet of the polishing lagoon (B3), therefore suggest a substantial risk of nutrient enrichment and ecological disturbance.

Nitrogen is an essential element for terrestrial and aquatic life; however, excessive nitrogen inputs can adversely affect the quality of receiving surface waters. Nitrogen enrichment is an important driver of eutrophication, a process involving the accumulation of nutrients in aquatic ecosystems that promotes excessive growth of algae and aquatic plants and may alter the composition and functioning of aquatic communities .

Organic and microbiological loads

Phosphate concentrations ranged from 11.32 to 227.55 mg L⁻¹ during the rainy season and from 20.86 to 180.51 mg L⁻¹ during the dry season. These values substantially exceeded the regulatory limits cited in the manuscript, namely 2 mg L⁻¹ under the Beninese standard and 3 mg L⁻¹ under the WHO guideline. The results are consistent with those reported by Phosphorus plays an important role in wastewater-treatment processes. Nevertheless, when discharged into natural environments at high concentrations, it can stimulate eutrophication of surface waters .

The recorded BOD₅ and COD values reported as 14,660 mg L⁻¹ and 5,521 mg L⁻¹, respectively were substantially higher than the recommended limits of 30 mg L⁻¹ for BOD₅ and 90 mg L⁻¹ for COD. These concentrations indicate a considerable organic load and are incompatible with the direct reuse of the effluent for irrigation.

The COD-to-BOD₅ ratio can be used to assess the biodegradability and characteristics of an effluent. In this study, the mean ratio was reported as 1.74, which is below 3 and therefore suggests that the effluent was readily biodegradable . This finding is consistent with a predominantly domestic origin of the wastewater treated at the faecal-sludge treatment plant.

However, the statement that the effluent therefore exhibited “significant nutrient pollution” is not consistent with the high COD and BOD₅ values. These results primarily indicate a

substantial organic load, although the effluent may also contain considerable inorganic contaminants, including nitrogen and phosphorus compounds. The interpretation should therefore distinguish clearly between organic pollution and nutrient-related or mineral pollution. The results are consistent with those reported by .

Nevertheless, the proposed treatment strategy should be reconsidered .

Because the effluent appears to contain a high biodegradable fraction, biological treatment may be appropriate; chemical pre-treatment may be required only if specific compounds inhibit biological processes or if the objective is to remove particular nutrients or refractory contaminants.

The concentration of *Escherichia coli* reached 2,710,000 CFU per 100 mL, substantially exceeding the WHO value of 1,000 CFU per 100 mL cited in the manuscript. This level represents a considerable public-health risk, particularly where the effluent is used to irrigate vegetables consumed raw. Faecal-coliform concentrations reached 4,270,000 CFU per 100 mL and also exceeded the limits reported for the Beninese standard and WHO guidance.

These organisms are indicators of recent faecal contamination and indicate that the effluent is unsuitable for irrigation without additional treatment . However, the presence of *E. coli* and faecal coliforms does not by itself confirm the presence of specific pathogens. Further testing would be required to determine whether pathogens such as *Salmonella* spp., *Shigella* spp., viruses or intestinal parasites are present.

The findings indicate that the treated effluent retained substantial physicochemical, organic, nutrient and microbiological pollution, particularly at the outlet of the polishing lagoon. Although the treatment process reduced suspended solids, the residual concentrations of ammonium, phosphorus, COD, BOD₅ and faecal indicators remained high and, in several cases, exceeded the applicable discharge or reuse guidelines. The microbiological results are of particular concern because the very high concentrations of *Escherichia coli* and faecal coliforms indicate a substantial risk of faecal contamination and limit the suitability of the effluent for unrestricted agricultural irrigation, especially for crops consumed raw. These findings highlight the need for additional treatment and routine monitoring before discharge into Lake Toho or reuse for irrigation. Further investigations should assess the effectiveness of additional treatment steps and determine the presence of specific pathogenic microorganisms and other contaminants of public-health and ecological concern. Such measures are essential to reduce potential risks to aquatic ecosystems and communities dependent on the receiving environment.

Conclusion

In light of the findings above, the study assessed the physicochemical and microbiological quality of effluent from the Adjagbo faecal-sludge treatment plant during different seasons. The results indicated that pH values were generally within the recommended range, while nitrate concentrations at the polishing-lagoon outlet were generally close to the irrigation-water guideline. However, the effluent remained heavily contaminated with suspended solids, organic matter, nutrients and faecal indicator bacteria.

The discharged effluent was characterised by high concentrations of total suspended solids, COD, BOD₅, ammonium, total phosphorus, *E. coli* and faecal coliforms. These concentrations exceeded the relevant discharge or reuse criteria cited in the manuscript, indicating that the current treatment process does not provide sufficient effluent quality for direct discharge or unrestricted irrigation reuse.

Seasonal effects were observed for selected parameters, including pH, BOD₅, COD, ammonium, total phosphorus and *E. coli*, depending on the sampling point. These findings underline the importance of routine monitoring across both rainy and dry seasons.

Additional treatment and strengthened effluent-quality control are therefore required before discharge into the Togba watercourse and its downstream connection with Lake Toho, or before any agricultural reuse. An integrated management approach covering the treatment plant, receiving watercourse and wider catchment should be implemented to protect local communities, downstream water uses and aquatic ecosystems.

Conflict of interest statement

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.