



Microbiological and physicochemical quality of some water points in the Nkolafamba Subdivision (Center Region, Cameroon)

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ABSTRACT

A study was carried out with the objective of evaluating the microbiological and physicochemical quality of surface water in the Nkolafamba subdivision, Center Region of Cameroon. Five sampling points were selected based on several criteria. The bacteria isolated were Mesophilic Aerobic Heterotrophic Bacteria (BHAM), bacteria that were witnesses to faecal contamination and *Pseudomonas*. Some abiotic parameters were measured using the usual techniques. The results show that some physicochemical parameters such as temperature was almost constant around 25 °C during the study period. There is also a pH which tended towards neutrality pH de (7,04 ± 1,03 U.C). However, it was noted that the stations are quite oxygenated (63,96%). Bacteriological analyzes revealed that an average value of 5.17 units (logUFC / 100mL) was recorded for BHAM. Overall, the abundances of total coliforms averaged 4.18 units (logUFC / 100mL). The waters of Nkolafamba also harbor a pathogenic microflora, with bacteria such as *P. aeruginosa* *P. pasteurella*, the abundance of which can sometimes reach 5,462 units (log CFU / 100mL). These germs can be the cause of eye infections or septicemia on users of these waters. These waters have a high bacterial load, and the microbiological pollution observed is predominantly animal. Some abiotic parameters such as pH, electrical conductivity, Suspended Solid (SS), nitrates and turbidity have influenced the distribution of these germs. The degradation of the quality of these waters is favoured by their proximity to sources of pollution, the runoff of contaminated water in the stations. These waters, without any treatment, are not recommended for human consumption according to the standards of the World Health Organization.

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Keywords: Abiotic variables, faecal coliforms, faecal streptococci, pathogenic bacteria, rivers.

INTRODUCTION

Water is an essential resource for human both for food needs and for various other uses (WHO, 2004). Yet, most of humanity does not have access to this resource. She suffers from

water stress with less than 1700 m³ of fresh water available per capita / year (WHO, 2004).

Of the 13,600 million km³ of water that cover the planet, only 0.014% is fresh water usable by human as surface water (WHO,

2004). In emerging countries in general and in Cameroon in particular, the massive resignation of the populations to use surface water (rivers, springs, etc.) is linked to the insufficient means of local municipalities to ensure an optimal distribution of drinkable water. Kueté et al. (2004) mention in this regard that these populations often face water supply problems in their distribution network. Indeed, the population explosion is causing a scarcity of sources of drinking water supply. The populations therefore, in the need to obtain drinking water, find themselves obliged to use surface water that they ignore the microbiological quality (WHO, 2004). However, this water has been qualified of dubious or contaminated quality by several studies and likely to cause many ills (Nola et al., 2010; Mounsang et al., 2013). Contamination of surface waters by pathogens is a pollution problem that dates back a long time (WHO, 2004). Pollution of water from faecal microorganisms appeared very early as soon as water was used as a vector for waste disposal (George and Servais, 2002). Human contamination occurs either by consumption of drinking water, by consumption of food contaminated by water, even during bathing or contact with water for recreational use (George and Servais, 2002).

Several studies have already been carried out on the microbiological quality of water in the Yaoundé region, as well as on the role of soil in the transfer of bacteriopolitants from the soil surface to underground water. They reveal that these waters are acidic, soft and weakly mineralized (Nola et al., 2000). They harbor a varied bacterial microflora made up of fecal bacteria, commensal bacteria, and opportunistic pathogenic bacteria, whose abundance dynamics undergo spatio-temporal variations (Nola et al., 2010). The population of these microorganisms is significantly influenced by physico-chemical factors such as gas content and certain dissolved ions, and meteorological factors such as precipitation and sunstroke (Nola et al., 2000; 2002). Other analyzes took into account the lotic and lentic environments in the Center, Littoral and West Cameroon region, and made it possible to

assess the state of health of these hydrosystems. Based on the structure of aquatic communities, the hydrosystems so far prospected concern, among others the Mfoundi (Foto Membohan et al., 2013) the municipal lake of Yaoundé (Zébazé Tougouet et al., 2011) the Obili lake (Ajeegah et al., 2018). This work reveals that urban and peri-urban waterways are subject to organic pollution of anthropogenic origin due to the promiscuity linked to the high density of populations in the watersheds and to the use of water bodies as receptacles of water waste.

Despite this work, very little information is available on the microbiological and physicochemical quality of the waters of the Nkolafamba subdivision, a small town located in the suburbs of the city capital and in full swing. In addition, little is known about the variation in the densities of some pathogenic bacteria present in surface waters often used by the populations that live there. Likewise, little data is available on the influence of physicochemical and hydrological factors on the variation in bacterial abundance of these rivers. The present work aimed at determining the microbiological and physicochemical quality of some rivers in the district of Nkolafamba.

Collection of data

Geographical setting

The city of Nkolafamba is located in the Center region of Cameroon, a suburb of the political capital Yaoundé, in the Mefou and Afamba department and in the Nkolafamba district. This small town is about 10km from Yaoundé between 3 ° 51 ' 32 " North latitude and 11 ° 39 ' 53 " East longitude. At the time of the 2005 census, Nkolafamba had 14,494 inhabitants and went back to around 40 small villages (RGPH, 2005). The climate is tropical and equatorial, with four seasons (2 dry and 2 rainy) of unequal duration. The average rainfall is around 1633 mm with average temperatures rarely exceeding 25 °C (Suchel, 1987). The hydrographic network of this area is organized around the Afamba river and its main tributaries: Mefomo, Lolo and Otoundoumba. The soils do not differ from those in the

Yaoundé region (Vallerie, 1973). Indeed, these soils are mainly of the domain of ferralitic soils. There are nevertheless two variants, namely red, clayey and acidic ferralitic soils (pH <5.5) and yellow clayey ferralitic soils very rich in iron (Vallerie, 1973). Figure 1 shows the study area and the location of the sampling stations.

Choice and geographic location of sampling points

In total, 5 sampling points were selected because of the importance of the frequency of water use, the interest given by the populations to these points, the proximity of a source of pollution, the desire to cover the entire study area and access: either 2 sampling points on the Afamba river, the city's main waterway, and 1 point on each of the Mefomo, Lolo and Otoundoumba tributaries. The geographical location of each sampling point, the codes adopted, and the altitude in relation to the sea are presented in Table 1. It appears that the study was carried out between 667 m and 719 m altitude, and between 3 ° 43 ' 11.1 " North latitude and 11 ° 33'33.3 " East longitude.

Sampling and method of analysis

Sampling

A 1m x1m quadrat so as to delimit the exact places where the watercourse is actually used by the populations was delimited at each sampling station to carry out the sampling and measure the hydrological properties of the watercourses (Bernard and Salle, 2010). In addition to hydrological parameters, physicochemical and microbiological parameters were considered. Samples for bacteriological analyzes were carried out in sterile 500 mL glass vials. The water samples for the physicochemical analyzes were taken in two batches of polyethylene bottles, a bottle of 1000 mL where the water was introduced without making bubbles, intended for the laboratory measurement of parameters such as oxygen dissolved, turbidity, colour among others. Another 250 mL vial in which dissolved CO₂ is fixed in the field before being determined in the laboratory by the volumetric method. All placed in a refrigerated enclosure and transported to the laboratory. The

physicochemical parameters considered in this study were measured in the field and in the laboratory using the techniques recommended by Rodier et al. (2009).

Analyzes of hydrological parameters

Water flow velocity

The water flow velocity (V) expressed in m / s was measured at each station by the indirect method which consists in determining, using a chronometer, the time (t in s) taken by a neutral dye not pollutant (methylene blue) to travel a known distance (d in m), without obstacle as previously defined (Rodier et al., 2009).

Wet section

A line transect was carried out at each of the sampling stations using two stakes planted at each of the banks of the stations at the edge of the wetland. These stakes perpendicularly support a graduated horizontal string. Then, a graduated ruler immersed vertically in the water allows you to determine the respective depths of water and sludge from one bank to the other, following the graduation of the string with a step of 50 cm. The data collected made it possible to represent and determine, by station, the area of the wetted section in (m²) by grid on graph paper (Rodier et al., 2009).

Debit

The water flow rate expressed in m³/s and was obtained from the product of the wetted section and the speed by the formula (Rodier et al., 2009), with: flow rate in m³/s, flow speed in m /s, wetted section in m².

Evaluation of physicochemical parameters

The physicochemical parameters were analyzed by the techniques recommended by Rodier et al. (2009). All the parameters considered were temperature, pH, conductivity, dissolved O₂, suspended solids, color, turbidity, nitrates, ammoniacal nitrogen, orthophosphates and dissolved CO₂.

The temperature, pH, conductivity and dissolved O₂ were measured *in situ* using a mercury column thermometer graduated to 1 / 10th of a degree Celsius, digital pH meter model brand SCHOTT GERATE CG 818, a portable TDS / conductivity meter from HANNA series HT 8733, and an oximeter from

HANNA, model HI 9146; the units are expressed in °C, UC, $\mu\text{S} / \text{cm}$ and in % O_2 saturation respectively. The measurement of the dissolved CO_2 content was carried out in two stages. On the field, the CO_2 was fixed by introducing into a 200 ml volumetric flask, 20 ml of sodium hydroxide (NaOH) N / 20 plus 2 to 3 drops of phenolphthalein, this mixture was made up with the sample of raw water up to the mark. The resulting mixture of pink color was stored in a 250 ml double-capped polyethylene flask and then returned to the laboratory. In the laboratory, 50 mL of this sample was titrated with N / 10 hydrochloric acid (HCl) until complete discoloration. The CO_2 content of the water expressed in mg / L, was then determined by the formula: $[\text{CO}_2] (\text{mg} / \text{L}) = (\text{control burette descent} - \text{sample burette descent}) \times 17.6$. Table 2 shows the physicochemical parameters measured in the laboratory, the method of analysis, and the reagents used, the wavelengths (λ) of reading with the DR / 2000 spectrophotometer.

Microbiological evaluation of the rivers of the city of Nkolafamba

Heterotrophic Bacteria Aerobic Mesophilic

BHAM species were isolated from the plain agar surface cast in a Petri dish by surface spreading techniques. 100 μL of the sample was taken using a sterile Hamburg strainer pipette and then deposited on the agar surface. The sample was subsequently plated using a sterile glass spreader (Marshal et al., 1991). The Petri dishes are then incubated at room temperature and the readings are taken gradually over 5 days (Holt et al., 2000).

Fecal and total coliforms

Fecal and total coliforms were isolated on ENDO-specific culture media by surface spreading and membrane filter techniques. The filter membrane technique is used when the spreading on the surface is negative (Diagnostic Pasteur, 1987, Marshal et al., 1991). Readings were taken after 24 to 48 hours of incubation. Fecal coliforms were incubated at 44 °C and total coliforms at 37 °C for 18 to 48 hours (Marshal et al., 1991). The enumeration was carried out by direct counting. (Holt et al., 2000).

Fecal streptococci

Isolation of faecal streptococci was carried out by the filter membrane technique on BEA agar (Bile Esculin Azide) at a temperature of 37 °C and the counts were carried out after 18 to 24 hours of incubation (Marshal et al., 1991).

Characterization of potentially pathogenic bacteria (genus Pseudomonas)

-Isolation and enumeration: *Pseudomonas* was chosen to verify the presence of bacteria endowed with pathogenicity and responsible for immediate public health problems (Mezaache, 2012). Isolation of bacteria of the genus *Pseudomonas* was done by the membrane filter technique on CN agar (Cetrimide) at a temperature of 37 °C for 18 to 24 hours (Marshal et al., 1991). After isolation and enumeration, strains with satisfactory cultural characteristics were collected and subjected to various other confirmatory tests.

-Biochemical identification of bacteria of the genus Pseudomonas: The identification of *Pseudomonas* has been based on a number of biochemical tests. After isolation of the bacteria, the colonies exhibiting the characteristics of bacteria of the genus *Pseudomonas* were purified by streaking on cetrimide medium. After subculture on PCA (Plate count agar) sloped and incubated at 37 °C for 18 to 24 hours, the biochemical tests were carried out according to the usual biochemical criteria, using the conventional gallery (Marshal et al., 1991). The tests considered were catalase, gas production, glucose utilization, lactose and mobility among others (Holt et al., 2000).

Determination of the probable origin of the microbiological contamination of the Nkolafamba Rivers

The relationship between the densities of faecal coliforms (CF) to those of faecal streptococci (SF) has made it possible to determine the probable origin of the pollution (WHO, 2004). Indeed, when $\text{CF} / \text{SF} \geq 4$, the pollution is probably of human origin; $2 \leq \text{CF} / \text{SF} < 4$ pollution is said to be predominantly human in a mixed population; $1 < \text{CF} / \text{SF} < 2$ pollution is probably linked to a mixed population, $0.7 < \text{CF} / \text{SF} < 1$ predominantly

animal water pollution in a mixed population, $CF / SF \leq 0.7$ pollution is said to be of animal origin (WHO, 2004).

Assessment of the importance of physicochemical and hydrological variables on the microbiological distribution of rivers

To determine the impact of physicochemical and hydrological variables on

the variation in microbial densities of Nkolafamba streams, spearman "r" correlation tests were performed using SPSS software. These tests were supplemented by analyzes of Mann Whitney and Kruskal Wallis comparisons between sampling periods, and between the different ones and by Principal Component Analysis (PCA).

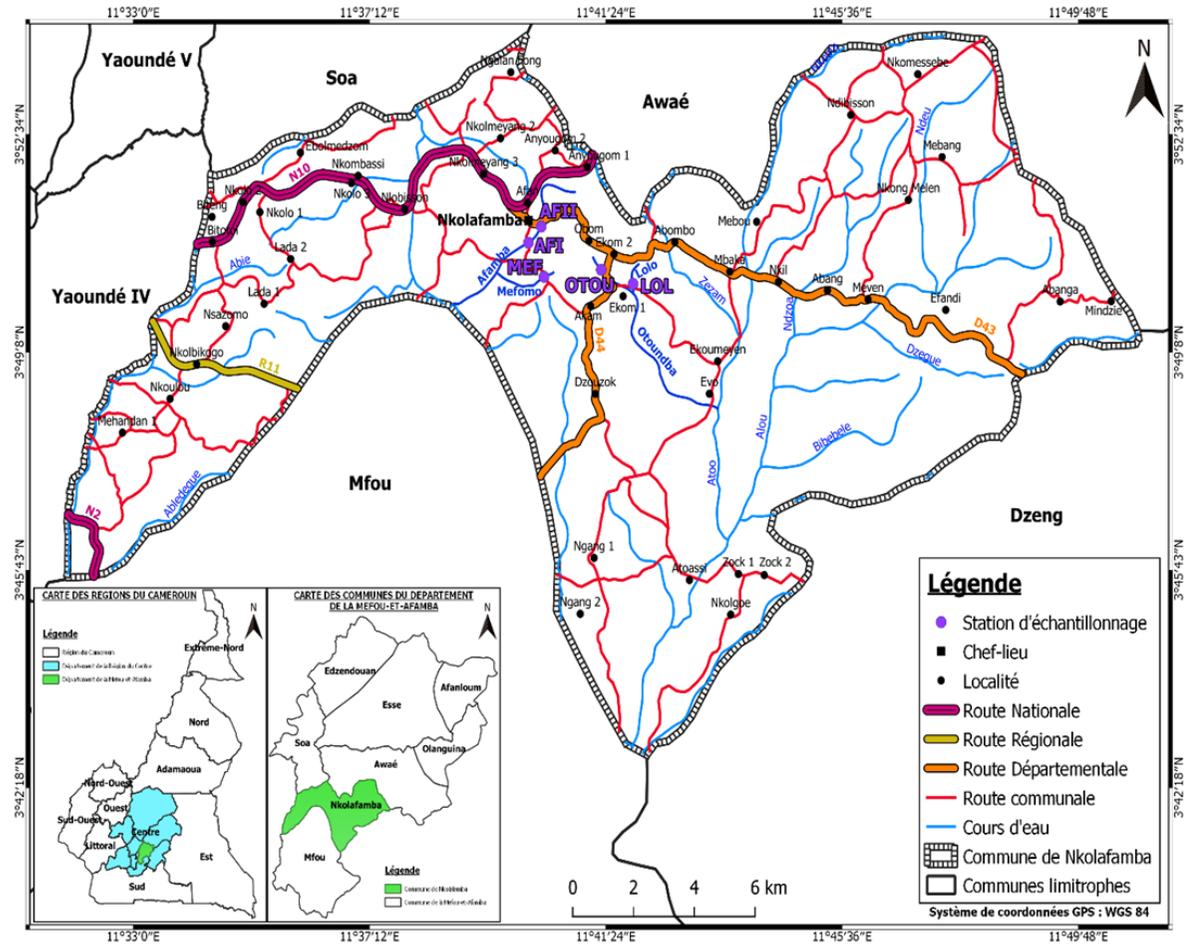


Figure 1: Presentation of the study area and the location of the sampling points.

Table 1: Geographical location, adopted code, and altitude of each sampling point.

Stations	Adopted Codes	Altitudes (m)	Geographical locations
Afamba I	AFI	670	03°43' 16,5``N 11°33' 16,9``E
Mefomo	MEF	698	03°43' 25,7``N 11°33' 33,3``E
Lolo	LO	699	03°43' 16,4``N 11°33' 20,2``E
Afamba II	AFII	667	03°43' 12,3``N 11°33' 21,9``E
Otoutoumba	OTOU	719	03°43' 11,1``N 11°33' 28,5``E

Table 2: Physicochemical parameters measured in the laboratory by spectrophotometer.

Parameters	Method	Reagents used	λ (nm)	Units
SS		-	810	mg/L
Color	Spectrophotometry	-	455	Pt/Co
Turbidity		-	450	FTU
Nitrate (NO_3^-)		NitraVer IV	570	mg/L
Phosphate (PO_4^{3-})		PhosVer	880	mg/L

RESULTS

Analyzes of hydrological variables

Water flow velocity

The highest value of the water flow velocity is recorded at the OTOU station (0.33m / s) followed respectively by MEF (0.22 m / s), AF II (0.155m / s), LO (0.153 m / s) and AF II (0.124 m / s) (Table 3).

Wet section

The wetted section is calculated by the product of the bed width and the average water depth. It varied during the study between 0.84 m² obtained at LO level and 5.22 m² recorded at OTOU. The beds of the five stations are covered with a significant amount of mud and the banks with phanerogam vegetation. Sludge depth was not taken into account because the area is free from industries that could release heavy metals into waterways. Figure 2 shows the bed widths and water depths obtained during the study period.

Flow rate

The maximum value was obtained at OTOU (1.76 m³ / s) and the minimum value is obtained at the level of LO (0.128 m³ / s) (Table 3).

Evaluation of physicochemical parameters

Physical parameters (MES, Temperature, Turbidity, Colour and TDS)

The physical parameters considered during this study varied throughout from campaign to campaign and from station to station and from sampling point to sampling point. The SM values obtained during the study period varied between 0 mg / L (April, Afamba1 station) and 46 mg / L (June at the

Lolo station) (Figure 3A). Solids content fluctuates around an average value of 19.48 ± 14.32 mg / L. The measured temperature values fluctuated between 23 and 28 °C. The highest value was recorded at the station on Mefomo in March. And the lowest value at the station on the Lolo in June. However, an average value of 25.48 ± 1.354 °C is noted (Figure 3B). The highest value for turbidity (97 FTU) was recorded in February at the station on the Lolo. The lowest value (34 FTU), was recorded in June at the same station (Figure 3C) with an average value of 59.933 ± 14.359 FTU. Overall, the water color values hovered between 22 and 437 Pt.Co with an average value of 117.96 ± 89.75 Pt.Co. The lowest value was recorded at Afamba1 and Otoundoumba stations in April and the highest value was recorded at the Lolo station in February (Figure 3D). The TDS values recorded during the study period varied between 15 and 87 mg / L. The highest value was obtained for the station on Lolo in March and the lowest value was recorded at the station on Mefomo in February. However, an average value of 27.266 ± 17.05 mg / L is noted (Figure 3E).

Chemical parameters

Overall, dissolved CO₂ values ranged from 0.2 to 7.04 mg / L. The highest value was recorded at Afamba1 station in June. The lower value was obtained at the station on Mefomo in June. However, an average value of $3,802 \pm 1,491$ mg / L is noted (Figure 4A). The dissolved oxygen contents fluctuated between 50.5 and 75.3%. They reached their maximum value at the station on Mefomo in March and

the minimum value was recorded at Afambal station in March and April. An average value of $63.96\% \pm 8.468$ was noted (Figure 4B). Ammoniacal nitrogen levels are found in the water in the form of traces with levels between 0.09 mg / L (in February at the station on the Mefomo) and 1.8 mg / L (in July at the station on Otoundoumba) (Figure 4C). The pH values fluctuated overall between 5.8 and 8.91 UC. The highest value was obtained at the station on Mefomo in February and the lowest value at the station on Otoundoumba in July (Figure 4D). Nitrate levels are highest in April at Afamba 2 station (20.8 mg / L). The lowest (0.5 mg / L) at the Afamba 1 station in March (Figure 4E). The electrical conductivity values oscillated between 32 and 173 $\mu\text{s} / \text{cm}$. The highest value was recorded at the station on the Lolo in February and June. The lowest value at Afamba 2 station in May (Figure 4F). The levels of orthophosphates in water show irregular variations, with values reaching 21.9 mg / L in March, at the station on the Mefomo. They were scarce throughout the month of April in the Afambal station (Figure 4G).

Microbiological analyzes of sampled water

Overall, the abundances of BHAM, faecal and total coliforms, faecal enterococci varied from station to station and during each campaign (Figure 10). In general, BHAMs were most abundant at all stations throughout the study period (Figure 5A). BHAM densities fluctuated between 4.36 and 6.03 units (log CFU / 100mL water). The lowest abundance was obtained at station AF1 in July and the highest abundance at the station on the Lolo during June. As for the total coliforms, their density varied from 3.69 to 5.71 units (log CFU / 100mL of water). The smallest value of 3.69 units (log CFU / 100mL)

Determination of the origin of microbiological pollution of water

The probable origin of the pollution was determined from faecal coliform to faecal streptococci (CF / SF) ratios (Table 4). The reports having thus been established by sampling station and by month, it can be seen that the pollution is predominantly of animal

origin in the Afambal stations during the months of February, April, May, June, July; Mefomo during all the months of the sampling campaign except June, Lolo during the months of February, March, April, June; Afamba 2 during every month and in Otoundoumba during the months of February, March, May and July. Microbial pollution is linked to a mixed population in Mefomo stations in April, Lolo during the months of May and July and in Otoundoumba station in June. Cases of pollution probably linked to human origin were not observed throughout our sampling campaign.

Assessment of the presence of a pathogenic species (genus *Pseudomonas*)

Qualitative analysis

Macroscopic examination of bacterial colonies on Cetrimide Agar showed three types. Small-sized white colonies, medium-sized white colonies, and large-sized white colonies fluorescent under ultraviolet radiation were observed. The results of the biochemical examinations carried out are presented in Table 5. It appears that the waters of the rivers analyzed mainly harbor *P. aeruginosa* and *P. maltophilia*.

Quantitative analysis

The densities of *Pseudomonas* colonies vary during the study period depending on the site and month of sampling. The highest value (5.462 log CFU / 100mL) and the lowest value (3.39 log CFU / 100mL) were recorded respectively in the AF II station in February and April respectively (Figure 6).

Assessment of the impact of hydrological and physicochemical parameters on the variation of microbial densities

Correlations between hydrological variables and bacterial densities

Significant ($P < 0.05$) and negative correlations were observed between the densities of fecal enterococci and the wetted section. The same observation was made between the total flora (BHAM) and the wetted section (Table 6).

Correlations between physicochemical parameters and bacterial abundances

Correlations between physicochemical parameters and the densities of the isolated bacteria were performed using Spearman's "r" correlation test. It emerges that very significant (P <0.01) and positive correlations were observed between the densities of total coliforms and parameters such as pH, electrical conductivity. The same observation was obtained between the density of faecal streptococci and the nitrate concentration (Table 7). Significant (P <0.05) and positive correlations were recorded on the one hand between fecal coliforms and SM content, between faecal streptococci and pH and between bacteria of the genus *Pseudomonas* and water turbidity (Table 7). A very significant (P <0.01) and negative correlation was noted between faecal streptococci and the content of NH₄⁺ media (Table 7). Significant (P <0.05) and negative correlations were observed between the densities of bacteria of the genus *Pseudomonas* and SS, between total coliforms parameters such as temperature and NH₄⁺ content (Table 7).

Comparisons between the variables considered

The comparison between the physicochemical and microbiological variables during the study period was carried out using the Kruskal-Wallis H test. From this test, it appears that the physicochemical variables such as: pH, electrical conductivity, nitrates, temperature, SS, have significantly varied during the months of sampling (P <0.05). The same observation was made for the densities of bacteria of the genus *Pseudomonas*.

The Mann Withney comparison test was performed between physicochemical

variables, bacterial densities and different months of sampling. It appears that significant differences (P <0.05) between the pH and the months of March, April, May, and June; between the electrical conductivities and the month of February; between nitrates and the months of March, May June; between temperature and the month of March; between MES and the months of March and April; between nitrates and the months of February, March, May, and June. The same observations were made between the abundances of bacteria of the genus *Pseudomonas* and those of February and March. The significance coefficients are presented in Table 8.

Affinities between biotic and abiotic parameters

The PCR applied to the various biological, physicochemical and hydrological variables shows a grouping of the parameters into 3 nuclei (Figure 7). Core 1 (N1) includes Afamba2 and Lolo stations in which BHAMs maintain strong correlations with nitrates, SM, color, and TDS. In the core (N2) encompassing the Otoundoumba and Afamba1 stations, a strong affinity is observed between temperature, flow, speed, wetted section, and dissolved oxygen. However, there is an absence of bacterial species. Regarding the nucleus (N3) encompassing the Mefomo station, there is a strong presence of bacterial abundances such as fecal enterococci, *Pseudomonas*, fecal and total coliforms all correlated with electrical conductivity and pH. The strong presence of microorganisms in this station can be explained by the acidic nature of the aqueous support and the rocky nature of the funds.

Table 3: Hydrological parameters of the sampling stations during the study period.

Parameters	Stations				
	AF I	MEF	LO	AF II	OTOU
Wet Section (m ²)	2.76	1.62	0.84	2.56	5.33
Speed (m/s)	0.155	0.22	0.153	0.124	1.76
outflow	0.42	0.35	0.128	0.317	1.76

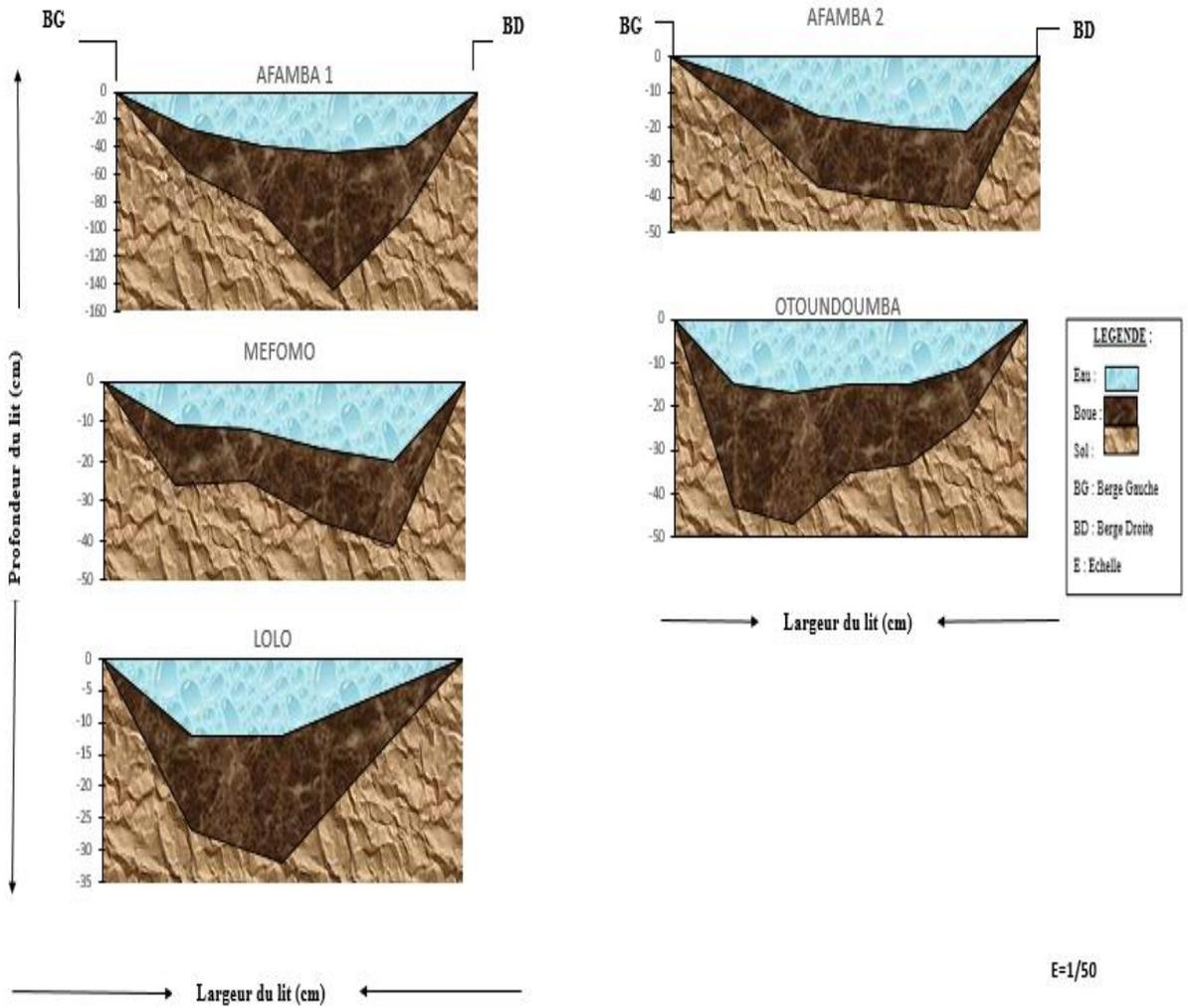


Figure 2: Wet Section of the sampling stations during the study period.

Table 4: presentation of faecal coliform reports on faecal streptococci by sampling station and by month.

Sampling Stations	February	March	April	May	June	July
AFAMBA 1	0	2,95	0	0	0	0
AFAMBA 2	0,060	0,092	0,083	0	0,142	0,142
MEFOMO	0,119	0	1,880	0	0,0714	0,0714
LOLO	0	0	0,004	1,728	0,051	0,051
OTOUNDOUMBA	0	0	2,25	0	1,333	1,333

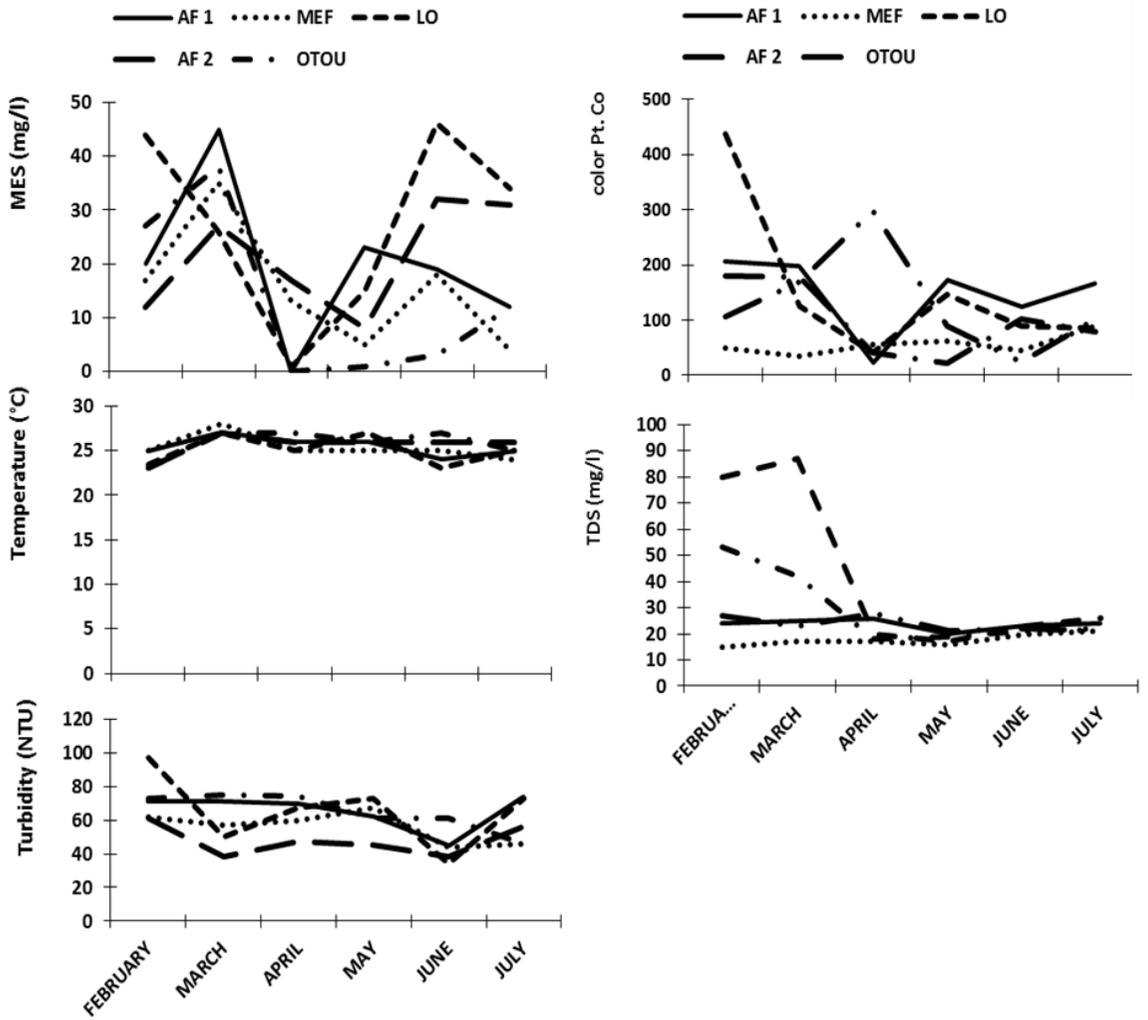


Figure 3: Spatio-temporal variations of the physical parameters measured during the study period (A: SM (suspended matter); B: temperature; C: turbidity, D: color; E: TDS (total dissolved solids).

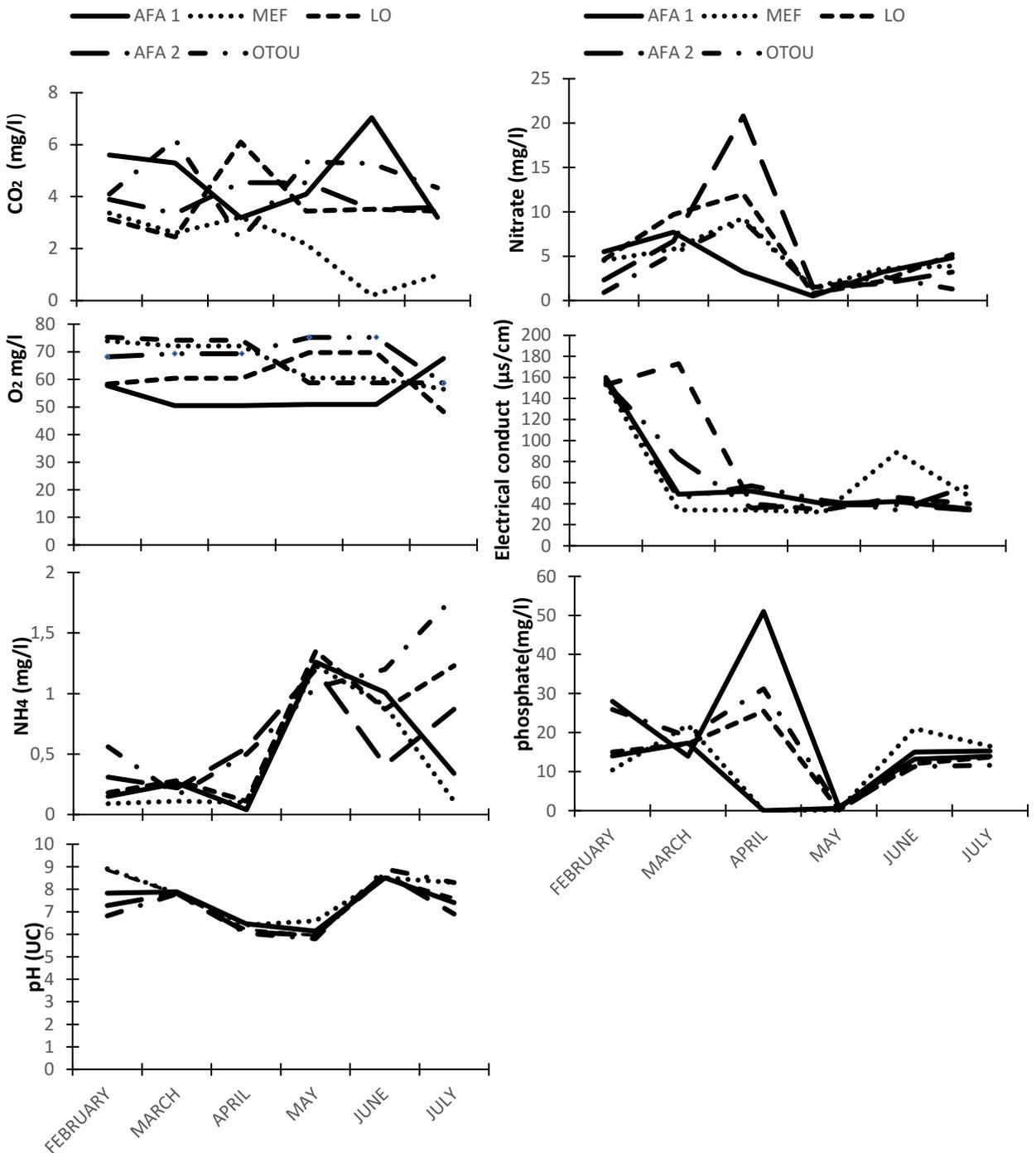


Figure 4: Spatio-temporal variations of the chemical parameters measured during the study period (A: dissolved CO₂; B: dissolved oxygen; C: ammoniacal nitrogen (NH₄⁺), D: pH; E: nitrates (NO₃⁻) E: electrical conductivity; G: orthophosphates (PO₄³⁻).

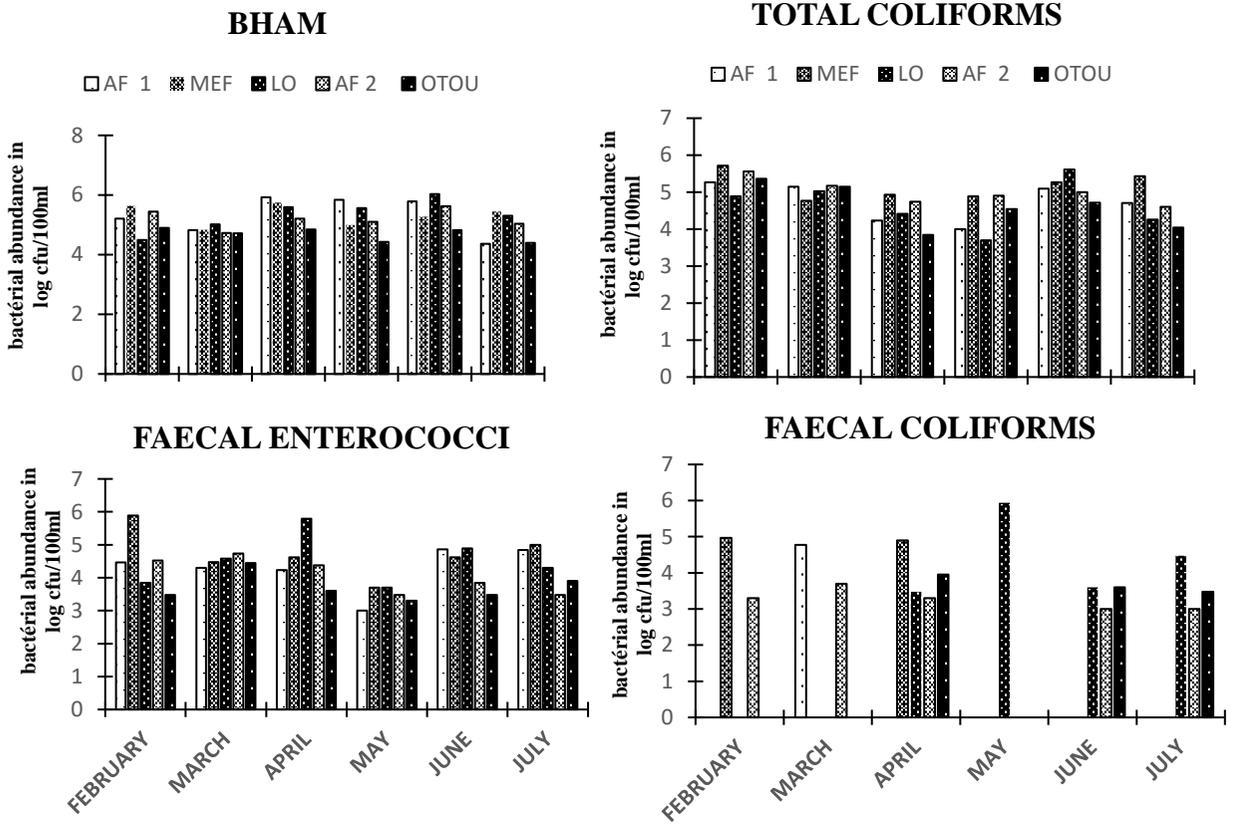


Figure 5: Spatio-temporal evolution of the abundances of bacteria isolated in the Afamba 1, Afamba 2, Lolo, Mefomo and Otoundoumba stations as a function of the months.

Table 5: Results of the identification tests of the strains of *Pseudomonas* isolated.

Identification test	Strains and months														
	Febuary			March		April		May		June		July			
	A	B	C	A	C	B	C	A	C	A	C	A	B	C	
Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
Mobility	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
Lactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
King A	+	+	-	+	-	+	-	+		+	-	+	+	-	
H ₂ S	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
Citrate	+	+	-	+	-	+	-	+	-	+	-	+	+	-	

Catalase	-	-	-	-	-	-	-	-	-	-	-	-	-	-
suspicious species	<i>Ps. aer</i>	<i>Ps. aer</i>	<i>Ps past</i>	<i>Ps aer</i>	<i>Ps aer</i>	<i>Ps past</i>								

(+): positive test; (-): negative test; *Ps. aer*: *Pseudomonas aeruginosa*; *Ps past*: *Pseudomonas pasteurilla*.

Table 6: Correlations between hydrological parameters and bacterial abundances.

hydrological ariables	microbiological Variables				
	FT	PS	SF	CF	CT
speed	-0,308	0,139	-0,123	-0,083	-0,048
outflow	-0,351	0,112	-0,207	-0,129	0,033
wet Section	-0,399*	0,008	-0,436*	-0,236	-0,136

*: P <0.05 P = degree of significance dof = 14, FT: total flora, PS: *Pseudomonas*, SF: faecal streptococci, CF: faecal coliforms, CT: total coliforms.

Table 7: Correlations between physicochemical parameters and bacterial density.

Physiochemical Variables	Microbiological Variables				
	FT	PS	SF	CF	CT
O ₂	-0,230	0,198	0,144	-0,161	0,062
Ph	-0,020	-0,256	0,428*	0,081	0,537**
TDS	-0,272	-0,130	-0,009	-0,148	0,133
Conductivity	-0,051	0,100	0,242	-0,070	0,481**
Colourr	-0,248	0,094	-0,054	-0,137	0,221
Turbidity	-0,293	0,373*	-0,251	-0,054	-0,320
Nitrate	-0,116	-0,051	0,526**	0,198	0,107
Phosphate	-0,259	0,021	0,278	-0,079	0,263
Temperature	-0,304	-0,344	-0,323	0,129	-0,411*
CO ₂	-0,038	0,072	-0,154	-0,038	-0,009
MES	-0,038	-0,401*	0,093	0,025	0,365*
NH ₄ ⁺	-0,117	-0,070	-0,562**	0,073	-0,416*

*: P <0.05 **: P <0.01 P = degree of significance dof = 5, FT: total flora, PS: *Pseudomonas*, SF: faecal streptococci, CF: faecal coliforms, CT: total coliforms **: degree of significance.

Table 8: P values indicating the significance thresholds for the KRUSKAL-WALLIS comparison H test between physicochemical parameters, bacterial abundances and months of sampling.

physicochemical and microbiological variables	Significance during the months
O ₂	0,749
Ph	0,001*
TDS	0,105
Conductivity	0,030*
Colour	0,306
Turbidité	0,186
Nitrate	0,04*
Phosphate	0,101
Temperature	0,016*
CO ₂	0,805
MES	0,016
NH ⁺ ₄	0,004*
BHAM	0,101
Pseudomonas	0,016*
Faecal Entérocoques	0,180
Coliforms faecal	0,509
Totals Coliforms	0,055

* significant at: P <0.05 P = degree of significance dof = 5.

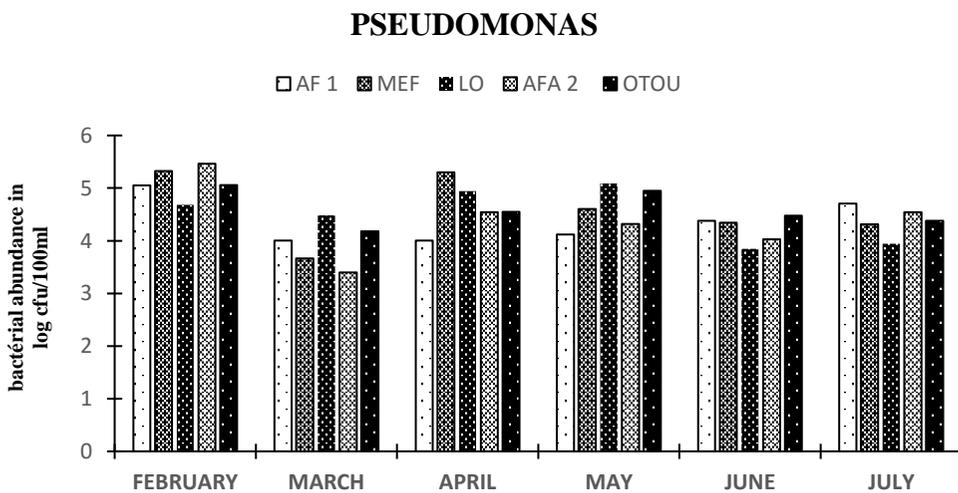


Figure 6: Spatio-temporal variations in the abundance of bacteria.

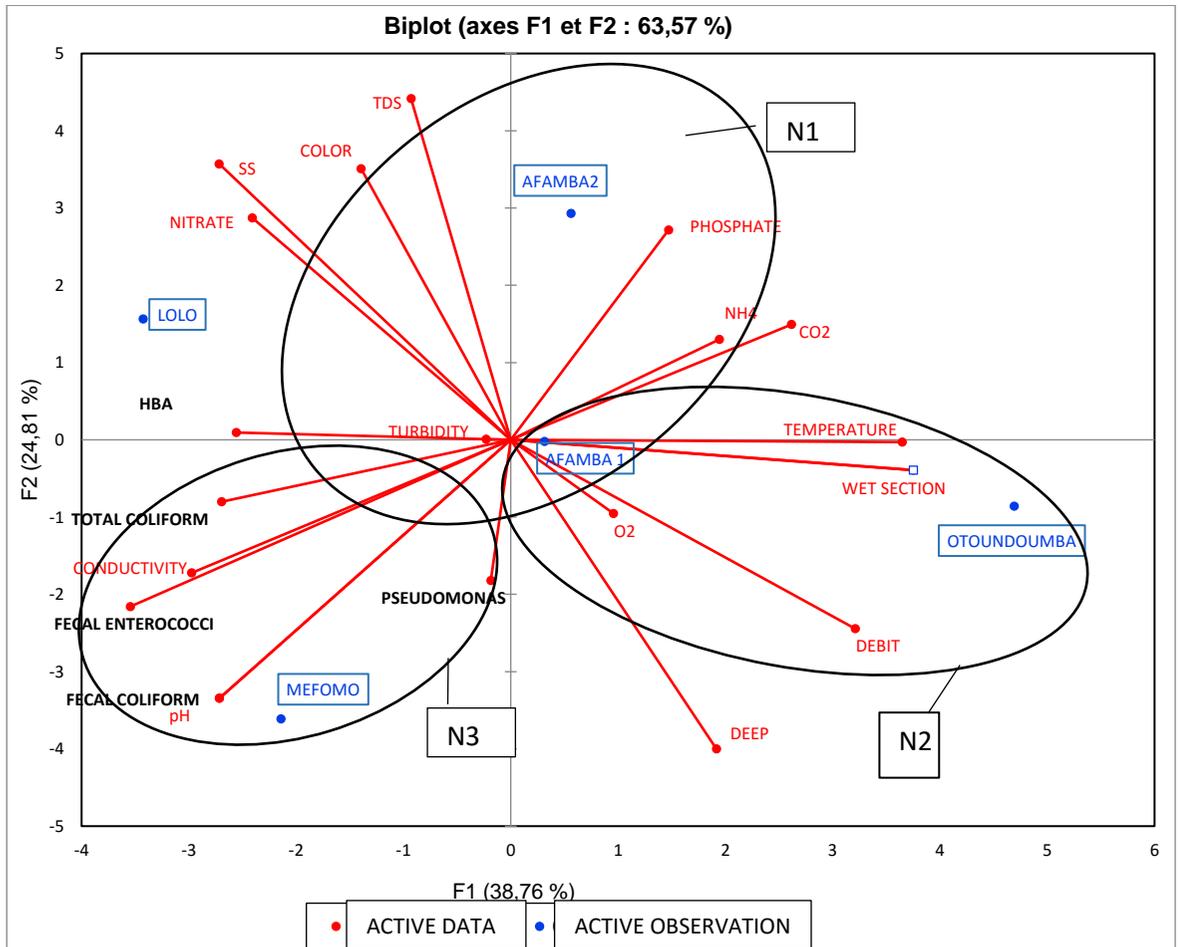


Figure 7: PCR values grouping the affinities between bacterial abundances and physicochemical and hydrological parameters.

DISCUSSION

Hydrological parameters

The low speed at Afamba 2 station (0.124 m / s) could be explained by its location in a marshy area. The increase in flow and in the wetted section at the Otoundoumba station and the low values of these variables observed at the Lolo station are thought to be due to lateral inflows from the various tributaries. Levêque and Balian (2005) point out in this regard that by receiving small tributaries, the wet section is called upon to increase.

Physico-chemical parameters

Temperature values vary little from station to station and at the same site, with an

average of 25.43 °C. This value is compatible with the activity of local organizations. These results are similar to those obtained by Tuékam Kayo (2007) in the waters of the Mfoundi watershed. This low thermal variation could be explained by the low conductivity of the soil (Ait, 2007).

An average pH value of 7.04 ± 1.03 U.C was recorded. The waters analyzed have a pH with an acidic tendency, although a value of 8.91 U.C was recorded in the Mefomo station in February. These values obtained would be linked to the low mineralization, the absence of anthropogenic activities, the activity of microorganisms present in the water and the nature of the soils crossed (Nola et al., 2002).

By considering the quality grid of Nisbet and Verneaux (1970), the stations would present overall a fairly satisfactory saturation in dissolved oxygen (63.96%) during the study period. This fairly good oxygenation of the water can be explained by the fairly large plant cover which promotes strong photosynthetic activity releasing oxygen, conducive to oxygenation of the water at the air / water interface. In addition, the strong presence of rafts in the watercourse promotes the resuspension of oxygen molecules (Stark, 2001). These results agree with the assertions of Nechad et al., (2014), Tchakonté (2016) according to which in forest areas, natural ventilation, the presence of rafts and meanders create conditions of turbulence and mechanical mixing favorable to natural ventilation.

The values of the CO₂ contents oscillate between 0.2 and 7.04 mg / L. According to Rodier et al. (2009), these levels are influenced by the climate and the seasons, as well as by the nature of the soil and vegetation. The low values of the electrical conductivity of the water from the stations can be explained by the low degradation of the organic matter present in the environment and would reflect the little polluted nature of these waters (Ajeegah et al., 2018). The low average values of SS (19.48 mg / L), color (117.96 Pt-Co) and turbidity (59.93 NTU) recorded in the stations are explained by the low load of the water in organic matter and the low intake of allogenic materials in water bodies. The low values of these parameters are similar to those obtained by Foto Menbohan et al. (2013) on the Nga and Tchakonté et al. (2016) in the peri-urban area of Douala. The SS values recorded would reflect an absence of anthropogenic pollution. The value of 46 mg / L observed in June at the Lolo station could be explained on the one hand by the nature of the substrate and on the other hand by the decomposition of the organic matter and in particular of the litter which releases the humic acids and colloidal substances. The maximum value of turbidity in February in Lolo could be due to the return of the rains. The relatively low concentrations of nitrate, (0.5-20.8 mg / L) of ammoniacal nitrogen (0.04 - 1.8 mg / L) and

orthophosphates (0 -21.9 mg / L) would be due to the low mineralization of waters and the little anthropized nature of the study area. This result is similar to that obtained by Tuekam Kayo (2007).

Biological parameters

In general, BHAM were present at all stations throughout the study period and dominate the identified bacterial community. Indeed, according to Levallois and Levesque (2003), the enumeration of the aerobic bacterial flora aims to estimate the density of the general bacterial population. The high abundance could be due to the fact that the environment of these stations is favourable for their development. In addition, the high bacterial load of BHAM recorded could also be due to contaminated runoff. According to Foster and Salas (1991) this factor favors the contamination of surface and underground water, causing bacteria to move. This contamination, however, depends on the pollutant load of the contaminant and the permeability of the overlying soil.

Among the main groups of bacteria indicative of faecal contamination, those represented are: total coliforms, faecal coliforms and faecal streptococci. The abundances of these groups of bacteria have varied from station to station and over time. The mean values of the abundances recorded are 4.93; 1.92 and 4.25 log CFU / 100 mL for fecal coliforms, total coliforms and faecal streptococci, respectively. These values are all above the standards set by the WHO. This therefore indicates a deterioration in the bacteriological quality of the water (WHO, 2004). This predominantly animal contamination occurs upstream in the Afamba stream which serves as a watering point for herds in the locality of Obala (Lekié Department, Center Region). The waters of Nkolafamba also harbor a pathogenic microflora, with in particular bacteria such as *P. aeruginosa*, *P. pasteurilla*, and the abundances of which can sometimes reach 5.462 log CFU / 100 mL. These germs can cause eye infections or sepsis on users of these waters (WHO, 2004). The permanent presence

of these pathogenic bacteria and their high abundance reflects the degree of pollution of these waters. Similar results have been obtained in particular by Mpakam et al. (2006) in Bafoussam where many pathogens such as *Escherichia coli*, *Salmonella*, *Shigella* and many other bacteria witnessing faecal contamination have been observed there. The same is true of the results obtained by Nanga et al. (2014) in Yaoundé and Ndjama et al. (2011) in Douala. These authors, having analyzed the drinking water, isolated bacteria indicative of faecal contamination and strict pathogenic and opportunistic bacteria.

Links between the evaluated parameters

The results of the correlations between the physicochemical and biological variables show that among the physicochemical parameters analyzed, certain variables significantly influenced the population and the distribution of bacteria throughout the study. Increasing the pH of the water significantly increases the abundance of fecal and total coliforms, and fecal enterococci. In this regard, Nola et al. (2002) are of the opinion that in a given medium, increases in pH sometimes favor the development of *Pseudomonas aeruginosa*, *Aeromonas hydrophila*, as well as the abundance of fecal coliforms and faecal streptococci.

The level of nitrate and SM which is significantly positively correlated with the abundance of faecal enterococci. The increase in the content of SM leads to a significant increase in the density of fecal coliforms and a significant decrease in bacteria of the genus *Pseudomonas*. This difference would result in the fact that bacteria react differently to organic matter and depending on the composition of the latter. Indeed, organic matter influences the availability of nutrients while serving at the same time as a source of energy and carbon for certain microorganisms (Nola et al., 2004).

Increasing the temperature significantly decreases the abundance of bacteria of the genus *Pseudomonas*. This could be explained by the fact that solar radiation has a bactericidal action on aquatic bacteria (Lopez-Pila and Szewzyk, 2000). They also have the effect of

warming the water and consequently contribute to the decrease in the number of microorganisms (Rodier et al., 2009). However, these results are similar to those of the work of Seni (2013) which showed that a high temperature would induce in bacteria alterations in cellular structures, thus promoting entry into a viable but non-cultivable state.

COMPETING INTERESTS

The authors declare that there is no competing interests.

AUTHORS' CONTRIBUTIONS

The coaches contributed to the design and planning of the experiments. SDB, AAT, CSM, YPY performed analyzes under the supervision of OVNE, MN. OVNE, SDB, RTK and DE wrote the manuscript. All authors have approved the final version of the manuscript.

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