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# Bacteriological Analyses and Physicochemical Parameters of Water from Highly Used Boreholes in the City of Ebolowa, Cameroon

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**Key words**

Bacteriological quality, Physico-chemical parameters, Borehole water, Ebolowa, Cameroon

**Abstract**

Rapid urbanization in Ebolowa (South Cameroon) has increased dependence on public and community boreholes for drinking water, yet their microbiological safety remains poorly documented. This study evaluated the bacteriological and physicochemical quality of water from four highly used boreholes in Ngalane, New Bell, Nko'ovos, and Ebolowa Si I. Boreholes were georeferenced using GPS and mapped with QGIS 3.34.1. Water samples were analyzed for total coliforms (TC) and *Escherichia coli*, while temperature, pH, total dissolved solids (TDS), and electrical conductivity were measured in situ. Differences among sites were assessed using one-way ANOVA with Tukey's test, and Spearman's correlation evaluated relationships between physicochemical parameters and bacterial loads. Ngalane showed the highest contamination, with *E. coli* and TC levels of 7,000 ± 1,000 and 35,666.67 ± 1,527.53 CFU/100 mL, respectively. New Bell showed moderate contamination, whereas Nko'ovos and Ebolowa Si I had no detectable *E. coli* and low TC levels. Significant spatial differences were observed ($p < 0.05$), with site-specific associations between physicochemical parameters and TC concentrations. These results reveal marked variability in groundwater quality and highlight the need for regular microbiological monitoring, borehole disinfection, and improved sanitation to protect public health.

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## 1. Introduction

Water resources comprise both surface water (rivers, lakes, reservoirs, and streams) and groundwater stored in phreatic, karstic, and alluvial aquifers. These resources are indispensable for ensuring freshwater availability and supporting socio-economic development, particularly in rapidly urbanizing regions (Bănăduc et al., 2022; World Health Organization [WHO], 2022). Water is essential for domestic activities, including drinking, cooking, bathing, and sanitation, as well as for agricultural and industrial production (Ahmed et al., 2025; WHO, 2022). According to the World Health Organization (WHO, 2022), safe drinking water is defined as water that poses no significant risk to human health throughout a lifetime of consumption. Achieving this objective requires compliance with microbiological, chemical, and physicochemical quality standards, while international standards such as ISO 24510, ISO 24511, and ISO 24512 provide guidance for the management and delivery of drinking water and sanitation services (Tsaridou & Karabelas, 2021). Despite substantial progress in improving access to drinking water worldwide, millions of people in low- and middle-income countries, including Cameroon, continue to depend on water sources that remain vulnerable to microbial contamination, making access to safe drinking water a persistent public health concern (United Nations Children's Fund [UNICEF] & WHO, 2021).

In Cameroon, groundwater extracted from boreholes has become an increasingly important source of drinking water, particularly in urban and peri-urban areas where piped water supplies are insufficient to meet growing demand. Although boreholes are generally regarded as safer than surface water and traditional wells, they remain susceptible to contamination, especially in areas characterized by inadequate sanitation infrastructure and uncontrolled urban expansion. Microbiological contamination of groundwater has been associated with numerous waterborne diseases, including cholera, typhoid fever, dysentery, and hepatitis A, thereby representing a major public health challenge (Fokou Bomgning et al., 2022; Sone et al., 2024). Several studies have evaluated groundwater quality in different regions of Cameroon; however, most have focused either on specific geographical areas or on limited physicochemical or microbiological parameters. Consequently, comprehensive information on groundwater quality in Ebolowa remains scarce (Nguimfack et al., 2022). Furthermore, few investigations have integrated microbiological and physicochemical analyses with a detailed spatial characterization of heavily used boreholes, limiting the understanding of environmental factors that may influence groundwater quality in this rapidly expanding city.

To address this knowledge gap, the present study provides an integrated assessment of the microbiological and physicochemical quality of water from heavily used boreholes in Ebolowa, complemented by a descriptive spatial analysis of borehole characteristics. Geographic Information System (GIS) tools were used to map borehole locations and document environmental features, including proximity to residential areas, borehole age, and disinfection practices. The primary objective of this study was to evaluate the microbiological quality of borehole water through the determination of total coliforms (TC) and *Escherichia coli*, together with key physicochemical parameters. By generating site-specific evidence on groundwater quality, this study seeks to support local authorities and public health stakeholders in strengthening water quality surveillance, improving groundwater management, and reducing the risk of waterborne diseases.

## 2. Materials and Methods

### 2.1. Study Area

The study was conducted in Ebolowa, the administrative capital of the South Region of Cameroon. Located in the Mvila Division, the city is bordered by Ambam to the south, Mengong to the north, Biwong-Bané to the west, and Mvangan to the east. According to the National Institute of Statistics, Ebolowa has an estimated population of approximately 180,000 inhabitants distributed over an area of 56 km² (National Institute of Statistics [INS], 2021).

Over recent decades, Ebolowa has experienced substantial demographic growth, leading to increased population density within the urban area. This accelerated urban development has resulted in the expansion of densely populated neighborhoods, potentially influencing both water demand and the vulnerability of groundwater resources to contamination. The population consists mainly of indigenous communities, particularly the Bulu ethnic group, alongside other socio-cultural groups, including the Bamiléké, Bamoun, and Eton communities, contributing to the city's demographic diversity (Ngoh, 2014).

The climate of Ebolowa is classified as tropical monsoon, characterized by alternating rainy and dry seasons. Average daily temperatures generally range between 27 °C and 30 °C, whereas nighttime temperatures vary between 19 °C and 22 °C (Météo Cameroun, 2020). However, climate variability associated with global climate change has increasingly affected the predictability of seasonal patterns and rainfall distribution (IPCC, 2023).

The relief of Ebolowa is characterized by gently undulating plateaus, forest-covered hills, savannah ecosystems, and narrow valleys. These environmental characteristics may influence groundwater recharge processes and the physicochemical and microbiological dynamics of borehole water.
This geographical, climatic, and demographic context provides the necessary framework for understanding potential factors influencing the quality of heavily exploited groundwater resources in the city.

### 2.2. Study Design and Ethical Authorization

A cross-sectional, descriptive, and analytical study was conducted from May 26 to June 26, 2025, to assess the physicochemical and microbiological quality of borehole water used by local populations in Ebolowa.
The study aimed to characterize water quality parameters and evaluate potential relationships between physicochemical characteristics and microbiological contamination, with particular emphasis on total coliforms (TC) as indicators of microbiological safety.
Authorization for field sampling and physicochemical measurements was obtained from the Sub-Prefect of the First District of Ebolowa following an official request submitted by the Director of La Grâce Private School in Ebolowa (Authorization No. 00000006/ACD/BAG). Laboratory analyses were authorized by the Director-General of the Pasteur Center of Cameroon in Yaoundé (Authorization No. 000823/2025/CPC/DS/DAF/CJ/SRH).

### 2.3. Sampling Sites and Selection Criteria

Sampling sites were selected using a purposive sampling approach based on their importance for community water supply.
Selection criteria included frequency of use, accessibility, and relevance as drinking water sources for local populations. Site selection was guided by approximately two months of field observations, complemented by information obtained from residents regarding water-use practices and historical utilization patterns.
Four heavily frequented boreholes located in the neighborhoods of Ngalane, Nko'ovos, Ebolowa Si I, and New Bell were selected as representative sampling points.

### 2.4. Sample Collection and Preservation

Water sampling was conducted over a one-week period. For each borehole, three samples were collected: one sample dedicated to physicochemical analyses and two samples intended for microbiological examination.
Sampling was performed each morning between 8:00 and 11:00 a.m., with a one-day interval between successive collections to ensure adequate sample management and traceability.
For microbiological analyses, water samples were collected aseptically into sterile 1-L bottles containing sodium thiosulfate to neutralize potential residual chlorine. Samples were labeled with the sampling location, date, and collection time.
Samples were transported under refrigerated conditions ($5 \pm 3$ °C) and analyzed within 24 hours after collection to preserve microbiological integrity.
Physicochemical measurements requiring immediate analysis were performed directly at the sampling sites. For laboratory physicochemical analyses, sterile 500-mL containers were used according to standardized water sampling recommendations (International Organization for Standardization [ISO], 2024).

### 2.5. Physicochemical Analysis

Physicochemical parameters were measured using calibrated analytical instruments, including pH meters, conductivity meters, multiparameter devices for temperature and total dissolved solids (TDS), photometers, and standardized reagents for ionic and nutrient determination.
Measurements were performed according to manufacturer instructions. Instruments were calibrated before analysis using appropriate reference solutions to ensure accuracy and reliability.
The investigated parameters included temperature, pH, electrical conductivity, and total dissolved solids.
Measured values were compared with Cameroonian drinking water quality standards (NC 207:2014-48), including recommended limits of $\leq 25$ °C for temperature, $\leq 1000$ µS/cm for electrical conductivity, and $\leq 1000$ mg/L for total dissolved solids.

## 2.6. Microbiological Analysis of Total Coliforms

### 2.6.1. Laboratory Processing and Analytical Framework

Total coliform (TC) analyses were performed at the Pasteur Center of Cameroon within the Hygiene and Environment Service (SHE). Microbiological analyses were conducted using membrane filtration methods according to standard procedures for drinking water quality assessment. Samples were analyzed in duplicate, and additional replication was performed when necessary to improve analytical reliability and reproducibility. The absence of detectable coliform bacteria was expressed as 0 CFU/100 mL, corresponding to acceptable microbiological quality according to Cameroonian drinking water standards (Agence des Normes et de la Qualité [ANOR], 2014). These criteria are consistent with international recommendations requiring the absence of fecal indicator organisms, particularly *Escherichia coli*, in drinking water (World Health Organization, 2022). Values reported as "ND" (Not Determined) indicate that no valid measurement was obtained for the corresponding parameter.

### 2.6.2. Culture Media and Preparation

Three culture media were used for bacterial detection, enumeration, and identification. Chromogenic Coliform Agar (CCA) was used as a selective and differential medium for the detection and presumptive identification of coliform bacteria, including *Escherichia coli*. The chromogenic substrates allow differentiation according to colony coloration, with *E. coli* generally producing blue-violet colonies and other presumptive coliform bacteria producing pink to red colonies. Tryptic Soy Agar (TSA) was used as a general-purpose isolation medium to support bacterial growth and obtain pure cultures from presumptive coliform colonies.Peptone Saline Buffer (PSB) was used as a diluent for sample preparation and serial dilution procedures.The combination of chromogenic media for differentiation and TSA for purification is consistent with recommended microbiological approaches for drinking water quality assessment (Azise et al., 2020).

### 2.6.3. Membrane Filtration and Incubation Procedures

The membrane filtration technique was applied for the concentration and enumeration of microorganisms present in water samples. Water samples were filtered through sterile membranes with a pore size of 0.45 µm. When required, serial decimal dilutions were prepared using Peptone Saline Buffer. Following filtration, membranes were transferred onto Chromogenic Coliform Agar plates and incubated at 36 ± 2 °C for 21–24 hours. Quality control procedures were implemented throughout the analytical process to minimize contamination risks and ensure reproducibility of results. The methodology followed internationally accepted procedures for microbiological examination of drinking water (Quansah et al., 2025).

### 2.6.4. Enumeration of Total Coliforms

Total coliform enumeration was performed by counting characteristic colonies developed on CCA medium after incubation. Pink to red colonies were considered presumptive non-*Escherichia coli* total coliforms, whereas blue-violet colonies were considered presumptive *E. coli* colonies. Only plates presenting countable colony numbers were considered for quantitative analysis.

Results were expressed as colony-forming units per 100 mL (CFU/100 mL) according to the following equation:

$$CFU/100mL = \frac{N \times 100}{V \times D}$$

where:

- **N** = number of colonies counted;
- **V** = filtered sample volume (mL);
- **D** = dilution factor.

### 2.6.5. Identification of Total Coliforms

Presumptive identification of total coliforms was based on colony morphology and coloration on CCA medium. Colonies displaying typical pink to red coloration were subcultured onto TSA medium to obtain pure isolates. After purification, isolates were subjected to biochemical confirmation using oxidase testing and the API 20E identification system. The API 20E system was used for biochemical characterization of presumptive coliform isolates according to the manufacturer's recommendations and established identification procedures (Mwakosya, 2025).

#### 2.6.6. Oxidase Test and Biochemical Confirmation

The oxidase test was performed to differentiate oxidase-positive bacteria from oxidase-negative Enterobacteriaceae, including members of the coliform group. Purified colonies obtained from TSA medium were subjected to oxidase testing. Oxidase-negative isolates were considered compatible with presumptive coliform bacteria and were selected for further biochemical identification. API 20E biochemical profiles were interpreted according to the manufacturer's identification scheme to confirm bacterial identity.

### 2.7. Quality Assurance and Quality Control

Quality assurance procedures were applied throughout all stages of the study, including sampling, transportation, physicochemical measurements, and microbiological analyses. Sampling materials were sterilized before use, and aseptic techniques were maintained during microbiological procedures. Physicochemical instruments were calibrated before measurements according to manufacturer recommendations. Microbiological analyses included duplicate measurements and additional repetitions when required to verify consistency and reproducibility. All procedures were performed following good laboratory practices.

### 2.8. Statistical Analysis

Data were initially entered and organized using Microsoft Excel and subsequently analyzed using IBM SPSS Statistics software (version 25). Descriptive statistics were calculated to summarize physicochemical parameters and microbial concentrations, including means and standard deviations. Differences in total coliform and *Escherichia coli* concentrations among sampling sites were evaluated using one-way analysis of variance (ANOVA). Tukey's post hoc test was applied when significant differences were detected. Relationships between physicochemical parameters (temperature, pH, electrical conductivity, and total dissolved solids) and microbial contamination indicators (TC and *E. coli*) were assessed using Spearman's rank correlation coefficient ($\rho$), considering the distribution characteristics of the data. A significance level of $p < 0.05$ was considered statistically significant, while $p < 0.01$ was considered highly significant. Values reported as "ND" corresponded to parameters for which statistical calculation was not possible because of insufficient variability or constant values among replicates.

### 2.9. Spatial Analysis and Mapping

The geographical distribution of sampling sites was analyzed using QGIS Desktop software (version 3.34.1). Spatial coordinates of borehole locations were integrated into a geographic information system to generate maps illustrating the distribution of sampling points within Ebolowa. This spatial representation facilitated visualization of sampling coverage and supported interpretation of potential spatial differences in water quality characteristics.

## 3. Results

### 3.1. Description of the Study Sites

Water samples were collected from four heavily used boreholes located in the neighborhoods of Ngalane, New Bell, Nko'ovos, and Ebolowa Si I in the city of Ebolowa. The geographical coordinates of each sampling site were recorded using a handheld Global Positioning System (GPS), enabling the spatial mapping of the investigated boreholes. In addition to geographic positioning, field observations were conducted to document the surrounding environmental characteristics, including the proximity of residential areas, sanitation conditions, borehole age, disinfection practices, and patterns of water use (Figure 1). As illustrated in Figure 1, all investigated boreholes were equipped with basic sanitation infrastructure, although important differences were observed in their management and environmental settings. Three of the four boreholes had been constructed within the last decade, reflecting the recent expansion of groundwater supply infrastructure in Ebolowa. In contrast, the New Bell borehole, approximately 15 years old, represented the oldest water point included in the study.

Marked differences were also observed in maintenance practices among the sampling sites. While the Ngalane, Nko'ovos, and Ebolowa Si I boreholes underwent routine disinfection at varying intervals (weekly, monthly, or every three months), no regular disinfection was reported for the New Bell borehole despite its intensive daily use. The absence of routine disinfection may increase the persistence and proliferation of microorganisms within the water distribution system, thereby compromising microbiological water quality.

The environmental context of the boreholes also varied considerably. Two boreholes were located in close proximity to residential buildings, a condition that may increase the risk of groundwater contamination through the infiltration of domestic wastewater or leakage from on-site sanitation facilities. Particularly noteworthy was the New Bell borehole, which is situated downstream of residential areas where surface runoff and household drainage may facilitate the transport of microbial contaminants into the aquifer. Such environmental conditions represent potential pathways for the introduction of fecal microorganisms into groundwater.

Conversely, although the Ngalane borehole is located farther from nearby dwellings, it exhibited environmental characteristics suggesting that groundwater quality may also be influenced by factors beyond the immediate surroundings. Historical contamination sources, legacy sanitation infrastructure, former agricultural activities, abandoned waste disposal sites, local hydrogeological conditions, soil permeability, and groundwater flow dynamics may all contribute to the long-term microbiological quality of borehole water.

Overall, these observations demonstrate substantial heterogeneity in environmental conditions and management practices among the investigated boreholes. Such differences are expected to influence groundwater vulnerability to microbial contamination and therefore provide an important context for interpreting the microbiological and physicochemical results presented in the following sections.

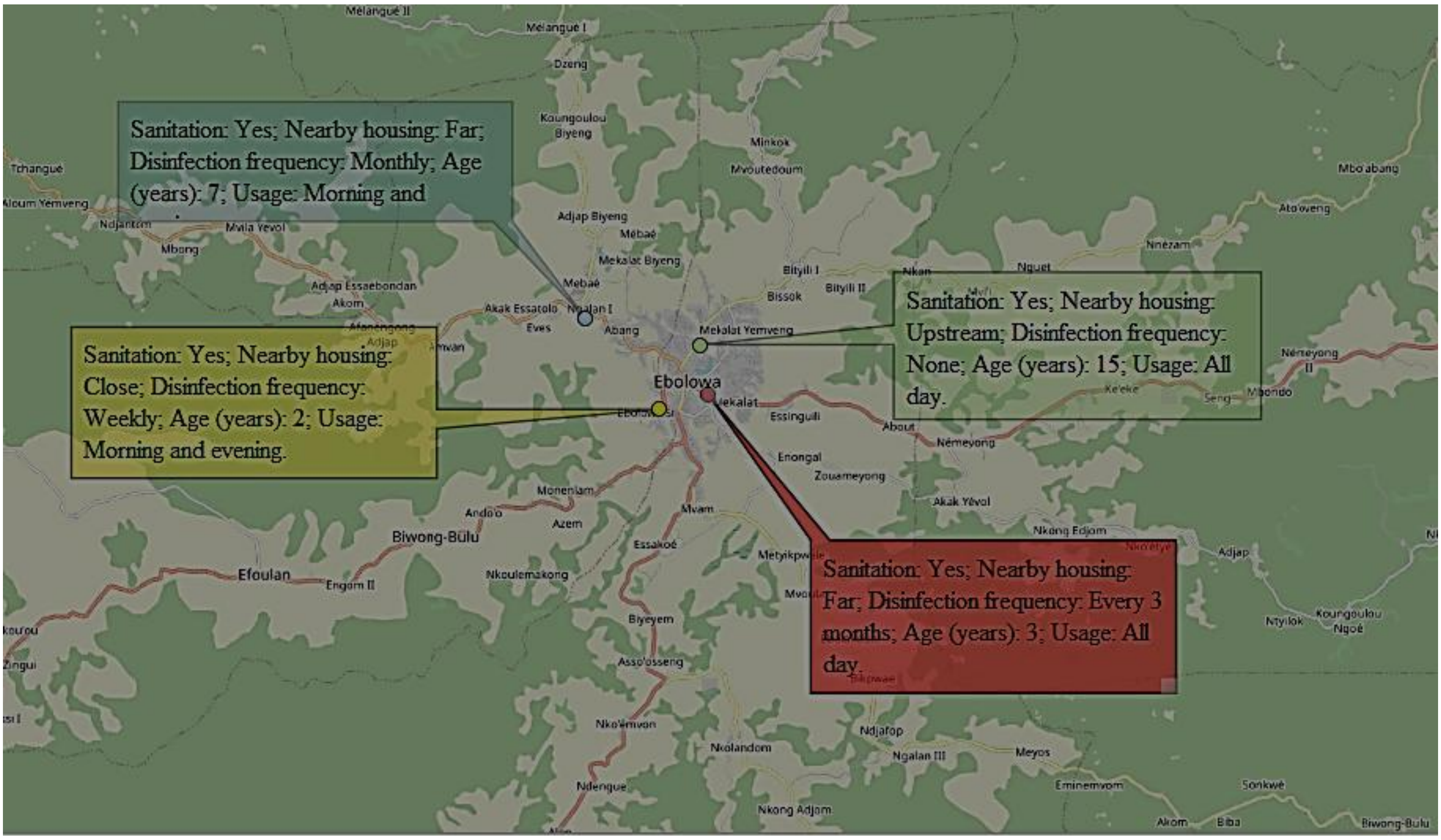


**Figure 1.** Spatial distribution and environmental characteristics of the investigated boreholes in Ebolowa, Cameroon**.** The map shows borehole location, sanitation status, proximity to residential areas, disinfection frequency, borehole age, and water-use patterns. Geographic coordinates were recorded using a handheld GPS and mapped using QGIS Desktop version 3.34.1. Source: Field survey, Ebolowa, Cameroon, 2025.2. Characterization of total coliforms

### 3.2. Microbiological Characterization of Borehole Water

The microbiological quality of borehole water was evaluated by determining the concentrations of total coliforms (TC) and *Escherichia coli* (E. coli), two internationally recognized indicators of microbial water quality and fecal contamination. The mean bacterial concentrations, expressed as colony-forming units per 100 mL (CFU/100 mL), together with their corresponding standard deviations, are presented in Table 1. Superscript letters indicate statistically significant differences among sampling sites, while the F- and *P*-values summarize the results of the one-way analysis of variance (ANOVA).

The results revealed marked spatial heterogeneity in microbiological contamination among the four boreholes. The Ngalane borehole exhibited by far the highest bacterial loads, with mean concentrations of 7,000 ± 1,000 CFU/100 mL for *E. coli* and

35,666.67 ± 1,527.53 CFU/100 mL for total coliforms. These values were significantly higher than those recorded at the other sampling sites ($P = 0.001$), indicating severe microbiological contamination and suggesting substantial fecal pollution of the groundwater. According to internationally accepted drinking-water quality standards, the detection of *E. coli* in any drinking-water sample indicates fecal contamination and renders the water unsuitable for human consumption without prior treatment. The exceptionally high TC concentrations further support the hypothesis of extensive microbial contamination within the aquifer supplying this borehole.

In contrast, the Ebolowa Si I and Nko'ovos boreholes exhibited excellent microbiological quality with respect to fecal contamination. *Escherichia coli* was not detected in any of the analyzed samples, while total coliform concentrations remained extremely low, averaging 0.67 ± 0.58 CFU/100 mL and 2.33 ± 0.58 CFU/100 mL, respectively. These findings suggest limited bacterial contamination and are consistent with the regular maintenance and disinfection practices implemented at these sites.

The New Bell borehole showed intermediate levels of contamination. Mean concentrations reached 200 ± 100 CFU/100 mL for *E. coli* and 1,500 ± 500 CFU/100 mL for total coliforms. Although considerably lower than those observed at Ngalane, these bacterial loads remain indicative of microbiological contamination and suggest that this groundwater source is also unsuitable for direct human consumption. The absence of routine disinfection, together with the borehole's location relative to surrounding residential areas, may partly explain the observed microbial contamination.

Statistical analysis confirmed highly significant differences in bacterial concentrations among the investigated boreholes. One-way ANOVA yielded $F = 142.9$ for *E. coli* and $F = 1438.4$ for total coliforms, with $P = 0.001$ in both cases, demonstrating that the observed spatial variation in microbiological quality was unlikely to have occurred by chance. Despite the relatively small number of replicates ($n = 3$ per site), the large effect sizes and low within-site variability resulted in clear statistical discrimination among sampling locations.

Overall, the microbiological results indicate substantial differences in groundwater quality across the study area. The distribution of bacterial contamination appears to reflect local environmental conditions and borehole management practices rather than a uniform pattern throughout the city. Boreholes subjected to regular maintenance and disinfection generally exhibited very low bacterial counts, whereas sites lacking routine sanitary management or potentially influenced by local contamination sources showed considerably higher microbial loads. These findings highlight the spatial variability of groundwater microbial quality in Ebolowa and emphasize the importance of continuous monitoring and site-specific management strategies to ensure the microbiological safety of drinking-water supplies.

**Table 1.** Mean concentrations (± standard deviation) of *Escherichia coli* and total coliforms (TC) in borehole water from the four sampling sites in Ebolowa.

| | SITES | | | | | |
|---|---|---|---|---|---|---|
| | **Ebolowa si I** | **New-Bell** | **Ngalane** | **Nko'ovos** | **F** | **P-value** |
| Means and Standard Deviations of *E. coli* Concentrations (CFU/100 ml) for Each Site | 0.00 ± 0.00$^{c}$ | 200 ± 100$^{b}$ | 7000 ± 1000$^{a}$ | 0.00 ± 0.00$^{c}$ | 142.9 | 0.001 |
| Means and Standard Deviations of TC Concentrations (CFU/100 ml) for Each Site | 0.67 ± 0.577$^{b}$ | 1500.00 ± 500.000$^{b}$ | 35666.67 ± 1527.525$^{a}$ | 2.33 ± 0.577$^{b}$ | 1438.4 | 0.001 |

Values sharing different superscript letters are significantly different according to Tukey's post hoc test following one-way ANOVA ($P < 0.05$). Results are expressed as colony-forming units per 100 mL (CFU/100 mL). Data are presented as mean ± standard deviation ($n = 3$). Source: Centre Pasteur du Cameroun, Yaoundé, 2025; field survey, Ebolowa, 2025.

#### 3.3. Relationships Between Physicochemical Parameters and Microbiological Contamination

To investigate the potential influence of water physicochemical characteristics on microbial contamination, Spearman's rank correlation analysis was performed between the measured physicochemical parameters (temperature, pH, total dissolved solids [TDS], and electrical conductivity [EC]) and the concentrations of **total coliforms (TC)** and ***Escherichia coli*** at each sampling site (Tables 2–5). Because of the limited number of observations ($n = 3$ per site), the correlation coefficients should be interpreted with caution and considered indicative of site-specific trends rather than definitive causal relationships.

#### 3.3.1. Ngalane Borehole

At the Ngalane borehole (**Table 2**), *E. coli* exhibited a perfect positive correlation with pH ($r$ = +1.000), suggesting that slight increases in pH were associated with higher *E. coli* concentrations. Conversely, moderate negative correlations ($r$ = –0.500) were observed between *E. coli* and temperature, TDS, and EC, indicating that increases in these parameters coincided with lower bacterial concentrations.
For total coliforms, a different pattern emerged. TC showed perfectly positive and statistically significant correlations with both TDS ($r$ = +1.000, $P < 0.01$) and EC ($r$ = +1.000, $P < 0.01$), whereas moderate negative correlations ($r$ = –0.500) were observed with temperature and pH. These findings suggest that greater water mineralization, reflected by increased TDS and electrical conductivity, was associated with higher total coliform concentrations at this site.
Given the exceptionally high bacterial loads observed at Ngalane, these relationships may reflect the combined influence of groundwater mineralization and localized contamination sources rather than the direct effect of individual physicochemical variables.

**Table 2.** Spearman correlation coefficients between physicochemical parameters and microbial indicators at the Ngalane borehole.

| | | T° (°C) | pH | TDS (mg/L) | EC (µs/Cm) |
|---|---|---|---|---|---|
| *E. coli* (CFU/100ml) | Correlation coefficient | -0.500 | +1.000 | -0.500 | -0.500 |
| | N | 3 | 3 | 3 | 3 |
| TC (CFU/100ml) | Correlation coefficient | -0.500 | -0.500 | +1.000** | +1.000** |
| | N | 3 | 3 | 3 | 3 |

#### 3.3.2. New Bell Borehole

Correlation analysis for the New Bell borehole (**Table 3**) revealed moderate positive associations between *E. coli* concentrations and temperature, TDS, and EC ($r$ = +0.500), while a moderate negative correlation was observed with pH ($r$ = –0.500). Although these correlations were not statistically significant, they suggest that warmer and more mineralized water may favor the persistence of *E. coli* at this site.
Total coliforms displayed stronger relationships with the measured physicochemical variables. Perfect positive and statistically significant correlations were observed with pH ($r$ = +1.000, $P < 0.01$), whereas perfect negative correlations were found with both temperature and EC ($r$ = –1.000, $P < 0.01$). A moderate positive correlation ($r$ = +0.500) was also observed between TC and TDS.
These results indicate that bacterial dynamics at the New Bell borehole are influenced by multiple physicochemical factors, although the direction of these relationships differs from those observed at Ngalane, highlighting the site-specific nature of groundwater microbial ecology.

**Table 3.** Spearman correlation coefficients between physicochemical parameters and microbial indicators at the New Bell borehole.

| | | T° (°C) | pH | TDS (mg/L) | EC (µs/Cm) |
|---|---|---|---|---|---|
| *E. coli (*CFU/100ml) | Correlation Coefficient | + 0.500 | - 0.500 | + 0.500 | + 0.500 |
| | **N** | **3** | **3** | **3** | **3** |
| CT (CFU/100ml) | Correlation coefficient | -1.000** | +1.000** | +0.500 | -1.000** |
| | **N** | **3** | **3** | **3** | **3** |

#### 3.3.3. Nko'ovos Borehole

No correlation analysis could be performed between *E. coli* and the measured physicochemical parameters at the Nko'ovos borehole because *E. coli* was not detected in any of the analyzed samples. Consequently, correlation coefficients were reported as **Not Determined (ND)**.
For total coliforms (**Table 4**), a perfect positive correlation was observed with temperature ($r$ = +1.000, $P < 0.01$), while strong positive correlations were found with both TDS and EC ($r$ = +0.866). In contrast, pH exhibited a strong negative correlation ($r$ = –0.866) with TC concentrations.
Although total coliform counts remained very low at this borehole, these correlations suggest that slight variations in groundwater physicochemical conditions may influence the occurrence of environmental coliform bacteria even in the absence of detectable fecal contamination.
**Table 4.** Spearman correlation coefficients between physicochemical parameters and microbial indicators at the Nko'ovos borehole.

| | | T° (°C) | pH | TDS (mg/L) | EC(µs/Cm) |
|---|---|---|---|---|---|
| *E. coli* (CFU/100ml) | Correlation coefficient | ND | ND | ND | ND |

| | | | | | |
|---|---|---|---|---|---|
| | **N** | **3** | **3** | **3** | **3** |
| CT(CFU/100ml) | Correlation coefficient | +1.000** | -0.866 | +0.866 | +0.866 |
| | **N** | **3** | **3** | **3** | **3** |

### 3.3.4. Ebolowa Si I Borehole

Similarly, *E. coli* was not detected in any sample collected from the Ebolowa Si I borehole, preventing calculation of correlation coefficients between this bacterium and the measured physicochemical parameters.
For total coliforms (Table 5), strong positive correlations were observed with temperature ($r$ = +0.866) and TDS ($r$ = +0.866), while a strong negative correlation was found with pH ($r$ = –0.866). In contrast, electrical conductivity showed virtually no association with TC concentrations ($r$ = +0.001), suggesting that conductivity had little influence on bacterial occurrence at this site.
The very low total coliform concentrations recorded at Ebolowa Si I indicate excellent microbiological quality despite these observed statistical relationships. The correlations therefore likely reflect natural physicochemical variability within the aquifer rather than conditions favoring substantial microbial proliferation.

**Table 5.** Spearman correlation coefficients between physicochemical parameters and microbial indicators at the Ebolowa Si I borehole.

| | | T° (°C) | pH | TDS (mg/L) | EC (µs/Cm) |
|---|---|---|---|---|---|
| *E. coli* (CFU/100ml) | correlation coefficient | ND | ND | ND | ND |
| | **N** | **3** | **3** | **3** | **3** |
| CT (CFU/100ml) | Correlation coefficient | +0.866 | -0.866 | +0.866 | +0.001 |
| | **N** | **3** | **3** | **3** | **3** |

Taken together, the correlation analyses demonstrate that the relationships between physicochemical parameters and microbial indicators varied considerably among the four boreholes. No consistent pattern was observed across all sampling sites, suggesting that groundwater microbiological quality is controlled by a combination of local hydrogeological conditions, environmental characteristics, borehole management practices, and potential contamination sources rather than by any single physicochemical parameter.
Among the measured variables, **TDS and pH exhibited the most consistent associations with total coliform concentrations**, although both the direction and strength of these relationships differed between sites. Likewise, the absence of detectable *E. coli* at Ebolowa Si I and Nko'ovos precluded correlation analyses at these locations and further highlights the marked spatial heterogeneity of groundwater microbial contamination within the study area.
Because the correlation analyses were based on only three observations per site ($n$ = 3), these findings should be interpreted cautiously. They primarily provide preliminary evidence of site-specific interactions between physicochemical characteristics and bacterial indicators, which should be confirmed through studies involving larger sample sizes and extended temporal monitoring.

## 4. Discussion

### 4.1. Microbiological Quality of Borehole Water in Ebolowa

The present study provides new insights into the microbiological quality of heavily used borehole water in Ebolowa by integrating bacteriological indicators with environmental observations and physicochemical characteristics. Overall, the findings revealed pronounced spatial heterogeneity in groundwater quality, with substantial differences in microbial contamination among the four investigated boreholes. These results suggest that groundwater quality in the study area is controlled by a complex interplay of environmental conditions, hydrogeological characteristics, and borehole management practices rather than by a single contamination factor.
The microbiological analyses identified Ngalane as the most contaminated site, exhibiting exceptionally high concentrations of both *Escherichia coli* (7,000 ± 1,000 CFU/100 mL) and total coliforms (35,666.67 ± 1,527.53 CFU/100 mL). The detection of *E. coli* is widely recognized as evidence of recent fecal contamination because this microorganism is predominantly associated with the intestinal tract of warm-blooded animals and humans. Likewise, elevated total coliform concentrations indicate severe deterioration of microbiological water quality and strongly suggest that the groundwater supplying this borehole is vulnerable to microbial intrusion. Although total coliforms include environmental species that are not exclusively of fecal origin, their occurrence at such high concentrations, together with the presence of *E. coli*, provides compelling evidence of significant sanitary impairment.
Interestingly, the severe contamination observed at Ngalane cannot be explained solely by the current environmental setting. Unlike some of the other investigated boreholes, Ngalane is located relatively far from residential buildings, suggesting that

additional factors contribute to groundwater deterioration. Historical contamination associated with former sanitation facilities, abandoned waste disposal sites, previous agricultural activities, or long-term infiltration of contaminated surface water may have altered the microbiological quality of the aquifer. Furthermore, hydrogeological characteristics such as preferential groundwater flow pathways, fractured subsurface formations, soil permeability, and aquifer connectivity may facilitate the transport and persistence of microorganisms independently of the present-day distance between boreholes and potential pollution sources. These observations highlight the importance of considering both historical land use and subsurface characteristics when assessing groundwater vulnerability.

In contrast, the Ebolowa Si I and Nko'ovos boreholes exhibited excellent microbiological quality. *Escherichia coli* was not detected in any of the collected samples, while total coliform concentrations remained extremely low. Although the presence of a small number of total coliforms does not necessarily indicate fecal pollution, these findings suggest that microbial contamination at these sites is minimal and that the groundwater remains comparatively well protected. The absence of detectable *E. coli* is particularly important because international drinking water guidelines recommend that this organism should be absent from water intended for human consumption. Consequently, these boreholes present substantially lower microbiological risks than the Ngalane and New Bell sites.

The New Bell borehole displayed an intermediate contamination profile characterized by moderate concentrations of both *E. coli* and total coliforms. Although bacterial counts were markedly lower than those recorded at Ngalane, they nevertheless exceeded acceptable microbiological standards for drinking water. This finding indicates that groundwater contamination occurs to varying degrees across Ebolowa and emphasizes that even moderate bacterial loads should not be overlooked, particularly when boreholes constitute the principal source of domestic water supply. The absence of routine disinfection at New Bell, combined with its location relative to nearby residential areas and drainage pathways, may contribute to the observed contamination pattern.

Statistical analysis further demonstrated that the differences in bacterial concentrations among sampling sites were highly significant. The large F-statistics obtained for both *E. coli* and total coliforms indicate that spatial variability greatly exceeded within-site variability. Although the number of observations per site was limited, the consistency of replicate measurements strengthens confidence in the observed spatial patterns. Nevertheless, these findings should be interpreted cautiously because the cross-sectional nature of the study does not account for potential seasonal fluctuations in groundwater quality.

Collectively, the microbiological results demonstrate that groundwater quality is highly heterogeneous within the city of Ebolowa. The coexistence of heavily contaminated and relatively protected boreholes illustrates the localized nature of groundwater pollution and underscores the need for site-specific surveillance rather than generalized assumptions regarding groundwater safety. Such spatial heterogeneity has been reported in numerous groundwater studies and reflects the combined influence of hydrogeological settings, environmental pressures, and local sanitary practices.

The findings are consistent with previous investigations conducted in Cameroon and elsewhere in sub-Saharan Africa, which have reported substantial microbiological variability among urban groundwater sources. Fokou Bomgning et al. (2022) documented widespread contamination of borehole water by indicator bacteria in Yaoundé, emphasizing that groundwater cannot be assumed to be microbiologically safe solely because it originates from underground aquifers. Similarly, Odoh (2024) demonstrated that urban expansion and inadequate sanitation infrastructure significantly increase the risk of groundwater contamination in rapidly growing African cities. The agreement between the present findings and previous studies reinforces the conclusion that protecting groundwater resources requires not only regular disinfection but also effective land-use planning and improved management of potential contamination sources.

### 4.2. Influence of Environmental Conditions and Borehole Management on Groundwater Quality

The marked differences in microbiological quality observed among the investigated boreholes indicate that groundwater contamination is strongly influenced by local environmental conditions and borehole management practices. Although all four boreholes were designed to provide safe drinking water, their sanitary status varied considerably, highlighting the importance of both anthropogenic and hydrogeological factors in determining groundwater quality.

Routine disinfection appeared to play an important role in limiting bacterial contamination. The Ebolowa Si I and Nko'ovos boreholes, which underwent regular maintenance and disinfection, exhibited either undetectable or extremely low concentrations of *Escherichia coli* and total coliforms. These findings suggest that appropriate sanitary management contributes substantially to preserving microbiological water quality. However, disinfection alone cannot guarantee microbiological safety when the aquifer itself is affected by contamination. This observation is illustrated by the Ngalane borehole, where severe microbial contamination persisted despite routine disinfection. Consequently, while periodic disinfection remains an essential preventive measure, it should be considered one component of a broader groundwater protection strategy rather than a standalone solution.

The New Bell borehole further emphasizes the importance of maintenance practices. Unlike the other investigated sites, this borehole reportedly did not undergo routine disinfection despite its continuous daily use. The moderate concentrations of *E. coli* and total coliforms detected at this site suggest that the absence of regular sanitary maintenance may have facilitated bacterial persistence within the borehole system. Nevertheless, the considerably lower bacterial loads observed at New Bell compared with Ngalane indicate that maintenance practices alone do not fully explain the observed differences, reinforcing the multifactorial nature of groundwater contamination.

The environmental setting surrounding each borehole also appears to have contributed to the observed spatial variability. Boreholes located close to residential areas may be exposed to contamination through the infiltration of domestic wastewater, leakage from septic tanks or pit latrines, and surface runoff carrying organic matter and microbial contaminants. Such contamination pathways are particularly relevant in rapidly expanding urban environments where sanitation infrastructure may be insufficient to protect shallow groundwater resources. In this study, the New Bell borehole is situated downstream of residential areas, creating conditions under which runoff and drainage water may transport contaminants toward the groundwater recharge zone.

However, the results also demonstrate that proximity to residential settlements cannot, by itself, explain groundwater contamination. The Ngalane borehole, despite being located relatively far from nearby houses, exhibited by far the highest bacterial concentrations. This finding strongly suggests that additional environmental and hydrogeological factors contribute to groundwater vulnerability. Historical contamination resulting from abandoned sanitation facilities, former waste disposal sites, or previous agricultural activities may continue to affect groundwater quality long after the original pollution sources have disappeared. Moreover, the geological characteristics of the aquifer—including soil permeability, the thickness of the unsaturated zone, groundwater flow pathways, and the presence of fractures or preferential infiltration channels—may facilitate the migration and persistence of microorganisms over considerable distances.

The contrasting microbiological profiles observed among the four boreholes therefore illustrate the complexity of groundwater contamination processes. Similar environmental conditions did not consistently produce similar microbiological outcomes, indicating that contamination results from the interaction of multiple site-specific factors rather than from any single determinant. This complexity underscores the importance of conducting comprehensive sanitary inspections alongside microbiological analyses when evaluating groundwater quality.

The findings of the present study are consistent with previous investigations conducted in urban groundwater systems. Fokou Bomgning et al. (2022) reported that borehole contamination in Yaoundé was closely associated with deficiencies in sanitary protection and local environmental conditions. Likewise, Odoh (2024) highlighted the influence of urbanization, population density, and inadequate sanitation infrastructure on groundwater microbial quality in West Africa. Together with the present results, these studies demonstrate that borehole safety depends not only on engineering design and routine maintenance but also on effective protection of groundwater recharge areas from anthropogenic contamination.

From a public health perspective, these observations have important implications for groundwater management in Ebolowa. Routine microbiological surveillance should be complemented by regular sanitary inspections of borehole surroundings, identification of potential contamination sources, and implementation of protective measures within groundwater recharge zones. Preventive interventions—including improved drainage systems, appropriate siting of sanitation facilities, periodic disinfection, and community awareness regarding groundwater protection—are likely to be more effective than relying solely on corrective actions after contamination has occurred.

Overall, the results indicate that maintaining the microbiological quality of borehole water requires an integrated management approach that combines effective borehole maintenance with long-term environmental protection and careful consideration of local hydrogeological conditions. Such a strategy is essential for reducing the risk of groundwater contamination and ensuring the sustainable provision of safe drinking water in rapidly urbanizing settings such as Ebolowa.

### 4.3. Relationships Between Physicochemical Parameters and Microbiological Contamination

The correlation analysis performed in this study revealed that the relationships between physicochemical parameters and microbiological indicators differed markedly among the investigated boreholes. These findings suggest that bacterial occurrence in groundwater is influenced by site-specific environmental conditions rather than by a universal physicochemical pattern. Because the analyses were based on a limited number of observations ($n = 3$ per site), the reported correlations should be interpreted as exploratory associations that provide insight into local groundwater dynamics rather than evidence of causal relationships.

Among the measured physicochemical variables, pH, total dissolved solids (TDS), and electrical conductivity (EC) showed the strongest associations with bacterial indicators, although both the magnitude and direction of these relationships varied between sampling sites. This variability reflects the complexity of groundwater systems, where physicochemical characteristics interact with hydrogeological conditions, groundwater residence time, aquifer mineralogy, and contamination sources to influence microbial survival and transport.

At the Ngalane borehole, *Escherichia coli* exhibited a perfect positive correlation with pH, whereas total coliforms were strongly and positively correlated with both TDS and EC. These findings suggest that increased groundwater mineralization coincided with higher bacterial concentrations at this site. Elevated TDS and EC may reflect the presence of dissolved inorganic ions and organic matter derived from natural weathering processes or anthropogenic inputs. Although these parameters do not directly promote bacterial growth, they may serve as indicators of groundwater influenced by infiltration processes that simultaneously transport dissolved substances and microorganisms into the aquifer. The exceptionally high bacterial concentrations observed at Ngalane therefore support the hypothesis that groundwater mineralization and microbial contamination may originate from common environmental sources.

In contrast, the New Bell borehole displayed a different correlation pattern. Total coliforms showed a strong positive association with pH, whereas *E. coli* exhibited only moderate correlations with temperature, TDS, and EC. These observations suggest that the physicochemical environment at New Bell differs from that of Ngalane despite the presence of microbiological contamination at both sites. Such differences may reflect variations in aquifer characteristics, recharge mechanisms, or local contamination pathways. They further demonstrate that groundwater microbial dynamics cannot be explained solely by physicochemical measurements but instead result from interactions among multiple environmental processes.

At both the Ebolowa Si I and Nko'ovos boreholes, *E. coli* was not detected in any of the analyzed samples, preventing statistical assessment of correlations with physicochemical variables. This absence of detectable *E. coli* is itself an important finding, indicating that these groundwater sources were not affected by recent fecal contamination during the study period. Nevertheless, low concentrations of total coliforms remained detectable, allowing exploratory correlation analyses. At both sites, total coliforms tended to increase with temperature and TDS while decreasing with pH. Although these relationships should be interpreted cautiously because of the small sample size and the very low bacterial counts, they may indicate that naturally occurring environmental coliforms respond differently to local physicochemical conditions than fecal indicator bacteria.

The observed positive associations between TDS and total coliform concentrations at several boreholes deserve particular attention. Total dissolved solids represent the combined concentration of dissolved minerals and inorganic salts in groundwater and are frequently used as an indicator of water mineralization. Elevated TDS values may reflect prolonged groundwater–rock interaction or the influence of anthropogenic inputs. When groundwater contamination results from infiltration of surface water or wastewater, dissolved substances and microorganisms may be transported simultaneously, producing positive associations between TDS and bacterial indicators. Similar relationships have been reported by Irvine et al. (2024), who observed positive correlations between groundwater mineralization and bacterial contamination in Benin. Although TDS itself does not directly determine bacterial growth, it may therefore act as an indirect indicator of groundwater vulnerability to contamination.

The influence of pH appeared less consistent across sampling sites. Positive correlations between pH and bacterial indicators were observed at some boreholes, whereas negative associations were recorded at others. Such variability is not unexpected because most coliform bacteria are capable of surviving within a relatively broad pH range commonly encountered in groundwater. Consequently, pH may influence bacterial persistence indirectly by modifying the physicochemical characteristics of the aquatic environment rather than acting as a primary determinant of microbial occurrence. Similar variability has been reported by Bagordo et al. (2024), who demonstrated that the relationship between groundwater pH and microbial contamination depends strongly on local environmental conditions.

Temperature also exhibited contrasting relationships with bacterial concentrations among the investigated boreholes. Although slightly higher temperatures were associated with increased total coliform concentrations at some sites, negative or weak correlations were observed elsewhere. These inconsistencies likely reflect the relatively narrow temperature range measured during sampling, which limits the ability to detect clear temperature-dependent microbial responses. Moreover, groundwater temperature is generally more stable than surface water temperature and may exert a weaker influence on bacterial dynamics than environmental contamination sources or hydrogeological characteristics.

Overall, the correlation analyses indicate that no single physicochemical parameter consistently explained the distribution of microbial contamination across the study area. Instead, the observed relationships varied according to local environmental and hydrogeological conditions, emphasizing that groundwater quality is controlled by multiple interacting factors. Physicochemical parameters should therefore be regarded as complementary indicators that can assist in identifying groundwater vulnerability but cannot substitute for direct microbiological analyses.

These findings highlight the importance of integrating microbiological monitoring with physicochemical characterization and sanitary inspections when evaluating groundwater quality. Such a multidisciplinary approach provides a more comprehensive understanding of contamination processes and supports the development of targeted management strategies for protecting drinking water resources. Future studies involving larger sample sizes, seasonal monitoring, and hydrogeological investigations would further clarify the mechanisms underlying the relationships observed in the present study and improve the understanding of groundwater microbial dynamics in tropical urban environments.

### 4.4. Public Health Implications, Study Limitations, and Future Perspectives

The findings of this study have important implications for public health and groundwater management in Ebolowa, where boreholes represent a major source of water for domestic activities and, in many cases, human consumption. The detection of *Escherichia coli* and elevated concentrations of total coliforms in some investigated boreholes indicates that certain groundwater sources are exposed to microbiological risks and may contribute to the transmission of waterborne diseases if consumed without adequate treatment. Although the presence of total coliforms does not necessarily confirm the presence of pathogenic microorganisms, their detection, particularly when associated with *E. coli*, reflects insufficient sanitary protection and highlights the potential risk of contamination by enteric pathogens.

The particularly high microbial loads observed at the Ngalane borehole represent a significant sanitary concern. The concentrations measured at this site greatly exceed acceptable microbiological criteria for drinking water and indicate a substantial deterioration of water quality. The presence of *E. coli* suggests recent fecal contamination, which may result from direct or indirect pathways of contamination, including infiltration from contaminated surface water, historical sanitation infrastructure, or preferential groundwater flow pathways. Under such conditions, relying solely on disinfection at the point of use may not provide sufficient protection, particularly if contamination is continuous or originates within the aquifer system itself.

The New Bell borehole also requires attention because of the combined effects of moderate microbial contamination and inadequate management practices. Although contamination levels were lower than those recorded at Ngalane, the detection of *E. coli* indicates that this water source does not meet microbiological safety requirements for direct consumption. The absence of routine disinfection, combined with the influence of surrounding residential activities, represents a potential pathway for continued contamination. Regular sanitary monitoring and the implementation of appropriate disinfection measures are therefore necessary to reduce health risks associated with this water source.

In contrast, the microbiological status of the Ebolowa Si I and Nko'ovos boreholes suggests a lower immediate health risk. The absence of detectable *E. coli* and the very low concentrations of total coliforms indicate effective protection against fecal contamination during the study period. However, these results should not be interpreted as evidence of permanent safety. Groundwater quality can change over time due to seasonal variations in rainfall, changes in land use, increasing urban pressure, or failures in borehole maintenance. Continuous monitoring remains essential to ensure long-term protection of these resources.

The study also highlights the importance of adopting an integrated groundwater protection strategy in rapidly urbanizing areas such as Ebolowa. Management actions should not be limited to periodic disinfection but should include systematic sanitary inspections, protection of borehole surroundings, improved drainage management, appropriate positioning of sanitation facilities, and identification of potential historical contamination sources. In particular, maintaining adequate separation distances between boreholes and possible pollution sources, such as latrines, wastewater discharge areas, and waste disposal sites, is essential for reducing groundwater vulnerability.

Despite providing valuable information on groundwater quality in Ebolowa, this study has several limitations that should be acknowledged. First, the number of sampling points and replicates was relatively limited, with three measurements per borehole. Consequently, the statistical correlations between physicochemical parameters and microbial indicators should be considered exploratory and require confirmation through larger datasets. Second, the study was conducted during a single sampling period and therefore does not capture seasonal variations in groundwater contamination. In tropical regions, rainfall intensity, groundwater recharge, and surface runoff patterns may strongly influence microbial transport and contamination dynamics. Long-term monitoring covering both wet and dry seasons would provide a more comprehensive assessment of groundwater quality.

Another limitation concerns the restricted range of microbiological indicators investigated. Although total coliforms and *E. coli* are widely used indicators of sanitary quality, they do not provide direct information on the presence of specific pathogenic microorganisms such as viruses, protozoa, or other bacterial pathogens. Future investigations incorporating additional microbial indicators and molecular approaches would improve understanding of the actual health risks associated with borehole water consumption.

Further research should also integrate hydrogeological characterization, including aquifer depth, groundwater flow direction, soil permeability, geological structure, and recharge mechanisms. Such information would help explain why certain boreholes located under apparently favorable sanitary conditions may still exhibit contamination, as observed at Ngalane. Combining microbiological monitoring, physicochemical analysis, geospatial assessment, and hydrogeological investigations would provide a more accurate framework for groundwater risk assessment.
Overall, this study demonstrates that borehole water quality in Ebolowa is highly heterogeneous and depends on the interaction between environmental conditions, contamination sources, and management practices. While some boreholes currently provide water of acceptable microbiological quality, others present significant contamination risks requiring immediate intervention. Strengthening groundwater surveillance, improving borehole management, and implementing preventive protection measures are essential steps toward ensuring safe and sustainable access to groundwater resources for the population of Ebolowa.

## Conclusion

This study assessed the microbiological and physicochemical quality of heavily used borehole water in Ebolowa, Cameroon, and revealed marked spatial variability among the investigated sites. The highest contamination levels were observed at Ngalane, where elevated concentrations of total coliforms and *Escherichia coli* indicated significant fecal pollution and potential health risks. In contrast, Ebolowa Si I and Nko'ovos showed satisfactory microbiological quality, while New Bell presented an intermediate contamination level associated with insufficient management practices.
The variability observed among boreholes highlights the influence of local sanitary conditions, environmental factors, and hydrogeological characteristics on groundwater quality. Physicochemical parameters showed site-specific relationships with microbial indicators, confirming that groundwater contamination results from complex interactions rather than a single controlling factor.
Regular microbiological monitoring, effective disinfection, improved borehole management, and protection of groundwater recharge areas are essential to reduce contamination risks and ensure the long-term safety of groundwater resources in Ebolowa. Further studies incorporating seasonal monitoring and hydrogeological investigations are recommended to better understand contamination pathways.

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## Competing interests

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