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Assessment of the Microbiological Quality of Well and Borehole Water in the Municipality of Sèmè-Podji, Benin

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ABSTRACT

Access to clean water and sanitation is essential for health. Its contamination is the main source of water-borne diseases such as typhoid fever and gastroenteritis, intestinal parasites, lead poisoning, cyanosis, fluorosis and diarrhoeal diseases in large proportion. In Benin, there is low coverage of the drinking water supply network (AEP) in rural and semi-urban areas. Drilling wells and boreholes are used by the majority of the population in uncovered areas for drinking water supplies. The lack of maintenance and the failure to respect the rules of hygiene with regard to these waters lead to their contamination. The purpose of this study is to assess the microbiological quality of well and borehole water in the municipality of Sèmè-Podji. A field survey has been carried out on the sources of pollution of these waters, the treatments which are carried out on the water before its consumption, the storage mode, the frequency and the mode of cleaning of the storage tanks on these waters. A total of 46 water samples, including 24 from wells and 22 from boreholes, were collected and analyzed for their microbiological quality using standardized methods at the Water and Food Quality Control Laboratory, affiliated to the DNSP and located in Xwlacodji. The analytical results showed a contamination of 77.65% by thermotolerant coliforms, 45.83% by *E. coli*, and 77.24% by Enterococci. As for common germs, 100% of the water analyzes were positive. Statistically, there is no significant difference between the microbial loads recorded in these waters, regardless of the arrangements made, from one district to another for thermotolerant coliforms, E. packages and Enterococci. As for common germs, a significant difference was observed between the waters of wells meeting the sanitation standards and the waters of wells not meeting these standards, but there are no more common germs in the waters of drillings not meeting the sanitation standards than in those of drillings meeting this safety perimeter. Water is therefore a potential source of water-borne disease epidemics, which must be addressed before water can be consumed in order to address the threat to public health.

Introduction

Human societies are facing a problem as vital and urgent as that of the late nineteenth century, which is to ensure that every human being has access to drinking water (Laimé, 2003). Freshwater, the main issue at stake, accounts for only 3% of the world's resource, some ¾ of which is stored as ice (Guidoum, 2017). Groundwater accounts for approximately 97% of total liquid continental freshwater (Malak and Romaissa, 2023). According to Merzoug *et al.,* in 2010, 75-90% of the world's population uses groundwater. Water availability per person per year is 3945 m^3 (UNESCO, 2004). The amount of fresh water on earth hardly increases, but human water needs since the beginning of this century have been seven times faster than population growth (Houndenou, 1992).

In Benin, according to the Direction de l'Hydraulique, the water resources potential is 13.106 billion m^3/year on average for surface water and 1.870 billion m^3/year on average for groundwater recharge (Kamagaté *et al.,* 2007). SONEB, the only national company producing and distributing drinking water, does not cover the whole territory. In order to satisfy their daily need for water, the population has no choice but to resort to water of questionable quality such as rain, well and stream water (Azonnakpo *et al.,* 2020).

In the municipality of Sèmè-Podji, the issue of access to drinking water and to hygiene and sanitation systems is a major concern for the entire population of the municipality. Of the 38 villages and city districts in the Commune, only 22 have water from the SociétéNationale des Eaux du Benin (SONEB) and the majority are concentrated in the districts of Agblangandan and Sèmè-Podji (Valette *et al.,* 2017).

The population of boroughs not fully covered by SONEB's Drinking Water Supply System (DW), in response to the shortage of drinking water, uses wells (traditional or modern), especially in areas where water is readily available and/or private boreholes, to supply drinking water.

Although these works have the advantage of solving the problem of water availability, the quality of this food is not often guaranteed, which has an impact on health. The aim of this work is to assess the microbiological quality of well and borehole water in the municipality of Sèmè-Podji.

Materials and Methods

Biological Material

Biological material consists primarily of twenty-three (23) water samples, including 12 well water and 11 well water samples.

Different stages of the research process

To achieve the objective of our study, the research was conducted in three phases: literature review, field work and laboratory analysis.

Literature Review

It has made it possible to gather prior information relating to the research subject, in order to better understand its aspects and contours. The documents consulted were, inter alia, briefs and theses, study reports, articles, books and websites.

Field work

The fieldwork was carried out in three phases: the exploratory phase, the investigation phase and the sampling phase.

The exploratory phase consisted of exploring the field of research, making direct observations in order to see the relevance of the problem raised. The survey sheet took into account information on water supply, the perception of drinking water quality by target households, factors that could contribute to water contamination at different water points. This sheet is often sent to heads of households.

During the sampling phase, 23 water samples for microbiological analysis were taken from 12 wells (6 of which comply with the sanitation standard in Benin (CODE DE L'HYGIENE PUBLIQUE EN REPUBLIQUE DU BENIN, 1987) and 6, not) and 11 boreholes (with 5 in compliance and 6 not in compliance with the standard). We used single-use sterilized sachets (WHIRL-PAK) with a capacity of 207 ml for the samples. Well water was collected with the wells of each household and placed directly in the sterilized sachets up to ¾ in accordance with aseptic rules. Drilling water sampling was carried out at the most widely used taps in each household.

Laboratory analysis

Microbiological analysis of water samples

For microbiological analyzes of water samples (drilling, wells), the following microorganisms were tested: Total Flora (banal microorganisms), thermotolerant *coliforms*; *Escherichia coli and fecal enterococci.*

Analysis methods

Two inoculation methods were used for the microbiological analyses. The incorporation method was used to count Total Flora and the membrane filtration method was used to count Thermotolerant *Coliforms*, *Escherichia coli* and *Faecal Enterococci*. The various stages in the preparation of the culture media were carried out in accordance with the manufacturers' instructions.

Total Flora Count

The culture medium used was Plate Count Agar, commonly known as PCA. Using a graduated pipette, we removed 1 ml of the sample from each sterile Petri dish. Next, 15 ml of the supercooled PCA agar was aseptically poured onto the sample. The whole mixture was carefully homogenised by rotating the Petri dish several times on the bench in the shape of a figure eight, taking care not to push the contents up into the lid. The plates were then incubated at 37°C for 24 to 48 hours. Yellowish-white colonies were observed.

Enumeration of thermotolerant Coliforms and *Escherichia coli*

The culture medium used was Rapid'*E.coli*2, known as REC 2. After filtering 100 ml of the sample through a membrane filter, the membrane was inoculated onto Rapid'*E.coli* 2 agar, which had previously been poured into petri dishes, avoiding the creation of air bubbles. The petri dishes were placed in an inverted position in an incubator at 44°C for 24 hours. The characteristic colonies were greenish-blue in colour. *Escherichia coli* can be identified by their blue colour and thermotolerant Coliforms by their green colour.

Enumeration of faecal Enterococci

Enterococci were counted on Slanetz-Bartley agar. The plates were incubated at 37°C for 24-48 hours. A 100 ml volume of water was filtered through a cellulose membrane. Characteristic colonies were purple to red in colour.

Statistical analysis

Data analysis and statistical processing are made possible by:

- MICROSOFT EXCEL software used for coding survey data and laboratory analysis results;
- MINITAB version 17 and GRAPHE PAD PRISME7 software for producing the various graphical documents;
- ANOVA test for analysis of variance;
- TUKEY'S multiple comparison test to determine differences between samples.

Differences were considered significant when $p<0.05$.

Results and Discussion

In this study, the results were interpreted using a two-class sampling plan. The tables below help to clarify the quality that a sample can have.

Microbiological analysis of well and borehole water samples

Well water samples

Total flora (common germs)

All the wells are of unsatisfactory hygienic quality (NS). Wells meeting sanitation standards in the Aholouyèmè district are more contaminated than those in the other districts. As for wells that did not meet sanitation standards, the Tohouè district had the highest levels of Total Flora, followed respectively by the Djèrègbé, Aholouyèmè and Ekpè districts. Nevertheless, wells that do not comply with sanitation standards are much more contaminated than those that do.

Thermotolerant coliform

It revealed the presence of thermotolerant *coliforms* (bacteria indicative of faecal contamination) in all the samples except well p4, which complied with the drinking water standard (<1 CFU/100mL). The highest rate of these germs was found in Aholouyèmè (1.5.104

CFU/100mL of water) in wells that did not comply with sanitation standards. However, there was no statistically significant difference (p >0.05) in germ contamination between wells that complied with sanitation standards and those that did not.

Escherichia coli

The wells that comply with sanitation standards are of unsatisfactory hygiene quality (NS), except for the well in the Djèrègbé district (p4), which complies with the potability standard, which recommends that drinking water should be free from *E coli*. The bacterial load recorded was higher in Aholouyèmè.

The water from wells that do not comply with sanitation standards is all of unsatisfactory hygienic quality (NS). The Aholouyèmè and Ekpè districts have the highest contamination rates, with 1.0.104 CFU and 5.6.103 CFU/100mL of water respectively, followed by the Djèrègbé and Tohouè districts. However, there was no statistically significant difference (p >0.05) in germ contamination between wells that complied with health standards and those that did not.

Enterococci

All the wells were of unsatisfactory hygienic quality (NS) for these germs. The Aholouyèmè district had the highest rate (2.4 x 103 CFU/100 mL), followed by the Epkè, Tohouè and Djèrègbé districts, for wells that complied with sanitation standards.

For those that did not comply, the Tohouè district had the highest contamination rate (2.2 x 103 CFU/100mL), followed by Djèrègbé (6.4 x 102 CFU/100mL), Aholouyèmè (3.8 x 102 CFU/100mL) and Epkè (0.8 x 102 CFU/100mL). However, there was no statistically significant difference (p>0.05) in germ pollution between wells that complied with health standards and those that did not.

Borehole water samples

Total flora (common germs)

All boreholes are of unsatisfactory hygienic quality (NS). Drilling that meets remediation standards is contaminated. Drilling that does not meet the sanitation standards is highly contaminated with Total Flora. However, there was no statistically significant difference (p >0.05) in contamination levels between boreholes that met and those that did not meet sanitary standards.

Thermotolerant coliforms

The Ekpè district had the highest rate of contamination (7.2.102UFC/100 mL), while the Aholouyèmè, Djèrègbé and Tohouè districts were free of faecal contamination (0UFC/100mL) in boreholes that complied with sanitation standards. The rate of thermotolerant coliform contamination is lower in boreholes that do not comply with sanitation standards. As a result, water quality is unsatisfactory as far as these germs are concerned.

Escherichia coli

The results of microbiological analysis of borehole water samples taken in the commune of Sèmè-Podji reveal a total absence of *Escherichia coli*.

Faecal Enterococci

Fecal enterococci are absent (<1UFC/100mL) in boreholes that comply with health standards at Ekpè and Tohouè. The same is true for those that do not meet these standards at Aholouyèmè and Ekpè. However, there was no significant difference (p >0.05) in germ pollution between boreholes that complied with health standards and those that did not.

Microbiological analysis of the water in the wells and boreholes investigated revealed that it was contaminated with most of the germs sought. The count of common germs, considered as a tool indicative of the general microbiological state of the samples submitted to the tests, is not satisfactory for the 23 water samples that were analyzed. The districts of Tohouè and Aholouyèmè have the highest microbial density levels followed by the districts of Ekpè, and Djèrébé whatever the type of water point. Similar observations were reported by Sohounou *et al.,* in 2015 and Vissin *et al.,* in 2017 in the same commune, which obtained banal sprouts with values above the Benin and WHO standard for drinking water (20UFC/mL) in the well water analyzed.

Kelomè *et al.,* in 2012 in Parakou and Tchaourou, Visudhiphan and Ghatak in 1971 in Bangui also found an unusual contamination of well and drilling water in banal sprouts.

Table.1 Microbiological quality of samples.

Value of N	Ratings
N < M	Satisfactory hygiene quality
N ≥ M	Unsatisfactory hygiene quality

M: the upper limit of the standard values
N: the enumeration value

Figure.1 Variation in Total Flora in well water samples

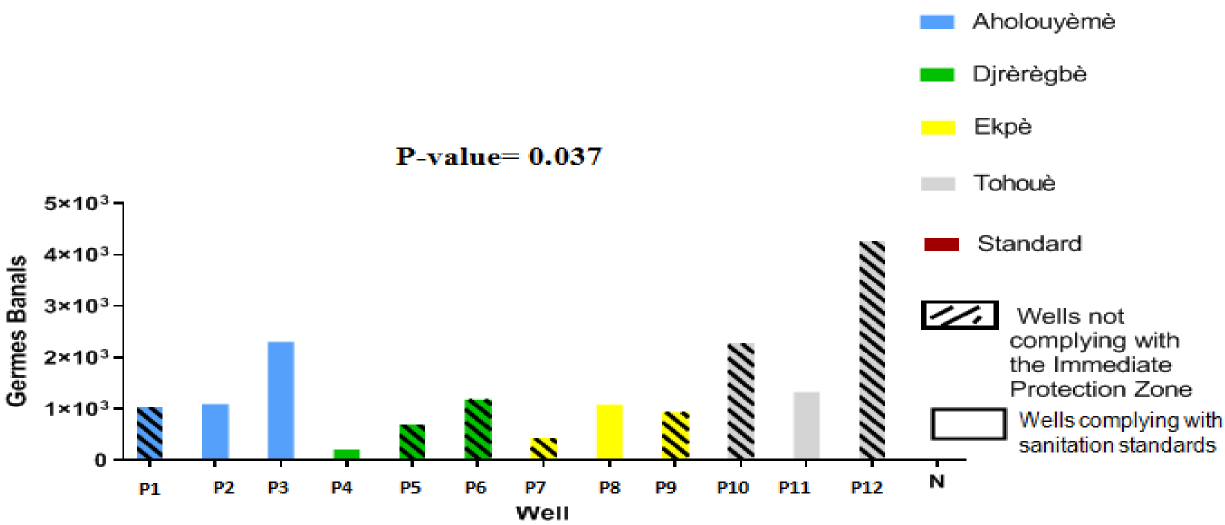


Figure.2 Variation in thermotolerant coliforms in well water samples

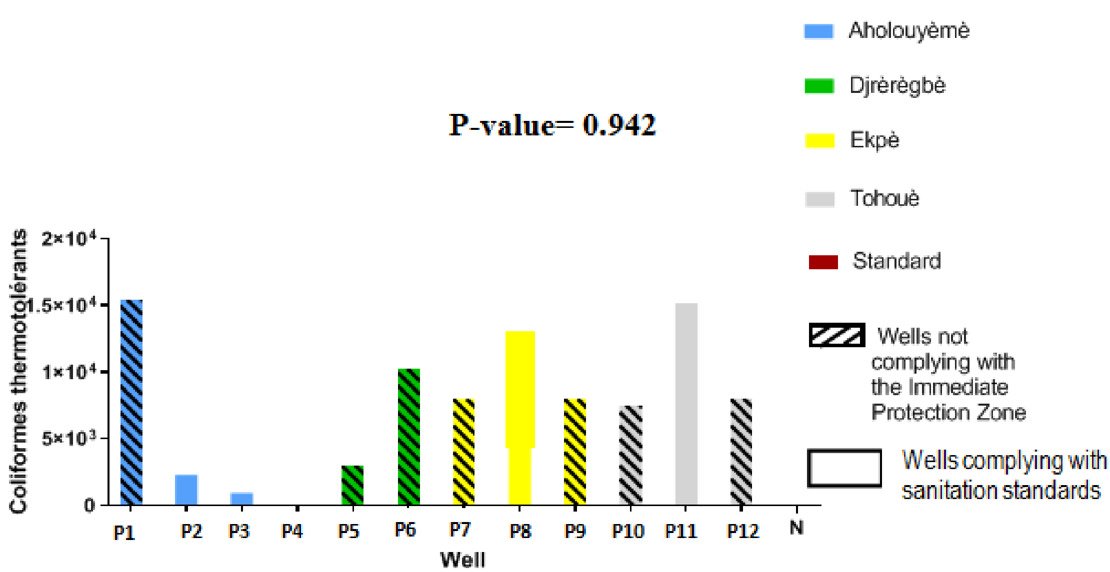


Figure.3 *Escherichia coli* variation in well water samples

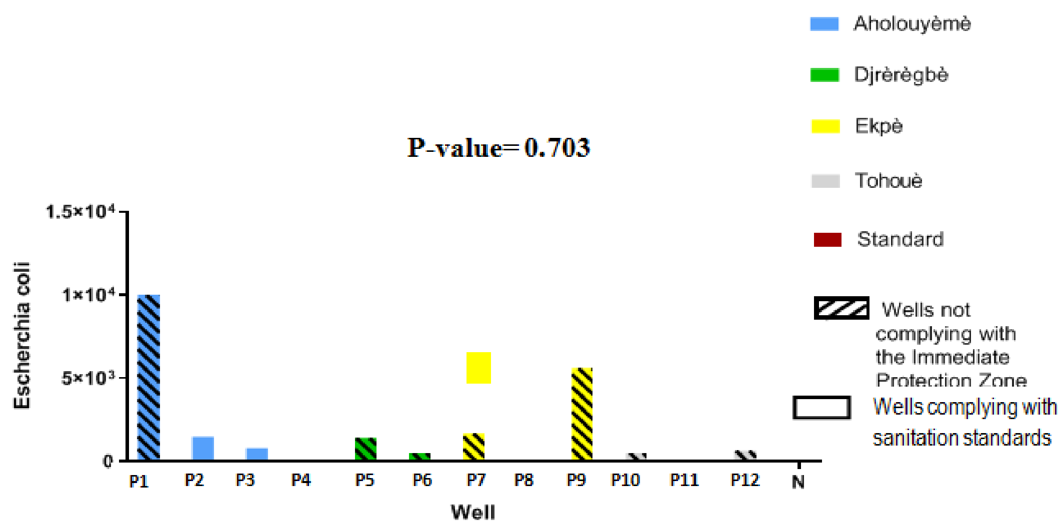


Figure.4 Change in Fecal Enterococci in Well Water Samples

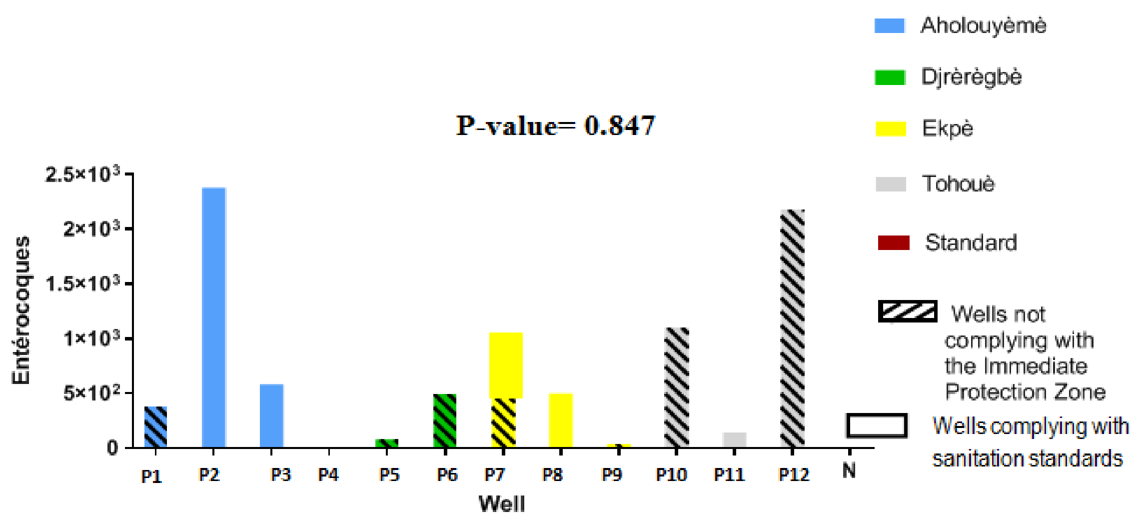


Figure.5 Variation of the Total Flora of Drilling Water Samples.

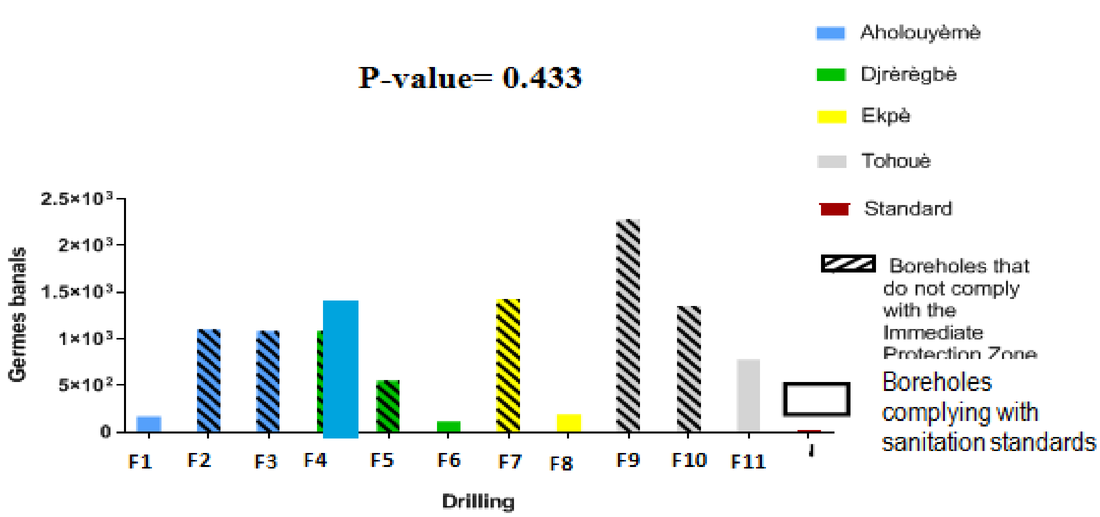


Figure.6 Variation of Thermally Tolerant Coliforms in Drilling Water Samples

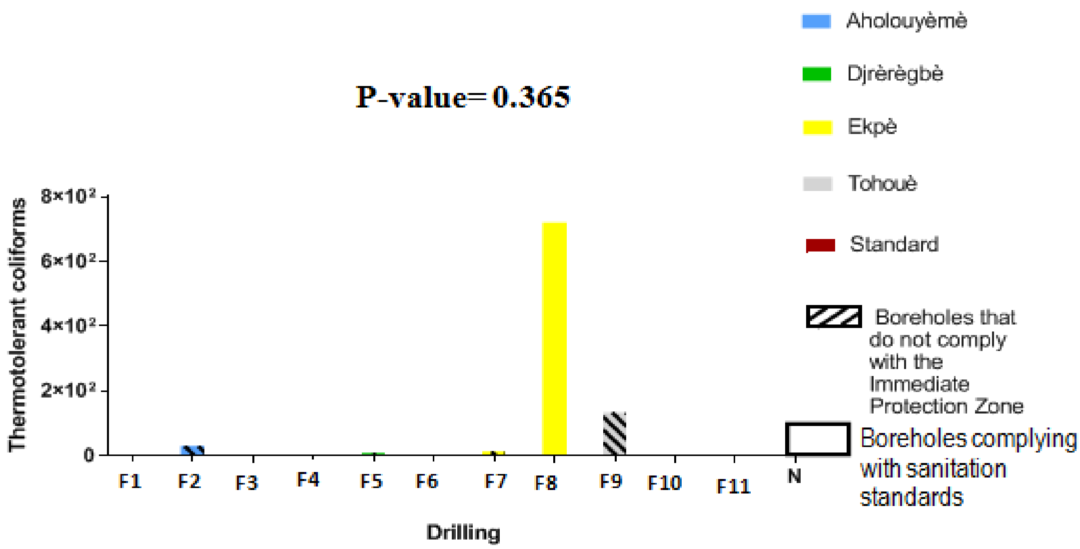


Figure.7 Variation of *Escherichia coli* in Drilling Water Samples

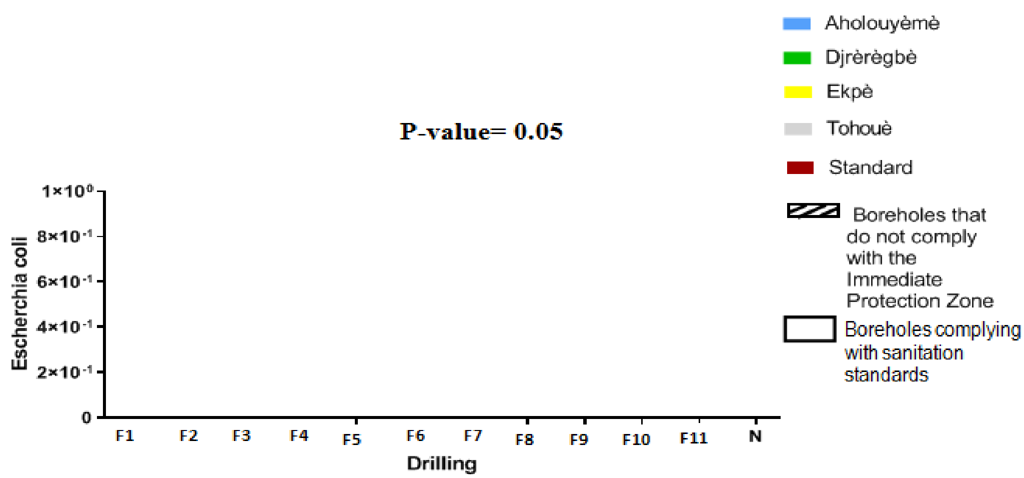
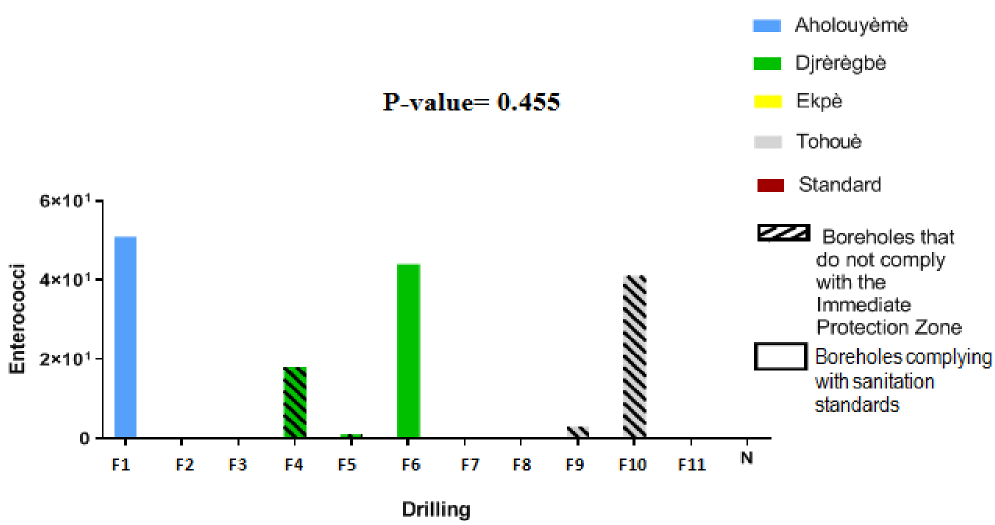


Figure.8 Variation of Enterococci in Drilling Water Samples



The actual presence of thermotolerant coliforms and sometimes *Escherichia coli*, which far exceed the maximum permitted values, reveal faecal contamination of wells and boreholes. The levels of thermotolerant coliforms in well and borehole water were 91.67% and 63.64% respectively; the most contaminated districts were Tohouè, Aholouyèmè, Djèrègbé and Ekpè. In addition, 91.67% and 0.00% of the water was contaminated with *E. coli*. In the same Commune, work by Dégbey *et al.,* in 2010 also identified excessive contamination of well water with thermotolerant coliforms and *E. coli*. The same finding was reported in Lomé in Togo (Soncy *et al.,* 2015) and Tangiers in Morocco (Lanjri *et al.,* 2014), concerning these germs.

The presence of *E. coli* raises suspicions of recent fecal pollution due to the infiltration of fecal matter into wells, especially during rainy periods, and the lack of hygiene by the population itself after having defecated. The results of the investigations carried out with regard to the risk factors of contamination (the presence of sources of contamination within 15 m of the wells: latrines and septic tanks, household waste, the method of tapping...), and the effect of the high temperature at the subsoil level really confirm the degree of bacteriological pollution observed. This contamination is also due to the hydromorphic nature of soils, which are very sensitive to the infiltration of polluted waters because of their soil characteristics (permeable soils, shallow water table, less than 2 meters in all seasons in some neighborhoods).

The borehole water analysed was less contaminated than the well water. However, there was no statistically significant difference between the levels of thermotolerant coliforms and *E. coli* in the well and borehole water analysed, regardless of the measures taken. In terms of drinking water quality standards, of the 23 water samples analysed, only 5 (21.74%) were recommended for human consumption in terms of faecal coliforms, and 12 (52.17%) were recommended for drinking water in terms of *Escherichia coli*.

The results of the *Enterococci* analysis showed that 78.26%, or 18 samples of our water, contained a number of Enterococci that far exceeded the maximum permitted values for drinking water (0 CFU of Enterococci in 100mL of drinking water). The most contaminated districts are Tohouè and Aholouyèmè, followed by Djèrègbé and Ekpè. The boreholes visited were more or less watertight in terms of microbial infiltration, which is similar to the results of Avocefohoun in 2018 in the commune of Djidja on the microbial quality of borehole water (Avocefohoun, 2018). However, there was no statistically significant difference between the rate of enterococci in water from wells and boreholes that complied with sanitation standards and in water from wells and boreholes that did not comply with these standards, in the municipality studied.

The remarkable presence of this germ would indicate faecal pollution (Rodier, 1978) of animal and human origin observed in the field (latrines, livestock practices, the presence of pigsties near certain water points, unhealthy cohabitation with domestic animals). It should be noted that 83.33% of the wells sampled were built with little technical skill; they had no anti-digging slab and virtually no internal lining. The absence of these safety features means that rainwater laden with huge impurities (human or animal faeces) can easily penetrate these wells. Our results confirm those of Dossa in 2002 in the city of Cotonou and in the municipality of Abomey-Calavi, which revealed that the well water sampled in southern Benin was contaminated by faecal enterococci, but are contrary to those of Kèlomè *et al.,* in 2012 in Parakou and Tchaourou, who reported only 10% contamination in the drinking water analysed. Of the 23 water samples analysed, only 5 (21.74%) were recommended for human consumption.

In the light of the results obtained, the water analysed is of unsafe hygienic quality, as the bacteriological contamination of the water from the wells and boreholes tested is widespread. 75.18% of the water tested was unsatisfactory from the point of view of hygiene or microbiological quality (95.83% of wells and 54.54% of boreholes). Overall, the quality of this water is poor, regardless of the measures taken. Well water is very heavily polluted with all the germs tested for, with over 94% of wells containing faecal germs. Borehole water is relatively less contaminated, but still contains almost 40% faecal contamination. The main causes are the presence of latrines less than 15m from water points, poor hygiene practices, lack of maintenance of wells and the hydromorphic nature of the soil. This water does not meet drinking standards and should not be consumed without prior treatment. Contamination of this water by faecal bacteria represents a major public health risk for consumers. This situation means that SONEB needs to improve its service to the locality.

Author Contributions

Gbèssohèlè Justin Behanzin: Investigation, formal analysis, writing—original draft. Alphonse Sako Avocefohoun: Validation, methodology, writing—reviewing. Atchadé Pascal Tchogou:—Formal analysis, writing—review and editing. Sagbo Damien Zannou: Investigation, writing—reviewing. Papin Sourou Montcho: Resources, investigation writing—reviewing. Parfait Sègla Alohoutade: Validation, formal analysis, writing—reviewing. Worou Nicodème Chabi: Conceptualization, methodology, data curation, supervision, writing—reviewing the final version of the manuscript.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethical Approval Not applicable.

Consent to Participate Not applicable.

Consent to Publish Not applicable.

Conflict of Interest The authors declare no competing interests.

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