

Research Article

Microbiological Analysis of a Week-Long Stored Water from Water Pump and Undeveloped Spring in New Binanuaanan, Pili, Camarines Sur

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Abstract: In Barangay New Binanuaanan, Pili, Camarines Sur, many residents not connected to the Pili Water District (PIWAD) depend on distant water pumps and springs for drinking water, which they store for later use. The safety of this stored water, particularly after prolonged storage, has not been previously assessed. This study aims to evaluate the potability of week-long stored water samples from two common sources: a private water pump (Sample 1) and an undeveloped spring (Sample 2). Using the Multiple Tube Fermentation Technique (MTFT), microbiological analyses were conducted by the Metropolitan Naga Water District to determine the presence of total and fecal coliforms. Sample 1 yielded 8.0 Most Probable Number (MPN)/100 mL for both fecal and total coliforms, while Sample 2 showed <1.1 MPN/100 mL for the same indicators. These values were compared to the Department of Health (DOH) standards for drinking water. The results indicate that only the spring water (Sample 2) remained potable after a week of storage, while the pump water (Sample 1) became microbiologically unsafe. This finding underscores the need for improved access to safe water sources and encourages further research on water quality and safe storage practices in underserved communities.

Keywords: *Microbiological Analysis, Multiple Tube Fermentation Technique (SMFT), water storage, potability*

INTRODUCTION

Water is one of the primary necessities of life. Despite its essential role in health and development, access to safe and potable water remains a pressing issue, particularly in rural and underserved areas. Globally, over two billion people consume water from sources contaminated with feces, significantly increasing their vulnerability to waterborne diseases such as diarrhea, cholera, dysentery, typhoid, and polio—resulting in approximately 485,000 diarrhoeal deaths annually (World Health Organization, 2022; United Nations, 2023). These challenges are most acute in developing countries, where the lack of modern water infrastructure and sanitation services contributes to poor health outcomes. Unsafe water, sanitation, and hygiene are responsible for over one million diarrhoea deaths annually, and it is estimated that up to 80% of diseases in these regions are linked to water-related problems (World Health Organization, 2019; United Nations, 2023).



In the Philippines, water security remains a significant public health concern. Approximately 57 million Filipinos—about 52 percent of the population—lack access to safely managed water services, with rural communities experiencing the greatest disparities (Water.org, 2023). In many of these underserved areas, residents depend on untreated or natural water sources such as wells, pumps, and springs. While these sources are essential for daily needs, they often pose hidden health risks, particularly when water is stored for extended periods without proper treatment or safe containment (UCANews, 2023; UNDP, 2023).

The barangay of New Binanuaanan in Pili, Camarines Sur, reflects this broader national issue. Many residents are not connected to the Pili Water District (PIWAD) and instead rely on private pumps and undeveloped springs for their drinking water (Pili Water District, n.d.). These sources are often located far from households, leading residents to store water for future use. However, this practice raises concerns about water quality, particularly the risk of microbiological contamination during storage (World Health Organization, 2022).

This study seeks to evaluate the potability of week-long stored water from two commonly used sources in Zone 2, New Binanuaanan—a private water pump and an undeveloped spring—through microbiological testing. Specifically, it aims to determine the total and fecal coliform levels in the stored samples and assess whether they meet safety standards set by the Department of Health (DOH). The study uses the Multiple Tube Fermentation Technique (MTFT), a recognized method for detecting coliform bacteria in water.

The novelty of this research lies in its focus on microbiological changes in stored natural water sources, an area that remains underexplored in rural water management studies in the Philippines. By identifying the potential health risks associated with stored water and evaluating its compliance with established safety standards, this study contributes new insights into water quality practices and encourages evidence-based interventions for safer water access in rural communities.

METHOD

This study uses a qualitative method through a case study approach, involving an intensive examination of the water sources in New Binanuaanan, Pili, Camarines Sur. The primary aim is to determine whether water stored for one week, originating from two distinct natural sources, remains potable. The results are based on laboratory analyses. The research methodology encompasses selecting two contrasting cases representing different water resources and evaluating their respective water quality over the one-week storage period. This integrates laboratory testing, including the assessment of microbiological parameters crucial for determining potability.

Two water samples—one from a pump and another from an undeveloped spring—were collected using sterile glass bottle containers. Labels and markers were used to indicate the date, time, and source of each sample. Sealable plastic bags were prepared for transport, and the necessary sampling forms were completed. The samples were placed in containers provided by the testing center



and kept chilled using ice packs to maintain the required temperature during transport to the Metropolitan Naga Water District.

On November 13, 2023, Sample 1 was collected from the pump outlet, ensuring a contamination-free process. Simultaneously, Sample 2 was obtained from the undeveloped spring, with precautions to prevent environmental contamination. Both samples were sealed, stored at consistent room temperature for one week, and submitted on November 20, 2023 to the Metropolitan Naga Water District, Production Department, Water Quality Division. Proper labeling, documentation, and handling protocols were followed. Laboratory results were released seven working days later.

Microbial Detection and Enumeration Procedures

To assess the microbiological quality of the water samples, the laboratory applied **membrane filtration** and **multiple-tube fermentation (Most Probable Number, MPN) methods** in accordance with **Standard Methods for the Examination of Water and Wastewater** (APHA, AWWA, & WEF, 2017) and the **World Health Organization Guidelines for Drinking-water Quality** (WHO, 2022).

1. **Membrane Filtration Method** – A 100 mL portion of each water sample was filtered through a sterile 0.45 μm membrane filter to retain bacterial cells. The membrane was placed on selective culture media (e.g., m-Endo agar for total coliforms, m-FC agar for fecal coliforms) and incubated at the appropriate temperature:
 - **Total coliforms:** 35 \pm 0.5 $^{\circ}\text{C}$ for 24 \pm 2 hours
 - **Fecal coliforms / E. coli:** 44.5 \pm 0.2 $^{\circ}\text{C}$ for 24 \pm 2 hoursColonies showing characteristic morphology were counted and reported as colony-forming units (CFU) per 100 mL.
2. **Multiple-Tube Fermentation (MPN) Method** – Serial dilutions of each sample were inoculated into lactose broth tubes with Durham tubes, incubated at 35 $^{\circ}\text{C}$ for 24–48 hours, and observed for gas production. Positive tubes were confirmed for coliforms using brilliant green lactose bile broth and EC broth. Results were interpreted using MPN statistical tables and expressed as MPN/100 mL.
3. **Heterotrophic Plate Count (HPC)** – Portions of each sample were spread onto R2A agar plates and incubated at 20–28 $^{\circ}\text{C}$ for up to 7 days to assess overall bacterial load.

These methods were chosen for their sensitivity and reliability in detecting microbiological contamination in drinking water.

RESULTS

This chapter presents the discussion, analysis and interpretation of the collected data from the microbiological analysis conducted to test the safety of the drinking water in two different sources in Zone 2, New Binanuaanan, Pili, Camarines Sur. Figure 1 shows the water pump which is the source of sample 1 in this study. This pump is owned by a family of seven in Zone 2, but the neighborhood also acquires drinking water from here.

It is a type of water hand pump that is operated manually. It is located within the residential areas of the community; therefore, it is much nearer and more convenient for their access. However, some people in the neighborhood who are not part of the family also acquires drinking water from here and store them in their own households.



Figure 1. Water pump/"Poso"



Figure 2. Undeveloped Spring/ "Bukal"

On the other hand, figure 2 shows the undeveloped spring which is the source of water for sample 2. It is located in the midst of a rice field, with trees taking cover. It has a long metal tube that transports water from the underground. The water flows from it nonstop, with no machine or hand-manipulation needed.

Although located in the same zone as the residents, these sources of water are being used even by the farther neighborhood, especially the undeveloped spring. Its public location, however far, is a good source of free water. On November 13, 2023, samples of water from these sources were gathered and contained in a sterile bottle. The bottles were filled completely with water, one from the water pump (Figure 3.a) and the other from the undeveloped spring (Figure 3.b). They were properly sealed (Figure 3.c) and labeled (Figure 3.d).



Figure 3.a. Sample 1 Procurement



Figure 3.b. Sample 2 Procurement



Figure 3.c. Sealed Samples



Figure 3.d. Samples Properly Labeled



The samples were stored for seven (7) days, from November 13 to November 20. On November 20, the samples were transported and submitted to the Metropolitan Naga Water District in Naga City for laboratory testing.

The results were released after seven (7) working days, on November 30, 2023. Since the data collection and testing took place in November 2023, and the present year is 2025, environmental and infrastructure changes to the water sources may have occurred. However, no follow-up sampling or updated environmental survey was conducted after the initial collection period. Therefore, any changes in the water sources—such as seasonal variations, repairs or upgrades to supply systems, or alterations in surrounding land use—were not factored into the current research findings. The analysis and conclusions presented are based solely on the conditions and microbiological quality of the water samples as they existed during the original sampling period in November 2023.

The table below presents the results of the microbiological analysis:

Table 1. Test Report of the Microbiological Analysis

| Laboratory No. | Sample/ Location | Sampling Date & Time | Results of Analysis | | | Remarks |
|----------------|--|-----------------------------|------------------------------|---|-------------------|---------|
| | | | Total Coliforms (MPN/100 mL) | Thermotolerant / Fecal Coliforms (MPN/100 mL) | HPC Test (CFU/mL) | |
| B23-11-345 | <i>Gripo</i> – Sample 1 | PUMP / Deep well | 11/20/23 6:00 PM | 8.0 | 8.0 | Failed |
| B23-11-346 | Water from <i>Bukal</i> – Sample 2 Burabod (<i>Bukal</i>) | FAUCET / Undeveloped Spring | 11/20/23 6:00 PM | <1.1 | <1.1 | Passed |

The water samples were analyzed using the Multiple Tube Fermentation Technique (MTFT) following the procedures outlined in the Standard Methods for the Examination of Water and Wastewater (20th Edition) and the WHO Guidelines for Drinking-water Quality. In this method, measured portions of each sample (10 mL, 1 mL, and 0.1 mL) were inoculated into a series of sterile tubes containing lauryl tryptose broth (LTB) with inverted Durham tubes to detect gas production. The inoculated tubes were incubated at 35 ± 0.5 °C for 24–48 hours, after which the presence of turbidity and gas indicated presumptive coliforms. From each presumptive positive tube, a loopful of culture was transferred aseptically into brilliant green lactose bile (BGLB) broth for confirmed coliform testing, incubated again at 35 ± 0.5 °C for 24–48 hours, and observed for gas production. To confirm fecal coliforms, positive tubes were inoculated into EC broth and incubated at 44.5 ± 0.2 °C for 24 hours, with the presence of gas confirming fecal coliform



contamination. The pattern of positive results across the three-dilution series was then compared to the Most Probable Number (MPN) statistical tables to estimate the number of coliform organisms per 100 mL of sample. This technique was selected for its reliability in assessing the microbiological safety of drinking water, and all media preparation, inoculation, incubation, and result interpretation followed strict aseptic procedures to prevent contamination (APHA, AWWA, & WEF, 1998; WHO, 2022).

DISCUSSION

The potability potential of Sample 1 and 2

The results show that Sample 1 which is the water from the water pump stored for a week failed the test, while Sample 2 which is the water from the undeveloped spring passed. The water samples were tested with the total coliform, and fecal coliform which presence of both identifies the quality of water.

Total Coliform

Sample 1 has 8.0 MPN/100mL while Sample 2 has <1.1 MPN/100mL. Most Probable Number (MPN)/ milliliters (mL) is a unit of measurement of bacteria density; thus, this result shows the presence and the amount of bacteria present per unit milliliter.

Coliform bacteria will not likely cause illness. However, their presence in drinking water indicates that disease-causing organisms (pathogens) could be in the water system. The total coliform group is a large collection of different kinds of bacteria. Total coliform bacteria are commonly found in the environment (e.g. soil or vegetation) and are generally harmless (Washington State Department of Health, 2023).

Thermotolerant / Fecal Coliforms

The result for the analysis of the amount of fecal coliform in sample 1 shows that it has 8.0 MPN/100mL, while sample 2 has <1.1 MPN/100mL.

Fecal coliforms are types of total coliform that mostly exist in feces. It is a sub-group of total coliform bacteria. They appear in great quantities in the intestines and feces of people and animals. The presence of fecal coliform in a drinking water sample often indicates recent fecal contamination, meaning that there is a greater risk that pathogens are present than if only total coliform bacteria is detected. It also poses an immediate health risk to anyone consuming the water (Washington State Department of Health, 2023).

Implications

The table below shows the Standard Values and Methods of Detection for Microbiological Quality of Drinking Water according to the Philippine National Standards for Drinking Water of 2017 ordered by the Department of Health.

The result shows that with the presence of total coliform group and fecal coliforms in sample 1 that is greater than the standard values set imply that the water is not potable. Therefore, it further imposes health risks for residents who consume it. On the other hand, sample 2 in which total coliform and fecal coliform



passed the standard values means that it is safe drinking water, even after one week of storage

Table 2. Standard Values and Methods of Detection for Microbiological Quality of Drinking Water

| Parameter | Standard Values | Methods of Analysis |
|--------------------------------------|----------------------|---|
| 1. Total Coliform | MTFT: <1.1 MPN/100mL | 9221 Multiple Tube Fermentation Technique |
| 2. Thermotolerant Coliform / E. coli | MTFT: <1.1 MPN/100mL | 9221 Multiple Tube Fermentation Technique |

It is vital to check the suitability of the water before its use or consumption. The presence of certain contaminants or bacteria in our drinking water can lead to health issues, such as diarrheal diseases, respiratory infections and numerous neglected tropical diseases.

CONCLUSION

Based on the microbiological analysis, the water sample from the water pump did not meet the safety standards for potability, while the sample from the undeveloped spring passed the test. This indicates that, after a week of storage, the water from the spring remained safe to drink, whereas the water from the pump was not suitable for consumption.

DECLARATIONS

This section should include as follow:

Competing interests: *“The authors declare that there is no conflict of interest”.*

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