Microbiological quality assessment of drinking water sources in Tonosí, Panama, using an advanced portable analytical technique

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Abstract

The microbiological assessment of water is crucial for public health. It is particularly significant in developing countries. This study assess the microbiological quality of water sources for human consumption in the District of Tonosí, Republic of Panama, using a portable advanced technique to detect fecal coliforms. Tonosí is a rural area, characterized by its tropical climate. Its main economic activities are agriculture and livestock. Eleven points were identified and sampled during the dry season of 2023. The Potatest 2 microbiological water quality laboratory was used to analyze the water samples. The procedure involved filtering water samples using a Membrane Filtration Unit (MFU), followed by incubation. Physicochemical parameters such as temperature, pH, residual chlorine, color, turbidity, and suspended solids were also measured. Fecal coliforms were found in most sampling points, exceeding acceptable limits according to the DGNTI-COPANIT Technical Regulation 21-2019 for drinking water in Panama. Parameters such as water temperature and pH were within regulatory standards, while some color and turbidity values slightly exceeded the limits, indicating the need for monitoring. Low residual chlorine levels suggested inadequate disinfection. The study emphasizes the importance of regular monitoring and inter-institutional collaboration for effective water quality management.

Keywords: microbiology, membrane filtration assembly, drinking water quality, Panama.

Avaliação da qualidade microbiológica de fontes de água potável em Tonosí, Panamá, utilizando uma técnica analítica portátil avançada

Resumo

A avaliação microbiológica da água é crucial para a saúde pública. É particularmente significativa em países em desenvolvimento. Este estudo avalia a qualidade microbiológica das fontes de água para consumo humano no Distrito de Tonosí, República do Panamá, utilizando uma técnica avançada portátil para detectar coliformes fecais. Tonosí é uma área rural, caracterizada por seu clima tropical. Suas principais atividades econômicas são a agricultura e a pecuária. Onze pontos foram identificados e amostrados durante a estação seca de 2023. O laboratório microbiológico de qualidade da água Potatest 2 foi utilizado para analisar as amostras de água. O procedimento envolvia filtrar amostras de água usando uma Unidade de Filtração por Membrana (MFU), seguida pela incubação. Parâmetros fisicoquímicos como temperatura, pH, cloro residual, cor, turbidez e sólidos suspensos também foram medidos. Coliformes fecais foram encontrados na maioria dos pontos de amostragem, excedendo os limites aceitáveis de acordo com o Regulamento Técnico DGNTI-COPANIT 21-2019 para água potável no Panamá. Parâmetros como temperatura da água e pH estavam dentro dos padrões regulatórios, enquanto alguns valores de cor e turbidez excederam ligeiramente os limites, indicando a necessidade de monitoramento. Baixos níveis de cloro residual sugeriam desinfecção inadequada. O estudo enfatiza a importância do monitoramento regular e da colaboração interinstitucional para uma gestão eficaz da qualidade da água.

Palavras-chave: microbiologia, montagem de filtração por membrana, qualidade da água potável, Panamá.

1. Introduction

Contaminated water remains a major global vector for waterborne diseases, particularly in regions with limited access to adequate water treatment and sanitation infrastructure. Furthermore, microbiological assessment is a cornerstone of water quality evaluation and public health, as it enables the detection of pathogenic microorganisms, including bacteria and viruses, that pose significant risks when water is consumed or used for domestic purposes (Canciu et al., 2021; Murei et al., 2024; Pandey et al., 2024).

The ingestion of water from water bodies contaminated by untreated or partially treated sewage effluents is strongly associated with a wide range of bacterial and viral diseases. These include cholera (*Vibrio cholerae*), typhoid fever (*Salmonella* serovar Typhi), diarrheal diseases caused by *Escherichia coli* and *Shigella* spp., as well as infections caused by enteric viruses such as adenoviruses, rotaviruses, noroviruses, and other caliciviruses and enteroviruses (Lanrewaju et al., 2022). These pathogens represent a persistent threat to public health, particularly in vulnerable populations.

Beyond ensuring drinking water safety, microbiological assessment supports the classification of source waters according to their suitability for multiple uses, including human consumption, agriculture, animal husbandry, and industrial applications (Shoushtarian; Negahban-Azar, 2020; Ahmad et al., 2021; Uddin et al., 2021; Yan et al., 2022). Continuous monitoring of microbiological indicators provides critical insights into the sanitary status of water bodies and informs the selection and optimization of appropriate treatment and management strategies.

In developing countries, the effective management of drinking water supply systems is highly dependent on systematic microbiological surveillance, as microbial contamination contributes not only to acute disease outbreaks but also to persistent endemic disease burdens (Fida et al., 2023). Consequently, microbiological risk assessment is an essential component of evidence-based decision-making processes aimed at preventing public health emergencies and ensuring long-term water security.

Conventional microbiological evaluation typically involves sterile collection, transportation, culturing, and laboratory-based analysis of water samples using culture-dependent methods, such as plate counts and selective media (Canciu et al., 2021). However, these approaches are often limited by logistical constraints, particularly in remote or resource-limited settings, where delays in sample transport and temperature fluctuations may compromise analytical reliability and data accuracy.

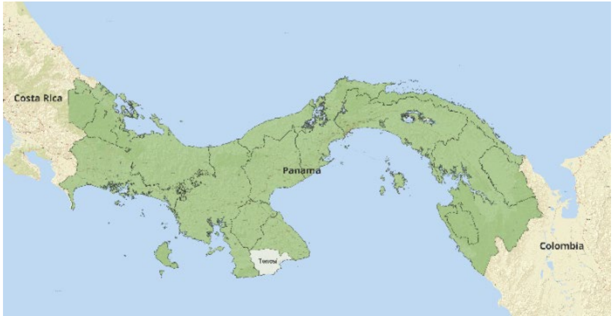
The present study aims to assess the microbiological quality of water sources intended for human consumption in the District of Tonosí, province of Los Santos, Republic of Panama, through the application of a portable microbiological assessment technique. This approach enables near real-time diagnostics, reduces sample degradation during transport, and enhances analytical robustness. The data generated provides actionable evidence to support public health protection, water safety management, and environmental monitoring, contributing to the development of more effective, resilient, and sustainable water quality surveillance frameworks.

2. Materials and Methods

2.1 Study area

The study comprises different townships within the district of Tonosí (Figure 1). Tonosí is located southwest of the Azuero Peninsula, between 7° 24' 0" north latitude and 80° 27' west longitude, with a population of 10,094 inhabitants and a territorial extension of 1291.8 km^2 (Instituto Nacional de Estadística y Censo, 2023), bordered to the north by the districts of Las Tablas and Macaracas, to the south by the Pacific Ocean, to the east by the district of Pedasí and the west by the district of Mariato, Veraguas, Republic of Panama (Instituto Nacional de Estadística y Censo, 2012, p. 2010).

(a)



(b)

Figure 1. (a) Republic of Panama. (b) Location of the district of Tonosí, Panama. Source: Authors, 2025.

The territory has a tropical climate, with average annual temperatures ranging from 26.8 °C to 29.7 °C, with maximums reaching up to 39.8 °C and temperatures dropping to around 15 °C in the highest altitude areas. On the other hand, precipitation varies between 132.8 and 205 mm per year, depending on the different meteorological stations in the area (Instituto de Meteorología e Hidrología de Panamá, 2023). However, climatic conditions present extreme changes in water availability and quality, observing scarcity in the dry season and floods in the rainy season.

The predominant economic activities in the District of Tonosí are agriculture and livestock. This region stands out nationally for its leadership in cattle breeding, rice cultivation, poultry, and pig breeding, occupying first, third, and fourth place in each of these areas at the national level (Evans, 2023).

Two fieldwork tours were carried out in different periods of the dry season, January and March 2023. Also, samplings were carried out in different townships of the district of Tonosí, and the types of water sources used by the communities for the supply and consumption of water resources were identified, such as private wells, public wells, and surface water catchments. The samples were taken directly from the region's taps of residences, schools, and shops.

Eleven sampling points were selected along the district of Tonosí, including the townships of Altos de Güera, El Cortezo, El Cacao, Flores, Cambutal, Guánico, Cañas, and Tonosí (the district's capital). Six of the sampling points correspond to groundwater sources, and the other five correspond to surface water sources.

2.3 Advanced portable technique for microbiology assessment

The advanced Portable Water Quality Laboratory (Microbiological) (Potatest 2, Palintest, Erlanger, KY, USA) was used (Figure 2). This equipment uses one of the detection methods of fecal indicator organisms established in the Guidelines for Drinking Water Quality of the World Health Organization (WHO), and complies with ISO 7704:2023 (World Health Organization, 2022), (ISO, 2023).

Figure 2. Portable Water Quality Laboratory (Microbiological). Source: Authors, 2025.

The water sample was filtered using a Membrane Filtration Unit (MFU) (Palintest, Erlanger, KY, USA). This unit consists of a stainless-steel cup, a funnel attached to a silicone base, a bronze disc for filter support, and a sterile 47 mm (0.45 µm pore size) membrane filter.

After filtration, the membrane filterswere cultivated in Membrane Lauryl Sulphate Broth (MLSB) (AVONCHEM-ACM-1820-O, Waterloo, St. West, Macclesfield). Only fecal coliforms were considered, so the portable incubator was adjusted to 44 °C for 18 hours to ensure adequate bacterial growth. During this period, fecal coliforms were multiplicated to form yellow colonies on the culture medium's red/pink background. These colonies are approximately 1 mm in diameter and capable of being visible to the naked eye (Palintest, 2011).

2.4 Quality assurance and quality control (QA/QC)

All procedures for sample collection, handling, and analysis followed the protocols established in the Standard Methods for the Examination of Water and Wastewater (American Water Works Association (AWWA) et al., 2022), with additional QA/QC measures implemented to ensure data reliability, minimize cross-contamination, and validate field-based results.

2.4.1 Sampling protocol

The water sample was taken for microbiological analysis under aseptic conditions to avoid contamination. As it is drinking water from taps, it was ensured that there were no leaks throughout the pipe to the outlet tap; any accessories were removed from the tap, such as the nozzle of the hose with filter; cleaning was carried out with 70% alcohol, the water was allowed to run for approximately 3 minutes to ensure a stable temperature, and a flow of water without residues stuck in the pipe, subsequently, water samples were taken. Sterile containers of 100 mL with and without sodium thiosulfate were used.

During the sample collection process, samples were kept at an appropriate temperature (<10 °C) until analysis. Once all the samples were obtained, they were left at room temperature to begin microbiological analysis with the portable equipment.

In addition, measurements were made of physicochemical and inorganic chemical parameters, such as temperature, pH, residual chlorine, color, turbidity, and suspended solids, with a water quality laboratory (DREL 2800, HACH, Loveland, CO, USA). These parameters are a fundamental part of environmental assessment and can influence the presence or absence of microbiology. Environmental conditions were also measured in the study area with a portable environmental meter (Kestrel 5200, Nielsen-Kellerman, Boothwyn, PA, USA).

2.4.2 Laboratory safety

The analysis was carried out in a mobile water quality laboratory using specific hygiene and safety measures within the laboratory and in the analysis procedure (Guevara et al., 2025). The mobile laboratory has an air purifier and a fume hood where microbiological analyses were performed.

In the laboratory, disinfection and cleaning measures were taken on the surfaces and surroundings of the work area. The effectiveness of disinfection was verified through a luminometer, Hygiena Touch luminometer (Hygiena, Camarillo, CA, USA), which measured the presence of biological ATP.

MFU was sterilized using 1 mL of methanol, which produces formaldehyde gas when it combusts. Formaldehyde has excellent bactericidal properties. The MFU is sterilized for each water sample. The Petri dishes were sterilized and left for 15 min at 121 °C. Also, all the receptacles used to prepare the medium and into which the medium is dispensed were also sterilized in deionized water for 10 minutes at 121 °C. The absorbent pads, the membrane filter, and the inner part of the Petri dish were handled with the utensils supplied by the portable Potatest 2 microbiological water quality laboratory, taking care not to touch them with the hands.

2.5 Regulations for verifying microbial water quality

The Guidelines for Drinking Water Quality: Fourth Edition, Incorporating the First and Second Addenda guide for water quality established by the WHO. It provides an authoritative basis for effectively considering public health in setting national or regional drinking-water policies and actions (World Health Organization, 2022), and DGNTI-COPANIT Technical Regulation 21-2019 Food Technology. Drinking water. Definitions and General Requirements establish the physical, chemical, biological, and radiological requirements that drinking water must meet. It applies to water supply systems in urban and rural areas and is required to be applied by law in the Republic of Panama (Table 1) (Ministerio de Comercio e Industrias, 2019).

Table 1. Permitted values for microbiological, physicochemical, and inorganic chemical values according to the DGNTI-COPANIT Technical Regulation 21-2019 for drinking water in the Republic of Panama.

Parameters	Units	Permitted value
Total coliforms	MPN/100 mL	<1.1
	CFU /100 mL	<1
Escherichia coli	MPN/100 mL	<1.1
	CFU /100 mL	<1
Color	Pt-Co	15
Turbidity	NTU	1
pH	pH units	6.5 - 8.5
Conductivity	µS/cm	850
Free residual chlorine	mg/L	0.3 - 0.8

Note: CFU = Colony Forming Units, MPN = Most Probable Number, Pt-Co = Platinum-cobalt color, NTU = Nephelometric Turbidity Unit, µS/cm = Microsiemens per centimeter, mg/L = Milligrams per liter. Source: Authors, 2025.

3. Results

Eleven sampling points were identified. Each point was sampled in duplicate as a quality control to verify the accuracy of the microbial presence in drinking water. Table 2 shows the townships, the ID given to each point, the type of water source, and the microbial presence in units of (CFU/100 mL) for January and March 2023.

Table 2. Results of the microbiological analysis of water sources for human consumption in Tonosí, Panama.

ID	Township	Water source	Fecal Coliforms (UFC/100 mL)	
			January 2023	March 2023
ADG-02	Altos De Güera	Groundwater	5	8
ADG-04	Altos De Güera	Surface water	41	67
ECZ-02	El Cortezo	Surface water	55	40
ECC-01	El Cacao	Surface water	0	6
FLR-01	Flores	Surface water	0	0
CMB-01	Cambutal	Surface water	14	0
CMB-03	Cambutal	Groundwater	2	3
GNA-01	Guánico	Groundwater	16	72
CNS-02	Cañas	Groundwater	1	1
CNS-04	Cañas	Groundwater	26	25
TNS-02	Tonosí	Groundwater	2	12

Source: Authors, 2025

The colonies' results ranged from 0 to 72 CFU/100 mL, and fecal coliforms were observed in most of the sampled points. The average of each point was taken to obtain representative values. For sample ADG-04, by March 2023, an extensive number of colonies had developed, making enumeration unfeasible. Consequently, the microbiological analysis team using the portable Potatest 2 water quality laboratory classified the result as Too Numerous To Count (TNTC). Therefore, only the countable value obtained from the duplicate sample was considered, corresponding to 67 CFU/100 mL.

Figure 3 illustrates the colony-forming units in one of the study samples. The yellow dots are the colonies of fecal coliforms, and the pink dots represent bacterial growth of another kind. However, another culture medium must be used to identify the other bacteria present, and another analysis methodology must be applied.

Figure 3. Colony Forming Units found in one of the samples. Source: Authors, 2025.

In situ parameters, including water temperature ranged between 27,3 °C - 35,2 °C, with similar values in both months, and the pH ranged between 6,75 - 8,17, within the range established in the DGNTI-COPANIT Technical Regulation 21-2019 (pH should be less than 8 for effective disinfection) (National Research Council, 1980).

Color, free residual chlorine, turbidity, and conductivity are presented in Figure 4.

Figure 4. Physicochemical parameters results for January and March 2023. (a) Color, (b) Free residual chlorine, (c) Turbidity, (d) Conductivity. Source: Authors, 2025.

The values over the green line indicate that the levels do not meet the permitted values for drinking water in Panama. The maximum value allowed for color is 15 Pt/Co, for turbidity 1 NTU, and for free residual chlorine ranges from 0.3 mg/L to 0.8 mg/L. Notably, all values should comply with those established in the technical regulations. However, the exceeded values are close to the permitted values. However, they are to be considered and implemented in monitoring.

Physicochemical parameters such as color, turbidity, and suspended solids demonstrate the presence of organic matter and bacterial growth in the water. However, drinking water quality could be more stable and vary daily depending on environmental and anthropogenic activities and the time between the contamination and the study (Küpper et al., 2022). On the other hand, the levels of free residual chlorine are almost zero. Chlorine is used as a disinfection product in Panama because it is low-cost, accessible, and efficient in eliminating disease-causing microorganisms without risking public health. However, chlorine is consumed as organisms are destroyed. Therefore, enough chlorine must be added (OMS, 2009).

4. Discussion

Latin American countries often face deficiencies in water management due to the lack of standardized protocols for disinfection and infrastructure maintenance, particularly in rural areas (Moreno et al., 2025). In the district of Tonosí, microbiological water quality problems stem from structural limitations in rural supply systems, which rely predominantly on rural aqueducts with restricted technical capacity, reducing the effectiveness of microbiological treatment. These challenges are further exacerbated by the dominant agricultural activities in the area, which contribute to high pollutant loads in nearby water bodies (Lin et al., 2022).

Regularly, Tonosí suffers from water shortages during the dry season, causing low water levels and forcing the population to choose diverse water sources without knowing the risks they may face. A study of Panama reveals that in areas where water pressure is inconsistent, irrigation affects water quality when pressure levels are lower, as it allows water from the environment to enter the pipes, raising the levels of total coliforms and E. coli bacteria and decreasing residual levels of free chlorine (Nelson; Erickson, 2016).

Fecal coliforms are indicators of quality. They are represented mainly by the microorganism Escherichia coli, usually found in the gastrointestinal tract of humans and warm-blooded animals (Paramo et al., 2018). The Guidelines for Drinking Water Quality by WHO indicates that not even one unit of E.coli is acceptable in the supply, distribution, and point-of-consumption system; therefore, its presence indicates recent contamination and possible risks to public health.

Multiple authors indicate that E. coli is associated with gastrointestinal diseases, such as diarrhea, vomiting, abdominal pain, and nausea, inhibiting nutrient absorption and malnutrition, especially for children (Bastaraud et al., 2020; Cevallos et al., 2018; Khan et al., 2018; Lin et al., 2022; Magana-Arachchi; Wanigatunge, 2020; World Health Organization, 2022). One of the diseases most associated with drinking water is diarrhea, which is the third leading cause of death among children under five years of age in the world (Sánchez, 2018). Furthermore, a document from the Ministry of Health reveals that one of the leading causes of morbidity in the Los Santos population is diarrhea and gastrointestinal diseases in children under one year of age (Perez, 2021).

Government strategies focused on new wells or storage structures without long-term methodologies, which could generate a major issue. Knowing the density of deaths worldwide through drinking water, the WHO recommends different methods to treat water, such as chlorination, ozonation, and filtration (World Health Organization, 2022). The problem of rural areas in Panama goes beyond building new water structures. It is necessary to place a supply plant with proper treatments, to have trained and constant personnel who carry out the due maintenance, and to inform the population of how the water arrives, including its quality and quantity. Inter-institutional collaboration and targeted capacity-building initiatives are fundamental in this regard.

5. Conclusions

The microbiological assessment of water quality in the District of Tonosí, Panama, reveals deficiencies in current water treatment and management practices, with direct implications for public health and the environment. The consistently low residual chlorine levels, combined with elevated turbidity, indicate inadequate disinfection, which facilitates bacterial persistence at multiple sampling points. These findings underscore the urgent need to optimize treatment protocols to ensure compliance with drinking water safety standards.

The successful application of advanced portable microbiological assessment tools demonstrated their effectiveness in generating reliable and timely data in remote and resource-limited settings, thereby supporting evidence-based decision-making. Moreover, the study highlights the strategic importance of inter-institutional collaboration and targeted capacity-building initiatives, which significantly enhanced local technical competencies for water quality monitoring and risk mitigation.

Collectively, the results advocate for the establishment of continuous surveillance programs and the implementation of integrated water management strategies, including the controlled use of disinfectants, improved waste management practices, and systematic infrastructure maintenance. The comprehensive documentation of methodologies and outcomes provides a scalable and replicable framework that can be adapted to similar regions facing comparable challenges. Consequently, this study contributes not only to improving water safety in Tonosí but also offers a sustainable model to support long-term water quality management and public health protection in broader contexts.

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7. Authors' Contributions

Natasha Alejandra Gómez-Zanetti: conceptualization, methodology, formal analysis, investigation, data curation, writing—original draft preparation, writing—review and editing, and visualization; *Dina Henríquez*: investigation, writing—review and editing, project administration, and funding acquisition*; Kathia Tamara Broce Mack.*: investigation, writing—review and editing, supervision, project administration, and funding acquisition; *Jorge Enrique Olmos Guevara*: investigation, writing—review and editing, and supervision; *Yazmin Lisbeth Mack-Vergara*: conceptualization, methodology, formal analysis, investigation, writing—original draft preparation, writing—review and editing, supervision, project administration, and funding acquisition.

8. Conflicts of Interest

No conflicts of interest.

9. Ethics Approval

Not applicable.

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Institutional Review Board Statement

Not applicable.

Informed Consent Statement

Not applicable.