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Water Quality Assessment: Physicochemical Properties and Microbial Characteristics of Water Across Different Sources

Rabin Paudyal[1*], Dolma Bhotia[1], Albina Shrestha[1], Rasmita Gautam[1]

[1]Department of Microbiology, Tri Chandra Multiple Campus, Ghantaghar, Kathmandu, Nepal.
These authors contributed equally

***Corresponding author:** Rabin Paudyal, Tri Chandra Multiple Campus, Ghantaghar. Kathmandu, Nepal. Email: rabin.poudyal@bimc.tu.edu.np

ABSTRACT

Objectives: To assess the quality of drinking water microbiologically as well as to monitor various physicochemical quality parameters.

Methods: The study was a laboratory-based cross-sectional study. Thirty samples of water were taken without contamination from different sources such as stone spouts, boring wells, pumps, dug wells, tap water and jar water. The samples were transported with a cold chain maintained and analyzed promptly. Physicochemical parameters were identified using methods indicated in APHA (2005). Spread plate technique and membrane filtration technique were conducted for total bacterial load count and total coliform load count respectively. Bacterial pathogens were isolated and identified through selective enrichment and culture on specific media as well as using biochemical characterstics. Kirby-Bauer disk diffusion technique was used to determine the antibiotic susceptibility pattern of the isolates which revealed varying rates of resistance among the isolates with some of them having multi-drug resistance.

Results: The findings indicated that the highest bacteria loads were on pump (1.43×10^6 cfu/ml) and stone spout water (1.02×10^6cfu/ml), whereas, jar water was not contaminated. In stone spout water counts of coliform were recorded at (70 cfu /ml).

Conclusion: The most frequently isolated pathogens were *E. coli* and *Klebsiella* spp Isolates showed resistance to amoxicillin which could mean there are threats to health and therefore better monitoring and treatments of water quality should be considered.

Keywords: Drinking water quality, physicochemical parameter, Coliform, AST, Kathmandu.

INTRODUCTION

World Health Organization (World Health Organization, 2022a) indicates that pollution of drinking water is responsible for 80% of disease and sickness around world. A study conducted by (Koju et al., 2015) in different water samples in Kathmandu Valley reported 80% of the total water samples to be contaminated with coliforms. In the same study, physicochemical parameters such as pH, conductivity, hardness, turbidity were above WHO permissible limit. Additionally, a study conducted by (Shakya et al., 2013) reported 61.4% distribution of coliforms . *E. Coli* was the most predominant pathogen isolated. Additionally, a study in Tokha, Kathmandu by (Shidiki et al., 2017) reported coliforms in 100% of water samples tested and that exceeding WHO standards.

A study in Bangladesh (Shamimuzzaman et al., 2019) also reported coliforms in 76.25% of the total water sample tested. So this study was conducted with an aim to compare various physiochemical and microbiological aspect of water which has utmost relevance to the context of Kathmandu Valley.

METHODS

Study type: A cross sectional study was conducted between April 2025 to June 2025. The study was carried

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out in the Microbiology Laboratory of Microbiological Research Organization (MiRON), Tinkune, Kathmandu.

Sample type and Size: 30 drinking water samples that consisted of stone spouts (7), Boring water (9), Hand pumps water (3), Tap water (3) and Jar water (3) were collected. Tap water samples, boring water samples and stone sprout samples were directly collected from the source in sterile Biological Oxygen Demand (BOD) bottles.

Sample Collection: For sample collection from well the BOD bottles were immersed inside the well and water sample was collected.

Physiochemical Analysis: pH and temperature was measured on site using standard calibrated pHmeter and thermometer. The samples were transported back to the laboratory using ice box. The conductivity of the water samples were observed using conductivitymeter. Turbidity was measured using nephelometer. Total hardness, ammonia, total iron were calculated using techniques as mentioned in (APHA (2005) Standard Methods for the Examination of Water and Wastewater. 21st Edition, American Public Health Association/American Water Works Association/ Water Environment Federation, Washington DC. - References - Scientific Research Publishing, n.d.)

Microbiological analysis: Enumeration of total bacterial load was carried out using spread plate technique on Plate Count Agar. Total Yeast and Mold Count was carried out using pour plate technique on Potato Dextrose Agar (PDA). Total coliform load was enumerated using pour plate technique on Violet Red Bile Agar (VRBA). For isolation of gram negative organism a loopful of water sample was streaked on McConkey Agar (MA). The McConkey Agar plate was incubated at 37 degree centigrade for 24 hours. The colonies such obtained were subcultured in Nutrient Agar and incubated as mentioned for MA. The isolated were identified using Gram Staining, Biochemical tests such as indole test, Methyl Red test, Voges Prosakaeur test, Citrate utilization test, Triple Sugar Iron agar test, Oxidative Fermantative test, catalase test, oxidase test and urease test. *Staphylococcus aureus* was selectively isolated using Mannitol salt Agar, *Pseudomonas aeuroginosa* was selectively isolated using cetrimide agar.

Antibiotic susceptibility testing: Antibiotic susceptibility testing of the isolates were carried out using Kirby Baeur disc diffusion technique following (CLSI, 2023).

RESULTS

A total 30 water samples were collected across a variety of sources. Boring water 9 (30%), Stone Spout 7 23%), Well 4 (14%), Jar 3 (10%), pump 4 (13%) and, public Tap water 3 (10%)

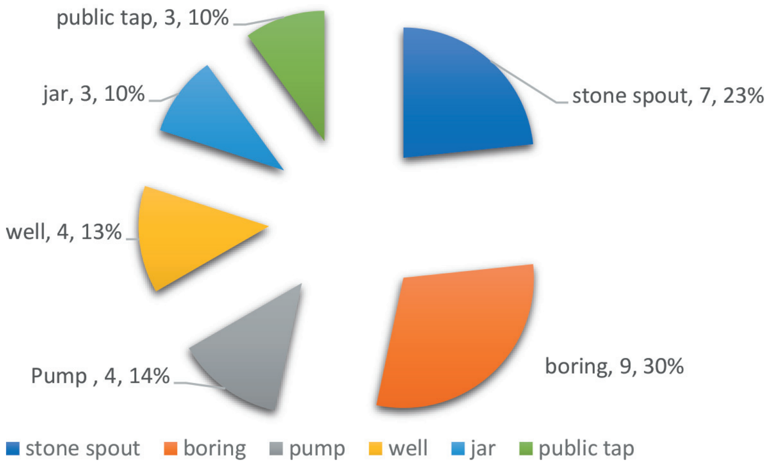

Figure: Distribution of water samples from various sources

Among the collected samples, the highest coliform load was seen on stones spout water samples (70) and least coliform load was seen on jar water (0).

Table 1: Distribution of coliform load in different water samples

S.N.	Types of samples	Total number of samples	Median of coliform load cfu\ml
1	Boring	9	50
2	Stones spout	7	70
3	Well	4	60.5
4	Pump	4	55
5	Tap water	3	16
6	jar	3	0

A total of 65 bacterial isolates were identified from different water sources, including stones spout, boring, well, pump, tap, and jar water samples. Among these, *Escherichia coli* (*E. coli*) which were the isolated species most frequently, 21.5% of the total bacteria detected. This was followed by *Klebsiella* (15.4%) and *Salmonella* (12.3%). Other bacteria such as *Pseudomonas*, *Citrobacter*, *Enterobacter*, *Proteus*, *Vibrio* spp, and *Shigella* were also present, with percentages ranging from (3.1) to (10.8%). The highest number of bacterial isolates was found in boring water (19 isolates), followed by pump water (16 isolates) and well water (12 isolates). Stones spout and tap water had lower bacterial counts, with 11 and 8 isolates respectively. *E. coli* has been considered to be of high health significance and moderate persistence (World Health Organization, 2022b). No bacterial isolates were found in jar water samples. This distribution indicates that water from different sources varies in bacterial contamination, with some pathogens more prevalent in certain types of water sources.

Table 2: Distribution of bacterial pathogens based upon sources of water.

Bacterial species	Stones spout n=(7)	Boring n=(9)	Well n=(4)	Pump n =(4)	tap n =(3)	Jar n=(3)	Total	Percentage (%)
E coli	3	5	2	3	1	0	14	21.5
Pseudomonas aeruginosa	2	1	1	1	1	0	6	9.2
Klebsiella spp	1	3	2	3	2	0	10	15.4
Salmonella spp	1	2	2	1	2	0	8	12.3
Shigella spp	0	1	0	1	0	0	2	3.1
Citrobacter spp	2	1	1	2	1	0	7	10.8
Enterobacter spp	1	1	1	2	1	0	6	9.2
Vibrio spp	0	2	2	1	0	0	5	7.7
Proteus spp	1	3	1	2	0	0	7	10.8
TOTAL	**11**	**19**	**12**	**16**	**8**	**0**	**65**	**100**

The comparative physicochemical analysis reveals that while pH, conductivity, and hardness generally complied with National Drinking Water Quality Standards (NDWQS) across all sources, iron and turbidity levels frequently exceeded safe limits. Boring water and stone spouts exhibited significant turbidity (mean > 4.7 NTU) and iron contamination (mean > 0.5 mg/L), indicative of geogenic leaching and surface runoff. In contrast, the unexpectedly high iron content in commercial jar water (2.03 mg/L) suggests critical failures in processing or storage. Additionally, the detection of elevated ammonia (mean 1.1 mg/l) in tap water serves as a concerning marker for potential sewage infiltration within municipal distribution lines. Collectively, these deviations highlight that despite acceptable mineral balance, the widespread presence of suspended solids and metallic impurities requires robust point-of-use filtration to mitigate aesthetic and health risks.

Table 3: Comparative Assessment of Physicochemical Parameters by Water Source

Parameters	NWQDS Limit	Stone Spout (Mean ± SD)	Boring (Mean ± SD)	Well (Mean ± SD)	Pump (Mean ± SD)	Tap (Mean ± SD)	Jar (Mean ± SD)
Temperature (˚C)	–	22.9	24.1	26.8	26.3	26.4	26.9
pH	6.5 - 8.5	6.57	6.64	6.65	6.63	6.81	6.87
Conductivity (μS/ cm)	1500	59.1	103.2	114.8	114.3	155.7	64.3
TDS (mg/l)	1000	36.9	64.5	71.7	71.4	97.3	40.2
Chloride (mg/l)	250	16.7	64.7	28.9	18.0	21.7	30.0
Hardness (mg/l)	500	108	114	226	220	153	184
Iron (mg/l)	0.3	0.52*	0.96*	0.79*	0.51*	0.71*	2.03*
Turbidity (NTU)	5 (10)	4.7	6.8*	6.8	0.8	5.3	6.0
Ammonia (mg/l)	1.5	ND	ND	0.34	0.60	1.1*	ND

ND: Not Detected   *: Higher than Limit Value

The water samples collected at higher temperatures (26–30°C) showed a significantly higher median coliform load (72 cfu/ml) compared to samples collected at lower temperatures (20–25°C), This indicates that increased temperature might be associated with elevated microbial contamination in the water sample.

Table 4: Distribution of total coliforms based upon temperature.

S.N	Temperature Range	Total No of samples	Median of coliform load (cfu/ml)
1	20-25	12	35.5
2	26-30	18	72

Among the 30 water samples analyzed, those with pH values within the WHO recommended range of 6.5 to 8.5 (Nepal Standard 2062) (covering pH groups 6-7 and 7-8) showed and a median of coliform load between 46 and 59 cfu/ml, indicating moderate microbial contamination within acceptable pH levels.

Table 5: Distribution of bacterial load and coliform load with respect to pH

S.N.	pH range	Total No. of samples	Median of coliform load (cfu/ml)
1	5-6	1	35
2	6-7	22	59
3	7-8	7	46

Out of 10 isolates of *Klebsiella* spp Ciprofloxacin (CIP) and Gentamicin (GE)were found to be sensitive against *Klebsiella* spp (6) isolates were found to be Sensitive to Cotrimoxazole (COT), 4 isolates were found to be resistant to Cotrimoxazole 7 isolates were found to be resistant to Tetracycline (TE) *Klebsiella* spp (10) were found to be resistant against Amoxicillin.Out of 6 isolates *Pseudomonas aeruginosa*, Amoxicillin was found to be not effective against *Pseudomonas aeruginosa,* 5 isolates were found to be sensitive to Gentamicin. *E coli* was found to be sensitive to Gentamicin and cotrimoxazole.

Table 6: Antibiotic Susceptibility Pattern of *Klebsiella* spp and *Pseudomonas aeruginosa*

Antibiotics	*Klebsiella* spp (n=10)		*Pseudomonas aeruginosa* (n=6)		*E. coli* (n=14)	
	S	R	S	R	S	R
Amoxicillin (AX)	0	10	0	6	4	10
Ciprofloxacin (CIP)	10	0	2	4	9	5
Gentamicin (GEN)	10	0	5	1	14	0
Tetracycline (TE)	3	7	2	4	8	6
Cotrimoxazole (COT)	6	4	5	1	10	4

DISCUSSION

The water samples collected from Kathmandu showed varied reliance on sources, with boring water (30%) and stone spouts (23%) being most common. This reflects the city's dependence on alternative supplies due to irregular municipal water distribution (Pandey et al., 2020). However, studies indicate that both groundwater and traditional sources are often contaminated by sewage leakage and poor protection measures (Shrestha et al., 2014; Khadka and Pathak 2016). The presence of jar, well, pump, and public tap water in smaller proportions highlights mixed usage patterns, each with specific contamination risks (Shrestha et al., 2017). These findings helps to keep emphasize the requirement of regular monitoring and improved water management to ensure safe drinking water in Kathmandu.

The distribution of bacterial load across water sources revealed that pump and stones spout water had the highest microbial contamination, with mean bacterial loads of 1.43×10^6 cfu/ml and 1.02×10^6 cfu/ml, respectively. These values exceed the recommended limits for safe drinking water by WHO, which suggest zero detectable fecal coliforms and very low heterotrophic plate counts to minimize health risks (WHO, 2017). Elevated bacterial counts in these sources may result from inadequate protection of water sources, surface runoff contamination, and poor sanitation infrastructure around the collection sites (Jahn et al., 2009).

Boring water, often sourced from deeper underground aquifers, showed significantly lower bacterial contamination (7.52×10^1 cfu/ml), consistent with the natural filtration provided by soil and rock strata. This aligns with previous findings that deep groundwater sources generally have better microbiological quality than surface or shallow water sources (Ashbolt 2004). Conversely, jar water samples showed no detectable bacterial contamination, likely reflecting post-collection treatment or proper storage conditions, corroborating studies that highlight the importance of water handling and storage in maintaining microbiological safety (Sobsey 2002).

The coliform bacteria where it is present means of critical indicator of fecal contamination and potential presence of pathogenic microorganisms (Edberg et al., 2000). The highest mean coliform load was found in stones spout water samples (73.71 cfu/ml), indicating substantial fecal contamination, possibly from nearby latrines, open defecation, or animal waste (Levy et al., 2012). Jar water, again, showed no coliform contamination, reinforcing the protective effect of proper storage or treatment.

The presence of coliform bacteria in public tap water (mean 18.67 cfu/ml) raises concerns regarding the efficiency of municipal water treatment and potential contamination in distribution systems. Leaks, backflow, and biofilm formation in pipes can contribute to such contamination even after treatment (Momba and Kaleni 2002).

Water temperature was found to significantly influence microbial loads, with samples collected at 26–30°C showing higher bacterial and coliform counts than those at 20–25°C. Higher temperatures facilitate bacterial replication and increase metabolic activity, leading to accelerated microbial growth (LeChevallier and Au 2004). Seasonal fluctuations in temperature can thus exacerbate water contamination risks during warmer months, emphasizing the need for heightened surveillance and preventive measures in such periods (Borchardt et al., 2003).

Water samples with pH within the recommended range of 6.5–8.5 showed moderate bacterial and coliform contamination. While pH affects microbial survival and chemical stability of water, it is not the sole determinant of microbiological quality (WHO 2017).

Escherichia coli was the most frequently isolated species, accounting for 21.5% of bacterial isolates, confirming the presence of fecal contamination and indicating a high risk for waterborne diseases (WHO, 2017). Other pathogens including Klebsiella, Salmonella, and Pseudomonas were also identified, posing additional health risks ranging from gastrointestinal infections to opportunistic infections in immunocompromised individuals (Momba and Kaleni 2002). The presence of these bacteria in multiple water sources underscores widespread contamination and the need for improved water sanitation and hygiene practices.

The absence of bacterial isolates in jar water samples indicates that water treatment or storage practices may significantly reduce microbial load, supporting interventions focused on household water treatment and safe storage as effective public health measures (Clasen, et al., 2007).

The antibiotic susceptibility patterns revealed multidrug resistance among isolates, particularly *Klebsiella* spp These isolates showed resistance to amoxicillin and tetracycline but remained sensitive to ciprofloxacin and gentamicin. Similarly, *Pseudomonas* spp was resistant to amoxicillin, with variable susceptibility to other antibiotics.

The emergence of antibiotic-resistant bacteria in environmental water sources is an increasing concern globally, as these bacteria can act as reservoirs for resistance genes that may transfer to human pathogens, complicating infection treatment (Berendonk et al., 2015). Kathmandu's water sources, thus, present potential public health risks not only from pathogenic contamination but also from the propagation of antimicrobial resistance.

The findings demonstrate that several water sources in Kathmandu do not meet microbiological safety standards, putting the population at risk of waterborne diseases. The high bacterial and coliform counts in pump and stones spout water highlight the urgency of source protection measures, including sanitary inspections, fencing, and community education on water handling.

Furthermore, the presence of antibiotic-resistant bacteria stresses the need for surveillance systems to monitor resistance patterns and implement antibiotic stewardship programs.

CONCLUSION

This study confirms that a significant portion of Kathmandu Valley's water sources violate WHO and national standards. The presence of pathogens like *Salmonella* and *E. coli* in traditional spouts and wells exposes a critical lack of source protection. Most alarmingly, the detection of antibiotic-resistant strains identifies these water bodies as active environmental reservoirs for antimicrobial resistance (AMR). This poses a dual threat: an immediate risk of waterborne epidemics and a long-term crisis of untreatable clinical infections. Consequently, urgent remediation is vital to prevent these community water sources from accelerating the spread of drug-resistant pathogens.

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CONFLICT OF INTEREST

The author declares no conflict of interest.

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