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# *Escherichia coli* Contamination in Shallow Groundwater and Associated Health Risks: A Case from Bedkot Municipality of a Terai Region, Nepal

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**Abstract:** Microbial contamination, particularly by *Escherichia coli* (*E. coli*), poses a major public health risk in Nepal's Terai region, yet localized data remain scare. This study evaluated *E. coli* contamination in shallow tube wells across urban, semi-urban, and rural areas of Bedkot Municipality during the pre-monsoon season, and assessed associated health risks according to the WHO guidelines. A cross-sectional design was employed in 2024, collecting 23 water samples from 40-foot-deep tube wells through a stratified random sampling: urban (n=10), semi-urban (n=6), and rural (n=7). For comparison, seven samples of chlorinated municipal water were analyzed prior to distribution. *E. coli* enumeration followed standard membrane filtration protocols. Results revealed a pronounced spatial gradient of contamination. Rural wells exhibited the highest mean *E. coli* concentration (13.8 ± 17.7 CFU/100 mL), with 57% of samples exceeding the WHO guideline (0 CFU/100 mL); 43% were classified as high risk and 14% as intermediate risk. Semi-urban wells showed a lower mean concentration (1.33 ± 1.5 CFU/100 mL), yet 66.7% exceeded guidelines, placing 33.3% at intermediate risk. Urban wells demonstrated minimal contamination (0.1 ± 0.3 CFU/100 mL), with only 10% non-compliance and at intermediate risk. All treated municipal water samples were free of *E. coli*, confirming the efficacy of centralized chlorination. These findings highlight heightened vulnerabilities in rural and semi-urban settings, likely linked to inadequate sanitation and land-use practices. Achieving SDG 6.1 in this region requires context-specific interventions, including sanitation improvements, wellhead protection, and expanded access to treated water.

## 1. Introduction

Access to safe drinking water is a fundamental human right and a cornerstone of public health, yet billions worldwide remain deprived of this essential service. The United Nations Sustainable Development Goal (SDG) 6 aims to ensure the availability and sustainable management of water and sanitation for all, with Target 6.1 specifically calling for universal and equitable access to safe and affordable drinking water by 2030 (Saidi *et al.*, 2022), (United Nations, 2015), (WHO, 2023). Despite progress, this target remains particularly challenging in low- and middle-income countries (LMICs), where microbial contamination of groundwater is persistent and widespread (Bain *et al.,* 2014), (Uprety *et al.,* 2020).

Microbial contamination, primarily through fecal matter, is a leading cause of global waterborne diseases, accounting for an estimated 1.7 billion diarrheal cases and nearly half a million deaths annually (WHO, 2023). *Escherichia coli* (*E. coli*), a commensal bacterium in the intestines of humans and warm-blooded animals, is a central indicator of fecal pollution due to its strong correlation with pathogenic organisms responsible for diarrheal diseases, cholera, and dysentery. Consequently, it plays a critical role in public health risk assessment and water quality monitoring (Haddou *et al.*, 2024), (Naily *et al.*, 2022), (Odonkor and Mahami, 2020), (Uprety *et al.,* 2020).

Nepal reflects these LMICs challenges. Although more than 90% of the population has access to basic water supply services, only about one-quarter benefit from safely managed sources, highlighting a critical gap between service coverage and water quality (National Planning Commission, 2020). This disparity is most acute in the Terai (Plain) region—Nepal's densely populated southern plains and agricultural hub. Here, groundwater from shallow tube wells is the predominant drinking source, but the alluvial aquifers are highly susceptible to microbial contamination due to shallow well construction, high population density, intensive agriculture, inadequate sanitation, and permeable soils (NSO, 2021), (Uprety *et al.,* 2020), (Smith, 2001). Diarrheal diseases, strongly linked to microbial exposure, remain a leading cause of child morbidity and mortality in the Terai (Dhimal *et al.,* 2022), representing a more immediate and widespread risk than chemical contaminants like arsenic (Mahato *et al.*, 2018).

Bedkot Municipality in Kanchanpur District, situated between the northern edge of the Terai plains and Siwalik hills, typifies the pressures facing rapidly urbanizing areas in the Terai. It encompasses urban, semi-urban, and rural communities, each exerting distinct stressors on groundwater resources (ENPHO, 2022). Approximately 68.6% of households rely on tube wells for drinking water (NSO, 2021), previous study has reported *E. coli* contamination in local sources (Uprety *et al.*, 2020). Sanitation infrastructure is underdeveloped: rural households depend largely on traditional pit latrines, while urban and semi-urban households use a mix of pit latrines and septic tanks. Poorly constructed and maintained facilities, often located near water abstraction points, significantly heighten *E. coli* contamination risks (ENPHO, 2022). These risks are further exacerbated by seasonal dynamics, where declining groundwater tables during the pre-monsoon season concentrate pollutants, and heavy monsoon rains mobilize contaminants into aquifers (Bain *et al.*, 2014), (Nayebare *et al.*, 2022), (Odonkor and Mahami, 2020).

Despite this heavy reliance on shallow groundwater, systematic assessments of *E. coli* contamination across settlement types in Bedkot Municipality are limited. A recent study focused on physicochemical water quality (Hamal *et al.*, 2025), but comprehensive data on *E. coli* contamination and the associated health risk during the critical pre-monsoon period are lacking.

This study addresses this gap by systematically evaluating *E. coli* contamination in shallow tube wells across urban, semi-urban, and rural areas of Bedkot Municipality during the pre-monsoon season and assessing the associated health risk based on WHO (1997) thresholds. The findings provide evidence-based insights to guide local water safety interventions and support Nepal's progress toward achieving SDG 6.1.

## 2. Materials and Methods

### *2.1 Description of Study Area*

The study was conducted in Bedkot Municipality, Kanchanpur, Nepal, which spans latitudes 28°32' N to 29°08' S and longitudes 80°03' E to 80°33' W (**Figure 1**). The municipality covers an area of 159.42 km² (Dahal and Timalsina, 2020) and features a tropical savanna climate (Köppen–Geiger classification) with an average annual temperature above 26 °C and annual rainfall between 1,800 and 2,000 mm. Elevation ranges from 192 to 1,401 meters above sea level (ENPHO, 2022).

Administratively, Bedkot comprises ten wards. The 2021 national census reported a population of 57,680 distributed across 12,595 households, with a population density exceeding 350 people per km² (NSO, 2021). Forest is the dominant land use (53.95%), followed by agricultural land (32.68%), riverine areas (5.23%), and built-up land (4.98%). The municipality's fragile geological formations contribute to heavy sediment loads during the monsoon, transported by a network of rivers and streams (Dahal and Timalsina, 2020). Groundwater is the primary drinking water source, accessed mainly through tube wells (hand pumps) at common depths of 20, 40, and 60 feet (ENPHO, 2022). Nearly all households (98.9%) depend on tube wells or piped water, while a small fraction rely on wells (0.5%), spouts (0.2%), bottled water (0.2%), and rivers/streams (0.2%) (NSO, 2021).

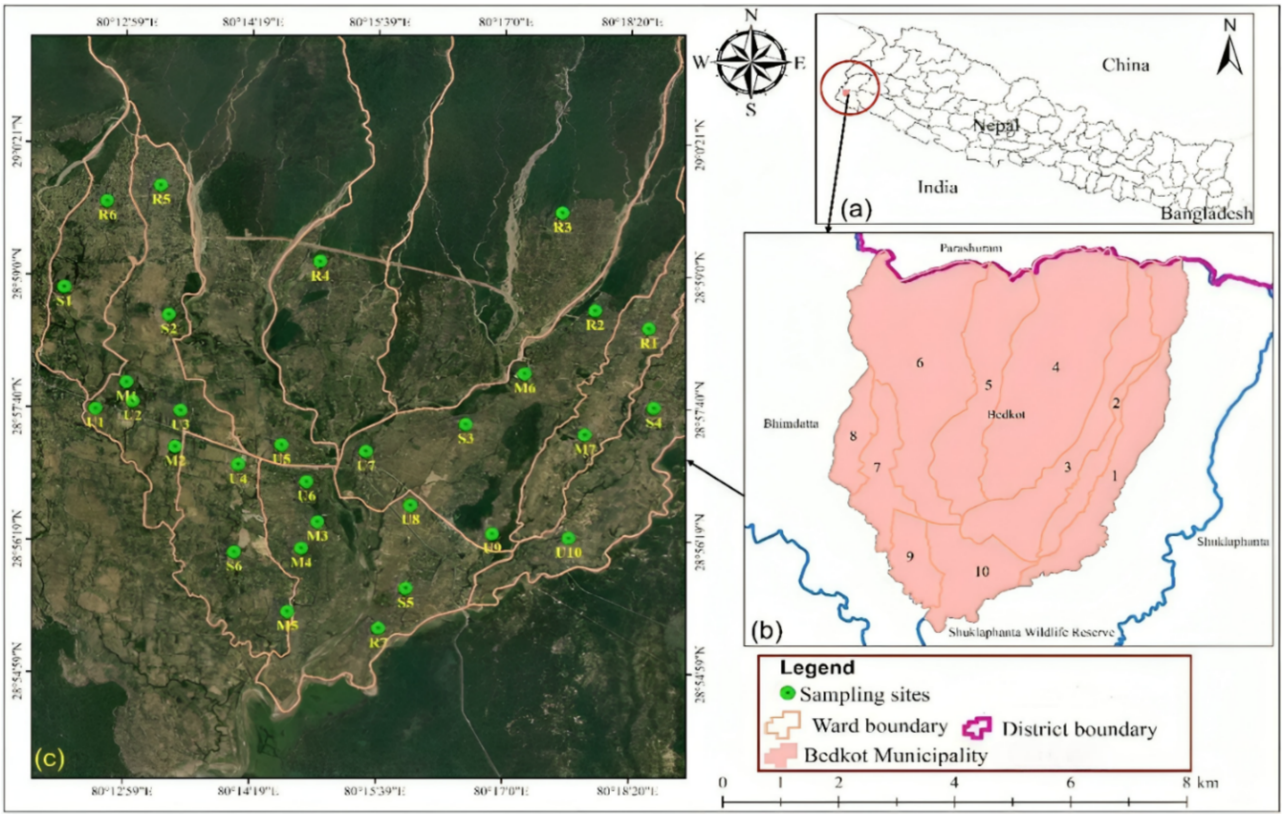


**Figure 1.** Location map indicating (a) the Map of Nepal (b) Bedkot Municipality (study area), and (c) sampling sites within the study area.

### *2.2 Study Design and Sampling Strategy*

A cross-sectional study was employed during the pre-monsoon season (April–May) of 2024 to assess *E. coli* contamination in drinking water. The study focused on privately owned, functional shallow tube wells of uniform depth (40 feet), which are widely used (with over 50% of households depending) and are highly vulnerable to contamination in the study area (ENPHO, 2022). A stratified random sampling strategy was employed to ensure representative coverage across urban, semi-urban, and rural settings. This was followed by purposive selection within each stratum to reflect local conditions, resulting in an unequal sample distribution.

A total of 23 tube well samples were collected: 10 from urban areas (U1-U10), 6 from semi-urban areas (S1-S6), and 7 from rural areas (R1-R7. A larger urban sample size (n=10) was chosen to account for higher population density and greater heterogeneity potential contamination sources (e.g., sewer leaks, septic tanks, waste disposal sites). The rural sample (n=7) was smaller, reflecting more uniform contamination drivers, such as agricultural runoff, livestock waste, and pit latrines (Ghosh *et al.*, 2023), (Smith, 2001). The semi-urban sample (n=6) represented areas with mixed contamination patterns.

Additionally, seven samples (M1–M7) were collected from randomly selected the municipal water supply system points, specifically after chlorination but before distribution to consumers. As 30.3% of households use municipal water as a secondary source (NSO, 2021), these samples (collected post-chlorination and pre-distribution) served as a benchmark for "safe" water distribution in the pipes network and provided an indicator to assess the efficacy of the centralized treatment process (WHO, 2011).

Consent was obtained prior to all sampling. Collections were conducted between 7:00 and 9:00 AM to minimize diurnal variability. Following APHA (2012) protocols, water samples was collected in sterile 125 mL bottles containing sodium thiosulfate to neutralize any residual chlorine. Wells and taps were flushed for five minutes, and sampling bottles were rinsed three times with source water before collection. Samples were immediately stored in a dark cooler with ice packs (~ 4 °C) and transported to the laboratory for analysis within six hours. This approach is consistent with methods used in similar groundwater quality studies in resource-limited settings (Ghosh *et al.,* 2023), (Uprety *et al.,* 2020).

### *2.3 Laboratory Analysis*

*E. coli* was enumerated using the membrane filtration technique (APHA, 2012). A 100 mL aliquot of each water sample was filtered through a 0.45 µm pore-size filter mounted on a funnel placed over a Buchner flask under vacuum pressure. The water samples were shaken before filtration. After filtering, the filter paper was carefully transferred to a sterile absorbent pad inside a sterile Petri dish using sterile forceps. The Petri dish was incubated in an inverted position at 37°C for 24 h. After incubation, the filter paper was examined under a microscope, and the number of colonies was counted, with the results expressed as colony-forming units (CFUs) per 100 mL of water. For quality control, duplicate analysis was performed for 15% of the samples. The culture medium was routinely checked using positive and negative control, and weekly verification of colony was conducted as described by Uprety *et al.* (2020).

### *2.4 Data Analysis and Health Risk Categorization*

Data were processed in Microsoft Excel 2019. Descriptive statistics (mean, standard deviation (SD), and range) were calculated for *E. coli* concentrations in each stratum. To evaluate health risks, *E. coli* concentrations were classified according to WHO (1997) thresholds: low risk (<1 CFU/100 mL), intermediate risk (1–10 CFU/100 mL), high risk (11–100 CFU/100 mL), and very high risk (>100 CFU/100 mL).

## 3. Results

Analysis of shallow tube well samples in Bedkot Municipality revealed widespread fecal contamination, with many sources failing to meet the WHO (1997) guideline zero detectable *E. coli* (0 CFU/100 mL). Non-compliance rates varied substantially across geographical settings. Semi-urban wells exhibited the highest rate of contamination, with 66.7% (4/6) exceeding permissible limits, followed by rural wells at 57.1% (4/7). In contrast, urban wells showed markedly better microbial quality, with only 10% (1/10) non-compliant.

All seven treated municipal water samples (100%) were fully compliant and free of *E. coli* (0 CFU/100 mL), confirming the effectiveness of centralized chlorination (**Figure 2**).

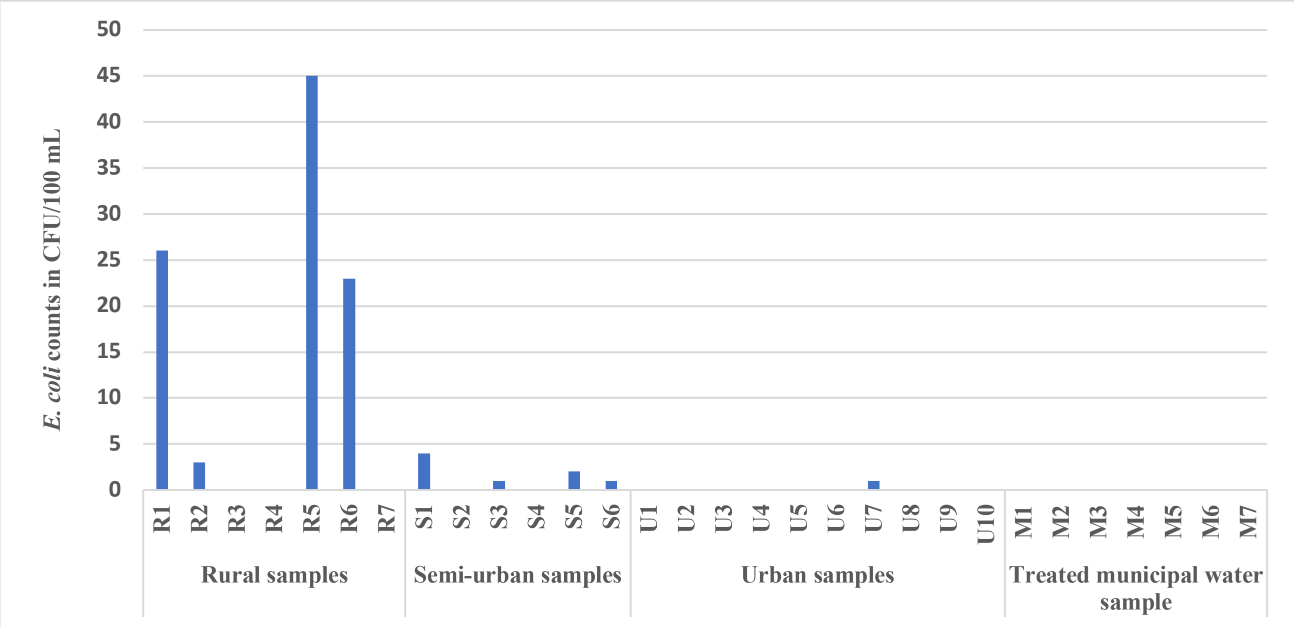


**Figure 2.** *E. coli* counts (in CFU/100 mL) in shallow tube wells of different settings and treated municipal water

A distinct spatial gradient in fecal contamination was observed. Rural wells recorded the highest mean *E. coli* concentration (13.8 ± 17.7 CFU/100 mL, range: 0–45), with high variability indicating a mix of safe and heavily contaminated sources. Semi-urban wells showed lower but still concerning levels (1.33 ± 1.5 CFU/100 mL, range: 0–4). Urban wells demonstrated minimal contamination (0.1 ± 0.3 CFU/100 mL, range: 0–1). All treated municipal water samples consistently recorded 0 CFU/100 mL (**Table 1** and **Figure 2**).

Risk classification according to WHO (1997) categories highlighted distinct exposure profiles. No samples exceeded the "very high risk" threshold (>100 CFU/100 mL). Rural communities faced the greatest risks: only 42.9% (3/7) of wells were "low risk," 14.3% (1/7) were "intermediate risk," and 42.9% (3/7) were "high risk." Semi-urban wells were comparatively safer, with 66.7% (4/6) "low risk" and 33.3% (2/6) "intermediate risk." Urban wells posed the least concern, with 90% (9/10) "low risk" and one well (10%, 1/10) in the "intermediate risk" category. All municipal water samples were classified as "low risk" (**Figure 3**).

**Table 1.** Descriptive analysis of *E. coli* counts (in CFU/100 mL) in shallow tube wells from different settings and treated municipal water.

| **Settings/ Sample Codes** | **Mean ± SD** (*E. coli* CFU/100 mL) | **Min** (*E. coli* CFU/100 mL) | **Max** (*E. coli* CFU/100 mL) |
|---|---|---|---|
| Rural (n =7; R1 – R7) | 13.8 ± 17.7 | 0 | 45 |
| Semi-urban (n= 6; S1 – S6) | 1.33 ± 1.5 | 0 | 4 |
| Urban (n = 10; U1 – U10) | 0.1 ± 0.3 | 0 | 1 |
| Treated municipal water (n = 7; M1 – M7) | 0 ± 0 | 0 | 0 |

Abbreviations: Min (Minimum); Max (Maximum); SD (Standard Deviation)

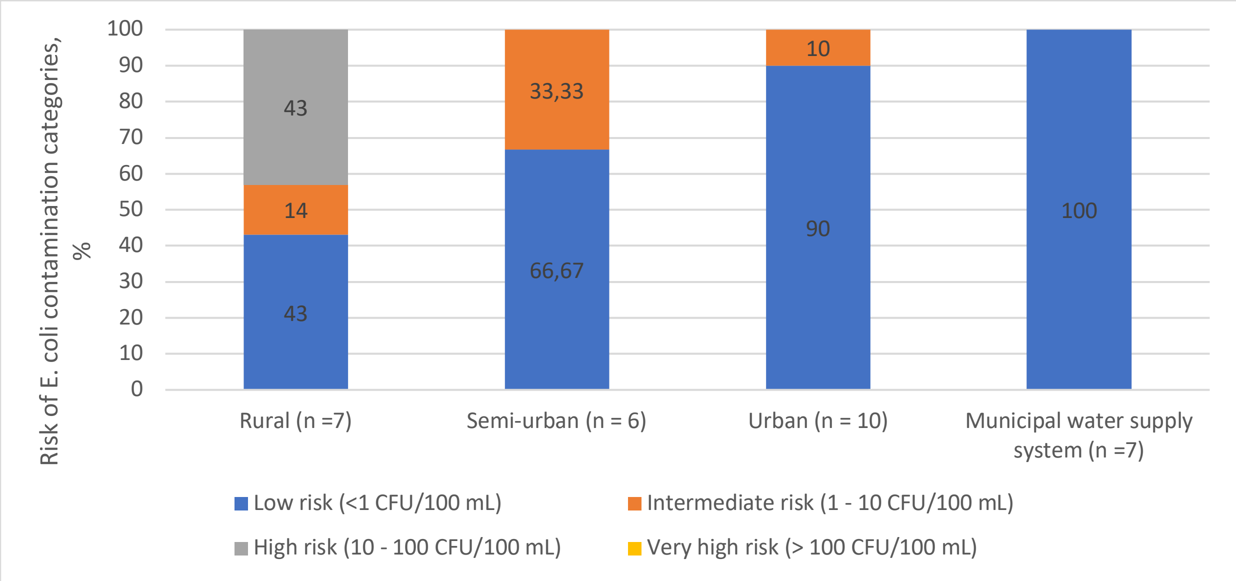


**Figure 3.** Health risk (in %) of *E. coli* contaminated in shallow tube wells from different settings and treated municipal water.

## 4. Discussion

This study reveals a pronounced prevalence and intensity of *E. coli* contamination in shallow tube wells, particularly in rural and semi-urban areas of Bedkot Municipality. In rural settings, over half (57.1%) of the wells were non-compliant with WHO guidelines, and 42.9% were classified as "high risk," with contamination levels reaching 45 CFU/100 mL. This presents a serious health threat, especially to vulnerable groups (Rai *et al.*, 2019). These findings align with studies from India, where rural groundwater contamination is linked to inadequate sanitation and agriculture (Megha *et al.*, 2015), and from across South Asia and Africa (Gautam *et al.,* 2025), (Ghosh *et al.,* 2023), (Sekgobela *et al.,* 2023). In Bedkot, probable contamination sources include poorly constructed pit latrines, insufficient separation between wells and sanitation facilities, and unsafe fecal sludge disposal (ENPHO, 2022).

Notably, semi-urban areas exhibited the highest rate of non-compliance (66.7%), exceeding rural levels. This likely reflects the rapid, unregulated expansion characteristic of peri-urban zones, where infrastructure and regulatory systems fail to keep pace with population growth (Dahal and Timalsina, 2020), creating a convergence of rural pressures (agricultural waste) and urban challenges (poorly managed septic tanks, inadequate sanitation in informal settlements) (Gautam *et al.*, 2025).

These risks are exacerbated by local hydrogeology. Sandy loam soils and shallow water tables in the Terai enable rapid microbial transport from surface sources, a vulnerability well-documented globally (Nayebare *et al.*, 2022), (Smith, 2001). Deficiencies in well construction—such as inadequate casing, cracked linings, and absent protective aprons—further heighten susceptibility to contamination (Luoto *et al.*, 2011), (Sekgobela *et al.*, 2023). In contrast, urban tube wells displayed markedly better water quality (90% low risk). This improvement likely reflects higher coverage of improved sanitation, denser paved surfaces that reduce infiltration, and better overall waste management. However, the one contaminated urban sample (U7) highlights the potential for "hotspots" due to infrastructure failures like sewage leakages (Gautam *et al.*, 2025), (Pant, 2011), underscoring that permeable soils mean even urban areas remain vulnerable (ENPHO, 2022).

Beyond infrastructure, climate and land-use are critical factors. Evidence shows rainfall can increase the likelihood of high *E. coli* contamination (Wu *et al.*, 2016). Similar seasonal influences are expected in Bedkot, where monsoon rains may amplify microbial transport, particularly under conditions of inadequate sanitation (Bain *et al.*, 2014), (Santos *et al.*, 2023).

A key positive finding was the complete absence of *E. coli* in all treated municipal water samples, highlighting the proven effectiveness of centralized chlorination in eliminating fecal pathogens and achieving compliance with WHO (2011) guidelines. This offers a scalable model for expansion to underserved populations.

The identified contamination gradient (Rural > Semi-Urban > Urban) mirrors Bedkot's developmental trajectory and underscores the necessity of tailored interventions. While rural areas require urgent sanitation upgrades, peri-urban zones demand integrated planning to address overlapping risks, and urban areas must maintain infrastructure resilience to prevent localized failures.

Comparisons with regional and international studies reinforce these findings. In Nepal's Terai region, thermotolerant coliforms and *E. coli* contamination have been widely reported in shallow wells (Gurung *et al.*, 2015), (Mahato *et al.*, 2018). Globally, rural sources are consistently more contaminated than urban ones (Santos *et al.,* 2023), though exceptions exist. For instance, peri-urban sources in Bangladesh and South Africa showed higher contamination than rural wells (Sekgobela *et al.,* 2023), (Wu *et al.,* 2016), while studies in Indonesia and India reported elevated contamination in urban settings (Gautam *et al.*, 2025), (Naily *et al.,* 2022). Compared to these contexts, Bedkot's contamination levels were relatively lower but still unsafe for consumption without treatment. The health implications are substantial, as consumption of this water poses a major risk for waterborne diseases, which remain a leading cause of morbidity and mortality in Nepal (Dhimal *et al.*, 2022), (Rai *et al.*, 2019). Children under five, the elderly, and immunocompromised individuals are particularly vulnerable to severe outcomes including diarrhea, cholera, typhoid, hepatitis, and malnutrition-related complications (Ghosh *et al.*, 2023), (WHO, 2023). This interplay of microbial contamination with poverty and limited healthcare access leads to significant socioeconomic costs (Dietler *et al.*, 2021), (Sarker *et al.*, 2021), reinforcing that microbial contamination poses the most immediate public health risk in the region, even as chemical risks persist (Hamal *et al.*, 2025).

**Implications for Policy and Practice**

The findings lead to the following evidence-based recommendations for Bedkot Municipality:

- Promote household water treatment and safe storage practices, particularly in rural and semi-urban communities (Luoto *et al.*, 2011).
- Establish routine, risk-based water quality testing of community and private wells, with public disclosure of results to inform user choices.
- Develop a municipal Water Safety Plan (WSP) focusing on wellhead protection, strict enforcement of safe distances between latrines and wells, and proactive sanitation upgrades (Diener et al., 2016).
- Prioritize the expansion of the treated piped water network to underserved rural and semi-urban areas, building on the proven efficacy of chlorination.
- Promote the construction of sealed latrines, improved septic systems, and better management of agricultural waste to reduce contamination at the source (ENPHO, 2022), (National Planning Commission, 2020).

## Conclusion

This study provides compelling evidence that a significant proportion of the population in Bedkot Municipality is exposed to fecal contamination through shallow tube well water. A clear spatial gradient of risk exists, with rural areas facing the greatest threat, followed by semi-urban zones, while urban areas show comparatively better—though not infallible—groundwater quality. The likely drivers of this contamination are multifaceted, stemming from inadequate sanitation and agricultural practices in rural areas, the transitional vulnerabilities of semi-urban expansion, and localized infrastructure failures in urban settings.

In stark contrast, the consistent absence of *E. coli* in treated municipal water confirms the critical role of centralized disinfection as an effective barrier. This underscores the expansion of this managed service as a primary solution. Overall, the findings highlight an urgent public health challenge that demands setting-specific interventions. Expanding access to safe municipal water, upgrading sanitation infrastructure, implementing comprehensive Water Safety Plans, and raising community awareness represent indispensable steps toward safeguarding drinking water and advancing tangible progress toward achieving SDG 6.1 in Bedkot and similar settings across the Terai.

## Limitations

Key limitations include the cross-sectional design, which captures contamination only during the pre-monsoon season and may not reflect seasonal dynamics (Smith, 2001). Furthermore, the relatively small sample size means the findings are indicative for the study area but may not be generalizable to the entire Terai region.

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**Data Availability Statement:** All datasets supporting the findings of this study are included in the manuscript.

**Disclosure statement:** *Conflict of Interest:* The authors declare that there are no conflicts of interest. *Compliance with Ethical Standards:* This article does not contain any studies involving human or animal subjects.

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