

Assessment Of Bacteriological Analysis Of Sewage Water, Beur, Patna

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Abstract

*The present study assessed the bacteriological quality of sewage water from the wastewater treatment plant at Beur. Wastewater samples were collected from primary, secondary, and tertiary treatment units to evaluate total and fecal coliform concentration, treatment efficiency, and temporal variability. Coliform enumeration was performed using the Most Probable Number (MPN) method, while IMViC biochemical tests were used for the isolation and identification of *Escherichia coli*. The results showed that bacterial concentration was highest in primary treated samples and gradually decreased in secondary and tertiary stages, indicating effective reduction of microbial contamination during treatment. Temporal variation in bacterial load was also observed during the study period. Statistical analysis revealed significant differences in coliform concentration among treatment stages. The study emphasizes the importance of wastewater treatment in reducing environmental pollution and protecting public health.*

Key Words: *fecal coliform, Coliform enumeration, biochemical tests*

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I. Introduction

Water Resources and Environmental Pollution Water is the most fundamental and essential natural resource required for sustaining all forms of life on Earth. It plays a critical role in various sectors including domestic consumption, industrial processes, and agricultural activities. Approximately 71% of the Earth's surface is covered with water, yet only 2.5% of this is freshwater, and merely 1% is accessible for human use (Gleick, 1996). The growing global population, coupled with rapid urbanization and industrialization, has led to an unprecedented increase in water demand, while simultaneously contributing to severe water quality degradation. Water pollution has emerged as one of the most pressing environmental challenges of the 21st century, affecting both developed and developing nations. The contamination of water bodies occurs through various anthropogenic activities including discharge of untreated sewage, industrial effluents, agricultural runoff containing pesticides and fertilizers, and improper waste disposal practices (Schwarzenbach et al., 2010). According to the United Nations World Water Development Report 2020, approximately 80% of global wastewater is discharged into the environment without adequate treatment, posing severe threats to human health and ecosystem integrity. In India, water pollution is particularly severe due to the high population density, inadequate sanitation infrastructure, and limited wastewater treatment facilities. The country generates approximately 62,000 million liters per day (MLD) of sewage from urban areas, while the installed treatment capacity stands at only 24,000 MLD, creating a treatment gap of about 61% (CPCB, 2021). This deficiency results in the discharge of vast quantities of untreated or partially treated sewage into rivers, lakes, and other water bodies, severely compromising water quality and public health. The state of Bihar, located in the fertile Indo-Gangetic plain, faces particularly acute water pollution problems. The Ganges River and its tributaries, which flow through the state, receive enormous quantities of untreated sewage from major urban centers including Patna, Bhagalpur, Munger, and other cities. Research conducted by the Bihar State Pollution Control Board has consistently shown that fecal coliform counts in the Ganges often exceed the permissible limit of 2,500 MPN/100 mL for bathing water by 10 to 100 times, with some locations recording counts as high as 240,000 MPN/100 mL (Dwivedi et al., 2018). These alarming levels of contamination pose serious health risks to the millions of people who depend on the river for drinking water, bathing, religious ceremonies, and agricultural activities. The situation in Bihar is further compounded by widespread open defecation in rural areas, inadequate sewage collection and treatment infrastructure in urban areas, and the discharge of industrial effluents without proper treatment. According to the Census of India 2011, only 23% of rural households in Bihar had access to improved sanitation facilities, resulting in approximately 40

million people practicing open defecation. This practice contributes significantly to the contamination of surface water and groundwater resources with pathogenic microorganisms.

Sewage Water: Composition and Characteristics

Sewage, also known as wastewater or domestic effluent, is the contaminated water generated from residential, commercial, institutional, and to some extent, industrial sources. It is essentially a dilute aqueous solution comprising approximately 99.9% water and 0.1% dissolved and suspended solids, including organic and inorganic materials, nutrients, pathogenic and non-pathogenic microorganisms, and various chemical compounds (Metcalf & Eddy, 2003)

The microbiological component of sewage

Important bacterial pathogens that may be present in sewage include *Salmonella typhi* and *Salmonella paratyphi* (causative agents of typhoid and paratyphoid fever), *Vibrio cholerae* (cholera), *Shigella* species (bacillary dysentery), *Campylobacter jejuni* (gastroenteritis), and various pathogenic strains of *Escherichia coli*. Viral pathogens include hepatitis A and E viruses, rotavirus, norovirus, adenovirus, and enteroviruses including poliovirus. Protozoan parasites such as *Giardia lamblia*, *Cryptosporidium parvum*, and *Entamoeba histolytica*, as well as helminth eggs from *Ascaris lumbricoides*, *Trichuris trichiura*, and hookworms, are also commonly found in sewage, particularly in regions with poor sanitation (Gerardi & Zimmerman, 2005). The presence and concentration of pathogenic microorganisms in sewage depend on the health status of the contributing population. In areas with endemic waterborne diseases, the pathogen load in sewage is considerably higher. For instance, studies in developing countries have found concentrations of *Salmonella* ranging from 10^2 to 10^4 per liter, *Giardia* cysts from 10^3 to 10^5 per liter, and helminth eggs from 10 to 10^3 per liter in raw sewage (Jiménez et al., 2010)

Coliform Bacteria as Indicator Organisms

The concept of using indicator organisms for assessing the microbiological quality of water was developed in the late 19th century and has since become the foundation of water quality monitoring worldwide. Indicator organisms are microorganisms whose presence, absence, or quantity in water provides information about the potential presence of pathogenic microorganisms and the degree of fecal contamination. First, coliforms are present in large numbers in the feces of humans and warm-blooded animals. Each gram of human feces contains approximately 10^8 to 10^9 coliforms, far exceeding the numbers of most pathogenic organisms. This abundance makes coliforms much easier to detect than actual pathogens, which may be present sporadically and in low numbers. Second, many coliform bacteria, particularly *E. coli*, demonstrate survival characteristics that are generally similar to or somewhat better than those of bacterial pathogens. They are generally more resistant to environmental stresses such as desiccation, temperature changes, and disinfection than most enteric pathogens. Therefore, the absence of coliforms provides a reasonable assurance of bacterial safety but may not guarantee the absence of these more resistant pathogens (Payment & Locas, 2011). Approximately 95% of fecal coliforms are *Escherichia coli*, with the remainder consisting mainly of thermotolerant strains of *Klebsiella*, *Citrobacter*, and *Enterobacter*. *E. coli* is considered the most specific indicator of fecal contamination. The World Health Organization (WHO) guidelines for drinking water quality state that *E. coli* or thermotolerant coliform bacteria must not be detectable in any 100 mL sample of water intended for drinking (WHO, 2017). The detection of *E. coli* in water indicates not only fecal contamination but also the possible presence of these pathogenic variants (Tallon et al., 2005).

Recent advances in microbiological techniques have led to the development of chromogenic and fluorogenic substrate methods that offer significant advantages in terms of speed and specificity. These methods employ media containing substrates that, when cleaved by enzymes specific to coliforms and *E. coli*, produce colored or fluorescent compounds. For example, the Colilert method uses ortho-nitrophenyl- β -D-galactopyranoside (ONPG) which produces a yellow color when cleaved by β -galactosidase (an enzyme present in coliforms), and 4-methylumbelliferyl- β -D-glucuronide (MUG) which produces fluorescence when cleaved by β glucuronidase (an enzyme specific to *E. coli*). These methods can simultaneously detect and differentiate total coliforms and *E. coli* within 18-24 hours, significantly reducing the analysis time compared to traditional methods (Fricker & Fricker, 1996).

II. Materials And Methodology

Study Area and Sample Collection

Water samples were collected from the Patna Beur Sewage Treatment Plant (STP), which is one of the major sewage treatment facilities serving Patna city, located at 25.6093° N latitude and 85.1376° E longitude. The treatment plant has a designed capacity of 120 million liters per day (MLD) and employs conventional activated sludge process for sewage treatment. Water samples were collected from three different treatment stages to

represent varying levels of treatment: (1) Primary treated water - collected from the outlet of primary clarification tanks after screening, grit removal, and sedimentation; (2) Secondary treated water - collected from the outlet of secondary clarifiers after biological treatment through activated sludge process; and (3) Final discharge water. Sample collection was conducted following standard protocols as described in Standard Methods for the Examination of Water and Wastewater (APHA, 2017).

III. Bacteriological Analysis

Most Probable Number (MPN) Method for Coliform Enumeration

The enumeration of coliform bacteria in water samples was performed using the Multiple Tube Fermentation Technique (Most Probable Number method) as described in Standard Methods (APHA, 2017). Inoculated tubes were incubated at $35^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$ for 24 hours and observed for gas production in Durham tubes and turbidity in the medium. Tubes showing both gas production (at least 10% of the Durham tube filled with gas) and turbidity were recorded as positive presumptive results. Tubes showing no gas after 24 hours were re-incubated for an additional 24 hours and examined again at 48 hours. Tubes negative after 48 hours were recorded as negative.

Positive presumptive tubes were confirmed using Brilliant Green Lactose Bile (BGLB) broth for total coliforms and EC broth for fecal coliforms. For total coliform confirmation, one loopful of culture from each positive presumptive tube was transferred to BGLB broth tubes and incubated at $35^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$ for 48 hours. For fecal coliform confirmation, culture from positive presumptive tubes was transferred to EC broth and incubated in a water bath at 44.5°C

$\pm 0.2^{\circ}\text{C}$ for 24 hours. Gas production in Durham tubes within 48 hours (for total coliforms) or 24 hours (for fecal coliforms) constituted a confirmed positive result.

The combination of positive and negative confirmed tubes across the dilution series was recorded, and the Most Probable Number of coliforms per 100 mL was determined using standard MPN tables (APHA, 2017). For samples requiring dilutions, the MPN value was multiplied by the appropriate dilution factor. Results were reported as MPN/100 mL with 95% confidence intervals.

IV. Isolation And Identification Of Escherichia Coli

Escherichia coli, as the most specific indicator of fecal contamination, was isolated and identified through a combination of selective culture on differential media and biochemical testing. Confirmed positive EC broth tubes (from fecal coliform testing at 44.5°C) were used as the source material for *E. coli* isolation.

MacConkey Agar, a selective and differential medium for isolation of coliform bacteria, was used as the primary isolation medium. The medium was prepared by suspending 50 g of MacConkey Agar powder (containing peptone, lactose, bile salts, neutral red, and crystal violet) in 1 liter of distilled water, heating to dissolve completely, autoclaving at 121°C for 15 minutes, cooling to approximately 50°C , and pouring into sterile Petri plates (approximately 20 mL per plate). Plates were allowed to solidify and stored inverted at 4°C until use (not exceeding one week).

Culture from confirmed positive EC broth tubes was streaked onto MacConkey Agar plates using sterile inoculating loops following the standard streak plate technique to obtain isolated colonies. Plates were incubated inverted at 35°C for 24 hours. After incubation, plates were examined for characteristic coliform colonies. Lactose-fermenting colonies (pink to red in color due to acid production) were considered presumptive *E. coli* or other coliform bacteria, while non-lactose-fermenting colonies (colorless or pale) were recorded but not further processed.

Well-isolated pink colonies from MacConkey Agar were selected (2-3 colonies per plate) and subjected to Gram staining to confirm the characteristic Gram-negative, rod-shaped morphology of coliform bacteria. Colonies showing Gram-negative rods were further tested using the IMViC tests (Indole, Methyl Red, Voges-Proskauer, and Citrate utilization tests) to differentiate *E. coli* from other coliforms.

The Indole test was performed by inoculating a pure colony into Tryptone Broth and incubating at 35°C for 24-48 hours. After incubation, 0.5 mL of Kovac's reagent was added to the tube. Development of a red color in the reagent layer indicated a positive result (indole production from tryptophan metabolism). *E. coli* characteristically produces a positive indole test.

The Methyl Red test utilized MR-VP broth inoculated with the test organism and incubated at 35°C for 48 hours. After incubation, 5 drops of methyl red indicator were added to the culture. Development of a distinct red color indicated a positive result (acid production from glucose fermentation with $\text{pH} < 4.4$). *E. coli* is methyl red positive.

The Voges-Proskauer test was performed using the same MR-VP broth culture used for the methyl red test. To a fresh aliquot of the 48-hour MR-VP broth culture, 0.6 mL of 5% alpha-naphthol solution and 0.2 mL of 40% potassium hydroxide were added, mixed well, and allowed to stand for 15-30 minutes with occasional shaking. Development of a pink to red color indicated a positive result (acetoin production). *E. coli* is typically VP

negative.

Citrate utilization was tested using Simmons Citrate Agar slants. The slants were inoculated by streaking the test organism on the slant surface and stabbing into the butt, then incubated at 35°C for 48 hours. Growth and development of blue color (from bromothymol blue indicator) indicated citrate utilization. *E. coli* is citrate negative (no growth or color change on citrate agar).

Isolates showing the characteristic *E. coli* pattern (Indole positive, Methyl Red positive, Voges-Proskauer negative, Citrate negative, or ++-- pattern) were confirmed as *E. coli*. The percentage of fecal coliform-positive samples that yielded confirmed *E. coli* was calculated for each water sample type.

The results of the present investigation on bacteriological analysis of sewage water at different treatment stages and its effect on wheat seed germination are presented in this chapter. The study was conducted over a period of five months (October 2025 to February 2026) and involved comprehensive microbiological analysis of water samples and systematic evaluation of germination parameters.

V. Results

Bacteriological Quality of Sewage Water

Total Coliform Bacteria

The concentration of total coliform bacteria in sewage water samples from different treatment stages showed substantial variation, reflecting the progressive removal of microbial contaminants through the treatment process. Table 1 presents the monthly variation in total coliform concentrations across different treatment stages during the study period.

Table 1: Total Coliform Bacteria Concentration (MPN/100 mL) in Different Treatment Stages

Sampling Month	Primary Treated Water	Secondary Treated Water	Discharge Water
October 2025	4.8×10^7	2.4×10^5	1.8×10^4
November 2025	5.2×10^7	2.8×10^5	2.1×10^4
December 2025	4.1×10^7	1.9×10^5	1.4×10^4
January 2026	3.9×10^7	1.7×10^5	1.3×10^4
February 2026	4.5×10^7	2.2×10^5	1.6×10^4
Mean ± SD	$4.5 \times 10^7 \pm 5.1 \times 10^6$	$2.2 \times 10^5 \pm 4.2 \times 10^4$	$1.6 \times 10^4 \pm 3.2 \times 10^3$

The data reveal a progressive reduction in total coliform concentrations through successive treatment stages. Primary treated water showed consistently high coliform counts, ranging from

3.9×10^7 to 5.2×10^7 MPN/100 mL, with a mean concentration of $4.5 \times 10^7 \pm 5.1 \times 10^6$ MPN/100 mL. These values indicate that primary treatment, which mainly involves physical removal of suspended solids, achieves limited reduction in coliform bacteria, estimated at approximately 40-50% based on typical raw sewage coliform concentrations.

Secondary biological treatment resulted in substantial reduction of coliform bacteria. The mean concentration in secondary treated water was $2.2 \times 10^5 \pm 4.2 \times 10^4$ MPN/100 mL, representing approximately 99.5% reduction compared to primary treated water. This reduction is attributed to the biological degradation of organic matter and die-off of bacteria in the activated sludge process, followed by sedimentation in secondary clarifiers.

Final discharge water, after tertiary treatment including disinfection, showed further reduction with mean coliform concentration of $1.6 \times 10^4 \pm 3.2 \times 10^3$ MPN/100 mL. This represents an additional 92.7% reduction from secondary treated water. However, it is noteworthy that even after complete treatment, the discharge water exceeded the Indian standard for unrestricted irrigation (≤ 1000 MPN/100 mL) by approximately 16-fold, though it met the standard for restricted irrigation (≤ 5000 MPN/100 mL for food crops eaten cooked).

Monthly variations in total coliform concentrations were observed across all treatment stages. The highest concentrations were generally observed in November 2025, while the lowest were in January 2026. These variations may be attributed to seasonal changes in sewage characteristics, temperature effects on treatment efficiency, and variations in hydraulic loading to the treatment plant.

Fecal Coliform Bacteria

Fecal coliform bacteria, being more specific indicators of fecal contamination than total coliforms, were enumerated separately using the elevated temperature test (44.5°C). The results are presented in Table 2.

Table 2: Fecal Coliform Bacteria Concentration (MPN/100 mL) in Different Treatment Stages

Sampling Month	Primary Treated Water	Secondary Treated Water	Discharge Water
October 2025	2.8×10^7	1.4×10^5	8.2×10^3
November 2025	3.1×10^7	1.7×10^5	9.5×10^3
December 2025	2.4×10^7	1.1×10^5	6.8×10^3

January 2026	2.2×10^7	9.5×10^4	6.1×10^3
February 2026	2.6×10^7	1.3×10^5	7.4×10^3
Mean \pm SD	$2.6 \times 10^7 \pm 3.6 \times 10^6$	$1.3 \times 10^5 \pm 2.9 \times 10^4$	$7.6 \times 10^3 \pm 1.4 \times 10^3$

Fecal coliform concentrations followed a similar pattern to total coliforms, with progressive reduction through treatment stages. The mean concentration in primary treated water was $2.6 \times 10^7 \pm 3.6 \times 10^6$ MPN/100 mL, constituting approximately 58% of the total coliform count, which is consistent with the expected proportion of thermotolerant coliforms in domestic sewage.

Secondary treatment reduced fecal coliform concentrations to $1.3 \times 10^5 \pm 2.9 \times 10^4$ MPN/100 mL, representing 99.5% removal efficiency, similar to that observed for total coliforms. Final discharge water contained $7.6 \times 10^3 \pm 1.4 \times 10^3$ MPN/100 mL fecal coliforms, showing 94.2% reduction from secondary treated water.

The ratio of fecal coliforms to total coliforms remained relatively constant across treatment stages (56-58% in primary treated water, 59-61% in secondary treated water, and 47-49% in discharge water), suggesting that treatment processes affect total and fecal coliforms proportionally, with slightly greater removal of non-fecal coliforms in the final disinfection stage.

Escherichia coli

Escherichia coli, being the most specific indicator of fecal contamination, was isolated and confirmed using MacConkey Agar and IMViC tests. The results are presented in Table 3.

Table 3: Escherichia coli Concentration (MPN/100 mL) and Detection Frequency

Treatment Stage	Mean E. coli (MPN/100 mL)	% of Fecal Coliforms that are E. coli	Detection Frequency
Primary Treated Water	$2.4 \times 10^7 \pm 3.2 \times 10^6$	92.3%	100% (15/15)
Secondary Treated Water	$1.1 \times 10^5 \pm 2.5 \times 10^4$	84.6%	100% (15/15)
Discharge Water	$6.4 \times 10^3 \pm 1.2 \times 10^3$	84.2%	100% (15/15)

E. coli was detected in 100% of samples from all treatment stages, indicating consistent fecal contamination. The mean concentration in primary treated water was $2.4 \times 10^7 \pm 3.2 \times 10^6$ MPN/100 mL, representing 92.3% of the fecal coliform count. This high proportion of E. coli among fecal coliforms confirms the predominant fecal origin of the sewage contamination.

Secondary treatment reduced E. coli concentrations to $1.1 \times 10^5 \pm 2.5 \times 10^4$ MPN/100 mL (99.5% reduction), while final discharge water contained $6.4 \times 10^3 \pm 1.2 \times 10^3$ MPN/100 mL

E. coli (94.2% reduction from secondary treated water). The proportion of E. coli among fecal coliforms decreased slightly through treatment (from 92.3% to 84.2%), possibly due to differential survival or removal rates of E. coli versus other thermotolerant coliforms.

Statistical Analysis of Bacteriological Data

One-way ANOVA performed on log-transformed bacterial count data revealed highly significant differences ($p < 0.001$) among treatment stages for all bacterial groups (total coliforms, fecal coliforms, and E. coli). Tukey's HSD post-hoc test indicated that all pairwise comparisons between treatment stages were significantly different ($p < 0.05$), confirming that each treatment stage achieved statistically significant reduction in bacterial concentrations.

Correlation analysis revealed strong positive correlations among the three bacterial groups: total coliforms vs fecal coliforms ($r = 0.97, p < 0.001$), total coliforms vs E. coli ($r = 0.96, p < 0.001$), and fecal coliforms vs E. coli ($r = 0.99, p < 0.001$). These high correlations indicate that the three bacterial groups behave similarly through the treatment process and that any of them can serve as a reliable indicator of treatment efficiency.

Summary of Results

The key findings of this study bacteriological quality can be summarized as follow:

- Total coliform bacteria concentrations decreased progressively through treatment stages from 4.5×10^7 MPN/100 mL (primary treated) to 2.2×10^5 MPN/100 mL (secondary treated) to 1.6×10^4 MPN/100 mL (discharge water).
- Fecal coliform and E. coli concentrations showed similar reductions, with final discharge water containing 7.6×10^3 and 6.4×10^3 MPN/100 mL respectively.
- Treatment efficiency was approximately 99.5% for secondary treatment and 92-94% for tertiary treatment.
- Final discharge water exceeded unrestricted irrigation standards but met restricted irrigation standards for microbiological quality.

VI. Discussion

The present investigation examined the bacteriological quality of sewage water at different treatment stages; The findings provide valuable insights into both the effectiveness of the Patna Beur sewage treatment plant in reducing microbial contaminants. This chapter discusses the results in the context of existing literature, explores the mechanisms underlying observed effects.

Total Coliform bacteria

The progressive reduction in total coliform bacteria through successive treatment stages observed in this study reflects the cumulative effect of physical, chemical, and biological processes employed in conventional activated sludge treatment. The mean total coliform concentration of 4.5×10^7 MPN/100 mL in primary treated water, representing approximately 40-50% reduction from raw sewage, is consistent with values reported in similar studies. Kumar et al. (2013) documented total coliform concentrations of 3.8×10^7 to 6.2×10^7 MPN/100 mL in primary treated sewage from a treatment plant in Haridwar, closely matching our findings. This limited reduction during primary treatment is expected, as the primary clarification process mainly targets suspended solids removal through physical sedimentation, with minimal direct impact on dissolved or colloidal bacteria.

The substantial reduction achieved through secondary biological treatment (99.5% removal, reducing concentrations from 4.5×10^7 to 2.2×10^5 MPN/100 mL) demonstrates the effectiveness of the activated sludge process in microbial removal. This removal efficiency is comparable to those reported in the literature. Toze (2006) noted that conventional secondary treatment typically achieves 1 to 2 log reduction (90-99% removal) of bacteria. Our observed reduction of 2.3 log units falls well within this range and slightly exceeds typical performance, possibly due to the relatively long hydraulic retention time (approximately 6-8 hours) and sludge age (8-10 days) employed at the Patna Beur plant, which allows extended contact between wastewater and activated sludge biomass.

The mechanisms of bacterial removal in activated sludge systems are multiple and synergistic. Physical mechanisms include adsorption of bacteria onto activated sludge flocs, followed by removal during secondary sedimentation. The extracellular polymeric substances (EPS) produced by microorganisms in activated sludge create a sticky matrix that can trap bacterial cells. Biological mechanisms include predation by protozoa, particularly ciliated protozoa that actively feed on bacteria. Bacteriophages (viruses that infect bacteria) present in sewage can also contribute to bacterial reduction. Additionally, natural die-off occurs as bacteria encounter unfavorable conditions including nutrient depletion, competition from the diverse activated sludge microflora, and exposure to toxic metabolites. The relative contribution of each mechanism varies depending on operational conditions, but collectively they achieve substantial bacterial reduction even without specific disinfection.

The final reduction achieved through tertiary treatment and disinfection (92.7% reduction from secondary treated water, from 2.2×10^5 to 1.6×10^4 MPN/100 mL) demonstrates effective but incomplete disinfection. Based on discussions with plant operators, the plant employs chlorination with a target residual chlorine concentration of 0.5-1.0 mg/L after 30 minutes contact time. The observed reduction of approximately 1.1 log units is somewhat lower than the 2-3 log reduction typically expected from chlorination. This could be attributed to several factors including chlorine demand from residual organic matter, presence of suspended solids that can shield bacteria from disinfectant contact, and possible regrowth of chlorine-resistant bacteria. Chaudhry et al. (2015) noted that chlorination effectiveness can be compromised when applied to effluent with high turbidity or organic content, as these constituents consume chlorine before it can effectively inactivate microorganisms.

Fecal Coliform and E. coli Concentrations

The proportion of fecal coliforms among total coliforms observed in this study (approximately 58% in primary treated water) is consistent with typical domestic sewage characteristics. This proportion remained relatively stable through primary and secondary treatment but decreased slightly in final discharge water (47-49%). This slight preferential removal of fecal coliforms during disinfection might reflect their slightly greater sensitivity to chlorine compared to some non-fecal coliforms, possibly due to differences in cell wall structure or enzymatic activities.

The high proportion of E. coli among fecal coliforms (84-92% across all treatment stages) confirms that the sewage contamination is predominantly of fecal origin, which is expected for domestic sewage. The consistency of this proportion through treatment stages suggests that E. coli and other thermotolerant coliforms have similar survival characteristics through the treatment process. This finding validates the use of E. coli as a reliable indicator of fecal contamination in treated wastewater, as it reflects the behavior of the broader fecal coliform group.

The final E. coli concentration of 6.4×10^3 MPN/100 mL in discharge water exceeds the WHO guideline of ≤ 1000 E. coli/100 mL for unrestricted irrigation by approximately 6-fold. However, it is well below the WHO guideline of $\leq 10^5$ E. coli/100 mL for restricted irrigation. From the Indian regulatory perspective, the discharge water meets the CPCB standard of ≤ 1000 fecal coliforms/100 mL for food crops eaten cooked, but exceeds the standard for crops eaten raw. These comparisons indicate that while the treatment plant achieves substantial microbial reduction, an additional treatment barrier would be needed if the effluent were to be used for unrestricted irrigation of crops consumed raw, such as salad vegetables.

Temporal Variability in Treatment Efficiency

The monthly variations in bacterial concentrations observed in this study, with highest values in November and lowest in January, likely reflect multiple interacting factors. Temperature is a primary driver of seasonal variation in both sewage characteristics and treatment efficiency. Lower temperatures during winter months (December-January) generally slow microbial metabolism in the activated sludge system, potentially reducing treatment efficiency. However, lower temperatures also reduce microbial reproduction rates in the sewage itself and enhance microbial die-off in clarifiers, which can result in lower bacterial concentrations in effluent. The observed pattern suggests that the latter effect (reduced bacterial survival) outweighed any reduction in treatment efficiency at lower temperatures.

Rainfall and associated stormwater infiltration into sewerage systems can significantly affect sewage quality and treatment plant performance. November typically experiences some residual monsoon rainfall in Bihar, which could dilute sewage (reducing bacterial concentrations per unit volume while increasing total flow) and cause hydraulic overloading of the treatment plant (reducing retention time and treatment efficiency). These competing effects could explain the higher bacterial concentrations observed in November. The dry winter months (December-February) typically see more stable sewage composition and hydraulic loading, potentially contributing to more consistent and effective treatment.

Variations in domestic water use patterns, holiday periods affecting institutional contributions, and changes in industrial discharges to the sewerage system can also contribute to temporal variability. Understanding and accounting for such variability is important for designing reliable treatment systems and setting appropriate discharge or reuse standards that provide adequate safety margins throughout the year. The progressive reduction in total coliform bacteria through successive treatment stages observed in this study reflects the cumulative effect of physical, chemical, and biological processes employed in conventional activated sludge treatment. The mean total coliform concentration of 4.5×10^7 MPN/100 mL in primary treated water, representing approximately 40-50% reduction from raw sewage, is consistent with values reported in similar studies. Kumar et al. (2013) documented total coliform concentrations of 3.8×10^7 to 6.2×10^7 MPN/100 mL in primary treated sewage from a treatment plant in Haridwar, closely matching our findings. This limited reduction during primary treatment is expected, as the primary clarification process mainly targets suspended solids removal through physical sedimentation, with minimal direct impact on dissolved or colloidal bacteria. The substantial reduction achieved through secondary biological treatment (99.5% removal, reducing concentrations from 4.5×10^7 to 2.2×10^5 MPN/100 mL) demonstrates the effectiveness of the activated sludge process in microbial removal. This removal efficiency is comparable to those reported in the literature. Toze (2006) noted that conventional secondary treatment typically achieves 1 to 2 log reduction (90-99% removal) of bacteria. Our observed reduction of 2.3 log units falls well within this range and slightly exceeds typical performance, possibly due to the relatively long hydraulic retention time (approximately 6-8 hours) and sludge age (8-10 days) employed at the Patna Beur plant, which allows extended contact between wastewater and activated sludge biomass.

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The final reduction achieved through tertiary treatment and disinfection (92.7% reduction from secondary treated water, from 2.2×10^5 to 1.6×10^4 MPN/100 mL) demonstrates effective but incomplete disinfection. Based on discussions with plant operators, the plant employs chlorination with a target residual chlorine concentration of 0.5-1.0 mg/L after 30 minutes contact time. The observed reduction of approximately 1.1 log units is somewhat lower than the 2-3 log reduction typically expected from chlorination. This could be attributed to several factors including chlorine demand from residual organic matter, presence of suspended solids that can shield bacteria from disinfectant contact, and possible regrowth of chlorine-resistant bacteria. Chaudhry et al. (2015) noted that chlorination effectiveness can be compromised when applied to effluent with high turbidity or organic content, as these constituents consume chlorine before it can effectively inactivate microorganisms.

VII. Conclusion

This study investigated the bacteriological quality of sewage water at different treatment stages at the Patna Beur Sewage Treatment Plant, the bacteriological analysis revealed progressive reduction in microbial contamination through successive treatment stages, Treatment Efficiency: The Patna Beur Sewage Treatment Plant achieves substantial reduction in bacterial contamination through its multi-stage treatment process.

Secondary biological treatment (activated sludge process) is the most critical stage, achieving approximately 99.5% removal of indicator bacteria. However, even after complete treatment including disinfection, the final discharge water contains bacterial concentrations exceeding WHO guidelines

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