# EVALUATION OF BACTERIAL AND FUNGAL LOAD IN WATER STORAGE TANKS IN ENUGU STATE UNIVERSITY OF SCIENCE AND TECHNOLOGY HOSTELS

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## ABSTRACT

This is a cross-sectional study evaluating bacterial and fungal loads in water storage tanks across selected hostels in the Enugu State University of Technology (ESUT). A total of 31 water samples were obtained and analyzed by serial dilution using pour plate method, by means of morphology, biochemical identification and microscopy, identifying six bacterial species; (*Escherichia coli*(19.4%), *Salmonella spp.*(16.1%), *Pseudomonas aeruginosa.*(9.7%), *Shigella spp.*(9.7%), *Staphylococcus aureus.*(9.7%), *and Klebsiella spp.* (6.5%) and three fungal species; (*Candida spp.*(9.7%), *Rhizopus spp.*(9.7%), *Mucor spp*.(16.1%). Quantitative analysis revealed bacterial loads ranging from 1.07 to 2.07 × $10^3$ CFU/ml and Fungi, from 3.8 to 4.9 × $10^3$ in Agbani hostels, with Parklane hostels exhibiting "Too Numerous to Count" levels in several samples. Fungal counts peaked at 6.2 × $10^3$ CFU/mL in Parklane Hostel 2. This project work had a duration of 2 weeks. The findings align with global research on waterborne pathogens and emphasize the need for regular water quality monitoring, improved tank maintenance, and public education on water safety.

## INTRODUCTION

Water storage tanks are integral components of water supply systems, especially in areas with irregular water availability. These systems are pivotal for maintaining water supply continuity but also present significant risks of microbiological contamination. The storage of water creates an environment conducive to chemical, physical, and microbial changes, which may compromise water quality and pose public health risks if not properly managed.

Water tanks often harbor diverse microbial communities, including bacteria and fungi. These microorganisms can form biofilms, deteriorate water quality, and potentially cause health issues. Bacterial contaminants are often linked to gastrointestinal illnesses, while fungal contaminants, although historically understudied, are increasingly recognized for their potential to cause opportunistic infections, allergies, and intoxications. The resilience of fungi under oligotrophic conditions (low nutrient levels) makes them a critical concern in stored water systems (Babič *et al.,* 2017).

Several factors influence microbial growth in water storage tanks, such as temperature fluctuations, residual disinfectant levels, organic matter, and structural materials of the tanks. In particular, fungi like Aspergillus and Penicillium thrive in these conditions, sometimes degrading materials like concrete and affecting tank integrity (Babič *et al*., 2017). Poor maintenance, prolonged storage times, and inadequate disinfection further exacerbate microbial proliferation (Zhou *et al.,* 2017).

The microbial contamination of water tanks is linked to outbreaks of waterborne diseases, particularly in high-density living areas like hostels. Beyond gastrointestinal illnesses, waterborne fungi have been associated with more complex health risks, emphasizing the need for their inclusion in water quality monitoring frameworks. Regular water quality assessments, proper tank maintenance, and adherence to global water safety guidelines are vital for mitigating risks (WHO, 2021).

Bacterial species such as Escherichia coli, Salmonella spp., and Pseudomonas aeruginosa are commonly identified in water storage tanks. These organisms are indicators of fecal contamination or inadequate disinfection processes. They are often associated with gastrointestinal illnesses and

other waterborne diseases (Zhou *et al* ., 2022). Fungi on the other hand in water systems are an emerging area of concern, with species such as Aspergillus, Penicillium, and Fusarium frequently identified in stored water. Unlike bacteria, fungi are resilient under oligotrophic (low nutrient) conditions, making them capable of thriving in chlorinated water. They can degrade tank materials, impact water aesthetics (taste, color, odor), and pose risks as opportunistic pathogens or allergens. Mixed bacterial and fungal biofilms further complicate water quality issues and cleaning processes (Babič *et al.*, 2017).

Access to clean and safe water is essential for maintaining public health and well-being. However, water storage systems, such as water tanks, can become reservoirs for microbial contamination if not properly maintained. In the Enugu State University of Science and Technology (ESUT) hostels, water tanks serve as a primary source of water for students, yet there is limited research on the microbial quality of this water. The presence of bacterial and fungal contaminants in these tanks could pose significant health risks, including waterborne diseases and infections.

This study seeks to address the gap in understanding the microbial load in water tanks at ESUT hostels by evaluating the levels of bacterial and fungal contamination. The findings will provide insights into the potential health implications for students and inform recommendations for improving water quality and hygiene practices in the hostels.

The aim of this study is to evaluate the bacterial and fungal load in water tanks at the hostels of the Enugu State University of Science and Technology (ESUT) to determine the microbial quality of the water and assess its potential health implications on the students.

## MATERIALS AND METHOD

### STUDY AREA

This study was conducted with water samples from Enugu State University Of Science And Technology (ESUT) hostels

## STUDY DESIGN

This study will employ a cross-sectional design which would require quantitative methods to evaluate bacterial and fungal load in water from water tanks

## STUDY POPULATION

This study includes water tanks in Enugu State University Of Science And Technology hostels in Parklane and Agbani. **SAMPLE SIZE**

Water samples were collected from 31 different water tanks (12 from Agbani and 19 from Parklane)

## SAMPLING TECHNIQUE

Water was aseptically sampled from 31 water tanks (19 from Parklane and 12 from Agbani) into sterile plastic containers. The water sources were differentiated by labelling the bottles. This helped in not mistaking one for another. The samples were instantly placed in a cooler with ice cubes as described by (Adeleye *et al.*, 2020) and transported to the laboratory for analysis.

## SAMPLE ANALYSIS

- **Total Bacteria Count**

Serial dilution of the water samples were carried out using a sterile pipette. An aliquot (1ml) of each water sample was homogenized with 9ml of distilled water in sterilized test tubes. Using standard pour plate method, 0.1ml of the serially diluted water were aseptically withdrawn from the test tubes for each sample and were introduced in sterile petri-dishes required for the Nutrient agar, Chocolate agar and Salmonella and Shigella agar. They were incubated in an inverted position in an incubator at 37°C for 24-48 hours and the viable colonies were counted as described by (Cheesbrough 2006). The isolates were characterized and identified based on their cultural, morphological (Gram Staining) and biochemical characteristics (Agu *et al.*, 2023). The total bacteria count was calculated as thus;

$$\text{CFU/ml} = \frac{N}{VD}$$

Where;

CFU/ml = Colony forming unit per milliliter

N = Number Of Colonies

D = Dilution Factor

V = Volume Of Culture Plated

- **Total Fungal Count**

Aliquot of 0.1 ml of dilution of the sample were inoculated into Sabouraud dextrose agar (SDA) and incubated at 28°C for 72 hours. Thereafter the developing colonies were counted.

- **Fungal Morphology**

Fungal growth on plate culture was observed; surface, spore and colour. Stained (lactophenol cotton blue) slide was examined using a microscope (×40) for structure of hyphae and details of sporulating structure (Agu and Chidozie, 2021).

- **DATA ANALYSIS**

The collected data were entered into Excel spreadsheet 2009 and summarized using table, percentages and mean. Additionally, the Pearson correlation coefficient used to analyze the relationship between Bacteria and fungal count.

## RESULT

In this study, the occurrence and load of fungi and bacteria in water samples from Agbani and Parklane hostels of ESUT were determined. 31 samples were collected and 9 isolates where isolated from the sample. These include 6 bacterial isolates and 3 fungal isolates. The bacterial isolates include: *Escherichia coli, Salmonella spp., Pseudomonas spp., Staphylococcus aureus, Shigella spp., Klebsiella spp.* The fungal isolates include: *Candida spp., Rhizopus spp. and Mucor spp.*

Results derived from the analysis of sample with bacteria and fungi isolated from each hostel sample is depicted in Table 1 below.

**Table 1:** Isolates found in the sample

| **Sample** | **Bacterial isolates** | **Fungal isolates** |
|---|---|---|
| Nomeks | A, B, G | D, E |
| Noca | A, B, C | E, F |

| | | |
|---|---|---|
| Diamond A | A, C, G | F |
| Diamond B | A, C, G, I | E |
| Hostel 1 | A, B, C, H | F |
| Hostel 2 | A, B, H, I | D, F |
| Hostel 3 | A, C, H | D, F |

**KEY:**

A = *Escherichia coli*

B = *Salmonella spp.*

C = *Pseudomonas aeruginosa*

D = *Candida spp.*

E = *Rhizopus spp.*

F = *Mucor spp.*

G = *Shigella spp.*

H = *Staphylococcus spp.*

I = *Klebsiella spp.*

Identified isolates from each sample for each hostel were evaluated to determine the load and the results are depicted in Table 2 below.

**Table 2: Total Bacterial And Fungal Count In Agbani hostels**

| **Samples** | **Bacterial count ($10^3$cfu/ml)** | **Fungal count ($10^3$cfu/ml)** |
|---|---|---|
| **NOCA** | | |
| NA 1 | 1.17 | 4.1 |
| NA 2 | 1.09 | 4.1 |
| NA 3 | 1.07 | 4.4 |
| **NOMEKS** | | |
| NK 1 | 2.05 | TFTC |
| NK 2 | 2.02 | TFTC |
| NK 3 | 2.07 | TFTC |

| | | |
|---|---|---|
| **DIAMOND A** | | |
| DA 1 | 1.30 | 3.5 |
| DA 2 | 1.32 | 3.5 |
| DA 3 | 1.35 | 3.8 |
| **DIAMOND B** | | |
| DB 1 | 1.07 | 4.3 |
| DB 2 | 1.02 | 4.9 |
| DB 3 | 1.04 | 4.3 |

**KEY:**

NA = Noca DA = Diamond A TFTC = Too few to count

NK = Nomeks DB = Diamond B

**Table 3: Total Bacteria And Fungal Count In Parklane hostels**

| **Samples** | **Bacterial count ($10^3$cfu/ml)** | **Fungal count ($10^3$cfu/ml)** |
|---|---|---|
| **HOSTEL 1** | | |
| H1 – A | 2.03 | TFTC |
| H1 – B | 2.01 | TFTC |
| H1 – C | TNTC | TFTC |
| H1 – D | 1.17 | NG |
| H1 – E | TNTC | NG |
| **HOSTEL 2** | | |
| H2 – A | 2.11 | 3.4 |
| H2 – B | 1.01 | 5.3 |
| H2 – C | TNTC | NG |
| H2 – D | 2.13 | TFTC |
| H2 – E | 1.79 | 4.2 |

| | | |
|---|---|---|
| H2 – F | 1.45 | 3.9 |
| H2 – G | 2.02 | 3.2 |
| **HOSTEL 3** | | |
| H3 – A | TNTC | NG |
| H3 – B | TNTC | NG |
| H3 – C | 2.03 | 3.3 |
| H3 – D | 1.76 | 4.5 |
| H3 – E | 1.09 | 4.7 |
| H3 – F | 2.11 | TFTC |
| H3 – G | TNTC | NG |

**KEY:**

H1 = Hostel 1 TNTC = Too numerous to count

H2 = Hostel 2 TFTC = Too few to count

H3 = Hostel 3 NG = No growth

**Table 3: Mean Bacterial and Fungal Count By Hostel Location**

| HOSTEL | Mean Bacterial Count ($10^{-3}$ CFU/ml) | Mean Fungal Count ($10^{-3}$ CFU/ml) |
|---|---|---|
| Agbani | 1.16 | 4.10 |
| Parklane | 1.66 | 4.06 |

**Table 4:**Morphological And Biochemical Identification Of Bacteria Isolates

| **Isolates** | **Surface** | **Color** | **Ele** | **Gram** | **Cat** | **Coa** | **Ind** | **MR** | **UR** | **VP** | **Oxi** | **Mot** | **Glu** | **Lac** | **Suc** | **Probable Organism** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| A | Smooth | Whitish | Convex | -Rod | + | - | + | + | - | - | + | + | + | + | + | *Escherichia coli* |
| B | Smooth | Greyish/Whitish | Convex | -Rod | + | - | + | + | + | - | - | + | + | - | - | *Salmonella spp* |
| C | Glistening | Cream | Flat | -Rod | + | - | - | - | + | - | + | - | + | - | - | *Pseudomonas spp* |
| G | Smooth | Cream | Raised | - Rod | + | - | + | + | - | - | - | - | + | - | - | *Shigella spp* |
| H | Smooth | Cream | Convex | + Cocci | + | + | - | + | - | + | - | - | + | + | - | *Staphylococcus aureus* |
| I | Glistening | Cream | Riased | -Rod | + | - | - | + | + | - | - | - | + | + | + | *Klebsiella spp* |

**KEYS:**

Ele = Elevation

Gram = Gram reaction

Cat = Catalase test

Coa = Coagulase test

Ind = Indole test

MR = Methyl-red test

UR = Urease test

VP = Voges-Proskauer test

Oxi = Oxidase test

Mot = Motility test

Glu = Glucose test

Lac = Lactose test

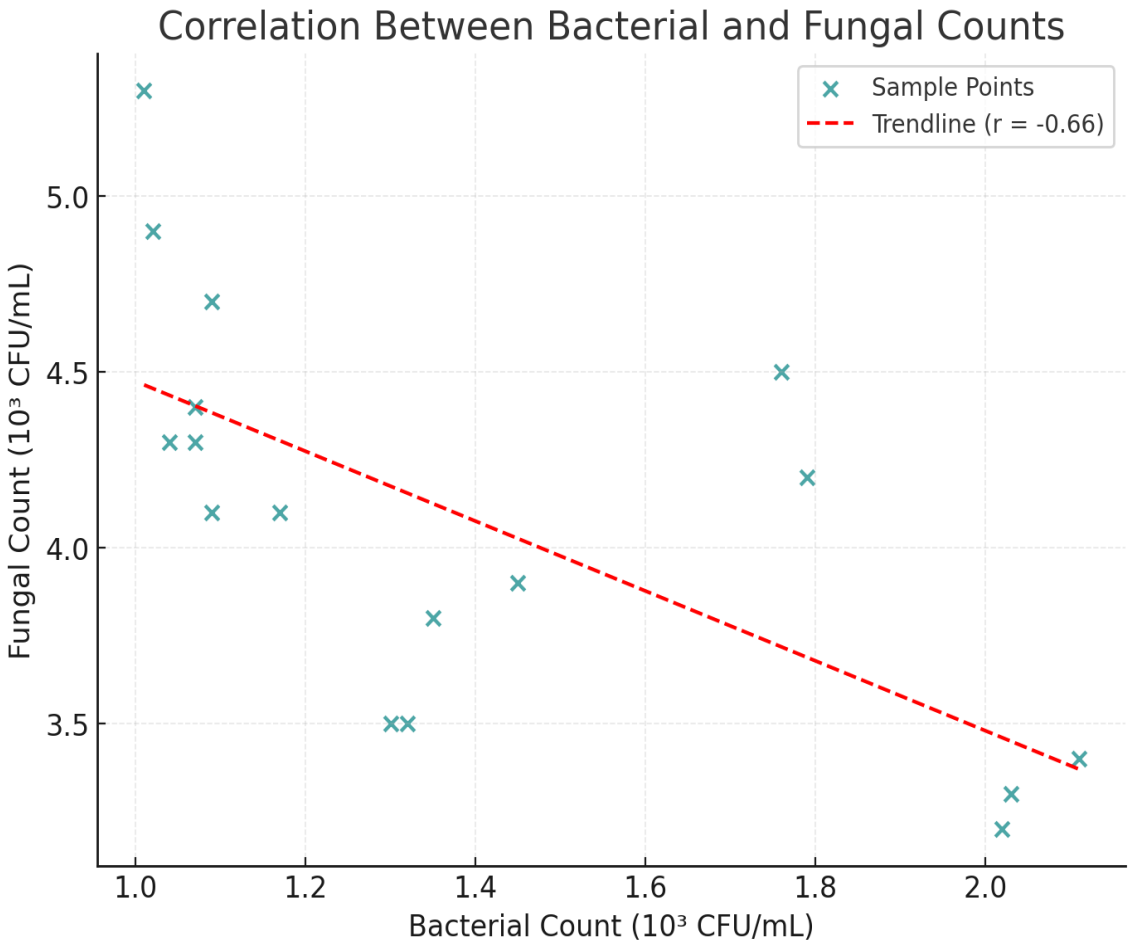
Suc = Sucrose test

**Table 5:** Fungal Morphology And Microscopy

| ISOLATES | COLONIAL MORPHOLOGY | MICROSCOPY | PROBABLE ISOLATE |
|---|---|---|---|
| D | On SDA, colonies are white and creamy with smooth surface. There appear to be circular in shape | They are oval shaped and are 3-6 micrometer in diameter. They appear to form | *Candida specie* |

| | | | |
|---|---|---|---|
| | | psuedohyphae which are elongated and with branching filaments that resemble true hyphae | |
| E | On SDA, they appear grayish white. They exhibit a root like growth pattern and appear to be growing downwards into the agar | They are aseptate and about 7micrometer in length. They appear to be branching with thick hyphae | *Rhizopus spp* |
| F | On SDA, they appear whitish, irregular and woolly. They appear to be growing upwards and outwards from the center | There is absence of hyphae. The show branching characteristics and are irregularly shaped | *Mucor spp* |

**Fig 1: Showing Correlation Between Bacterial And Fungal Count In Sample**




## DISCUSSION

The Bacterial and fungal counts of isolates from the water samples in Agbani and Parklane hostels are shown in table 1 and table 2. In Agbani hostel, the total bacterial count ranges from 1.02 x$10^3$ to 2.07 x$10^3$ cfu/ml, where **Sample NK3** recorded the highest bacteria count of 2.07 x$10^3$ and **Sample DB4** recorded the lowest bacteria count of 1.02 x$10^3$. Fungal counts ranged from 3.8 x$10^3$ to 4.9 x$10^3$cfu/ml. Parklane recorded bacterial counts ranging 1.01 x$10^3$ to 2.13 x$10^3$ and Fungal count of 3.2 x$10^3$ to 6.2 x$10^3$.

The bacterial isolates identified include *Escherichia coli, Salmonella spp., Pseudomonas spp., Staphylococcus aureus, Shigella spp., and Klebsiella spp.*. These organisms are associated with varying degrees of waterborne diseases and contamination:

- ***Escherichia coli* (19.4%)** was the most frequently isolated bacterial organism similar to other studies (Adeleye *et al.*, 2022). Its presence suggests fecal contamination, posing risks of gastrointestinal infections.
- ***Salmonella spp. and Pseudomonas spp*. (16.1% each)** are associated with diseases such as typhoid fever and nosocomial infections, respectively. Pseudomonas spp. is also known for its environmental resilience.
- ***Shigella spp. and Staphylococcus spp. (9.7% each)*** are less frequent but significant, as Shigella is a major cause of dysentery, while Staphylococcus can lead to skin and soft tissue infections.
- ***Klebsiella spp.* (6.5%)** is a common pathogen in immunocompromised individuals and is an indicator of biofilm formation in water systems.

The fungal isolates identified were Candida spp., Rhizopus spp., and Mucor spp.:

- ***Mucor spp. (16.1%)*** was the most prevalent fungal species, often associated with allergic reactions and mucormycosis in immunocompromised individuals.
- ***Candida spp. and Rhizopus spp.* (9.7% each)** are opportunistic pathogens. Candida is a significant concern due to its association with candidiasis, while Rhizopus is linked to zygomycosis.

**Correlation Between Bacterial and Fungal Loads**

The negative correlation (− 0.66) from **Fig 1,** between bacterial and fungal counts indicates an inverse relationship, suggesting that factors favoring bacterial proliferation may inhibit fungal growth, and vice versa. This could be due to competition for nutrients, environmental factors, or the presence of antimicrobial compounds produced by some microbes.

**Health Implications**

- Waterborne Diseases:

Pathogens such as E. coli, Salmonella spp., and Candida spp. are associated with diarrhea, typhoid fever, and fungal infections, respectively. The detection of these organisms poses a significant public health risk (Patrick et al., 2019).

High microbial loads also increase the likelihood of antimicrobial resistance, complicating disease management

- Vulnerable Populations:

Individuals with compromised immune systems are particularly at risk from fungal contaminants like Candida spp. and bacterial pathogens (Patrick et al.,).

## CONCLUSION

The evaluation of bacterial and fungal loads in water tanks at ESUT hostels reveals significant microbial contamination, emphasizing the urgent need for improved water management practices. The findings demonstrate the widespread presence of pathogenic bacteria such as Escherichia coli, Salmonella spp., and Shigella spp., indicative of fecal contamination and poor sanitation. Similarly, fungal species like Candida spp. were detected, highlighting the role of environmental factors and inadequate tank maintenance in fostering microbial growth.

This study aligns with global findings on water contamination, reinforcing the importance of routine water quality assessments and educational initiatives to enhance public awareness and hygiene practices. By addressing these challenges, ESUT can ensure safer water supply systems and better health outcomes for its students

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