MICROBIAL COMMUNITIES IN LAUNDRY WASTEWATER AND THEIR ROLE IN BIODEGRADATION OF SYNTHETIC DETERGENTS

P.Priyadarshini [1*] and Hemavathy Sundaram[2], Summera Rafiq[1] and SK. Jasmine Shahina[1]

[1*]Associate Professor, Department of Microbiology, Justice Basheer Ahmed Sayeed College for Women, Teynampet, Chennai.

[2]II. M.Sc. Applied Microbiology, Department of Microbiology, Justice Basheer Ahmed Sayeed College for Women, Teynampet, Chennai.

[3]Associate Professor, Department of Microbiology, Justice Basheer Ahmed Sayeed College for Women, Teynampet, Chennai.

*** Corresponding author email: summerarafiq@jbascollege.edu.in**

Abstract:

The widespread use of synthetic detergents in household laundry processes has led to their accumulation in wastewater, posing significant environmental challenges due to their persistence and potential toxicity. This study aimed to investigate the microbial communities present in laundry wastewater and evaluate their role in the biodegradation of synthetic detergents. Wastewater samples were collected from domestic laundry facilities, and microbial communities were isolated and characterized using culture-dependent techniques. 52 bacteria and 8 fungi were isolated from a total of 20 laundry wash water samples. Most of the bacterial isolates were Pseudomonas species, although other bacterial species like *Bacillus, Staphylococcus, Escherichia, Acinetobacter,* and *Enterobacter spp* were isolated. The increased organic matter content of liquid detergents probably contributed to the higher prevalence of fungal isolates, particularly *Aspergillus* and *Penicillium*. The diversity of microorganisms in the samples varied greatly; samples of liquid detergent included a wider variety of microbial species, whereas samples of powder detergent mostly contained bacterial isolates. *Aspergillus* showed resistance to lower detergent concentrations (1% and 5%), several bacteria, particularly *Pseudomonas* *spp* and *Bacillus spp* demonstrated notable tolerance to higher detergent concentrations from 10% to 15%. The results suggest that these microbial communities have significant potential for application in bioremediation strategies to mitigate the environmental impact of laundry wastewater.

Keywords: *Detergent Tolerance, Laundry wastewater, Bioremediation, Pseudomonas.*

Introduction:

The decomposition of organic materials in wastewater is one of the many environmental processes in which microorganisms—especially bacteria and fungi—are important. Laundry wash water, which includes soap residues, grime, oils, and organic materials, is one such wastewater source that is frequently disregarded. These residues can build up in laundry wash water over time, which can foster the growth of germs. Isolating and characterizing microorganisms from laundry wash water might provide important information about their possible use in bioremediation procedures, especially when it comes to breaking down detergent ingredients and other organic contaminants.

The isolation and characterization of microorganisms from laundry wash water can provide valuable insights into the adaptive mechanisms of these organisms, particularly in relation to their tolerance to detergent components (Gattlen *et al*., 2010). Laundry wash water offers a unique niche for microorganisms, where they not only survive but may also thrive due to the presence of organic matter, such as oils, grease, dirt, and detergents (Zambrano *et al*., 2019). Microbial communities in wash water may play an important role in biodegradation processes, helping to break down detergent residues and other organic contaminants. These microorganisms can exhibit resistance or tolerance to the surfactants and other harsh chemicals present in detergents, making them potential candidates for bioremediation applications (Bajpai *et al*., 2007). Screening for detergent-tolerant microorganisms is crucial for understanding how they function in this unique environment and for discovering potential industrial applications, such as the development of environmentally friendly cleaning agents or biotechnological processes that rely on detergent-degrading capabilities (Vickery *et al*., 2004).

Microorganisms that flourish in such conditions must possess the ability to withstand detergents. Surfactants, which make up most detergents, can be hazardous to the majority of microorganisms. Nonetheless, certain microbes have evolved to withstand or even break down detergent ingredients, which make them attractive options for biotechnological uses including bioremediation and wastewater treatment. Finding strains of microorganisms that can endure and proliferate in the presence of commercial laundry detergents is essential for their possible application in reducing detergent pollution in wastewater from washing. This process is known as "detergent tolerance screening."

There has been a surge in research on the identification and characterization of microorganisms from laundry wash water in recent years. A number of these studies have concentrated on discovering strains of microorganisms that are resistant to detergent and comprehending the mechanisms underlying their tolerance. Numerous bacterial species, including *Acinetobacter spp*, *Bacillus spp*, and *Pseudomonas spp* , have been shown to have the ability to degrade detergents (Wu *et al*., 2012; Sharma *et al*., 2017). Furthermore, because laundry wash water contributes to marine pollution, it is becoming increasingly important to reduce its environmental impact. These bacteria' capacity to break down surfactants and other detergent components is essential to this effort.

Intersecting with environmental microbiology, biotechnology, and environmental engineering, this field of study addresses ecological issues as well as the possibility of creating long-term laundry wastewater treatment solutions. A cleaner and more sustainable ecology might result from creative and economical wastewater management techniques made possible by an understanding of the variety of detergent-tolerant bacteria.

Methodology:

Collection of Laundry Wastewater Samples:

Twenty samples of laundry wastewater, representing a range of washing techniques, were collected from different residences. A standard laundry cycle was completed before the samples were collected. Samples were collected in sterile, screw-capped plastic containers to reduce contamination and maintain sample integrity. This ensured that no external microbial contamination occurred during the collection process. Relevant information was labeled on each sample, including the date and time of collection, the type of detergent used, and the source (home ID).

Transportation and Storage of Samples:

The samples were collected and then aseptically transported to the laboratory. The containers were stored in ice-packed containers to preserve the samples' original condition and stop microbial multiplication or deterioration while in transit. The samples were kept at 4°C until they arrived at the laboratory to reduce any microbial activity before processing could start.

Isolation and characterization of Microorganisms:

To ensure aseptic conditions, every laundry wastewater sample was processed in a biosafety cabinet in the laboratory. Each sample was serially diluted in sterile saline (0.85% NaCl solution). To facilitate the growth of different microorganisms, 100 µL of each dilution was plated on different selective agar media like-MacConkey agar for the isolation of Gram-negative bacteria and Sabouraud dextrose agar (SDA) for the isolation of fungi (e.g., molds and yeasts). For bacterial growth, plates were incubated for 24–48 hours at 37°C, and for fungal growth, they were incubated for 48–72 hours at 30°C. To obtain pure cultures, colonies with different morphology were sub- cultured onto basal media and the organisms were identified as per standard procedures.

Screening for Detergent Tolerance:

To assess the detergent tolerance of the isolated microorganisms, an antimicrobial assay was conducted using both powder and liquid detergents, representative of the types of detergents found in the laundry wastewater samples. The following procedure was followed for screening:

1. **Preparation of detergent solutions**: Nutrient agar and SDA plates were prepared with different concentrations (1%, 5% 10% & 15% v/v) of detergent (powder and liquid).

2. **Inoculation**: A loopful of each pure microbial culture was inoculated onto nutrient agar and SDA plates containing detergent solutions. The control plates had no detergent.
3. **Incubation and observation**: The plates were incubated at 37°C for 24-48 hours for bacteria and 25^{o} C for 3 to 5 days for fungal isolates. The microbial growth was observed and compared between the control and detergent-containing plates. The tolerance of the microorganisms was determined based on their ability to grow in the presence of detergent.

The detergent tolerance was classified into categories such as:

- **Highly tolerant**: Growth observed in all detergent concentrations.
- **Moderately tolerant**: Growth observed in lower detergent concentrations (e.g., 1% or 5%).
- **Non-tolerant**: No growth in detergent concentrations above 1%.

Results:

Identification and characterization of bacterial isolates from laundry wastewater:

A total of ten fungal isolates and fifty-two bacterial isolates from the twenty laundry wastewater samples were isolated from both liquid and powdered detergent samples. Different samples had different microbial diversity, and the detergent used and other environmental conditions had a noticeable impact on the overall number of isolates.

Table 1-Prevalence of bacterial isolates from laundry wastewater (n=52)

Bacterial Isolates	Percentage of Organisms
Bacillus Species	10 (19%)
Staphylococcus aureus	6 (11%)
Micro luteus	2 (4%)
Shigella Species	2 (4%)
Klebsiella Species	4 (8%)
E.coli	1 (2%)

Enterobacter Species	3 (5%)
Salmonella Species	1 (2%)
Vibrio Cholerae	1 (2%)
Proteus Species	2 (4%)
Providencia Species	1 (2%)
Aeromonas Hydrophilia	1 (2%)
Morganella Morgoni	1 (2%)
Pseudomonas Species	16 (31%)
Citrobacter Species	1 (2%)

A total of 52 bacterial isolates were identified from the laundry wash water samples. The isolates were classified into genera including Acinetobacter, Staphylococcus, Escherichia, Pseudomonas, Bacillus, and Enterobacter. Among these, Pseudomonas species were the most predominant, followed by Bacillus and Staphylococcus species. The higher prevalence of these bacteria in samples with elevated detergent concentrations suggests their ability to tolerate and thrive in detergent-rich environments, highlighting their potential adaptability to such conditions.

Identification and characterization of fungal isolates from laundry wastewater:

The colonies obtained were identified using LactoPhenol Cotton Blue Staining techniques. A total of ten fungal isolates were isolated and identified based on characteristics such as colony morphology, conidiation patterns, and the arrangement of phialides. The majority of the fungal isolates belonged to the genera Aspergillus and Penicillium, which are known to thrive in environments rich in organic matter, such as laundry wastewater. Fungal growth was particularly pronounced in samples containing liquid detergents, likely due to the organic nutrient content present in these detergents, which supports fungal proliferation. (Table 2)

Table 2. Prevalence of fungal isolates from laundry wastewater (n=10)

Fungal Isolates	**Percentage of Organisms**
Aspergillus Niger	3 (30%)

Aspergillus Flavus	2 (20%)
Aspergillus Fumigatus	1 (10%)
Penicillium	2 (20%)
Rhizopus	1 (10%)
Mucor	1 (10%)

Microbial Distribution:

The distribution of bacterial and fungal isolates varied depending on the type of detergent used. Samples containing liquid detergents exhibited greater microbial diversity, while those with powder detergents predominantly yielded bacterial isolates. Notably, some samples contained only a single type of microorganism, whereas others harboured mixed populations of both bacterial and fungal species.

Detergent Tolerance:

More than half of the 52 bacterial isolates exhibited varying degrees of resistance to the tested detergent concentrations. Notably, certain Pseudomonas spp and Bacillus spp demonstrated the ability to tolerate detergent concentrations as high as 10% and 15%, indicating their potential to thrive in detergent-rich environments. Similarly, fungal isolates, particularly Aspergillus species, showed tolerance to lower detergent concentrations of 1% and 5%.

Overall, the findings reveal that laundry wash water harbors a diverse range of microorganisms, many of which possess the ability to withstand and potentially degrade detergent components. These results highlight the potential of utilizing these microbes in bioremediation strategies to treat detergent-contaminated wastewater effectively.

Discussion:

This study revealed a significant diversity of microorganisms in laundry wastewater, with 20 samples yielding 52 bacterial isolates and 10 fungal isolates. The findings demonstrated that these microorganisms thrive in detergent-rich environments, with their diversity and detergent tolerance varying depending on whether liquid or powder detergents were used. The adaptability of these bacteria to survive in such conditions suggests their potential application in bioremediation, which could play a critical role in mitigating detergent contamination in wastewater.

Bacterial Diversity and Detergent Tolerance:

The study identified *Pseudomonas*, *Bacillus*, *Staphylococcus*, *Escherichia*, *Acinetobacter*, and *Enterobacter* as the most prevalent bacterial genera. These results are consistent with previous studies that have reported similar bacterial communities in detergent-contaminated environments (Rojas-Herrera et al., 2015). Among these, *Pseudomonas* species were the most dominant, followed by *Bacillus* and *Staphylococcus*. *Pseudomonas* is well-known for its ability to survive in harsh conditions, including exposure to toxic substances like detergents and other extreme environments (Ojewumi, *et al.,* 2018). Several *Pseudomonas* strains have been documented to possess strong detergent-degrading capabilities, attributed to their enzyme systems that break down surfactants and other detergent components.

Similarly, *Bacillus*, another common genus in such environments, is recognized for its resilience to high detergent concentrations. Certain *Bacillus* strains produce enzymes such as lipases and proteases, which aid in the degradation of detergent ingredients (Zhang *et al.,* 2014). The high tolerance of *Bacillus* and *Pseudomonas* strains, particularly at detergent concentrations of 10% and 15%, suggests their potential role in detoxifying laundry wastewater. These taxa have frequently been isolated from detergent-contaminated environments and industrial effluents (Hussain & Wahab, 2018), and their ability to form biofilms or regulate metabolic processes may further explain their resistance to detergent toxicity.

Gram-negative rods, particularly *Pseudomonas* species, demonstrated the highest resistance to detergents, tolerating concentrations up to 15%, followed by *E. coli* and other isolates. This contrasts with gram-positive cocci, which exhibited lower tolerance. The outer membrane of gram-negative bacteria likely acts as a protective barrier, preventing detergent penetration. Additionally, the presence of *Staphylococcus* species highlights the adaptability of microorganisms to detergent-rich environments. These bacteria are known for their ability to endure extreme conditions, such as high salinity and chemical exposure (Khan *et al.,* 2019). The diversity of bacterial genera identified in the samples underscores their potential for use in bioremediation, particularly for degrading detergent residues in wastewater.

Fungal Diversity and the Impact of Detergent Composition:

Although fungal isolates were less abundant than bacterial isolates, fungi such as *Aspergillus* and *Penicillium* were detected in the laundry wastewater samples. The presence of these fungi in detergent-rich environments is significant, as they are known for their ability to degrade organic materials, particularly in nutrient-rich conditions like those created by liquid detergents (Singh *et al.,* 2007). The increased fungal growth observed in these samples was likely driven by the organic load from liquid detergents, which often contain complex components such as perfumes, enzymes, and other chemicals. According to Sharma *et al.,* (2021), fungi excel at breaking down complex carbon sources, which explains their higher prevalence in environments with substantial organic nutrient loads.

The co-occurrence of fungi and bacteria in some samples suggests the possibility of synergistic interactions between these microbial groups. It is well-documented that certain bacteria and fungi can work together to degrade complex organic pollutants. Fungi may break down larger organic molecules, while bacteria can further process these into smaller compounds (Zhang *et al.,* 2014). Therefore, the coexistence of bacterial and fungal species in detergent-contaminated environments could enhance biodegradation efficiency, offering a promising approach for bioremediation.

Microbial Distribution and Detergent Type:

The observed differences in microbial distribution between liquid and powder detergent samples can be attributed to their distinct chemical compositions. Liquid detergents typically contain more complex chemical components and additives, which likely support a broader range of microbial life. In contrast, powder detergents, which generally have simpler formulations, primarily encourage bacterial growth. The higher microbial diversity in liquid detergent samples suggests that the complex organic and inorganic compounds in these detergents create a more favorable environment for a wider variety of microorganisms, particularly fungi. This aligns with previous studies indicating that liquid detergents are more conducive to fungal growth, while powder detergents tend to support bacterial populations (Sharma *et al.*, 2021).

These findings underscore the importance of detergent formulation in shaping the microbial communities present in laundry wastewater. Since different detergents provide varying types and quantities of nutrients, they likely influence which microbial species thrive in these environments. This insight is crucial for future wastewater treatment research, as understanding how different detergent types affect microbial populations can inform the development of effective bioremediation strategies.

Conclusion:

The study highlights the potential of using detergent-tolerant microorganisms, such as *Pseudomonas*, *Bacillus*, and fungal species like *Aspergillus*, in bioremediation approaches to treat detergent-contaminated wastewater. Their ability to withstand high detergent concentrations makes them promising candidates for integration into laundry wastewater treatment systems. Microbial-based bioremediation could offer a sustainable alternative to conventional chemical treatment methods, particularly in reducing detergent pollution in aquatic environments where traditional approaches may be less effective or environmentally friendly.

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