Microbial Populations Occurrence in the Domestic Wastewater and Food Industry Effluents

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ABSTRACT

Background and Objective: The development of sustainable pollution control necessarily involves the microbial community of the polluted environment. The current investigation attempts to isolate a population of bacterial colonies, identification of microbes from domestic greywater and wet grinding and pickle industries' effluents. **Materials and Methods:** Samples of domestic greywater (GW) and effluents from the wet grinding industry (WGI) and pickle industry (PI) for the isolation and identification of bacterial and fungal colonies. Standard procedures were performed, including Gram staining and biochemical tests. Analysis of the ANOVA revealed significant variation ($p \leq 0.05$) in the Colony Forming Unit (CFU) of wastewater samples. **Results:** Abundant microbial load of microbial community was found in the raw pickle industry effluent samples, followed by wet grinding industry effluent samples. Whereas, grey water was determined with a relatively low microbial population. However, fecal contamination indicator of *Coliform* bacteria was noticed in greywater alone. Yeast colonies were noticed in untreated wet grinding industry effluent. An organic acid-rich pickle effluent was detected with *Citrobacter* species. **Conclusion:** Microbial communities inventory in wastewater and food industry effluents revealed the biological quality of wastewater and further would provide the method to develop proper management principles and guidelines, for the improved water quality.

KEYWORDS

Microbial community, colony forming unit, wastewaters, biochemical tests, opportunistic pathogens, biological water quality

INTRODUCTION

Microbes a diverse group of microbes including bacteria, viruses, protozoa and fungi[1] are generally found ubiquitous including the contamination sites and possess unique degradation properties, thereby having a crucial role in the occurrence of biogeochemical cycles[2]. Most of the microbes utilize the organic substances of the substratum and a few specific microbes utilize the chemical substances of the substratum upon which they occur. Harmful microbes dwelling in the contaminated site emit unwanted noxious and toxic substances and gases, create a foul smell[3] and eventually has deteriorating the

environment and human health[4,5]. The source of water pollution, such as wastewater from domestic and industries is discharged into the environment without proper treatment, creating waterborne pathogens and diseases. Such organisms cause a serious global water quality problem[6]. The pathogenic microbe's growth rate is based on nutrient contents found in wastewater[7], competition among the inhabiting microbes to their coexisted microbes[8].

Around the world, more than 2.1 billion people lack access to safe water and their effect creates nearly 88% of diarrhea[9,10], also this condition causes several types of body ailments in human beings[11]. In this context, some of the physical, chemical, biological and disinfection methods are adopted for the treatment of domestic and industrial effluents[12]. The treated water qualities are monitored through physicochemical and biological water quality parameters and these parameters are regularly monitored to ensure that the treated water meets the standards for sustainable use[13].

Report on microbial community enumeration from various kinds of wastewater includes grey water[14,15], poultry industry[16], dairy effluent[17,18], pharmaceutical industry[19], coffee processing industry[20], caper processing industry[21], pepper processing industry[22] and hydrocarbon contaminated soil[23]. Advanced technologies of enzyme-based or electronic devices using flow cytometry are employed to determine the microbial quality of the water[24,25]. Therefore, understanding the microbial community in polluted sites is crucial for developing effective strategies for remediation and ensuring the safety of the ecosystem. The objective of this current work was to assess the microbial quality of greywater and food industrial effluents, viz., wet grinding and pickle industrial effluents using a Colony Forming Unit (CFU), isolation and identification of bacteria and fungi.

MATERIALS AND METHODS

Study area: The wastewater samples were collected from Thiagarajar College Campus Sewage Treatment Plant STP and the wet grinding food industry and pickle industry, all three units from Madurai, Tamil Nadu, India. Raw samples were collected and the source and the sampling period are detailed below.

Greywater: Wastewater released mainly from the student hostels, kitchen, mess and canteen (Latitude 9.913622° and Longitude 78.147989°), samples collected in the months of July, 2015, November, 2015 and March, 2016.

Wet grinding industry effluents: Manufacturing of batter for the popular south Indian food items of *idili, dosai* and *vadai* at large scale (Latitude 9°55'17.3964" and Longitude 78°8'49.8444"), samples collected in the equal monthly intervals between April, 2017 and March, 2018.

Pickle industry effluents: Green pickle production using green vegetables, at a large scale (Latitude 10.0474° N and Longitude 78.0904° E), samples collected in equal monthly intervals between June, 2019 and February, 2020 and between March, 2021 and May, 2021

Sample collection: Raw samples were collected during every sampling period in clean plastic bottles according to the standard procedure[26], from the sources of collection points. The wastewater samples were stored at 4°C in the laboratory, for further analysis.

Water quality parameters: Physicochemical water quality of untreated wastewater was discussed in the previous publication[27,28].

Isolation of bacteria: The bacterial organisms used in this study from the effluent samples were isolated by spread plate technique on the agar media after serial dilution. One milliliter of sample was mixed with 9 mL of sterile distilled water, followed by serial dilution with sterilized distilled water in the range of 10^{-3}-10^{-6}. The diluted samples of 0.1 mL were spread with nutrient agar. The plates were incubated and monitored for growth[29]. Morphologically identical colonies were segregated and the streak plate technique was used for the establishment of pure culture establishment and following the repetition several times, pure culture plates were obtained.

Bacterial identification: Bacterial culture was streaked over nutrient agar plates and incubated at 37°C for 12 hrs in order to obtain individual colonies. Bacterial culture was obtained by inoculating the mother culture onto the freshly prepared nutrient broth. The inoculum was further used for Gram's staining, a biochemical test for the identification of bacterial colonies[29], using selective media.

Fungal isolation: Isolation of fungi was performed by serial dilution and spread plate method. One milliliter of effluent samples was serially diluted into 10^{-6} to 10^{-8} and then smeared over the rose bengal agar (RBA) medium The fungal isolates were subsequently sub-cultured on RBA plates and the pure culture was obtained using the standard procedure[29].

Identification of fungal colonies
Colony characterization: The colony morphology was determined by macroscopical observation of colour, shape, size and type of colonies, observed using a high-resolution magnifying lens. Further, stained using lactophenol cotton blue, the isolated microbial colonies were observed under the microscopic field to study the hyphae morphology[30].

Data analysis: The number of colonies that emerged in the serial dilution plates were counted and calculated Colonies Forming Units (CFU) by using the following formula:

$$CFU/mL = \frac{\text{No. of colonies} \times \text{Total dilution factor}}{\text{Volume of the cultured plate (mL)}}$$

Statistical analysis: One-way ANOVA method was computed using SPSS software (version 16.0), to compare the log-transformed CFU values ($p \leq 0.05$) obtained during the sampling periods in the experiment.

RESULTS
Estimation of bacterial colonies: The principal pathway for disease that causes microorganisms to enter the human body is through water, considered to be a hotspot for microbial growth and reproduction. The untreated pickle industry (PI) effluents were enumerated with a significantly higher population of bacterial colonies, as compared to other effluent samples, during the study period (Fig. 1). Greywater (GW) samples were found with a very less number of bacterial colonies and wet grinding industry (WGI) effluent was estimated to moderate number of bacterial colonies. Water samples taken in the months of July, August and September were found to have a large population when compared with the rest of the month of all three untreated water samples (Fig. 1). Comparably less significant bacterial populations were found in February and March month sampling (Fig. 1).

Isolation of bacterial colonies: Two gram-positive bacilli, two gram-negative cocci and three gram-negative bacilli isolates were found in untreated GW (Table 1). Three groups of Gram-negative bacilli, five Gram-positive cocci and three Gram-positive bacilli bacteria were isolated in untreated WGI (Table 2). Two Gram-positive bacilli, nine Gram-negative bacilli and two Gram-positive cocci were found in the untreated PI effluents (Table 3). A total of 7, 11 and 13 bacterial colonies were detected, respectively from the GW, WGI and PI effluent samples.
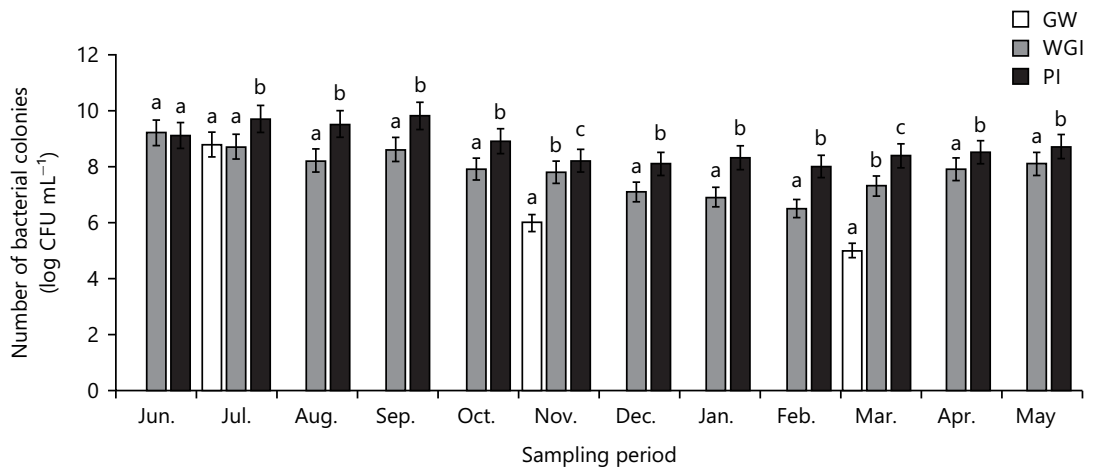
Fig. 1: Bacterial colony counted from the untreated wastewater from grey water (GW-July'15, Nov'15, Mar'16), wet grinding industry (WGI-Apr'17 to Mar'18) effluents and pickle industry (PI-Jun'19 to Feb'20, Mar'21 to May'21) effluent during the experimental period

Different alphabets in the bar represented the statistically significant ($p \leq 0.05$, n = 3)

Biochemical test: Totally fourteen biochemical tests were performed for the identification of bacterial genus and their results were represented in Table 1-3, respectively for the wastewater samples of GW, WGI and PI. Based on the results, the untreated GW isolate had identified as two *Bacillus* sp., *Pseudomonas* sp., *Micrococcus* sp., *Staphylococcus* sp., *Flavobacterium* sp. and *Escherichia coli* (Table 1). Among them, *E. coli* produces a green metallic sheen in EMB agar medium. Two *Pseudomonas* sp., two *Staphylococcus* sp., two *Bacillus* sp., *Enterobacter* sp., *Streptococcus* sp., *Micrococcus* sp., *Pediococcus* sp. and *Lactobacillus* sp., were identified from the untreated WGI effluent samples (Table 2). *Flavobacterium* sp., three *Pseudomonas* sp., *Xanthomonas* sp., *Enterobacter* sp., *Citrobacter* sp., *Klebsiella* sp., *Staphylococcus* sp., *Micrococcus* sp., *Citrobacter* sp., *Enterobacter* sp. and *Serratia* sp., were detected from the effluent of PI (Table 3).

Isolation of fungal colonies: Morphological characteristic features of isolates from untreated GW, WGI and PI effluent samples also their Key identification feature using the microscopic field have been shown in Table 4-6. A total of nine fungal colonies were isolated from the untreated GW samples (Table 4) and that includes three morphologically different *Aspergillus* sp., *Chrysosporium* sp., *Rhizopus* sp., *Fusarium* sp., *Mucor* sp., *Geotrichum* sp. and *Alternaria* sp. In untreated WGI effluent samples had *Mucor* sp., *Saccharomyces* sp. and 2 morphologically different *Aspergillus* sp. and a total 4 fungal colonies were frequently isolated during the study period (Table 5). A total of nine fungal colonies occurred in the untreated PI samples (Table 5) including *Mucor* sp., six morphological different *Aspergillus* sp. and 2 morphologically different *Penicillium* sp. (Table 6).

DISCUSSION

Pathogenic microbes play a vital role to deteriorate the biological quality of the effluents. Microbial isolates from the GW, WGI and PI effluent samples were detected with distinct microbial colonies based on their nature of effluent composition and also found with different CFU values. This phenomenon is due to the nature of effluents from the different food industries, clearly indicating the organic substances and contaminants present in the effluents, supporting the findings of the previous report[31]. Likewise, the existence of microorganisms is strongly dependent on the ambient temperature and oxygen level and nutrients.

Table 1: Biochemical test confirming the presence/absence of bacterial species occurred in the greywater samples, collected from the STP, Thiagarajar College, Madurai

Gram +/- & Shape	Gas formation (lactose broth)	Motility	Catalyst	Oxidize	Amylase activity	Protease activity	Indole test	Methyl red test	VP test	Citrate utilization	Triple sugar ion test				Oxidation fermentation (paraffin)		Urease production	H$_2$S production	Bacteria
											Slant	Butt	H$_2$S production	Gas production	Open tube	Closed tube			
+Rod	-	+	+	-	+	+	-	+	-	-	K	A	-	-	-	-	-	-	*Bacillus sp.*
+Rod	-	+	+	-	-	+	-	-	-	-	K	A	-	-	-	-	-	-	*Bacillus sp.*
+Cocci	-	-	+	-	-	-	-	+	-	-	K	K	-	+	-	-	-	-	*Micrococcus sp.*
-Rod	-	-	+	+	-	-	-	-	-	-	K	A	-	-	+	-	-	-	*Flavobacterium sp.*
+Cocci	-	-	+	+	+	-	-	+	-	+	K	A	-	-	-	-	-	-	*Staphylococcus sp.*
-Rod	-	+	+	+	+	+	+	-	-	-	K	K	-	+	+	-	-	-	*Pseudomonas sp.*
-Rod	+	+	+	-	-	-	+	+	-	-	K	A	-	+	-	+	-	-	*Escherichia coli*

K: Alkaline reaction and A: Acid reaction

Table 2: Biochemical test confirming the presence/absence of bacterial species occurred in the effluent samples, collected from the wet grinding industry

Gram +/- & Shape	Gas formation (lactose broth)	Motility	Catalyst	Oxidize	Amylase activity	Protease activity	Indole test	Methyl red test	VP test	Citrate utilization	Triple sugar ion test				Oxidation fermentation (paraffin)		Urease production	H$_2$S production	Bacteria
											Slant	Butt	H$_2$S production	Gas production	Open tube	Closed tube			
+Rod	-	+	+	-	+	+	-	+	-	-	K	A	-	-	-	-	-	-	*Bacillus sp.*
+Cocci	-	-	+	+	-	-	-	+	-	-	K	K	-	+	-	-	-	-	*Micrococcus sp.*
+Cocci	-	-	+	-	+	-	-	+	-	+	K	A	-	-	-	-	-	-	*Staphyllococcus sp.*
+Cocci	-	-	+	+	+	+	-	+	-	+	A	K	-	+	-	+	-	-	*Pediococcus sp.*
-Rod	-	+	+	+	+	+	+	-	-	-	K	K	-	+	+	-	-	-	*Pseudomonas sp.*
+Rod	-	-	-	-	+	+	-	+	+	+	K	A	-	-	-	-	-	-	*Bacillus sp.*
+Cocci	-	-	+	-	+	+	-	-	-	+	K	A	-	-	-	-	-	-	*Staphyllococcus sp.*
-Rod	-	+	+	-	-	+	-	-	+	-	K	K	-	-	-	-	-	-	*Enterobacter sp.*
+Rod	-	-	+	-	-	-	-	-	-	-	A	A	-	-	-	-	-	-	*Lactobacillus sp.*
+Cocci	-	-	+	-	-	-	-	+	-	+	K	A	+	-	+	+	+	-	*Streptococcus sp.*
-Rod	-	+	+	+	+	+	+	-	-	-	K	K	+	-	+	-	+	-	*Pseudomonas sp.*

K: Alkaline reaction and A: Acid reaction

Table 3: Biochemical test confirming the presence/absence of bacterial species occurred in the effluent samples, collected from the pickle industry

Gram +/-& shape	Gas formation (lactose broth)	Motility	Catalyst	Oxidize	Amylase activity	Protease activity	Indole test	Methyl red test	VP test	Citrate utilization	Triple sugar ion test				Oxidation fermentation (paraffin)		Urease production	H_2S production	Bacteria
											Slant	Butt	H_2S production	Gas production	Open tube	Closed tube			
+Cocci	-	-	+	-	-	-	-	+	+	+	K	A	-	-	-	+	-	-	Staphylococcus sp.
-Rod	-	-	+	+	-	-	-	+	-	-	K	A	-	-	+	-	-	-	Flavobacterium sp.
-Rod	-	+	+	+	+	+	+	-	-	-	K	A	-	+	+	-	-	-	Pseudomonas sp.
-Rod	-	+	+	+	-	-	+	-	-	-	K	A	-	+	+	-	-	-	Pseudomonas sp.
+Cocci	-	-	+	-	+	+	-	-	+	-	K	A	-	-	-	-	-	-	Micrococcus sp.
-Rod	+	-	+	-	+	-	-	+	+	-	A	A	-	-	+	-	+	-	Klebsiella sp.
-Rod	+	+	+	-	-	+	-	+	-	-	A	A	-	-	+	-	-	-	Enterobacter sp.
-Rod	-	+	+	-	+	-	-	-	-	-	K	A	-	-	-	-	+	-	Xanthomonas sp.
-Rod	-	+	+	+	-	+	-	-	-	-	A	A	-	-	-	+	-	-	Pseudomonas sp.
+Rod	-	-	+	+	+	+	-	+	-	-	K	A	-	-	+	-	-	-	Bacillus sp.
-Rod	-	+	+	-	-	+	-	-	+	-	K	A	-	+	+	-	+	-	Serratia sp.
+Rod	-	+	+	-	+	-	-	-	-	-	K	A	-	-	-	-	-	-	Bacillus sp.
-Rod	+	-	+	-	-	-	-	+	+	+	A	A	-	+	-	-	-	-	Citrobacter sp.

K: Alkaline reaction and A: Acid reaction

Table 4: Morphological characteristics of fungal isolates from greywater

Genera	Colony morphology	Key identification feature using the microscopic field
Aspergillus sp.	Colonies consisted with white basal mycelia, covered by black conidial heads at later stages	Radiate conidial heads are broad, globose, dark brown and splitting into many loose columns at later stages. Smooth-walled conidiophore stipes, septate, brown to black coloured vesicle. Biseriate, globose and metulae found with rough walls phialides
Aspergillus sp.	Colonies was granular, flat, often with radial grooves. Initial it looks yellow colour but quickly it changed the colour bright to dark yellow to green	Conidial heads were typically radiate and splitting to form columns in later stage. Condium are globose to subglobose, pale green colour, biseriate, sometime uniseriate, Conidiophore stipes was hyaline and coarsely roughened and it was frequently noticeable near the vesicle
Aspergillus sp.	Colonies appeared as sandy brown with a yellow to deep dirty brown	Conidia are globose to ellipsoidal, small, Conidia with biseriate head have a metulae and phialides
Chrysosporium sp.	Fungal colonies appeared a granular, cottony and raises folded in appearance. Colonies was white cream with yellow colour	Hyphae septate, Conidia are hyaline, broad-based, one-celled and smooth wall. These conidia were broader than the vegetative hyphae and formed terminally on pedicels, along the sides of the hyphae and intercalary positions. The conidia usually had an annular frill which was ruminants of the hyphal wall that remains after detachment from the hypha
Rhizopus sp.	Colonies was developed quickly and covered an agar surface with a thick cottony. It was white at initial and turn to grey or yellowish brown in later stage due to sporulation	Stolon, pigmented rhizoids, one celled sporangiospores, apophyses and columeneela were found collapsed, formed with umberella shaped structure
Fusarium sp.	Colonies appeared cottony white and it turns a pink colour in later stage	Hyphae was found with cross walls and were hyaline. Short conidiophores and uncomplicated. Sickle shaped macroconidia were largely found
Alternaria sp.	Colony was flat, downy to woolly and it was covered by grayish, short, aerial hyphae. The surface is greyish white colour to dark greenish black in later stage	Fungal hyphae had a septate and brown in colour. Conidiophores also septate and brown in colour, simple and large conidia, zigzag appearance, acropetal chains and may produce germ tubes. Ovoid to obclavate, darkly pigmented, muriform, smooth
Mucor sp.	Mycelia emerged like cotton fibres to fluffy. Initially white in colour and turns into greyish brown in later stage	Broad and non-septate Hyphae with thin wall sporangia were observed. Brown colour Sporangiophore forms into long, branched spherical structure
Geotrichum sp.	Colonies was emerged fast and flat hyphae with white to cream colour	Hyphae were hyaline, septate, branched and break up into chains of hyaline, smooth, arthroconidia one-celled, subglobose to cylindrical with double septum

Table 5: Morphological characteristics of fungal isolates from wet grinding industry effluent

Genera	Colony morphology	Key identification feature using the microscopic field
Aspergillus sp.	Coloniesconsisted with white basal mycelia, covered by black conidial heads at later stages	Radiate conidial heads are broad, globose, dark brown and splitting into many loose columns at later stages. Smooth-walled conidiophore stipes, septate, brown to black coloured vesicle. biseriate, globose and metulae found with rough walls phialides
Aspergillus sp.	Colonies appeared as sandy brown with a yellow to deep dirty brown	Conidia are globose to ellipsoidal, small, Conidia with biseriate head and seen with metulae and phialides
Mucor sp.	Mycelia emerged like cotton fibres to fluffy. Initially white in colour and turns into greyish brown in later stage	Broad and non-septate hyphae with thin wall sporangia were observed. Brown colour Sporangiophore forms into long, branched spherical structure
Saccharomyces sp.	Larger colonies with creamy nature. Colonies were larger than bacterial colonies	Small round shape colonies of smooth texture with glistening surface entirely elevated and with raised margin also smooth and texture

Table 6: Morphological characteristics of fungal isolates from pickle industry effluent

Genera	Colony morphology	Key microscopic field identification features
Aspergillus sp.	Colonies appeared as deep green with yellow colour in the center	Septate hyphae, radiate conidial heads with loosely columnar. Smooth conidiophores found with globose to subglobose, colorless to pale brown, uniseriate vesicles found with phialides cover on the upper surface
Mucor sp.	Mycelia emerged like cottony to fluffy. Initially white in colour, it turns into greyish brown in later stage	Sporangiophore long branched, spherical non-septate hyphae, brown colour
Aspergillus sp.	Colonies was granular, flat, often with radial grooves. Initial it looks yellow colour but quickly it changed the colour bright yellow to green	Radiate conidial heads found with splitting to loose columns in later stage. Coarsely roughened condium with globose to subglobose structure, pale green colour, biseriate or uniseriate and found with near the vesicle
Aspergillus sp.	Colonies was typically plain green with a dark red-brown tinge	Conidial heads are columnar, globular, small and biseriate, brownish colour, smooth-walled stipes changed a rugged wall in later stage
Aspergillus sp.	Colony appeared to be varyingly coloured from pale green, to greenish–beige	Radiate conidiophores are hyaline, septate, plae brown, smooth, brittle nature. Vesicles found with phialides and small, metulae
Penicillium sp.	Colonies showed flat, filamentous and velvety, woolly in texture with olive green at center and white at the periphery	Septate hyphae, conidia found with simple, round, unicellular, metulae found with flask-shaped phialides and branched
Aspergillus sp.	Colonies consisted with white basal mycelia, covered by black conidial heads at later stages	Radiate conidial heads are broad, globose, dark brown and splitting into many loose columns at later stages. Smooth-walled conidiophore stipes, septate, brown to black coloured vesicle. biseriate, globose and metulae found with rough walls phialides
Aspergillus sp.	Colonies appeared light green colour with fluffy hyphae	Hyphae septate and hyline. Conida found with simple globose, uniseriate and splitting to form loose columns
Penicillium sp.	Colonies was appeared blue-green in colour with a yellowish pigment. Pigment was appeared after several days that diffuse throughout the medium	Filamentous hyphae with conidia, colorless, slender, tubular, branched and septate hyphae. Conidia found with long, cottony or fluffy in texture

The severely pathogenic nature of Colic bacteria was identified in GW effluent. As this type of microbe is generally encountered in fecal contaminants and also causes a number of infectious illnesses[32,33]. *Pseudomonas* species were found as the common biological indicator of the contaminated water, In this experiment, *Pseudomonas*, a potential contaminant[34] was detected from the two food industrial effluents samples (Table 2). *Staphylococcus* species, a Gram-negative bacteria were frequently occurred in all three untreated wastewater samples and that isolate was acted as a food poisoning microbe and considered an opportunistic pathogen in human health problems[35]. *Streptococcus* species were found in WGI effluent. They are considered wastewater indicator organisms[36]. Flavo bacterium species were noticed in both GW and PI wastes and this pathogen was previously found in industrial wastewater[37].

The three different *Bacillus* sp., isolates were found in the WGI effluent sample due to the presence of high carbohydrate level present in the WGI effluents. Likewise, *Bacillus* species have a unique starch-degrading feature, denoted by Yezza *et al.*[38] and Shofiyah *et al.*[39] Some strains of *Pediococcus* sp., have been found in the untreated WGI effluent samples during the study period and this result is in accordance with the similar isolates found as food spoilage properties[40]. Pseudomonas, *Xanthomonas* sp. and *Citrobacter* species were found most frequently in untreated PI effluent samples, a similar group of microbial consortium used in the degradation of oil and organic acid-rich effluent[41]. *Aspergillus* sp., *Penicillium* sp., *Fusarium* sp. and yeast are often detected form polluted water environments[42] which deteriorate the biological water quality. Among the three untreated wastewater samples used in this experiment were enumerated with similar fungal colonies. Larger volumes of yeast colonies were noticed in the WGI effluent, as it contained higher starch content and yeast colonies fermenting properties, emanating foul odors.

Biological indicators have received increasing attention in several wastewater treatments and reuse procedures[43]. Hence, harmful microbes in the reused wastewater or improperly treated wastewater are likely to persist in the land and soil environment, which can enter into agricultural edible crops, thereby getting entered into the food chain. This deleterious effect poses a greater challenge due to domestic and industrial water pollution. The "*Field-to-Fork chain*" principle is essential to adapt the proper recycling and proper monitoring at each stage of water treatment, which will be contributing towards safeguarding consumers, thereby the outbreaks of food-borne illnesses could be prevented[44]. Therefore, essential means of monitoring wastewater from domestic and industries become inevitable to determine the biological quality, which would help to develop proper management principles and technologies, to achieve the acceptable water quality.

The complex microbial ecosystems found in the wastewater, are involved in the degradation of organic matter, nutrient removal and disease surveillance. Microbial community identification can be used to detect and track the presence of pathogens and indicator organisms in wastewater. However, only a small fraction of them can be cultured in the laboratory and that restricts the accuracy of microbial identification. Further, microbes consist of antibiotic-resistant genes and which determine the impact of wastewater discharges on the spread of resistance in the environment. This information is valuable for implementing measures to mitigate the spread of antibiotic resistance. It empowers operators and researchers to optimize treatment processes, enhance water quality and safeguard public health. This analytical result would be applicable in the devising of a suitable recycling treatment process in the purification of water, which will serve the precious natural resource of inland fresh water and also in a cleaner environment and sustainable living.

CONCLUSION

Untreated GW, WGI and PI effluents contained significant concentrations of harmful bacteria and their CFU values were investigated. This highlights the need for proper treatment of wastewater before discharge to prevent contamination and potential health risks. Additionally, *Escherichia coli* and *Staphylococcus* species were discovered in untreated grey water. Both species have to be eliminated through appropriate recycling. Pathogenic *Staphylococcus* sp., *Staphylococcus* sp. and *Enterobacter* sp., bacteria were present in untreated WGI effluent. Similar microorganisms, including as *Klebsiella* sp., *Serratia* sp. and *Xanthomonas* species, are also found in untreated PI effluent. Different *Aspergillus* sp. and *Penicillum* species were found in both food industry effluents. Opportunistic pathogen of *Geotrichum* sp., *Fusarium* sp., *Mucor* sp., *Alternaria* sp., and *Rhizopus* sp., were found in grey water. These microbes must be closely monitored before being released into an aquatic habitat on land. Failure to do this could have serious health consequences for humans and wildlife.

ACKNOWLEDGMENTS

SIGNIFICANCE STATEMENT

The microbial community plays a vital role in the biodegradation of pollutants and its composition and diversity can provide valuable information about the environmental conditions of polluted sites. Microbial community monitoring and management are essential to prevent the spread of harmful microorganisms and protect public health also their result is necessarily important to wastewater treatment for the removal of pathogenic microbes. Thereby, maintaining the biological quality of recycled wastewater has become the foremost requirement that fulfills the principle of the freshwater management system.

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