Impact of meteorological conditions on the water quality of wastewater treatment systems: a comparative study of phytoremediation and membrane bioreactor system

Khush Bakht Andleeb and Imran Hashmi

ABSTRACT

Two demonstration wastewater treatment systems, i.e. a phytoremediation system and a membrane bioreactor (MBR) system, were studied for a six-month period from August 2016 to January 2017. The phytoremediation system consists of wetland cells implanted with diverse phytoremediation macrophyte species at NUST H-12 sector Islamabad, Pakistan, while the MBR system comprises primary clarifiers, membrane tanks and bio tanks that treat domestic wastewater through hybridization of biological and biomechanical techniques. The phytoremediation system receives domestic wastewater at the rate of 283 m$^3$/d, and greater hydraulic efficiencies were achieved because of compartmentalization and higher aspect ratios, whereas the MBR system receives 50 m$^3$/d. The present study was conducted to analyze and compare the correlation between water quality parameters of wastewater treatment systems and meteorological conditions. Statistically significant correlation was exhibited between eight water quality parameters (pH, EC, turbidity, dissolved oxygen, total suspended solids (TSS), chemical oxygen demand (COD), biological oxygen demand (BOD) and total coliforms (TC)) and meteorological factors (ambient temperature and relative humidity). Predominant species isolated and identified through polymerase chain reaction and 16S rRNA sequencing from wastewater of the phytoremediation system and sludge of the MBR system belong to the phylum Proteobacteria with relatively higher abundance of Enterobacter, Shigella, Escherichia and Salmonella genera.

INTRODUCTION

The main objective of domestic wastewater treatment methods is to produce effluents with lowest organic contents that renders it harmless to the receiving agricultural land or water bodies. Water is referred to as polluted when it is being impaired by the release of anthropogenic contaminants. The resulting polluted water undergoes a marked shift in its ability to support its affiliated biotic communities and does not remain potable for human use (Sharma & Dubey 2011). The per capita annual water availability of Pakistan has decreased from 5,000 m$^3$ in 1951 to 1,038 m$^3$ in 2010, which is slightly higher than the internationally acceptable water scarcity level of 1,000. According to a report by the International Monetary Fund (IMF), Pakistan is now designated as the third most water-stressed country globally because its per capita yearly water availability is 1,017 m$^3$ (Haydar et al. 2015). In order to cope with the water crises, effective water management strategies should be adopted to reuse wastewater in a sustainable way.

Phytoremediation systems are natural/biological wastewater treatment arrangements that are becoming a significant substitute for conventional wastewater treatment systems (activated carbon absorption, activated sludge, trickling filters, anaerobic systems) because of their proficiency, reduced energy requirements, and fewer establishment and management requirements (Adrados et al. 2014; Luo et al. 2016; Victor et al. 2016; Zhou et al. 2017; Xu et al. 2017). However, membrane bioreactor (MBR) technology has emerged as an efficient technology with a smaller ecological footprint, lower sludge production and improved treated water quality. Various studies have

Impacts of meteorological factors on water quality parameters are documented in various studies, and significant impacts and correlations have been identified. Zhang and his coworkers explored the correlation among various physicochemical parameters (suspended solids (SS) and total dissolved solids (TDS)) with meteorological parameters in a reservoir in China. They concluded that the three meteorological parameters exhibited positive correlation with SS. Moreover, significant positive and negative correlations among water quality parameters (chemical oxygen demand (COD), biological oxygen demand (BOD), TDS, total nitrogen, total phosphorus and electrical conductivity (EC)) and meteorological factors were exhibited (Zhang et al. 2017). More recent reports deal with such aspects (Baig et al. 2017; Major & Ciesliński 2017).

The role of bacteriological communities in wastewater treatment systems is irrefutable. Various studies have reported that the diversity of microorganisms in the phytoremediation system is critical for its proper functioning in terms of pollutant degradation and maintenance (Bekwe et al. 2005; Kadlec & Wallace 2008; Sleytr et al. 2009; Oopkaup et al. 2016). Concerning the MBR system, the profound role of bacterial communities, more specifically quorum quenching bacteria, in membrane fouling has been reported extensively. In addition to that, the role of microbes in nitrogen, phosphorus and organic compound removal was also recognized (Ma et al. 2015; Karray et al. 2016; Sato et al. 2016; Oh et al. 2017).

The main aim of the study was to analyze and compare the correlation between eight water quality parameters of the phytoremediation and MBR setup (pH, EC, turbidity, dissolved oxygen (DO), total suspended solids (TSS), COD, BOD and total coliforms (TC)) with meteorological factors (ambient temperature and relative humidity) and to compare the diversity of microbial communities in both treatment systems through polymerase chain reaction (PCR) and 16SrRNA sequencing.

**METHODOLOGY**

**Systems description**

**Phytoremediation setup**

The phytoremediation arrangement consists of vertical subsurface flow constructed wetlands located in NUST H-12 campus, Islamabad, Pakistan, receiving domestic wastewater from academic blocks, residential apartments, hostels and schools. The mean inflow of wastewater into the scheme is being maintained at 283 m$^3$/d for a population of about 6,000 inhabitants. Operation started in 2013 with funding from UNESCO and it is being maintained by PMO NUST.

The phytoremediation treatment system comprises pretreatment via a sedimentation tank (1,066.8 cm long, 365.76 cm wide and 183 cm deep) with a total capacity of 71 m$^3$. After the sedimentation tank, the setup consists of eight ponds (1,524 cm long, 670 cm wide and 213 cm deep) planted with *Typha latifolia*, *Pistia stratiotes* and *Centella asiatica*, having a total capacity of 218 m$^3$. The setup has the capacity for treating 283 m$^3$/d of water. The overall capital cost incurred on the treatment facility was 6.5 million PKR. The overall cost for operation and maintenance of the treatment facility is majorly related to the salaries of hired human resource, as in the four years of operation no major cost has been incurred in any other category of operation and maintenance. In the year 2018, the total amount spent on salaries of human resource was 0.465 million PKR. Reed beds are filled with substratum composed of fine and coarse gravel and are coated with geotextile membrane. Details of ponds are represented in Table 1.

**MBR setup**

The MBR setup at NUST was established with funding from NUST research and development fund and Samsung corporation, Korea, and came into operation in 2015. It has the

<table>
<thead>
<tr>
<th>Pond no.</th>
<th>Plant species</th>
<th>Length (cm)</th>
<th>Width (cm)</th>
<th>Depth (cm)</th>
<th>Total capacity (m$^3$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>P 1</td>
<td><em>Typha latifolia</em> (Typha)</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 2</td>
<td><em>Pistia stratiotes</em> (Water lettuce)</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 3</td>
<td><em>Centella asiatica</em> (Penny wort)</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 4</td>
<td><em>Centella asiatica</em> (Penny wort)</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 5</td>
<td>Blank</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 6</td>
<td>Blank</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 7</td>
<td><em>Pistia stratiotes</em> (Water lettuce)</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
<tr>
<td>P 8</td>
<td>Aeration pond</td>
<td>1,524</td>
<td>671</td>
<td>213</td>
<td>155</td>
</tr>
</tbody>
</table>

| Categorization of phytoremediation setup |
capacity to treat 50 m³/d of wastewater. The total capital cost of the NUST MBR plant was 40 million PKR and the maintenance cost is approximately 90 million PKR, which basically includes the salaries of staff and maintenance of equipment. The arrangement comprises a primary clarifier followed by five bio tanks and a membrane tank, having total capacities of 1,500, 12,000 and 2,350 m³ respectively.

**Sampling and analyses**

Water samples of influent and effluents of the phytoremediation and MBR setups were collected three times in a month from August 2016 to January 2017 from the inlet and outlets of the phytoremediation system and MBR system. Samples were collected in sterile bottles and were immediately transferred to a laboratory for further analysis. The following parameters were analyzed: temperature, pH, EC, DO, turbidity, TSS, TDS, COD, BOD and TC using standard laboratory procedures (APHA/AWWA/WEF 2012). pH was recorded by following the potentiometric method and using a HACH 156 pH meter. EC and TDS were determined by using an Ino Lab 720 conductivity meter and the measuring unit was μS/cm, while turbidity was measured using a HACH 2100P meter. BOD was measured using the five day BOD test by using the dilution method, while COD was determined through the closed reflux titration method. TC was determined by the membrane filtration technique and was carried out on EMB agar media (APHA/AWWA/WEF 2012).

**Acquisition of meteorological data**

Daily mean data of ambient temperature and relative humidity were acquired from August 2016 to January 2017. Effects of individual meteorological parameters on the water quality parameter of the setups were analyzed for six months.

**DNA extraction and PCR amplification**

Samples for bacterial isolation were collected from the surface and sediments of Ponds 2 and 7 of the phytoremediation setup, implanted with *Pistia stratiotes* (water lettuce) and the activated sludge of the MBR system. Bacterial strains were isolated using the method followed by Darveau and his coworkers in 1983 (Darveau & Hancock 1983). Genomic bacterial DNA was extracted using the Invitrogen Pure Link Genomic DNA Mini Kit following the manufacturer’s instructions (Cat no. K1820-01, USA).

**Table 2 | Selected primers for PCR amplification**

<table>
<thead>
<tr>
<th>Primers</th>
<th>Sequence (5’-3’)</th>
<th>Targeted genes</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>518F</td>
<td>CCAGCAGCCGCGGTATACG</td>
<td>16S rRNA</td>
<td>Waheed et al. (2013)</td>
</tr>
<tr>
<td>800R</td>
<td>TACCAGGTATCTAATCC</td>
<td>16S rRNA</td>
<td></td>
</tr>
</tbody>
</table>

PCR amplification of 16S rRNA gene of bacterial strains was performed using primers 518 F and 800 R (Table 2), and was carried out in a 50 μL reaction mixture containing 25 μL of Taq PCR master mix (Bio Basic, Canada) with forward and reverse primer having a total volume of 4 μL and 20 μL nuclease-free water. The volume of DNA template in the total reaction mixture was 1 μL. The PCR mixture was placed in a thermocycler (Extra Gene 9600) for amplification. It was run for 5 min at 95 °C for template denaturation followed by 40 cycles for template amplification, consisting of three steps: 95 °C for 1 min for DNA denaturation into single strand, 61 °C for 1 min for the primers to anneal to their complementary sequences on either side of the target sequence, 72 °C for 1 min for extension of the complementary DNA strand from each primer and final elongation at 72 °C for 10 min.

**16S rRNA sequencing**

PCR products were kept in an ice box and preserved isolates were sent to the Genome analysis department, Macrogen, Seoul, South Korea, for 16S rRNA sequencing. Obtained sequences were trimmed through Bioedit software and were analyzed through the BLAST tool of the National Center of Biotechnological Information (NCBI). After proper detection of the obtained species, accession numbers were obtained from the NCBI GenBank library. FASTA sequences were run in MEGA 7 software to get the phylogenetic tree, which showed linkages between the isolated strains and those at NCBI GenBank.

**Statistical analysis**

Results were analyzed by using graphical tools in Microsoft Excel 2016. Correlation was applied by using the Microsoft Excel data analysis tool to test the significant and non-significant effects of meteorological parameters on physicochemical and biological water parameters.
RESULTS AND DISCUSSION

Monthly variations in water quality parameters with ambient temperature (°C)

Temperature has a notable impact on the pollutant removal efficiencies as well as total pollutant loads of both treatment techniques, as biodegradation, biomass and bacterial community structure is highly dependent on temperature oscillations (Meng et al. 2014; Faulwetter et al. 2009).

pH varied from 7.07 to 7.77 throughout the experimental period. The highest pH was noted in December, i.e. 7.77, while the lowest was observed in September, i.e. 7.07, as depicted in Figures 1 and 2(a). The pH at the outlet of the MBR system was observed to be higher than the inlet and outlet of the phytoremediation system, as in the MBR system an alkaline environment was maintained to buffer the hydrogen ion concentrations created in the nitrification process, which in turn leads to an increase in the pH of the final effluents (Iorhemen et al. 2016).

Negative correlation was observed between ambient temperature and pH of the inlet (r = −0.63) and outlets (r = −0.56) for the phytoremediation setup and the outlet of the MBR system (r = −0.77) as depicted in Figure 1(a).

The pH of treated water was found to be within the permissible limits set by the EPA (6-10) to be acceptable for agricultural purposes.

Non-significant variation in EC was observed with months for both treatment units and its concentration in the treated water was found to be within the permissible limit of 3,500 mg/L set by the EPA to be fit for agricultural purposes. The concentration of DO in the influent sample was very low (up to 0.08 mg/L in August), which is because of the higher microbial content, which consumes oxygen for degradation. An important improvement in the water quality of the outlets was significant. The oxygen content in the water sample of the outlet of the MBR was higher than the rest of the samples. A DO value of 5.21 mg/L was observed at temperatures as low as 18.25 °C in the MBR system. At higher temperatures, reduction in DO levels up to 0.08 mg/L was noted, which indicates that warm water did not endure DO at higher concentrations. Significant negative correlation (r = −0.93) of EC with temperature was noted in the case of the phytoremediation setup while positive correlation (r = 0.25) between EC and ambient temperature was observed for the MBR configuration (Figure 1(a)). A negative correlation was exhibited between DO and the ambient temperature of the inlet and MBR outlet (r = −0.93) and a positive correlation with the phytoremediation outlet (r = 0.45) was identified, as depicted in Figure 1(d). A study was conducted by Akratos et al. (2009) to study the effects of temperature on the DO of the system and have reported higher DO values in winters, when solubility of oxygen in water was higher, and lower values were stated in summers because of its least solubility (Akratos et al. 2009).

Ambient temperature exhibited significant negative correlation with turbidity of the inlets (r = −0.95) as observed in Figure 1(c). However, significant positive correlation (r = −0.98) was noted between TSS and the ambient temperature of the inlets (Figure 1(e)). Not much significant variation of turbidity and TSS values of the outlets of both systems with ambient temperature was identified. Increase in ambient temperature resulted in increase in temperature of the water, which enhanced the self-diffusion coefficient of the water. Therefore, higher concentration of TSS at elevated temperature may be attributed to this diffusion coefficient. This is in line with the study carried out by Ahsan and his coworkers in 2005 (Ahsan et al. 2005).

Negative correlation existed between ambient temperature and COD, BOD concentrations of the influent sample (r = −0.96 for COD and r = −0.63 for BOD) and effluents of the phytoremediation system, i.e. r = −0.53. However, no correlation was exhibited between ambient temperature and concentrations of BOD in the MBR effluent (Figure 1(f) and 1(g)). Studies have reported that the COD concentration in the system is because of organic matter and the concentration of this organic matter is highly dependent on the microbial activities of the aerobic and anaerobic bacteria (Vymazal 2009).

Increase in ambient temperature was positively associated with increase in the total coliform concentration of the inlet (r = 0.60). However, decrease in temperature had a positive impact on the total coliform concentration of the phytoremediation system with the exception in winters where the highest coliform concentration was noted. Coliform concentrations at the outlet of the MBR system remain consistent and no significant impact of temperature on TC concentration at the MBR outlet was observed (Figure 1(h)).

Monthly variations in water quality parameters with relative humidity (%)

Water vapor content is the total percentage of the saturated vapor pressure of water at a given temperature. Various factors influence the water vapor content of the atmosphere, and these are air temperature, wind direction and also nearby water bodies, which have an influential impact on water vapor content. With variations in the relative humidity (RH), friction velocities in the water content will be altered and lower humidity decreases particle cohesion.
hence the total amount of pollutant holding capacity of the water will be altered (Csavina et al. 2014).

Significant variations in the pH of both setups were observed with fluctuations in the relative humidity. Highest pH was observed in December for both setups, when the relative humidity was least, while the lowest pH was noted in September when the RH was highest. Significant negative correlation was observed between the RH (%) and pH.
and pH of the outlets of both setups ($r = -0.94$ for PS and $r = -0.87$ for MBR). This whole trend is depicted in Figure 2(a).

Positive correlation was observed between the EC of the inlet and relative humidity ($r = -0.8$). While negative correlation was noted between the EC of the outlets of both
systems and relative humidity ($r = -0.69$ for the phytoremediation system and $r = -0.33$ for the MBR) as depicted in Figure 2(b). Concerning DO, a positive correlation was observed between relative humidity and the inlet and outlet of the phytoremediation system. However, the DO of the outlet of the MBR system exhibited a slight negative correlation with relative humidity ($r = -0.49$), as shown in Figure 2(c). The maximum value of DO in winter and the increase in the concentration with relative humidity can be attributed to the prevailing wind conditions, which permitted the increase in the solubility of oxygen. This is in line with the previous studies (Fishar 1999; Ali 2008).

A significant increasing trend was observed between the TSS of the inlet and relative humidity ($r = 0.64$), while relative humidity had not shown any significant impact on the TSS concentrations of the outlets of both systems, as depicted in Figure 2(e). A significant declining trend was noted between the turbidity of the inlet and relative humidity ($r = -0.71$), while non-significant correlation was noted between the turbidity of the outlets of both systems and relative humidity, as depicted in Figure 2(d).

With the increase in relative humidity, a decrease in COD and BOD concentrations of the inlets was observed ($r = -0.53$ and $r = -0.92$); however, concerning the outlets of both setups, a non-significant correlation existed between relative humidity and COD and BOD concentrations (Figure 2(f) and 2(g)). Further, the reported work of the previous studies conducted by Sankararajan and his coworkers in 2017 indicated a decrease in BOD concentration up to 5% with an increase in relative humidity through genetic programming (Sankararajan et al. 2017).

Strong positive correlation was exhibited between total coliform concentration of the inlet and outlets of the phytoremediation setup and relative humidity ($r = 0.7$) while concentration of TC at the outlet of the MBR setup was not influenced by relative humidity (Figure 2(h)).

**PCR amplification and 16S rRNA sequencing of isolated bacterial strains**

The DNA of the isolated strains were extracted through a kit method and were confirmed by running it on agarose gel. Extracted DNA of the isolated strains was further subjected to the PCR amplification process for genus identification.

518 Forward and 800 Reverse primers were used to amplify the 500 bp fragment of 16S rRNA genes of isolated bacterial strains. PCR amplification products of 500 bp were obtained for all the isolates. Amplified genes of strains KB1-KB19 were visualized by 1% agarose gel, stained with loading dye, and observed under UV transilluminator.

PCR products were sent to the genome analysis department, Macrogen. Sequences that were obtained were trimmed through Bio edit software and were identified through the BLAST tool of NCBI. After getting the accession number, a phylogenetic tree (Figure 3) was constructed, which demonstrated the relatedness and linkages of different bacterial strains. The assessment of bacterial communities in the phytoremediation setup has been addressed by several researchers (Baptista et al. 2003; Nicomrat et al. 2006) that have categorized the microbial communities in constructed wetlands (CW) for domestic wastewater and mentioned that these systems are reliant on microbial compositions for optimum wastewater treatment. Dominant bacterial species isolated from the phytoremediation setup belong to the phylum **Proteobacteria** and the species identified were *Enterobacter cloacae*, *Enterobacter kobei*, *Enterobacter hormaechei*, *Enterobacter asburiae*, *Enterobacter aerogenes*, *Gamma proteobacterium*, *Francobacterium pulversis*, *Citrobacter freundii*, *Shigella dysenteriae*, *Escherichia albertii* and *Escherichia coli* (Table 3). This is perfectly in line with the study conducted by Calheiros and his coworkers in 2009, which involved the identification of bacterial communities from wetlands, and the results revealed *γ-Proteobacteria* being the most dominant phylum responsible for removal of phenols and organic compounds from wastewater (Calheiros et al. 2009). Previous studies have reported that aerobic autotrophic ammonia oxidizing bacteria, denitrifying bacteria and methanogens belong to the phylum **Proteobacteria**, and have an impressive role in pollutant removal from wetlands (Ibekwe et al. 2003; Gorra et al. 2007; Tietz et al. 2007). Calheiros and his colleagues in 2010 have worked on the bacterial community dynamics of horizontal subsurface flow constructed wetlands (HSFCW) and have identified **Firmicutes**, **Actinobacteria**, **α**, **β**, and **γ-Proteobacteria** as being the most dominant ones (Calheiros et al. 2010).

All bacterial species isolated from activated sludge of the MBR setup belong to the phylum **Proteobacteria**. This is in accordance with the study conducted by Sato and his coworkers in 2016 that identified **α**, **β**, and **γ-Proteobacteria** as the most dominant species in the pilot scale MBR technique during operation with a relevant abundance of 37%, and identified some species as organic substance consumers. The activated sludge of the MBR system provides higher nutritious carbon and other features necessary for the development of a wide variety of microorganisms; therefore, it has been recognized as an ideal source for isolation of precise bacteria that are capable of enzyme degradation or production potentials.
Some previous studies have reported Proteobacteria to be present in bulk in activated sludge, and associated with the secretion of EPS that is mainly responsible for formation of the bio cake layer on the membrane surface (Hu et al. 2012; Zhang et al. 2012; Hu et al. 2015).

Conclusions

Phytoremediation and MBR methods are efficient and reliable approaches for the removal of pollutants from domestic wastewater and its reuse for irrigation purposes.

The main conclusions are as follows:

1. Higher efficiency of the MBR technique than phytoremediation. Climatic parameters have strong positive and negative correlation with water quality parameters. pH, turbidity, EC, COD, BOD and TSS of the phytoremediation system were negatively correlated with ambient temperature while DO and TC were positively correlated. BOD was non-significantly correlated with meteorological factors for the MBR setup. DO, COD, turbidity and TC were positively correlated with relative humidity for the MBR technique. However, pH, DO, EC and turbidity were negatively correlated with relative humidity for the MBR technique.

2. The predominant species identified from the wastewater of the phytoremediation configuration belong to the phylum Proteobacteria (Enterobacter cloacae, Enterobacter kobei, Enterobacter hormaechei, Enterobacter...
Table 3 | Source, scientific name along with the accession number of the isolated and identified species from the phytoremediation system (PS) and MBR sludge

<table>
<thead>
<tr>
<th>Strain ID</th>
<th>Source</th>
<th>Species identified</th>
<th>Accession number</th>
</tr>
</thead>
<tbody>
<tr>
<td>K.B 1 PS</td>
<td>PS</td>
<td>Enterobacter cloacae</td>
<td>KY751345</td>
</tr>
<tr>
<td>K.B 2 MBR sludge</td>
<td>Pantoea dispersa</td>
<td>KY751346</td>
<td></td>
</tr>
<tr>
<td>K.B 3 PS</td>
<td>PS</td>
<td>Salmonella enterica</td>
<td>KY751347</td>
</tr>
<tr>
<td>K.B 4 PS</td>
<td>Enterobacter hormaechei</td>
<td>KY751348</td>
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</tr>
<tr>
<td>K.B 5 PS</td>
<td>Escherichia coli</td>
<td>KY751349</td>
<td></td>
</tr>
<tr>
<td>K.B 6 PS</td>
<td>Escherichia coli</td>
<td>KY751350</td>
<td></td>
</tr>
<tr>
<td>K.B 7 MBR sludge</td>
<td>Shigella dysenteriae</td>
<td>KY751351</td>
<td></td>
</tr>
<tr>
<td>K.B 8 PS</td>
<td>Escherichia coli</td>
<td>KY751352</td>
<td></td>
</tr>
<tr>
<td>K.B 9 MBR sludge</td>
<td>Enterobacter hormaechei</td>
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<tr>
<td>K.B 10 PS</td>
<td>Franconibacter pulsarius</td>
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<tr>
<td>K.B 11 PS</td>
<td>gamma proteobacterium</td>
<td>KY751355</td>
<td></td>
</tr>
<tr>
<td>K.B 12 PS</td>
<td>Citrobacter freundii</td>
<td>KY751356</td>
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<tr>
<td>K.B 13 PS</td>
<td>Enterobacter asburiae</td>
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<td>K.B 14 PS</td>
<td>Enterobacter aerogenes</td>
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<td>K.B 15 PS</td>
<td>Escherichia coli sp.</td>
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<td>K.B 16 PS</td>
<td>Shigella sp.</td>
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<tr>
<td>K.B 17 PS</td>
<td>Shigella dysenteriae</td>
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<tr>
<td>K.B 18 MBR sludge</td>
<td>Salmonella waycross</td>
<td>KY751362</td>
<td></td>
</tr>
<tr>
<td>K.B 19 PS</td>
<td>Escherichia albertii</td>
<td>KY751363</td>
<td></td>
</tr>
</tbody>
</table>

asburiae, Enterobacter aerogenes, gamma proteobacterium, Franconibacter pulsarius, Citrobacter freundii, Shigella dysenteriae, Escherichia albertii and Escherichia coli. Predominant bacterial species identified from the activated sludge of the MBR setup were Salmonella enterica, Pantoea dispersa, Shigella dysenteriae, Enterobacter hormaechei and Salmonella waycross, and they too belong to the phylum Proteobacteria.

In conclusion, phytoremediation is a suitable technology for treating domestic wastewater, land limitation is a major issue that needs to be solved; however, lesser energy requirements and low capital cost are the bigger advantage for the decision makers to take into consideration. The MBR method is reliable in terms of better effluent water quality, lesser sludge production and lower land requirements; however, operational cost and maintenance requirements remains a bottleneck in the overall performance that needs to be figured out.

In Pakistan, though some companies have started the implementation of wastewater treatment technologies they still have not been implemented on a very large scale because of the electricity shortages, less skilled man power and ineffective utilization of the natural resources. Pakistan has to bear health cost of Rs. 114 billions for waterborne diseases. Through this project, introduction of aquatic plants, microbial strains and other technical tools to treat wastewater will be utilized by the industrialists and other stakeholders to treat effluent before final discharge. This academic research will spur wastewater treatment businesses in the country.

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