



## Phytoremediation Potential of *Vetiver Zizanioides*: A Green Technology to Remove Pollutants from Pampa River by Hydroponic Technique

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### ABSTRACT

Pampa river is the third longest and one of the most stressed rivers in Kerala. Hence, many researches have been done highlighting the pollution status of Pampa river. In this study, phytoremediation, the cost-effective and eco-friendly technique was employed for the removal of contaminants from this river. By planting *Vetiver zizanioides* for 6 months by hydroponic technique, the amount of lead, mercury, and most probable number of coliform was found to be decreased by 100%. A large decrease of dye content was also noticed by this technique.

**Key words:** Microbiological contamination, Hydroponic technique, Phytoremediation, *Vetiver zizanioides*, Pollutant removal efficiency.

### 1. INTRODUCTION

Water is the most important resource, around which the life on earth revolves. Even though earth holds 70% of surface water, unfortunately, only <1% of the water supply can be used for drinking. The dumping of pollutants directly or indirectly to the water bodies has increased significantly in the recent times due to the growth of human population, industries, agricultural activities, etc. Water pollution occurs due to the addition of a lot of undesirable materials into the aquatic systems.

Pampa river is one of the most important rivers in the South Western Hills of Kerala. The famous shrine of Sabarimala, one of the most popular pilgrim centers in South India is situated in the hills of Pampa plateau. Lack of sanitary latrines, lack of facilities for sewage collection and treatment, accumulation of wastes discharged from hotels, and commercial establishments located at Sabarimala are the major sources for the pollution of Pampa river [1]. The pollution is mainly due to human excreta and biodegradable wastes such as used leaves, vegetable wastes, discarded clothes, and food wastes [2]. Indiscriminate disposal of used plastic bottles form the major portions of the nonbiodegradable waste. The polluted Pampa river water has become host to many waterborne diseases in the District of Pathanamthitta and Alappuzha. Dumping of waste from slaughterhouses and chicken corners into the Pampa river is another major issue of pollution.

Erumely is a place where the famous “Petta Thullal” is taking place. During Petta Thullal each devotee paints his body with green-, black-, and red-colored kumkum. This kumkum will contain toxic heavy metals such as mercury and lead as well as carcinogenic synthetic dyes [3]. Petta Thullal takes place almost every day from the first of the Malayalam month of Vrichikam till a couple of days before the closing of Sabarimala temple on Makaram. The devotees wash their bodies in Pampa River after the Petta Thullal.

The effects of the volume of the deadly cocktail of pollutants would intensify and transform Pampa river into a hot spot of pollution over the coming years. The hazardous pollution in Pampa river invited the attention of researchers across the country, and their research papers highlighted its volume of contamination. Unfortunately, no natural remedy was suggested for this burning issue.

Conventional methods of water purification are uneconomic and disrupt the structure and fertility of the soil. Here, we are suggesting an eco-friendly and economic green technology for water purification. We focused on phytoremediation technique using *Vetiver Zizanioides* (VZ), commonly called as “Ramacham” by hydroponic technique. Since VZ is a versatile plant capable of growing in different conditions, it can be effectively employed for the remediation of pollutants from contaminated water.

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## 2. EXPERIMENTAL

Water samples were collected from two selected stations of Pampa river. Station I was the region where devotees wash their body after Petta Thullal. One of the major problems encountered in this station was the production of large volumes of highly colored wastewater. Station II was a place near Sabarimala temple where pilgrims use water for their sanitary purpose.

### 2.1. Water Sampling and Characterization

Bottles were thoroughly cleaned with distilled water and rinsed with water samples before sampling. About 1.5 L of water samples from the stations were collected, without air bubbles, in 2 L sterile bottles between 9.00-9.30 am. The preservation of samples in the laboratory was performed as per standard procedure [4].

Chemical and biological parameters were measured by standard methods. Mercury and lead present in water were analyzed using inductively coupled plasma-atomic emission spectroscopy (ICP-AES). Synthetic dye content in water sample was carried out by ultraviolet (UV)-visible spectral analysis in the range of 400-600 nm.

### 2.2. Phytoremediation of Waste using VZ

Phytoremediation refers to the natural ability of certain plants called hyperaccumulators to bioaccumulate, degrade or render harmless contaminants in water, air or soil. Contaminants such as metals, pesticides, solvents, and crude oil and its derivatives have been mitigated in phytoremediation projects worldwide. This method does not require any expensive equipment or highly experienced personnel and hence easy to implement [5].

### 2.3. Plant Material

Healthy VZ clumps were selected from vetiver nursery. 3 weeks old VZ slips of roughly same size were used. They were removed from the propagating field, and the roots were cleaned carefully to remove any adhering soil. The shoots were cut back approximately in 10 cm height to reduce transpiration and root length 10 cm were used for phytoremediation experiment.

### 2.4. Pot Experiment

Pilot studies have been carried out in the laboratory, using containers of 10 L capacities to carry out the experiment. A preliminary trial was conducted over 6 months. Clumps of VZ were grown in wastewater collected from station I and II with floating island technique in containers supported by thermocol. Each container has one clump and their axis was kept submerged in the wastewater and allowed to grow for 6 months. The pots were placed on a level concrete platform which was not shaded throughout the day to ensure good shoot growth. All the pots received the same amount of sunlight and heat during the trial.

Control (wastewater from stations without plant) was also kept under the same condition.

### 2.5. Characterization of Water during Pot Culture Study

During the growth of VZ small aliquots of water samples from each container was drawn after every 2 months, and chemical and biological parameters were measured by standard procedure.

## 3. RESULTS AND DISCUSSIONS

UV-vis and ICP-AES analysis of water sample collected from station I confirmed the presence of high load of synthetic dye and heavy metals (lead and mercury) in it. Microbiological analysis of water sample collected from station II confirmed the contamination of coliform bacteria.

### 3.1. Phytoremediation of Wastewater using VZ

#### 3.1.1. Phytoremediation of mercury in station I

Mercury settle down and remains in sediments, and hence, is considered as the most dangerous pollutant. Before planting VZ, the amount of mercury in station I was  $2.5 \text{ mgL}^{-1}$  during the pilgrimage season. The permissible limit of mercury in drinking water is  $0.002 \text{ mgL}^{-1}$  [6]. After planting VZ, the mercury level decreases for every 2 months as shown by Figure 1. After 4 months of planting, 100% reduction of mercury level was found, compared to that in the control pot. Similar results were obtained by Truong by planting VZ in the Lake and Yolo countries of Northern California [7].

#### 3.1.2. Phytoremediation of lead in station I

Initially, the concentration of lead in station I was  $1.03 \text{ mgL}^{-1}$ . The permissible limit of lead in water is  $0.015 \text{ mgL}^{-1}$  [8]. After planting VZ, there was a reduction in the concentration of lead in every 2 months which is evident from Figure 2. 100% reduction of lead was found after 4 months of planting, compared to that in the control pot. This result was in close agreement with the study conducted by Wilde and coworkers. They planted VZ in synthetic wastewater for 4 months whereby the concentration of lead was found to be below detectable limit [9]. VZ is tolerant to extremely high levels of Pb in soil [10]. Most of the absorbed Pb in VZ plants tends to accumulate in the roots, and only a small portion moved to shoots [11].

#### 3.1.3. Phytoremediation of synthetic dye

UV-vis spectral analysis in the range of 400-600 nm was taken after every 2 months of treatment with VZ. The decrease in the amount of dye in the water sample within 2-6 months as confirmed by UV-vis spectra is shown in Figure 3. This clearly suggests that VZ is highly beneficial for phytoremediation of dye in wastewater.

The removal of color from dye bearing wastewater by VZ may be due to its large-sized root system and luxuriant growth in the medium.

3.1.4. Phytoremediation of coliform

About 100% removal of most probable number of Coliform from wastewater was observed by planting VZ for 6 months. In the control sample which was kept idle for 6 months, there has been a steady increase of coliform bacteria is shown in Figure 4. This may be due to the favorable condition for the growth of coliform in stagnant water. Similar work has been carried out in Australia and found that VZ successfully removed the waste products from the septic tank effluent [12].

3.1.5. Chemical analysis of VZ after phytoremediation

VZ grew well in the wastewater with no visible signs of adverse effect and root and shoot grow efficiently.

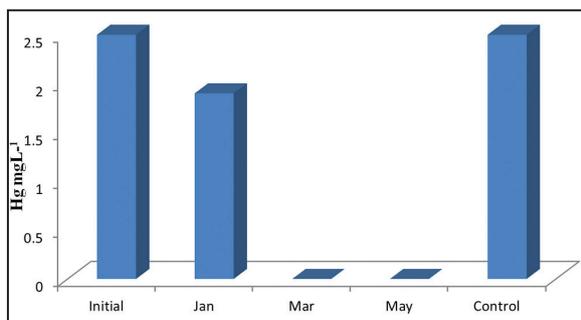


Figure 1: Variation of Hg (mgL<sup>-1</sup>) after planting *Vetiver zizanioides*.

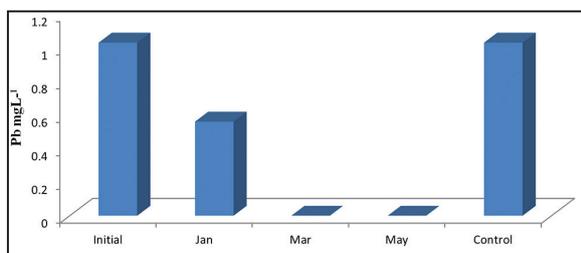


Figure 2: Variation of Pb (mgL<sup>-1</sup>) by planting *Vetiver zizanioides*.

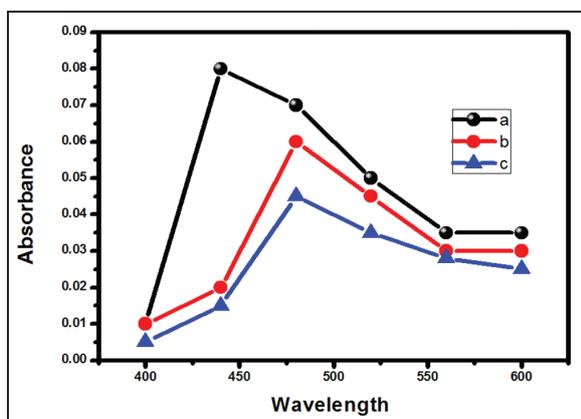


Figure 3: UV-vis spectral analysis analysis of dye content after, (a) 2 months, (b) 4 months, (c) 6 months of planting *Vetiver zizanioides*.

Figure 5 shows the growth performance of VZ after 6 months of planting.

Variation of the color of wastewater after planting VZ with an interval of 2 months is shown in Figure 6. The reduction in the color intensity was due to the high dye absorption efficiency of VZ.

3.1.6. Scanning electron microscopy (SEM)

SEM result of VZ root and shoot with reference shows a modification of the surface. This is because of the

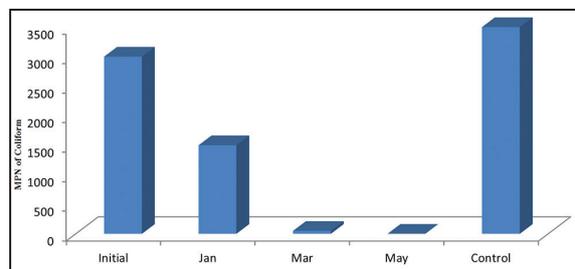


Figure 4: Variation of most probable number of coliform after planting *Vetiver zizanioides*.

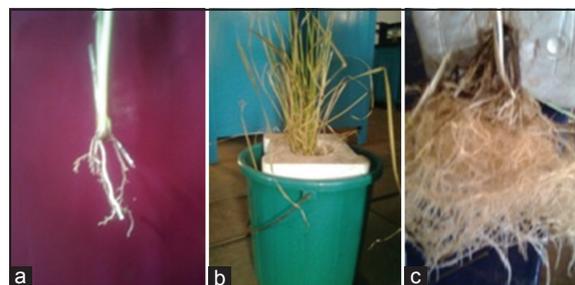


Figure 5: Growth performance of *Vetiver zizanioides* shoot and root in the wastewater. (a) At starting period, (b) shoot after 6 months, (c) root after 6 months.



Figure 6: Water samples from the control pot and treatment pot with 2 months interval. (a) Initial, (b) after 2 months, (c) after 4 months, (d) after 6 months of planting *Vetiver zizanioides* (VZ)

absorption of heavy metals such as mercury and lead. The sample root surface shows greater variation than reference root. This indicates that the pollutant was largely absorbed in the root biomass. There is a little variation in shoot surface as compared to the reference shoot. This may be due to the accumulation of heavy metals in root biomass with little transportation to shoot (Figure 7).

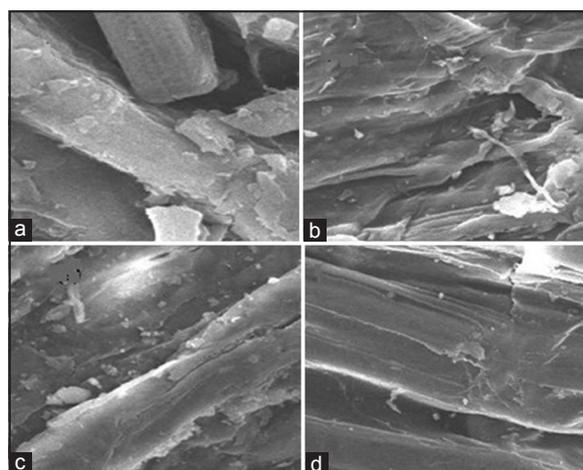
### 3.1.7. Gas chromatography with mass spectrum (GC-MS) analysis

The chloroform extract after removing the wax content, of powdered VZ root and shoot planted in the medium and control were used for the GC-MS analysis. The wax content was found to be more in the plants grown in the polluted medium. The reason was due to the fact that the plant has to produce more wax to face the stress condition in the polluted water [13]. GC-MS study of VZ revealed qualitative

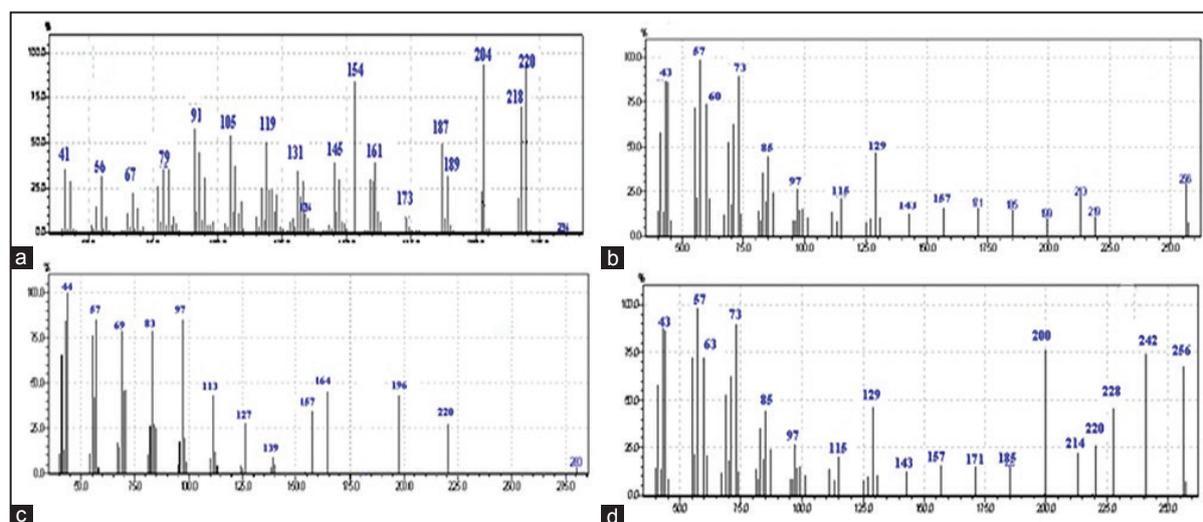
analysis of different constituent before and after planting the polluted medium. The chemical samples chloroform identities of a number of compounds from root and leaf extract were determined by matching their recorded mass spectra with data bank spectra (NIST and WILLEY libraries) provided by the instrument. The compounds identified in the root extract before planting in the contaminated water at retention time 14.8 were terpinen-4-ol (mol.wt.154), vetivone (mol.wt.218), khusimone (mol.wt.204), and khusimol (mol.wt.220). Similar components were reported by the GC-MS analysis of VZ roots grown under different cultivation conditions [14]. The above components were found to be absent in the root extract after planting VZ in the polluted water. This may be due to the fact that metabolism plays a significant role in phytodegradation of contaminants as well.

The components identified in the leaf extract of VZ at retention time 14.8 before planting in the contaminated water were 2,4-Pyrimidinedione (mol.wt.157), 2-Piperidinone (mol.wt.113), 7-Methylcaprolactam (mol.wt. 127), 2-Methyl-2-(3-oxobutyl)-1,3-cyclohexanedione (mol.wt.196), and 4,7-decadiynoicacid (mol.wt.164). In the extract of leaf after planting in the polluted medium, several new components such as pentadecanoic acid (mol.wt.242), hexadecanoic acid (mol.wt.256), tetradecanoic acid (mol.wt.228), and dodecanoic acid (mol.wt.200) were observed (Figure 8).

Phytodegradation can result in the formation of toxic intermediate chemicals from the original contaminant or result in the creation of fewer toxic compounds, thus having a beneficial effect. Plants absorb contaminants through root systems and store them in the root biomass and transport them to the stem or leaves.



**Figure 7:** Scanning electron microscopy analysis of *Vetiver zizanioides* root and shoot. (a) Sample root, (b) reference root, (c) sample shoot (d) reference shoot.



**Figure 8:** Gas chromatography with mass spectrum of root extract. (a and b) And shoot extract (c and d) of *Vetiver zizanioides*. (a) In control, (b) in wastewater, (c) in control, (d) in wastewater.

#### 4. CONCLUSION

The phytoremediation efficiency of VZ was evaluated by planting it in wastewater of Pampa river by a floating platform technique in the lab. It has been shown that VZ is highly capable of reducing coliform bacterial counts by 100% for 6 months of planting. Mercury and lead in station I was found to be decreased by 100% by planting VZ for 4 months. From the UV-vis spectral analysis, a reduction in dye removal was also observed by planting VZ for 6 months in the wastewater.

The ability of VZ to absorb harmful heavy metals, bacteria, and dye should be of great concern to all because fresh water and effective sanitation are basic necessities of all human beings. The fact that VZ reduces disease-causing bacteria should be a reason enough to consider it as a solution for environmental remediation of pollution. In addition, no harm can be caused by testing it in the environment, as greater harm is occurring by doing nothing.

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#### \*Bibliographical Sketch



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