Pollutant Removal in Wastewater by Vetiver Grass in Constructed Wetland System

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Abstract

Constructed wetland technology is one of the emerging and acceptable technologies because it can effectively remove all most all types of pollutants from waste waters without harming the environment. The objective of the present study was to find out the effectiveness of vetiver grass (Vetiveria zizanioides L. Nash) in the pollutant removal from waste water in constructed wetlands. The vetiver plants (Vetiveria zizanioides L.Nash) (ODV-3) were planted (Test group and control group) in the constructed wetland and after 90 days, the test group was divided into three (T1, T2, T3) and were treated with waste water (50% dilution) from automobile service station (W1), spray painting workshop (W2) and sewage (W3) respectively, and allowed to grow for further 15 days. At the end of the experiment (on the 15th day of waste water treatment), the treated water from the tanks was collected and analyzed for various chemical attributes. The plants were uprooted, and the plant biometric parameters and nutrient content were also determined. The chemical characteristics of the wastewaters analysed show that all the wastewaters were contaminated, and automobile service station effluent was heavily polluted. More than 50% percentage removal of pollutants especially nutrients after 15 days treatment of waste waters in constructed wetlands was observed, and it showed the efficiency of the vetiver variety for improving the water quality. In the entire test group plants studied the important biometric parameters and total wet biomass was increased compared to that of the control plants after the effluent treatment period. All the wastewaters supplied enough nutrients to the plants and T3 plants showed highest growth which was treated with sewage water. Therefore this phytoremediation study using Vetiveria zizanioides L. Nash (ODV-3) in constructed wetlands clearly showed the effective removal of pollutants from the wastewaters. Increase in growth parameters and nutrient content in the roots and leaves of the plants is an indication of the efficiency of absorption and accumulation of nutrients by the plants from the wastewaters and highly tolerant nature of vetiver against the wastewater induced stress. From the present study it was evident that vetiver grass is an ideal candidate for waste water treatment using constructed wetland technology.

Key words: Biometric parameters, constructed wetland, nutrients, pollutant removal, vetiver, waste water.

1. Introduction

One of the major global threats that our environment is facing today is environmental pollution, increasing with every passing day and causing grave and irreparable damage to the atmosphere. Pollution of soil and water with waste waters of different characteristics is a common practice. Waste water treatment before disposal is the only remedy for this problem. For the treatment of these waste waters rich in nutrients and other toxic chemicals has mainly been done using conventional wastewater treatment systems such as activated sludge and biological nutrient removal technologies or otherwise by several chemical methods. These technologies are very expensive and dependent on electrical energy and skilled personnel or impossible to carry out, as the volume of contaminated material is very large (Salomons *et al*., 1995).

The constructed wetland systems have proved to be an adequate technology for the treatment of a wide variety of waste water in urban, suburban and rural areas of many developed countries (ITRC, 2003). On the contrary, only few CWs are installed to study the potential applicability of this technology in India till date. It is based on the principle of attached growth biological reactors similar to conventional trickling filters with a combination of aerobic and anaerobic zones created around plant roots. The contaminants present in the wastewater are treated as they seep through the root-zone of the plants by a combination of plants, soil, and bacteria and hydraulic flow systems resulting in physical, chemical, and microbiological processes. Oxygen present in the zones closer to the roots facilitates the degradation of wastewater. A wide variety of microorganisms present in the root-zone of the plants results in efficient degradation of organics (Gupta *et al.*, 2013).

Vegetation is important for all phytoremediation applications, either in soil or in wetlands. The plant accepted as safe for phytoremediation and assisted works; it should have certain qualities or characteristics. It is necessary to use plants that tolerate high levels of toxic pollutants (Troung *et al.*, 1995). The scientific research conducted in the last ten years has clearly demonstrated that vetiver grass (*Vetiveria zizanioides* L. Nash) is also one of the most effective and low-cost natural methods of environmental protection (Anon, 1997).

Vetiver grass is a versatile hardy plant having stuff and erect stems, deep, extensive, fast growing and penetrating root systems and are highly tolerant to adverse climatic conditions [frost, heat, wave, temperature (5-55^{0}C), drought, flood and inundation], edaphic conditions, and highly tolerant to elevated levels of heavy metals, herbicides, pesticides, and high efficiency in absorbing dissolved nitrogen, phosphorous, sulphate, As, Cd, Cu, Pb, Hg, Ni, Se and Zn (Zhen *et al*.,1997; Troung *et al*., 1995 and Knoll, 1997). Research in China indicated that vetiver could remove dissolved nutrients and reduced algal growth within two days under experimental conditions (Anon, 1997).

2. Materials and Methods

2.1. Plant Materials

The Vetiver plants (*Vetiveria zizanioides* L.Nash) (ODV-3) were procured from the Herbal Garden of Aromatic and Medicinal Plants Research Station, Kerala Agricultural University, Odakkali, Ernakulam district in Kerala, South India. Nair *et al*. (1983) reported that the ODV-3 is high yielding variety among the other varieties in Kerala. The vetiver plants were removed from the propagating soil and surface sterilized with distilled water to remove any adhering soil. Then the tops and roots of the vetiver sprouts were pruned to 10 cm and 5 cm respectively.

2.2. Waste waters used for the experimental study

The waste waters were collected from three selected stations in Thiruvananthapuram city and were used for phytoremediation studies in constructed wetlands using vetiver plants. These includes wastewaters collected from an Automobile Service station at Kaniyapuram (W1); Automobile spray painting workshop at Kazhakkuttom (W2) and the raw sewage from the Valiyathura sewage farm (W3) in Thiruvananthapuram district, Kerala.

The wastewaters used for the experimental study were diluted (50%) with tap water. For the determination of Biological Oxygen Demand, two sets of each sample were collected in the labelled BOD bottles and one set was fixed with Winkler's reagent. All these samples were brought to the laboratory and analyzed for various chemical characteristics.

2.3. Methodology

The various chemical attributes of waste water samples were analyzed following the procedures in APHA (2012) and by Trivedy & Goel (1998). The biometric parameters of the experimental plants were determined following the standard procedures of Sadasivam and Manikam (1996) and Gupta (1998). The concentration of nutrients (sodium and potassium) was determined using a Flame photometer (ELICO, Model CL 360). All the bio-chemicals and chemicals used for the estimations were of analytical grade.

2.4. Experimental Design

The wetlands were constructed according to UN-HABITAT (2008) in plastic tanks having 50 litres capacity with drainage facility by filling the base with gravel (20-40mm) up to 8 cm height. The next layer was filled with gravel (5-10mm) to a height of 3 cm. Then the thick sand was used to fill the next layer to a height of 25 cm and again the gravel (5-10mm.) was used to fill the top layer of 3 cm height. The tap water was poured into the tanks so that the water column was 10cm above the soil bed and kept for 1 week for acclimatization. Then drained out all the water from the tanks and six clumps of vetiver grass, (*Vetiveria zizanioides* L. Nash, ODV-3) were planted in each tank and were maintained in the controlled condition for growth. All the plants were watered daily with sufficient amounts of tap water for growth and acclimatization. After one month growth, the tanks were loaded with tap water until the water level was 10cm higher than the soil surface to create a constructed wetland (CW) system.

The vetiver plants were allowed to grow under controlled conditions for three months to attain minimum maturity. After 90 days, the test plants were divided into three groups (T_1, T_2, T_3) and were treated with waste water (50% dilution) collected from automobile service station, spray painting workshop and sewage which was directly loaded to the respective tanks. The test groups and control group plants were allowed to grow for further 15 days. The total experimental period was for 105 days. Replicates of the experimental groups were also maintained. The wastewater treatment period was for 15 days.

At the end of the experimental period (on the 15^{th} day of waste water treatment), the treated water from the tanks were collected in sampling bottles and the physico-chemical parameters were analyzed. The entire study group plants were uprooted and the morphological changes (plant height, number of leaves, leaf area, and root length) and the total wet biomass of different test group plants were determined. Then the plant parts (leaves and roots) of the different test group plants were separated and the concentration of different physio-biochemical parameters such as total chlorophyll, carotenoids, proteins and carbohydrates were determined. The oven dried plant materials were used for the analysis of nutrients such as total nitrogen, phosphorous, sodium, and potassium.

3. Results and Discussion

3.1. Water and Waste water Characteristics before treatment

The results of the chemical characteristics of tap water and waste waters are given in Table. 1.

Table.1. Chemical characteristics of tap water and waste water.

Parameters	Tap Water	W1	W2	W3
TDS (mg L^{-1})	57	30408.21	2140.64	1641.76
BOD (mg L^{-1})	BDL	5.26	6.47	44.91
COD (mg L^{-1})	BDL	320	498	1290
NO_3-N (mg L^{-1})	BDL	0.27	0.39	0.97
NO_2-N (mg L^{-1})	BDL	BDL	BDL	BDL
TN (mg L^{-1})	BDL	8.6	9.5	23.67
NH_4-N (mg L^{-1})	BDL	3.9	5.1	17.61
Total Phosphorus (mg L^{-1})	BDL	2.07	1.57	4.01
Inorganic Phosphorus (mg L^{-1})	BDL	1.37	1.04	2.43
Silicate (mg L^{-1})	6.5	13.66	9.41	11.41
Chloride (mg L^{-1})	56.6	97.71	621.44	83.47
Sulphate (mg L^{-1})	68.7	93.41	39.41	71.28
Ca (as $CaCO_3$) (mg L^{-1})	36	65.4	557.6	110.4
Mg (as $CaCO_3$) (mg L^{-1})	24	39.2	42.6	50.4
Na (mg L^{-1})	5.9	237.6	66.4	69.4
K (mg L^{-1})	0.7	40.6	47.6	50.1
Iron (mg L^{-1})	0.62	6.2	0.61	1.61

BDL- Below Detectable Limit

The results show that the various chemical parameters of tap water used in the control wetland showed that all the chemical parameters are within the permissible limit of Indian Environmental Standards and Environment Protection Act (Raman and Devotte, 2006 and EPA, 2002). In different waste waters analysed majority of the water quality parameters *i.e.* TDS, COD, TN, NH_4-N, Ca, Mg etc. are above the permissible standard limits for safe disposal to inland water bodies/ land as per the Indian Environmental Standards (Raman and Devotte, 2006) and EPA (2002). All the other parameters analysed were within the standard permissible level but the continuous discharge of these effluents can cause serious effects on land or receiving water body.

3.2. Changes in the waste water characteristics after treatment

The results of the analysis of various chemical characteristics of water and waste waters after the experimental period were given in Table 2.

Table.2. Chemical characteristics of tap water and waste water after treatment

Parameters	Tap Water	W1	W2	W3
TDS (mg L^{-1})	59.9	777.56	396.02	329.99
BOD (mg L^{-1})	0.91	0.94	1.17	7.45
COD (mg L^{-1})	2.12	60.8	90.78	223.04
NO_3-N (mg L^{-1})	BDL	0.052	0.074	0.18
NO_2-N (mg L^{-1})	BDL	BDL	BDL	BDL
TN (mg L^{-1})	0.13	3.68	4.14	10.26
NH_4-N (mg L^{-1})	BDL	1.75	2.21	7.84
Total Phosphorus (mg L^{-1})	0.021	0.51	0.38	0.97
Inorganic Phosphorus (mg L^{-1})	BDL	0.42	0.312	0.721
Silicate (mg L^{-1})	0.36	7.22	4.95	5.92
Chloride (mg L^{-1})	2.64	45.49	277.97	37.91
Sulphate (mg L^{-1})	BDL	30.42	12.37	22.23
Ca (as $CaCO_3$) (mg L^{-1})	1.62	23.54	190.42	37.64
Mg (as $CaCO_3$) (mg L^{-1})	1.21	13.48	14.52	16.56
Na (mg L^{-1})	BDL	88.76	25.42	27.11
K (mg L^{-1})	BDL	16.41	19.01	19.59
Iron (mg L^{-1})	0.91	1.77	0.17	0.44

BDL- Below Detectable Limit

Total dissolved solids (TDS) are the amount of solids which are dissolved in water. It was found that after the experimental period the TDS of the waste waters were reduced. The percentage reduction in TDS content were recorded as 97.44 %, 81.51% and 80.0% in W1, W2 and W3 waste waters respectively. The solids present in the effluents are removed by the wetland system by processes like straining, sedimentation, impaction and interception. In addition, during the particle transport through the filtering media, there could be other removal mechanisms such as adhesion, chemical and physical adsorption, and flocculation, depending on characteristics of the particles and media (Koottatep *et al.*, 2010). The removal of solids takes place through a complicated set of internal processes including production of transportable solids by wetland biota, low water velocity, vegetation, substrate and filtration (Reed *et al.*, 1995).

Biological Oxygen Demand (BOD) is an indicator of pollution in water. It determines the amount of DO used up for the oxidation of degradable biological waste materials present in water. The results (Table 1 & 2) show that the BOD content of the raw waste waters was higher before the treatment in constructed wetland system. But after the treatment it was reduced by 82.13%, 81.92% and 83.41% in the service station effluent, spray painting spent wash and raw sewage respectively. BOD reduction in wetland is achieved by aerobic bacteria attached to the media and to the plant roots (Shivhare and Roy, 2013).

Chemical Oxygen Demand (COD) determines the amount of DO used for the chemical oxidation of wastes in water. High COD indicates that high degree of pollution load in water. From the results it is observed that, constructed wetland technology using vetiver grass is a very effective method of treating waste waters containing higher COD. Because when the treatment period was over, the COD of the waste waters reduced considerably. The percentage reduction was recorded as 81.0%, 81.77% and 82.71% in the waste waters such as W1, W2 and W3 respectively. According to Zirschky (1986) and Vipat *et al.* (2008) BOD and COD associated with settleable solids in waste water is removed by sedimentation while that in colloids and soluble form is removed as a metabolic activity of microorganisms and physical and chemical interaction within the root zone/substrate.

Nitrates are one of the important limiting nutrients. Presence of nitrate indicates organic pollution in water. The results show that the NO_3-N concentration in the waste waters were reduced to a notable extent by the treatment with vetiver grass and showed a removal efficiency of 80.74%, 81.02% and 81.44% in the W1, W2 and W3 waste waters respectively. Nitrogen undergoes several transformations in wetlands, including ammonification, nitrification, denitrification, volatilization, adsorption and plant and bacteria uptakes and these mechanisms are considered as the key process for nitrogen removal from wetlands (Shivhare and Roy, 2013). According to Billore *et al*. (2002) plant absorption is the dominant mechanism of nitrogen removal. The present study also shows that vetiver plants in the constructed wetlands removes the NO_2-N contents in all waste waters by 100%.

By comparing the values of TN content in the waste waters such as service station effluent, spray painting spent wash and raw sewage indicated that the wetland treatment with the vetiver grass is very efficient in removing the TN content from the different waste waters. The results show that the vetiver has a removal efficiency of 57.21%, 56.42% and 56.65% in the W1, W2 and W3 wastewaters respectively.

The ammonia-nitrogen content in the waste waters also reduced considerably due to the vetiver wetland technology by 55.13%, 56.67% and 55.48% in service station effluent, spray painting spent wash and raw sewage respectively. According to Shivhare and Roy (2013) about 35% of total ammonia oxidized gets converted into nitrites and nitrates and this indicates low activity of autotrophic ammonia oxidizing and nitrate oxidizing bacteria. Hence the oxidized ammonia is either getting converted into nitrogen gas or consumed by plant uptake or by volatilization through aeration but volatilization requires high pH (>11) and high DO in the system. Hence plant uptake is the major mechanism of ammonia removal in this system.

Other important nutrients like inorganic phosphorus and total phosphorus were also found decreased on treatment with vetiver wetland technology, and the percentage reduction was recorded for inorganic phosphorus as 69.34%, 70.01% and 70.33% and for total as 75.36%, 75.77% and 75.81% in service station effluent, spray painting spent wash and raw sewage respectively. Phosphorus removal can be achieved in CWs by adsorption and precipitation, and a small amount is also taken up by plant growth (Hoffman *et al*., 2011). The phosphorus in the wastewater may be removed through sedimentation and burial adsorption and precipitation, and exchange process between soil and overlying water column (Boonsong and Chansiri, 2008).

The constructed wetland with vetiver grass showed a removal efficiency of 47.14%, 47.40% and 48.12% for silicates, and 67.43%, 68.61% and 68.81% for sulphates in service station effluent, spray painting spent wash and raw sewage respectively. The silicates and sulphates in the different waste water treated are also effectively removed by vetiver plants.

The study also revealed that the waste waters containing chlorides, calcium and magnesium can also be treated effectively using constructed wetland technology with vetiver plants. The chloride contents in the waste waters were reduced by 53.44%, 55.27% and 54.58%, the Ca contents by 64.01%, 65.85% and 65.91% and the Mg contents were reduced by 65.61%, 65.92% and 67.14% in the service station effluent, spray painting spent wash and raw sewage respectively.

As in the case of other nutrients the constructed wetland with vetiver grass removes incoming sodium and potassium contents in the influents with a percentage removal efficiency of 62.64%, 61.72% and 60.94% and 59.58%, 60.10% and 60.90% respectively in the service station effluent, spray painting spent wash and raw sewage. The marked increase in the performance is due to the increase of root system in the vegetation that is responsible for the uptake of nutrients. The study also shows that the iron present in the waste waters is also effectively removed by vetiver plants. After the treatment period the iron content in the waste waters were reduced by 71.45%, 72.13% and 72.67% in service station effluent, spray painting spent wash and raw sewage respectively.

Vegetation and the rhizosphere play an important role in the wastewater treatment. The breakdown of contaminants and the treatment of wastewater are achieved by the controlled seepage of the waterborne pollutants through a root-zone of plants. Organic pollutants are broken down as a food source for the extraordinary variety of microorganisms that are present in the soil and plants. In fact, plants need all the nutrients for their metabolism to grow and to reproduce (Baskar *et al*., 2009). The vetiver grass has thick massive root system and therefore the surface area is also very high. It provides enough space for the microbes to multiply and the wide spreading roots taken up nutrients and other contaminants as a part of their uptake mechanisms and incorporates it in their biomass.

3.3. Changes in the Biometric parameters of Vetiver plants

3.3.1. Changes in the plant morphology and biomass

Characteristics of waste waters may differ with their source of generation. So the response of the plants grown in waste water contaminated sites may also differ. The morphological characteristics such as plant height, root length, number of leaves and total wet biomass of the plants are given in Table.3.

Table.3. Changes in the Biometric parameters and Total biomass of Vetiver plants

Groups	Plant Height (cm)	Root Length (cm)	No. of Leaves	Total wet Biomass (g)
C	180.6	190.2	26	428.5
T1	174.6	186.3	28	421.8
T2	180.9	192.3	28	437.6
T3	182.3	192.4	28	440.6

From the results it was clear that the different waste waters influenced the growth of the plants. The comparison with control plants, the test group plants showed variations in their growth. The plants treated with service station effluent (T1) showed comparatively lesser growth than the others and it is evident from the height, root length and low biomass content. It may be due to the stress induced by various pollutants in service station effluent (Singh, 2005). The variations in morphology were resulted from the stress induction due to this waste water flooding. But this decrease was not that much significant when compared to control plants. It proves the fact that the vetiver plants are very tolerant, even in metal polluted sites.

The waste water from spray painting workshop showed moderate growth and biomass production. But the sewage treated vetiver plants showed higher growth and biomass production when we compare them with the control plants. It may be resulted due to the sewage water application. The studies by Boonsong and Chansiri (2008) reported that sewage waters were rich in nutrients like N, P and K, because of the presence of organic matter. He also reported that, the intake of these macronutrients by highly tolerant plants resulting in fast growth even in metal contaminated sites. This also is in agreement with the results of our study.

3.3.2. Changes in the concentration of Pigments and Biomolecules

Chlorophylls, carotenoids and pheophytin are the important photosynthetic pigments found in leaves. The pigment concentrations of both the control and test groups were estimated and given in the Figure 1. The important biomolecules such as carbohydrates and protein of the test group and control were estimated and are given in Figure 2.

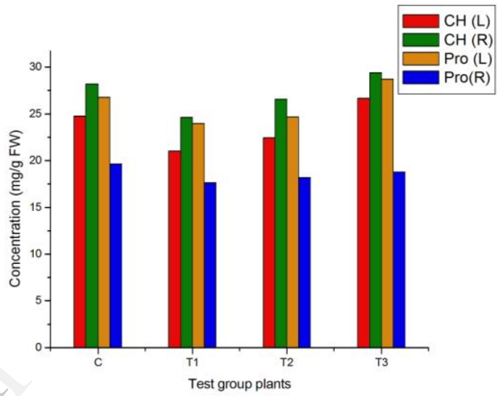

Figure 1. Changes in pigment content in vetiver plants

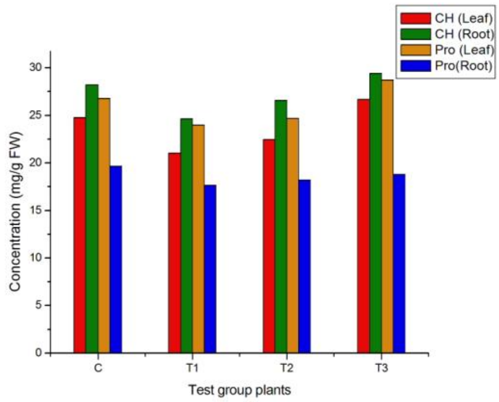

Figure 2. Changes in carbohydrate and protein content in vetiver plants

The results show that both the concentration of pigments and biomolecules in the test group plants varied considerably with the characteristics of the applied effluent. When we compare the concentration of pigments and biomolecules in the T1 and T2 test group plants with that of the control plants, it is clear that the values are reduced slightly and the reduction was higher in T1 plants which were treated with service station effluent. But in the case of T3 group (treated with sewage) all the pigments and biomolecules are increased compared to the control plants. Here also the reduction is not very significant. It is evident from the results that among three waste waters, W1 (service station effluent) induced more stress on the vetiver plants, but they tolerate it to a good extent because of their good defence mechanism. In the case of T3 group, the sewage was rich in all nutrients and the plants well nourished with the sewage and grown well.

3.3.3. Changes in the concentration of Nutrients

Nitrogen, phosphorus, sodium and potassium are very important among the macronutrients in the plant body. The results obtained after the experimental period of the control and test group plants are given in Figure 3.

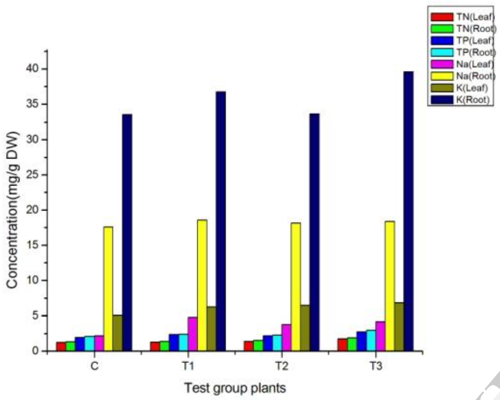

Figure 3. Changes in Nitrogen, Phosphorus, Sodium and potassium contents in vetiver plants.

From the results it was clear that, as compared to control all the nutrients in the test groups were increased (both in leaves and roots) when the experimental period over. This indicates that all the three effluents supplied enough nutrients to the plants and the plants absorbed them from the soil solution to a greater extent. It was also observed that all the four nutrients were found higher roots than in leaves.

The total nitrogen (TN) and total phosphorus (TP) content in the experimental groups were in the order T3>T1>T2>C and it was same in the case of effluents added. That means sewage was rich in TN and TP than the other two and thus the T3 plants absorbed higher amounts of nitrogen and phosphorus than the others and grown well and established better root system. It is evident from the plant's biometric parameters also.

It was also observed that among the three test groups, T1 reported higher concentration of sodium and T2 showed lower concentration than the T1 and T3. This may be due high concentration of sodium in the effluents treated. That is in W1 (service station effluent) the sodium content was very higher than the other two wastewaters. As in the case of TN and TP the potassium (K) content was more in T3 group plants i.e. the group treated with sewage. The potassium content estimated in the experimental group plants were in the order T3>T2>T1>C.

4. Conclusion

In this phytoremediation study by constructed wetland technology using vetiver grass, there is a remarkable reduction in total dissolved solids, biological oxygen demand, chemical oxygen demand, nitrates, phosphates, silicates, sulphates, chlorides, calcium, magnesium, sodium, potassium and iron in different waste waters. The treated water can be directly disposed into any surface water body or land as the concentration of the pollutants are below the standard permissible limits of effluent discharge. Therefore *Vetiveria zizanioides* L. Nash is a good candidate for remediation of waste waters by constructed wetland technology.

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