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Evaluation of the Effectiveness of Vetiver Plant with the Hydroponic Culture to Remove Anionic Surfactant LAS from Hospital Laundry Wastewater

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Abstract

Background & Objective**:** Phytoremediation is an economically, environmentally, and scientifically efficient technique suitable for developing countries, and it is a worthwhile business. One of the special plants for remediation of these effluents is the vetiver plant. The purpose of this study was to determine the effectiveness of vetiver plant with hydroponic culture to remove the LAS wastewater from the hospital's laundry.Materials and methods: This was a descriptive-analytical study. Sampling was carried out using special sampling containers for hospital laundry wastewater. In this study, the concentration of LAS parameter was measured. The concentration of anionic surfactants was measured with methylene blue active substances or MBAS method. All sampling conditions and other tests were performed according to the guidelines of the book standard method. After various tests, the obtained results were analyzed using Statistical software Excel and SPSS; the average values for each parameter were calculated, compared to the standard efficiency of the remediation process, and plotted on the corresponding graphs.Results: In this study, the root efficacy of vetiver plant in treatment of hospital laundry wastewater was investigated. The highest removal efficiency of LAS in the pilot of vetiver hydroponics culture with 0.5, 1.5, 2, 5, 10, and 20 mg/L concentrations of the hospital laundry wastewater in two days was 70, 80, 72, 60, 50, and 40 percent, respectively. The concentration of LAS in the laundry room varied from 10 to 40 mg/L.Conclusion: The overall results show that the efficiency of this method in degradation and reduction of LAS of the wastewater of hospital laundry has been decreased with increase in surfactant concentration. Increase of the concentration of surfactants resulted in reduction of oxygen levels, as well as reduction in activity of bacteria and their destruction, and reduced the effectiveness of vetiver plant in LAS treatment.

Keywords: Vetiver, Hydroponic, Surfactant, Laundry.

Introduction

The wastewater treatment is a very important process all around the world, and includes high costs in different countries and cities. Therefore, selecting a suitable technology, which is compatible with climate, economy, and social conditions is a significant issue (Ghaderi, 2004)

Using wastewater treatment systems with a low-level technology and without consuming energy, such as artificial wetlands, not only reduces economic costs but also helps environmental reformation. In artificial wetlands physical, chemical, and biological processes are used for purification of different sewage pollutants such as organic materials, detergents, nitrogenous and phosphorous compounds, heavy metals and suspended solids (Reed and et al., 1995; Brown & Reed, 1994).

Consequently, choosing the suitable plant helps a lot to increase the efficiency of the refining system. One of these plants which has been considered by experts in purification is vetiver. Owing to its unique ecological and morphological characteristics, as well as the high resistance against climate changes, long-term drought, floodwater, temperature variations between -14 C and 55 °C, pH between 3.3 and 12.5, heavy metals, and insecticides, it has many uses (Truong and et al., 2008; Barakati and et al., 2011).

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It was Gerick who first proposed the term "hydroponic". In California, he had succeeded in cultivating plants without the use of soil, from early growth to fruiting stage in a trading scale. This term was derived from two Greek words. Hydro (water) and Ponro (labor) literally means laboring something in water (Son and et al., 2016) more precisely, it is a method of growing plants without soil. In this method, some material are used to maintain the root system, and the nutrition of plant is done by nutrient solution, which is added to the substrate. The Material used as the plant growth substrate could be either an organic material such as peat humus and tree bark or an inorganic material such as perlite, vermiculite and rock wool (Arzani, 2007).

In general, hydroponic cultivation, despite the need for sufficient expertise and relatively high initial investment, compared to soil cultivation, has many benefits including high efficiency, fewer human resources, easy work, no need to crop rotation, plant growth uniformity, better control of pests & disease, minimum water loss, no competition between the plants for water and nutrients, the possibility of providing nutrients according to the needs of plants and use less chemical fertilizer and therefore having healthier crops. Because of the specific climate conditions and water resources limitations, Iran is one of those countries that needs a fundamental restructuring of the cultivation system and in doing so, the development of cultivations such as hydroponic cultivation can be an appropriate approach (Delshad, 2005).

In 1995, Bavor reported that over 90% of artificial wetlands had the potential to reduce the concentrations of organic matter, suspended solids and typical bacteria, while they have low utilization requirements. Moreover, these systems have increasingly been considered for complementary treatment of treated wastewater along with conventional wastewater treatment technologies. (Bavor and et al., 1995).
Detergents are divided into four anionic, cationic, non-ionic and amphoteric groups which the anionic ones have widespread application than the others. Based on the active surfactant agents, they have two different kinds of hard and soft. LABS or Linear alkyl benzene sulfonate soft detergents are dissoluble. On the other hand, ABS or Branched alkyl benzene sulfonate hard detergents are indissoluble in the environment (Ying, 2006).

Linear alkyl benzene sulfonate has been used as an ingredient in household detergents such as washing powder and dishwashing liquids and other types of domestic cleaners. The toxicity of detergents is mostly related to their alkyl and aryl groups. By skin defatting and its stimulation, anionic surfactants cause redness, pain and general dermatitis, and even thickening, cracking, and blistering of the skin. Oral use also causes diarrhea, bowel swelling, and occasionally nausea. Upon oral intake, detergents are absorbed in the stomach and intestines, but there is no low cutaneous absorption. They can cause allergies and eye, skin and mucous membranes irritations (Eaton and et al., 2005). Due to their physicochemical properties, detergents are widely used in many branches of technology and research in many fields like pharmaceuticals, cosmetics, textile industry, agriculture, biotechnology, etc. Detergents remain as a superficial layer on the water surface, thereby, they form an ill-scene environment, reduce gas exchange, and jeopardize the health of the aquatic animals by reducing dissolved oxygen. These compounds further result in water taste and odor changes, produce surface foams, disturb water treatment processes, increase water treatment costs, and lead to the death of aquatic organisms. These substances produce stable foam on water surface at concentrations higher than 1mg/L. The growth of aquatic plants and algae increases dissolved oxygen consumption in water which leads to mortality of fish. Other consequences of these substances include degradation and destruction of ecosystems, disruption of coagulation, sedimentation and water filtration, eutrophication due to the increased phosphates, lack of proper degradability, and physiological reactions in contaminated water consumers. A large amount of foam formed on water surface prevents the light passing through the water and inhibits the vital phenomenon of photosynthesis. Detergents are able to change the state and quality of proteins and, as a result, deactivate viruses, disrupt bacterial metabolism and retard their functions. Detergents rupture the membranes of microorganisms giving rise to the destruction of enzymes, and also retard and disturb the activities of enzymes affecting bacterial respiratory functions (Hosseini and et al., 2007; Chapman & World Health Organization, 1996; Nori and shahriari, 2005; Roshany and et al., 2007).

In 1989, EPA recommended a maximum secondary concentration of 0.5 mg/L for foaming agents. In 1984, WHO announced that no foaming agent should exist in untreated water? A maximum surfactant amount of 0.2 mg/L has been determined for drinking water. There are higher values for the standard cationic surfactants. Roshany and et al., 2007. The Institute for Standardization and Industrial Research in Iran (1996) determined a maximum permissible level of 200mcg/L for detergents in drinking water (Nordin & Amalina, 2006).

LAS is the largest group of anionic surfactants that could decompose by bacteria up to 90-97%, amounting 3-21 mg/L in domestic wastewater, which decomposes slightly in anaerobic conditions (Sharvelle and et al., 2007; Fan and et al., 2009).

Another study demonstrated that LAS detergents could decompose up to 98% in sewage treatment systems. Adverse effects of detergents on wastewater treatment include reduction of sewage treatment efficiency and increase of bacteria in the effluent. They also concluded that anionic detergents have a congesting effect on household and industrial sewages and due to their ability in producing surface foams, they can cause direct toxic effects on many organisms. Furthermore, they found out that many bacteria including Acinetobacter-johnsoni and Pseudomonas-beteli, can decompose 96.4-97.2% of linear detergents in 10 days. These two bacteria can only decompose in aerobic conditions (Amirmozafari and et al., 2007).

According to the findings of the Ali Abedini and Yadollah Yamini research in 2005, the data gathered from the analysis of the water samples show that the concentration of surfactants in the southern part of the Caspian Sea Basin has not been critical. But given the fact that surfactants may be a booster for the toxicity of other types of pollutants, for instance, heavy metals and hydrocarbons, it is important to look out for the concentrations of these pollutants in the Caspian Sea (Abedini, 2005).

The results obtained from the different areas of Anzali Wetland imply that the surfactant's concentration has a large difference in various studied locations. Statistical data show that the concentration of surfactants in the different locations had been less than 0.05. In particular, the eastern part of the Anzali Wetland, which is the main recipient of the most polluted water runoff from Pir-Bazar, has the highest concentration, while the Caspian Sea had the least amount of concentration. The comparison of the seasons showed that winter and spring had the most and least amount of surfactant, respectively (ozmat and et al., 2005).

Lattyak and Charles et al. examined the removal of detergents along with the elimination of COD in wetlands and found that only 40-70% of detergents underwent normal decomposition. The low removal efficiency was related to the measurement of non-decomposable detergents (Sharvelle and et al., 2007).

Laundry units at hospitals are one of the wastewater production sources, where detergents, disinfectants, and bleaches are used to wash and disinfect the clothes of patients, bed sheets, blankets, and other items. Typically, 250g of detergent powder, 200g of bleach powder and half a liter of Javelle water (for bed sheets, hood and white cloth) are used per 40L of water (if the capacity of washing machine be 10kg) (Zotesso and et al., 2017). Since growing the plant in the water can eliminate the pollutants by phytoremediation and, according to the studies carried out in this regard, there is no comprehensive research about the LAS removal from the hospital's laundry wastewater by the artificial wetland method with hydroponic cultivation. This study aimed to determine vetiver plant's root efficiency with hydroponic cultivation for the removal of LAS anionic surfactant from the hospital's laundry wastewater.

Materials and Methods

This experimental pilot study was carried out at Sari Faculty of Health during summer and autumn 2017. Four polyethylene pilot tanks with capacity of $20 \times 40 \times 60$ cm^3 were prepared and filled with soil mixed with sand, gravel and clay. Irrigation was performed with subsurface continuous system in this research. The mixture was stored in two 120L tanks for initial settlement; then, it completely homogenized with a mixer and entered into the four pilot tanks through the faucet with a 0.6 L/h discharge rate. In order to evaluate the efficiency of Vetiver plant in the treatment of wastewater, LAS parameters in the inflow and outflow were measured according to the standard method instructions. Finally, the efficiency of Vetiver plant in pollutant removal in different concentrations and retention times was calculated and was tabulated and shown on the graph by the EXCEL software. No plant was used in the first pilot as a control, but 10 vetiver plants per pilot were used in the second pilot. Wastewater was sampled (120 L) from the laundry wastewater and the effluent at Sari Imam Khomeini Hospital. Pilots were located in the laundry wastewater treatment plant of Sari Faculty of Health; then, both pilots were filled with the wastewater up to the bed level. The inflow and outflow concentrations of pollutants were tested in these pilots.

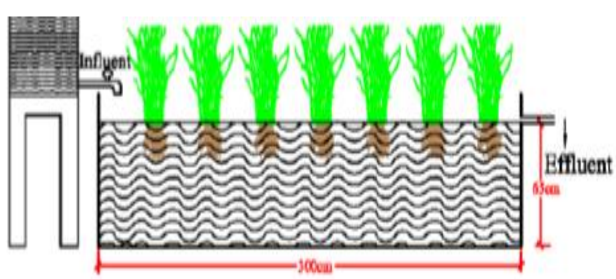

Fig. 1: polyethylene pilot tanks

Preparation of half-strength Hoagland solution

The experimental plantlets were transferred to the Faculty of Health, Mazandaran University of Medical Sciences. In order to adapt to the new environment and to prepare for planting, the plantlets were maintained hydroponically in the half-strength Hoagland solution with the following formula under artificial light for a period of 2-5 weeks. The half-strength Hoagland solution (1 L) was formulated as below:

- KH_2PO_4 0.067 g
- KNO3 0.253 g
- Ca (NO3)2 .4H2O 0.590 g

- MgCl2 .6 H2O 0.200 g

After incubation in the half-strength Hoagland solution for 2-5 weeks, the plants were weighed and transferred to the conditions provided in both artificial and natural light settings. Inflow and outflow from pilots were sampled once every two days.

Preparation of MBAS test reagents and solutions to determine the concentration of LAS anionic surfactant

Preparation of Phenolphthalein's reagent: Dissolve 0.1g of solid Phenolphthalein in 80mL in Ethyl alcohol and then dilute the solution to 100mL with distilled water.

Sodium hydroxide normal 1 solution: Dissolve 10g of solid NaoH in distilled water and then dilute the solution into a 250mL volumetric flask.
Sulfuric acid normal 1 solution: Dilute 2.7cc of concentrated solution of sulfuric acid normal 36.8 into a 100cc volumetric flask with distilled water.
Preparation of methylene blue solution: To test the amount of LAS (mg/L), we dissolve methylene blue (100mg or 0.1g) in 100mL of water, and transfer 30mL of which to a 1000mL flask, followed by addition of water (500mL). Afterwards, we add 6 normal sulfuric acid (41mL) and monohydrate sodium hydrogen phosphate (50g), dissolving well by stirring, and diluting the solution to 1000 ml with distilled water.
Preparation of washing solution: First, pour 41mL of sulfuric acid normal 6 into 500mL of distilled water in a 1000cc flask. Then, add 50g of sodium monosodium phosphate ($NaH_2PO_4H_2O$), dissolve well by stirring, and dilute to 1000mL with distilled water.

Spectrophotometric determination of LAS concentration by using methylene blue active substance (MBAS)

To conduct the experiment, sulfuric acid normal 1, phenolphthalein, and sulfuric acid were first prepared followed by formulation of methylene blue, and chloroform. The surfactants were extracted and measured by methylene blue and spectrophotometric method according to APHA 1989. The extraction was carried out with a decanter funnel with a mixture of 12.5cc of methylene blue solution and 5cc of chloroform, which was stirred vigorously for 30 seconds to separate liquid and organic phases.

The extraction process was repeated 2 times each with adding 5mL of chloroform. The chloroform layer was poured into a second funnel containing 25mL wash solution and stirred vigorously for 30 seconds. Afterwards, two more separation processes were performed with 5mL of chloroform per separation. The organic phase was transferred to a 50mL volumetric flask by means of a Pyrex funnel and silk filter, and the final solution volume was adjusted to 50mL with chloroform. Then, the sample absorption was read by spectrophotometry at a wavelength of 652nm. In this study, detergents were measured by Methylene Blue Active Substances (MBAS) method according to the instructions given in the standard method handbook. This method is a colorimetric analysis test based on the formation of a blue salt in the reaction of methylene blue with anionic surfactants. This salt is soluble in chloroform and the intensity of the color obtained in the organic phase is proportional to the initial concentration of detergent in water or wastewater after the extraction process. Then, the intensity of the final color is measured by spectrophotometer at 652 nm wavelength (American Public Health Association, 1989).

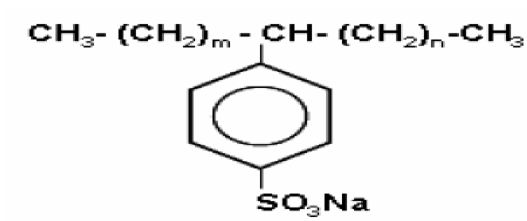

Fig. 2: Structural and chemical formula of linear alkyl benzene sulfonate (LAS)

Results

According to table.1, after reading the sample outputs of pilots by the spectrophotometer at a wavelength of 625nm, LAS concentrations were embedded on the device absorption curve, and we used the linear regression equation to determine the concentration.

Example: After calculating the amount of absorption of a sample which was equal to 0.1, we substituted numbers 0.25-0.5 into the concentration equation of unknown substance and obtained their value.

Table 1. Concentration of Produced LAS and the Correspondent Absorption with spectrophotometer at a Wavelength of 652 nm

Concentration	0.05	0.25	0.5	1.5	2.5	5
Absorption	0.02	0.07	0.125	0.5	0.8	1.5

After diluting and determining the concentration of laundry wastewater by using the calibration curve, it was transferred into the control pilots and the Vetiver plant was irrigated regularly. The results of inflow and outflow concentrations and the efficiency of each concentration were measured and recorded. In order to determine the concentration of LAS for 2 mg/L and higher, diluting and condensing were done.

Table 3. Efficiency of the Vetiver Plant in Eliminating the Various Concentrations of Surfactant Compared to Vetiver-Less Control Pilot, with a Retention Time of 2 Days

	Concentration of inflow LAS	Output of control pilot	Efficiency of control pilot	Output of vetiver pilot	Efficiency of the vetiver root
1	0.5	0.35	30%	0.15	70%
2	1.5	0.9	40%	0.3	80%
3	2.5	1.5	40%	0.7	72%
4	5	4	25%	2	60%
5	10	8	20%	5	50%
6	20	18	10%	12	40%

1. LAS outflow concentration was 0.5 mg/L with an initial concentration of 0.15 mg/L, and a Vetiver efficiency of 70% for LAS removal.
1. LAS outflow concentration was 1.5 mg/L with an initial concentration of 0.3 mg/L, and a Vetiver efficiency of 80% for LAS removal.
2. LAS outflow concentration was 2.5 mg/L with an initial concentration of 0.7 mg/L, and a Vetiver efficiency of 72% for LAS removal.
3. LAS outflow concentration was 5 mg/L with an initial concentration of 2 mg/L, and a Vetiver efficiency of 60% for LAS removal.
4. LAS outflow concentration was 10mg/L with an initial concentration of 5mg/L, and a Vetiver efficiency of 50% for LAS removal.
5. LAS outflow concentration was 20mg/L with an initial concentration of 12 mg/L, and a Vetiver efficiency of 40% for LAS removal.

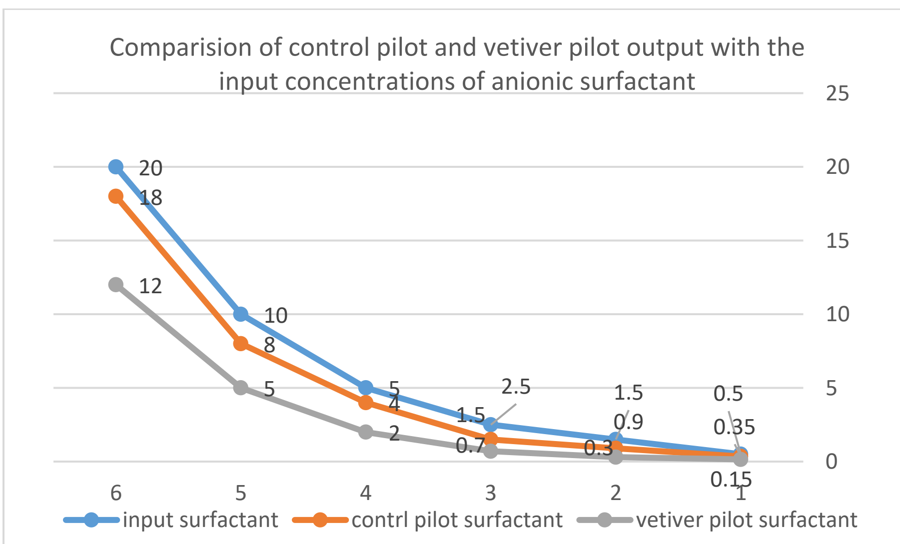

Fig. 3: Comparison of control pilot and vetiver Pilot output with the input concentrations of anionic surfactant

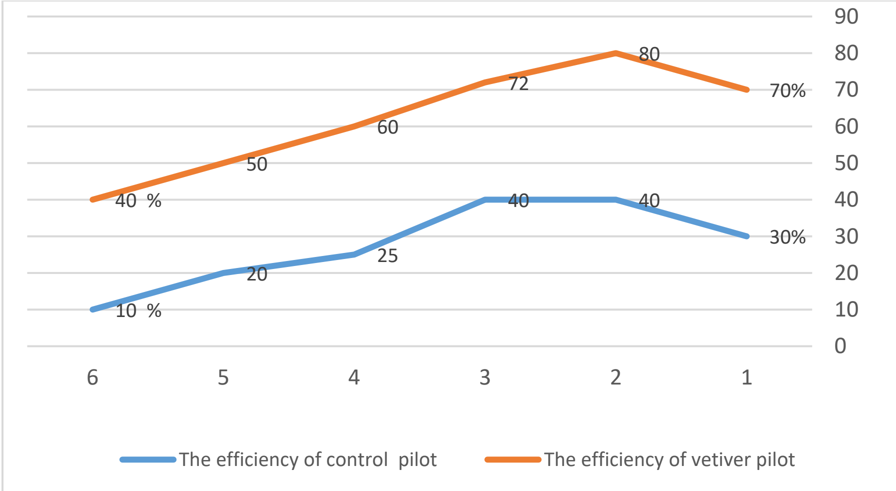

Fig. 4: Efficiency of the control pilot and the plant pilot in eliminating the various concentrations of anionic surfactant

The concentration of LAS in hospital laundry wastewater was measured by the MBAS method in the range of 10-40 mg/L and more. To create different concentrations of LAS, a laundry wastewater was diluted with different volumes of distilled water. Then, the solutions were stored in 20L and 120L tanks. After mixing with wastewaters from other hospital wards at the entrance to the treatment plant, laundry wastewater's concentration was 0.3-1 mg/L, with the efficiency of over 97%. After the aeration pool, in the effluent settling section of the refinery, this amount has dropped to 0.3-0.1 mg/L. For LAS input concentrations of 0.5, 1.5, 5, 10 and 20 mg/L with retention times of 2 days, these numbers were decreased to 0.15, 0.3, 0.7 and 2mg/L, respectively, by the root of vetiver plant's effect. Using phytoremediation by vetiver grass, LAS concentration with retention times of 2 days was reduced from 10 mg/L to less than 5 mg/L. Also, the percentage of 40 has been obtained for the efficiency of the Vetiver plant in eliminating the anionic surfactant (LAS) from the hospital's laundry wastewater, with a retention time of 2 days and 20 mg/L concentration.

Discussion and Conclusion:

As a result, with the analysis done in this study, the control pilot's efficiency range with a retention time of 2 days, was 10-40%, the Vetiver pilot's efficiency range with a retention time of 2 days, was 40-80%.

Table.4. Comparison of Output Surfactant's Concentrations from Control Pilot and Vetiver Plant Pilot According to Detergent Discharge into Environment Standards

usage of irrigation and agriculture	Absorbent Drain well	Discharge of Surface water	the concentraition of LAS from control pilot's output	the concentraition of LAS from vetiver pilot's output	the concentraition of input surfactant	
1.5	0.5	0.5				
✓	✓	✓	0.35	0.15	0.5	1
✓	✓	✓	0.9	0.3	1.5	2
✓	×	×	1.25	0.7	2.5	3
×	×	×	3	2	5	4
×	×	×	7	5	10	5
×	×	×	15	12	20	6

1. This plant at concentrations higher than 5mg/L of surfactant did not reach the standard output surfactant concentration which is below than 0.5-1.5 mg/L. The effluent ejected from the Vetiver pilot cannot be discharged to the surface water of the absorbing wells and has no use for agricultural and irrigation and also, is not environment-friendly.
   Considering the unique characteristics of Vetiver plant, including its high resistance to adverse environmental conditions and a proper efficiency, this plant is very suitable for final filtration of the wastewater. However, the results of this research show that phytoremediation of the laundry wastewater by cultivating Vetiver in hydroponic method will not be suitable because of high pollution rate, non-aeration and creating an anaerobic process. In this study, Vetiver plant has been growing exponentially in wetland with

anionic surfactant (LAS) which had a concentration range between 0.2-20 mg/L and its efficiency of purification has not been adequate. In wetlands with concentrations higher than 20 mg/L, the growth of Vetiver plant would stop and after a week, its leaves, crown, stem and eventually root would be rotten and gradually decomposed.

2. Because of high contamination, untreated sewage can cause many environmental problems. Because this is an aerobic process, if the levels of pollution is high, phytoremediation process does not persist and will abolish. Therefore, it is not suitable for the treatment of laundry wastewater. It only can be used for refining the already treated wastewater in cases where a better quality of effluent is needed.
3. Results of ANOVA test indicate that BOD5 concentration in the outflow of vetiver plant pilot is significantly lower than its outflow concentration in the control ($P < 0.05$). It is also lower than the inflow concentration of vetiver pilot. In this study, control pilot's efficiency range for its process of dissolving and eliminating the anionic surfactants (LAS), with a retention time of 2 days, was 10-40%, and the Vetiver pilot's efficiency range was 40-80% which indicates that hydroponic system is not the best method for decomposing surfactants.
4. In a study by esmaeili & Dr. dianati et.al, the removal percentage of detergents in the artificial subsurface wetland with soil bed was 70-92%, which is a relatively good efficiency (Esmaeili and et al, 2018).
   In a study by Dr. Mahvi et.al on the sewage system of Ghods Town by using lagoon method in six steps, samples were taken and the amount of detergent measured at the inlet and outlet of the phytoremediation and acquired these results: the elimination efficiency was 90-95%, the detergent effluent discharges were less than the standard limit (1.5 mg/L), and the BOD's exclusion rate was 91.9%. They implied that the wastewater treatment plant is working with proper efficiency (Mahvi and et al., 2004).
5. Consequently, hydroponic cultivated Vetiver plant is not a proper plant for wastewater treatment. The laundry sewerage, first needs to be filtered by the sludge aeration process and then, go through the artificial wetland of Vetiver plant.
6. Also, vetiver cultivated in the artificial wetland within 5 months (mid-spring until late summer) with a suitable irrigation reached the highest growth rate (1.5 - 1.8 m in height). It is, therefore, very useful as livestock fodder after harvesting and chopping.

Suggestions

More than all the provinces in Iran, Mazandaran province with 650 natural wetlands and 17500 hectares' area of natural wetlands, has complete natural systems, but in order to use natural regeneration systems, sewage treatment, and new, integrated wetland systems, have not been a major activity and only limited studies have been done on natural ecosystems of northern regions of Iran. On the other hand, disturbing phenomena such as the intrusion of various agricultural pesticides, the entry of toxic sewage and industrial wastewater, and other adverse factors have seriously threatened the potential of these systems.

Due to the regional and climate conditions and the existence of appropriate substrates for natural regeneration systems, it is necessary that more steps to be taken. In developing countries, many hospitals do not have sewage treatment plants and the hospital wastewater is often spilled to the urban sewage without any treatment. Urban wastewaters are finally discharged into rivers, lakes and oceans without advanced refinement. Moreover, the unmanaged sewage treatment is not risk free. The sludge and effluent produced in the process of hospital wastewater treatment can be a potential risk of transmission of infectious diseases and chemical threats. Therefore, it is necessary to have precise and correct information about the status of sewage discharge from hospitals and take necessary actions to prevent the penetration of untreated hospital wastewater into the environment and think about those approaches which can obstruct its leakage into surface and underground waters, because after water-pollution the disease spread among people. And it's urgently needed to build artificial wetlands for filtration of a variety of sewage and wastewater.

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