International Research Journal of Engineering and Technology (IRJET) e-ISSN: 2395-0056
Volume: 06 Issue: 06 | June 2019 www.irjet.net p-ISSN: 2395-0072

Ascertaining the Removal Efficiency of Lemna Minor in Froth Diminution by using Phytoremediation

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Abstract - *The frothing over the wastewater can be reduced by treating the wastewater with duckweed (Lemna minor). The L.minor is used as a phytoremediation agent over the wastewater to reduce the concentration of froth forming pollutants in water. The wastewater is taken in three tubs and the L.minor is inoculated onto the samples. The removal efficiency of the plant is determined for 1, 3, 5 and 7 days of treatment. The initial and final water samples after treatment were analyzed for Linear alkylbenzene sulphonate (LAS), Biochemical oxygen demand (BOD), Chemical oxygen demand (COD), Phosphate, Nitrates and the removal efficiency were found to be 96.39%, 98.466%, 98.499%, 69.485% for sample-1, 92.67%, 92%, 93.10%, 70.173%, 98.944% for sample-2 and 97.669%, 98.5%, 98.62%, 69.391%, 98.5% for sample-3. All the samples are kept for 7days of Hydraulic retention time (HRT). The dissolved oxygen (DO) of water is increasing with the detention period and the final concentration is between 6-8.5 mg/l for all samples. From the results, the pollutants uptake of L.minor is more. So, this can be used as a phytoremediation agent to reduce the concentration of froth forming pollutants in water.*

Key Words: Hydraulic retention time (HRT), *Lemna minor*, linear alkyl benzene sulphonate (LAS), Phytoremediation, Anionic surfactants, Duckweed

1. INTRODUCTION:

Water is the common liquid that covers about 71.4% on Earth's surface and it is a precious natural resource with the almost fixed quantum of availability. With continuous growth in the Country's population, the amount of wastewater is increasing but the availability of fresh water is decreasing. The continuous discharge of industrial effluents, untreated sewage water into the water bodies which contains a lot of surfactants, phosphates, etc..,

The froth is created when the surface tension of water is reduced and the air is mixed with it. The froth forming pollutants are nutrients (Nitrogen, Phosphorus and Carbon) and cations (Sodium, Potassium, and Magnesium). These pollutants come from Sewage, Agricultural runoff, fossil fuels, industry effluent, detergents, and other household wastes. The phosphorus in the water settles down as sediment when the high-intensity wind comes in pre-monsoon phase cause the water to churn and release phosphorous from sediments. During heavy rains, fresh water enters the lakes with force and stirs it, causing froth to build.

It causes breathing difficulties, irritation on the skin, besides spreading an unbearable stench. As the froth is toxic and it is carcinogenic. The water should be limited to have nutrients, heavy metals, etc. The problem can be avoided by Phytoremediation technique of using the living plants to clean up the water.

The *Lemna minor* belongs to duckweed family is used as a phytoremediation agent in this study. The *L.minor* have the capacity to reduce BOD, COD, Anionic surfactants, heavy metals, nutrients, etc. The macrophyte degrades the pollutants by breaking the complex molecules and incorporate them into their body this helps them to grow.

2. OBJECTIVES:

The objective is to evaluate the removal efficiency of *Lemna minor* in reducing the concentration of anionic surfactants in various study areas. The main objectives of this study are:

- To evaluate the suitability of *Lemna minor* in reducing the frothing and determining the sources of pollutants which are responsible for the froth in the wastewater.
- Finding the efficiency of the plant and comparing the removal efficiency in 0, 1, 3, 5, 7days of treatment.
- To compare the Growth with water quality parameters to know the uptake of the plant in all the samples.
- Assessing the removal mechanism of *Lemna minor* in all the three samples.

3. LITERATURE REVIEW:

3.1 Removal efficiencies of nutrients from wastewater treated with duckweed (*Lemna minor L.*)

Yagmur Uyasal and Oya Zeren (2003) examined the nutrient removal from wastewater treated with duckweed (*Lemna minor*) was evaluated utilizing a research facility scale pilot framework comprising of two ponds. The *Lemna*

minor was grown by supplying a laboratory prepared synthetic wastewater at the rates of 4000 mg/m^2/d COD, 3520 mg/m^2/d BOD_5, 740 mg/m^2/d NH_3-N and PO_4-P. The detention period was 3 and 6 days for pond1 and pond2 respectively. The water quality parameters were analyzed, gives COD, BOD_5, NH_3-N and PO_4-P, and the removal efficiencies were found to be 88.1%, 88.8%, 85.4% and 37.5% for Hydraulic retention time (HRT) of 3days, however 92.2%, 94.7%, 75.3% and 50.0% for HRT of 6days. From these outcomes, it is clear that duckweed is quite successful in removing nutrients from wastewater.

The use of macrophytes in wastewater treatment is very effective, especially in developing countries. These systems are relatively low in cost compared to conventional methods and can be used for the treatment of industrial wastewaters (e.g. food industry) without mechanical or chemical equipment. To maximize its yields the duckweed should be harvested daily. Compared to the traditional lagoons, these systems are free of mosquitoes and do not emit bad odors. Furthermore, these plants create a dense and green cover on the water surface, and can even be used for recreational purposes.

3.2 Inhibition effect of the anionic surfactant SDS on duckweed, *Lemna minor* with considerations of growth and accumulation

Neylan dirilgen and Nilsun since (1994) worked on the inhibition of growth and accumulation effects of sodium dodecyl sulfate (SDS) on *Lemna minor* L. were tested at l-300 ppm levels both in the full-strength and l/IO-diluted Jacob's supplement media under ceaseless light at 26-30° C for a 7-day test period. The growth of *Lemna minor L.* was increased by increasing concentrations of SDS between 1 and 40 ppm and inhibited thereafter, while no response was observed when the concentration reached a value of 100 ppm. On the contrary, the growth in the l/10-diluted medium was observed to be slightly enhanced up to concentrations of 40 ppm, but the SDS concentration between 40-80 ppm experiencing inhibition of growth, thereafter. The SDS concentration between 1-90 ppm the accumulation of SDS by *Lemna minor* L. (expressed on a dry weight basis) increased with increasing concentrations of SDS, both in l/10-diluted media and full-strength. The linear and nonlinear regression analysis performed to determine the correlation of increasing SDS concentrations with growth and accumulation. The statistical significance of observed differences in full- strength and diluted media were also investigated. The SDS concentration in the range of 1-90ppm subjected to increment in pH of both full-strength and diluted media during the experiment period later it remains constant at higher SDS concentration. This tells that when SDS subjected to biodegradation it results in increment in pH of the medium.

4. METHODOLOGY:

4.1 Frothing:

Frothing is nothing but foaming. This is treated as pollution. There is no single reason for frothing. Following are multiple reasons to form a froth on the water like Sediments of phosphate, surfactants, filamentous bacteria, Methane gas, etc..,

1. Filamentous bacteria: They are due to fats, oils, and grease. These bacteria make wastewater to form froth. The wastewater with this bacteria makes froth into brown color.

2. Altitude: The phosphate in wastewater deposited as sediments when the water flows from upstream to downstream because of the altitude the froth occurs.

3. High BOD and methane: Decomposition of algae, fish& macrophytes leads to increment in biological oxygen demand (BOD) of the water body. These organic compounds were released into water act as bio surfactants which can be degradable over a time.

4. Phosphate Sediments: When the sewage from houses is dumped without any treatment contains a high amount of phosphate they get deposited over time. The sediments of phosphorous get raised when the high intensity of rain hit the wastewater or when an upstream contains high altitude. The water gets churned and those sediments get mixed which reduces the surface tension. The less surface tension leads to frothing or foaming over the water.

5. Synthetic detergents: which are made up of surfactants are the main reason for the frothing. Few surfactants are not biodegradable.

6**.** Fats, oils, grease, bio surfactants which comes from residential and industrial produce foam.

4.2 *Lemna minor* Macrophyte*:*

Specie outline:

They belong to the family of Lemnaceae.

- L.minor is a macrophyte with one, two or four leaves each having a single root. The root is 1-2cm long, leaves 1-8 mm long and 0.6-5 mm broad, light green in color with three veins and small air spaces to assist flotation.
- This belongs to the family of duckweed, Contains very small flowers. L.minor has higher growth at a temperature between 6 to 33 degrees.
- Duckweed has higher growth, it up takes more nutrients, can be handled easily, high protein content and low fiber content.
- Optimal growth at pH between 6.5 to 8.

- Conductivity > 2000 µS/cm and salinity >1517 mg/l inhabit plant growth.
- Lemna minor is more tolerant to SDS than Azolla.

Mechanism of *Lemna minor*:

The O_2 produced during photosynthesis is conveyed through duckweed roots into the water and supports the formation of the 10cm thick aerobic layer in the root zone. This layer favors oxidation of rising gases (reduces anaerobic fermentation of organic matter) thus preventing odor diffusion.

The Lemna minor is a floating plant which is made out of D-galacturonic corrosive (64%), galactose, arabinose, xylose, and D-apiose. Polysaccharides may also consist of arabinose or xylose which treats the wastewater. Pectin is a polymer of galacturonic acid present in this macrophytes removes the heavy metals in water.

4.3 Samples and their Locations:

The wastewater samples are tested to measure water quality. Samples collected 15cm below the surface of the wastewater stream and tested within 12hrs.

Sample-1: The sewage coming from upstream contains households and small scale industries. The frothing intensity of this stream is very less at the starting but day by day it is increasing.

Location: Vasant Vihar, Vizianagaram

Sample-2: The pond where the sample collected is totally eutrophicated. The surface is covered with macrophytes. The nearby houses sewage water is entered into this pond.

Location: Vasanth Vihar, Vizianagaram

Sample-3: This Lake is severely polluted with animal waste, industrial waste. The partially or untreated sewage is diverted into this lake which is a major cause for frothing of this lake. Pig waste is dumping into this lake which is increasing the pollution load of the lake. The PCB of Telangana is striving to eliminate the dumping of untreated sewage into the water body.

Location: Ramakrishna Puram Lake (RK Puram Lake, Kukatpally, Hyderabad

4.4 Process of treatment:

The treatment process starts with the collection of macrophytes and ends with the testing of the wastewater after 1, 3, 5, 7days of treatment. The detailed steps of treatment are as follows:

1. Collection of Lemna minor plants from a nearby pond. To avoid any changes in the treatment the plants should be washed with distilled water or potassium permanganate.
2. Then samples collected from all the three sources are poured into an empty tub.
3. Growth of the plant should be measured every time before testing. The plant's weight should be measured every time.
4. After measuring the initial weight of the plant. They are inoculated onto the samples.
5. The testing for 1, 3, 5, 7 days should be done to determine the efficiency of the plant.
6. The weight of the plant is double in the 2nd day of treatment and pollutants concentration is decreased.
7. After testing for water quality the removal efficiency of the plant is determined.
8. The parameters are compared to know the uptake and removal efficiency of the plant.

5. RESULTS AND DISCUSSION:

The Samples were collected from regions where frothing is visible. The frothing is due to many reasons but in this study the ability of *Lemna minor* in treating the surfactants is determined. The removal efficiency of that macrophytes is determined for every sample.

5.1 Growth rate determination:

The weight of plants before and after the treatment is measured to know their uptake with respect to the pollutants.

Table 1: Growth of L.minor in dry weight basis

Dry weight (grams)			
Period	Sample-1	Sample-2	Sample-3
0day	8.75	8.75	8.75
1day	8.995	9.17	9.1
3days	17.357	18.62	18.2
5days	22.96	21.4	23.52
7days	23.24	22.685	23.87

Anionic surfactants determination:

The anionic surfactants determined by using Methylene blue activated substances (Standard Methods for the Examination of Water and Wastewater, 1998)

Calibration:

Table 2: Absorbance results for varying concentrations of MBAS

MBAS Concentration(mg/l)	Absorbance
0	0
20	0.197
40	0.264

60	0.35
80	0.422
100	0.55

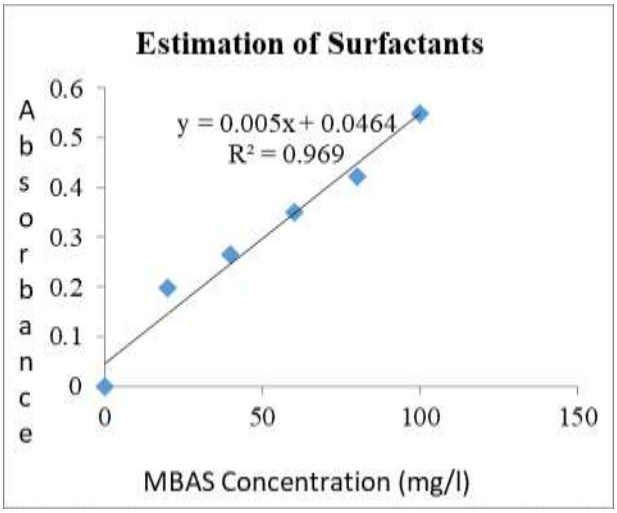

Chart 1: Calibration curve for absorbance and concentration

5.2 Analysis and Discussion:

The wastewater is increasing day by day this leads to pollution of the environment. As we can see the results they show that there is an increase in percentage removal and decrease in surfactant concentration.

Analysis of results of all samples:

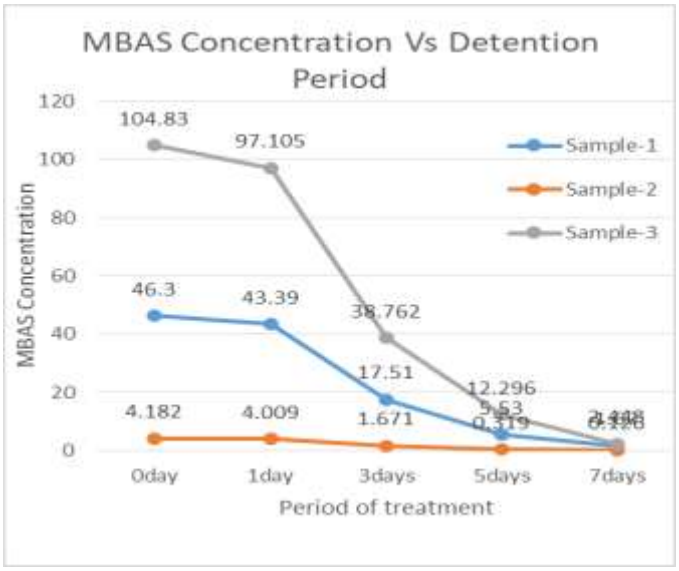

Chart 2: MBAS Concentration variation with Detention period

The results show that the *Lemna minor* removal efficiency increases with the concentration of anionic surfactant. As the sample-2 is pond water which contains very less amount of surfactant concentration. The pond water is mostly eutrophicated contains macrophytes. So, the LAS compound is biologically degradable, the concentration reduces over time. That's why the pond water has very less concentration. The pond water contains very less concentration of 4.182 mg/l MBAS as LAS. The removal efficiency of 4.136%, 60.047%, 86.67%, 92.67% achieved for 1, 3, 5 and 7 days of treatment with *Lemna minor*. Raw domestic wastewater has surfactant concentration from 1 to 20mg/l with approximately 60% are of anionic type (APHA 1989).

The wastewater collected from a stream in Vizianagaram (Sample-1) has about 46.3mg/l surfactant concentration. The *Lemna minor* shows a good response to that water. Most of the treatment is completed at the detention period of 3 days. After 3days the growth is also increasing and percentage removal of pollutant is also increasing. The removal efficiency of 6.28%, 62.18%, 88.05%, 96.39% for 1, 3, 5, 7days.

The sample-3 has very high concentration compared to all the three samples which are of 104.83 mg/l as LAS. The concentration of anionic surfactant inversely proportional to removal efficiency. The removal efficiency achieved for sample-3 of 7.3695%, 63.023%, 88.27%, 97.669% for 1, 3, 5, 7days.The macrophytes remove the maximum amount within 3days of treatment. When compared to 5th and 7th day there is a slight difference. All the three samples which are treated with *Lemna minor* for about 7days removed maximum contaminant in the wastewater. That water can be reused for irrigation purpose. The toxic levels of LAS to aquatic organisms Is in range of 0.1 to 8.2 ppm. The macrophytes can be used for treating or removing the anionic surfactants concentration. Because they remove below the permissible limit of surfactant concentration in wastewater. For the drinking water, the allowed surfactant concentration of 0.2 to 1 mg/l. So, this can be used as drinking water after disinfection.

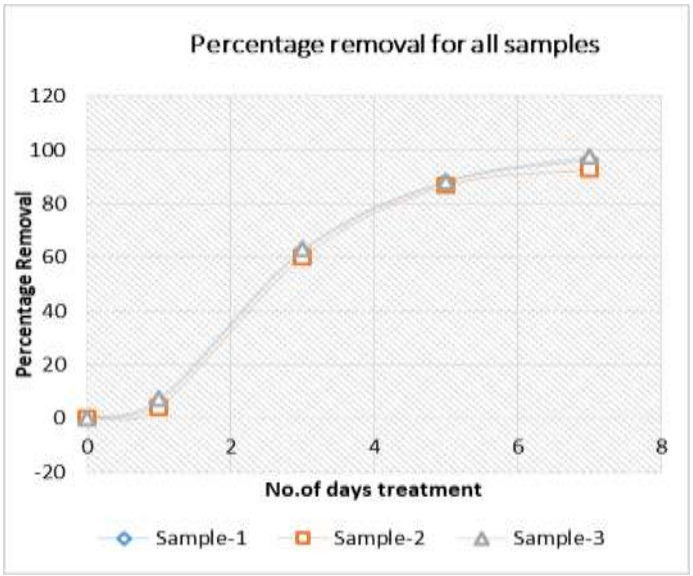

Chart 3: Percentage of removal for all samples

Above graph depicts that, Percentage removal for sample-3 is high at 7days of treatment. All the samples show a similar response to *Lemna minor*. The removal efficiency increases linearly with the days of treatment.

Table 3: Percentage removal for water quality parameters

Sample	Parameter	1day	3days	5days	7days
Sample-1	COD	11.552	89.001	91.251	98.449
	BOD	9.211	87.241	98.326	98.466
	Nitrates	6.250	81.000	98.125	98.250
	Phosphates	8.247	37.320	51.340	69.485
Sample-2	COD	9.290	88.200	92.200	93.100
	BOD	8.327	88.800	91.800	92.000
	Nitrates	7.193	83.133	98.550	98.944
	Phosphates	8.242	38.148	53.148	70.173
Sample-3	COD	12.012	90.203	90.424	98.622
	BOD	9.205	88.030	98.413	98.500
	Nitrates	7.050	81.250	98.460	98.500
	Phosphates	8.261	38.087	52.435	69.391

From the results, all the sample are suitable for biological treatment as their BOD/COD is greater than 0.3. So, they can be easily degradable. The Macrophyte selected to remove anionic surfactants is worked well in removing the organic matter in the water which has removal efficiency of about 92 - 98% in all the samples. When the organic matter decreases there is a chance in the increment of oxygen levels in the water.

The dissolved oxygen levels are raised which shows that the duckweed we used is good at removing the organic matter. The graphs demonstrate that the nutrient-like Nitrate is removed greater than 95%. There is a less removal efficiency observed in phosphate in all samples of about 67 to 69% which is very less compared to all the other parameters. The Sample-2 which is wastewater has a high amount of nutrient content this is because the pond water is totally eutrophicated.

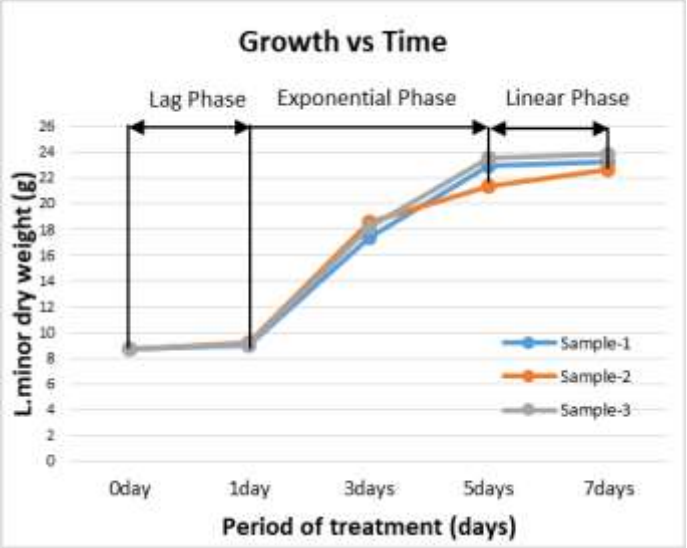

Chart 4: Growth of Lemna minor with the days of treatment

The above graph is about how *L.minor* growth is present in all the samples. As we can see there are three phases in the growth: Lag phase, exponential phase, linear phase. Lag phase occurs at the starting and last, in a day, the growth in all sample is very less. This occurs because, during this phase, the organism prepares to grow. Unseen biochemical changes like cell separation and differentiation of tissues occur during this time. From all the samples the L.minor adapt easily in pond water (sample-2) that's why the weight got increased well within a day.

During the exponential growth phase, there is an increment in the growth of the plant. In this phase the plants grow well, producing new fronds and they also divide rapidly to take advantage of fresh medium. The *L.minor* in sample-2 shows that it adapts easily to that environment so the growth is little faster when compared to all other samples. The macrophyte has more growth in this phase. The sample-1 and sample-2 show good increment because they contain a high amount of nitrates, phosphates.

The nutrients concentration in the medium decreases because the plants uptake them and grow in numbers. So, the Linear phase starts after the exponential phase. During this phase growth is slow but the death rate increases. As a result, the initiation of new fronds and the senescence and death of old ones start to come into equilibrium. For duckweeds, this phase typically occurs as nutrients content reduces, that low concentration leads to a reduction in the growth. The macrophyte in all the samples shows little change in dry weight.

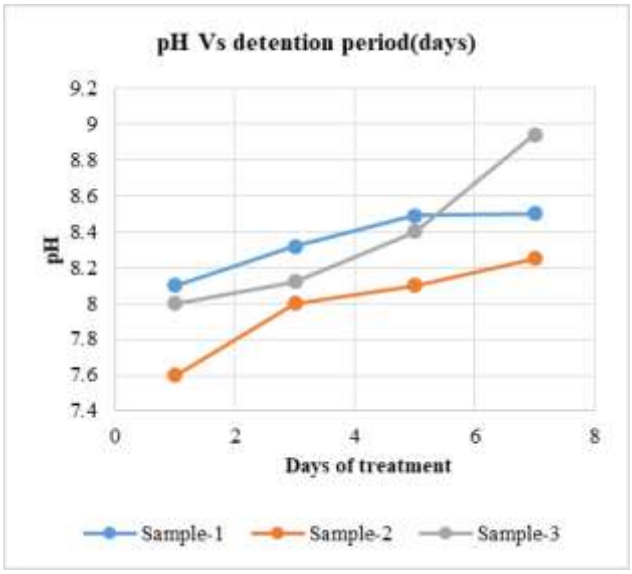

Chart 5: pH variation with the detention period for all samples

As the detention period increases the pH of the water is becoming alkaline this is a drawback but that can be leveled by the addition of some other lower pH chemicals with the water after the treatment. The *Lemna minor* has good removal efficiency achieved within 5days of treatment. But the seven days of treatment is suggested to reduce the concentration of pollutants.

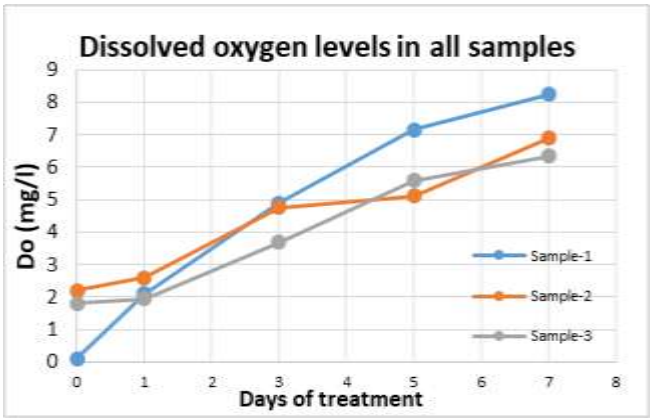

Chart 6: DO Vs Hydraulic retention time for all samples

Initially, the dissolved oxygen is high for pond water but later on, the oxygen level is increasing with the detention period. The Sample-1 DO concentration is very high after 5, 7days is 7.15, 8.25 mg/l. Finally, after the 5days of treatment the DO level is going to 5-7mg/l for all samples which is a safe concentration for aquatic life when disposed into water bodies

6. CONCLUSION:

- The Anionic surfactants are the major reason for frothing in wastewater. The wastewater, when treated with *Lemna minor*, has removal efficiency of 92-97% for Anionic surfactants as MBAS.
- The macrophyte also increasing the dissolved oxygen levels in the water. For the Hydraulic retention time (HRT) of 7 days the DO concentration in sample-1, sample-2, sample-3 is 8.25, 6.9, 6.35 mg/l.
- The phosphate is also one of the major reason for frothing in wastewater but the *Lemna minor* capacity in removing the phosphate is low when compared to other parameters of about 67-70%.
- The macrophyte worked well in reducing the BOD, COD, and Nitrate. The nutrient uptake is high for 3days treatment because the growth of the plant has doubled in 2 days and that is the reason why the removal efficiency also increased when tested for 3 days of treatment.
- The pH of water becoming alkaline (pH>7) when subjected to long detention periods that can be reduced by the addition of lower pH chemicals.
- By analzsing all the results, the *Lemna minor* has very good removal efficiency. So, this macrophyte can be used to remove froth forming pollutants in water.

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