PHYTOREMEDIATION OF WASTEWATER WITH *THALIA GENICULATA* IN CONSTRUCTED WETLANDS: BASIC POLLUTANTS DISTRIBUTION

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ABSTRACT

Constructed wetlands (CW) are efficient wastewater treatment technologies with low energy consumption. A constructed wetland with horizontal sub-surface flow was designed at a pilot scale involving Thalia geniculate as vegetation, with a wastewater loading rate of 204±66 L/day, using gravel as inert medium with a porosity and density of n= 56.3 ± 3.5 and 1666.7 ± 119.3 kg/m3, respectively. The reactor allows the biological treatment of 0.85 ± 0.05 and 0.66 ± 0.05 m3 of wastewater, with 4.2 days as a hydraulic retention time, favoring the removal of 85% of the average values of BOD with a k of -0.43 days-1. The pollutant analysis showed a pH value of 7.5 ± 0.1 in the reactor. The temperature (30.44 to 28.32°C), the electrical conductivity (4010 to 2922 µS/cm), the turbidity (144 to 17 UTN) and the bacterial biomass (30000 to 2646 mg/kg) decreased substantially from inlet to oulet across the reactor. The efficiency of the wastewater treatment in the CW is notable, nevertheless, keeping the appropiate hydraulic retention time is important in order to fully comply with the maximum permissible limits of 30 mg/L established in the Mexican environmental legislation (NOM-001-SEMARNAT-1996).

Keywords: wastewater treatment, macrophytes, subsurface flow, pollutant removal efficiency.

1 INTRODUCTION

Constructed wetlands (CW) is not a new technology around the world. They have been studied because of its efficiency to remove organic matter through microbial degradation and settling of colloidal particles, pathogen elimination in domestic water and alternatives in construction design of wastwaster treatment [1]–[7]. Nevertheless, in Mexico and Latin America the applications of CW technology has been incipient, despite the technology has demonstrated the versatility of applications in small and medium urban areas, easy installation, operation and maintance, with highly competitive costs [8]. The most common type of constructed wetlands are the free water surface constructed wetlands (FWS-CW) and the horizontal subsurface flow constructed wetlands (HF-CW).

In this context, it is estimated that CW plants require two or three growing seasons to achieve the maxium removal effiency [9]. The vegetation in the CW is important because the pollutant removal through direct assimilation into their tissues provides an adequate medium for microbial activity through the transport of oxygen to the rhizosphere, stimulating the aerobic degradation of organic matter and nitrifying bacteria growth [10], [11]. It has also been shown that the efficiency removal of contaminants depends on the support material and the hydraulic retention time (HRT) [12]. The communities interested in adopting this technology must develop CW based on local parameters [13], since the design can be influenced by hydrometeorological factors, which must be taken into account for the operation of the system [14], [15].

In an experimental study with vertical flow constructed wetlands (VF-CW) a bacterial culture of fungi and actinomycetes was used [4]. It was concluded that microorganisms play a key factor in the decontamination process and can be an indicator in the removal of

WIT Transactions on Ecology and the Environment, Vol 228,
www.witpress.com, ISSN 1743-3541 (on-line)
doi:10.2495/WP180071

chemical oxygen demand (COD), biochemical oxygen demand (BOD), total nitrogen (TN), total phosphorus (TP) and total suspended solids (TSS). A train of treatment was also evaluated, consisting in three CW connected in series involving a sub-superficial flow (HF CW), using vegetation such as reed (*Scirpus americanus*), enea (*Typha latifolia*) and water lilies (*Eichhornia crassipes*). The system was stabilized in 44 days, having an HRT of 15 days in each CW, reporting high removal efficiencies of COD (71%), calcium (91%), chloride (77%), nitrite (82%), ammonium (99.9%), phosphate (77%) except for nitrate ion (36%) and electrical conductivity (93%) [9].

The effect on the removal of ammoniacal nitrogen in contaminated water was also studied from the Erh-Ren River in southeastern Taiwan [16]. The evaluation was performed through an experimental control and coupled HF and FWS, CW systems connected in series. Wetland vegetation such as Chinese grass (*Pennisetum alopecuroides L.*) and Pacific Island grass (*Miscanthus floridulus*) were involved in a HF-CW and FWS-CW respectively. These species did not survive to the winter due to the low temperature and the high salinity of the water, due to the salt intrusion from the sea water into the river. As a consequence, common reed (*Phragmites australis*) in both CW systems was shown, with an initial density of 2 plants per m^2 and growing around 100 plants per m^2 after 3 months. The control system during the experiment worked without species, concluding that season time affected the CW performance, particularly for the removal of ammoniacal nitrogen.

Mexico has 110 CW systems throughout the country, three of them in the state of Tabasco, and only one operates in the Municipality of Centro at 64% of its capacity, because it was designed to treat a flow of 0.125 m^3/s and currently operates with a flow of 0.080 m^3/s [17]. In Tabasco-Mexico, the coverage of water treatment is low since 60 out of 93 wastewater treatment plants are known for their defficient performance operation. There is a notable predominance of conventional technologies and primary wastewater system, whose treatments are inefficient, and the costs have not yielded the expected results.

In this respect, it is necessary to look for economic treatments that are easy to operate and appropriate for the climatic conditions and natural resources of the region, presenting technical and economical advantages over chemical treatment methods [17]. The objective of the current research work was to evaluate the phytoremediation potential of wastewater with *Thalia geniculata* in a CW, analizing the basic pollutan distribution and degradation at a pilot scale. CWs built in Tabasco consider HF and FWS system operated mainly with introduced species, such as *Thypa latifolia*. In the case of *Thalia geniculata*, it is also a native specie from Tabasco, but not studied so far and present advantages from introduced species in CW systems.

2 MATERIALS AND METHODS

2.1 Location of the pilot-scale CW

The experimental HW-CW was installed at the Division Academica de Ciencias Biologicas (DACBiol), which is a campus from the Universidad Juarez Autonoma de Tabasco. The vegetation was collected in swampy areas from the Municipality of Centro, Tabasco.

2.2 CW design characteristics

The reactor is 2.5 m long x 1.2 m wide x 1 m high [18]. The preparation began with the cleaning of the corrosion areas using an anticorrosive primer (white enamel finish layer) was applied in the external part. In the internal part, waterproofing of acrylic paste based on resins and mineral fillers (1 cm thick) was applied. Afterwards, an elastomeric waterproofing layer was applied with a textile fiber reinforcing the internal part to avoid possible filtrations (five layers were placed). Once the reactor was waterproofed, all accessories, 1-inch hydraulic polyvinylide (PVC) pipes and fittings (valves, elbows, T's, connectors, etc.) were installed for the supply and distribution of the wastewater. For natural aeration, internal sampling points were placed. Finally, 50 cm of mixed gravel was placed in the reactor, and then proceed for the stabilization phase of vegetation.

2.3 Planting and stabilization of vegetation

The vegetation was placed into the gravel support medium. The stem size on the surface was 10 cm long and roots were placed 15 cm below the surface [8]. The reactor was fed with clean water at the beginning, maintaining a level of 40 cm of water for stabilization of the vegetation [19]. Thereafter, wastewater from the carcamus of the DACBiol was added to the CW. The stabilization phase in CW lasted six months from February to July 2016.

2.4 Hydraulic retention time, removal efficiency and degradation rate

In the reactor, a mixed gravel support medium (crushed rock from the Teapa River, southern region of Tabasco) was placed and the hydraulic retention time (HRT) was calculated with the operation flow of the wastewater [8]

$$HRT = n\ d\ A / Q, \tag{1}$$

where n is the porosity, d is the heigh of the support medium, A is the cross section of the reactor and Q is the water flowrate.

$$\eta = [(C_1 - C_2) / C_1] \text{ x } 100, \tag{2}$$

where η represents the removal efficiency in %, C_1 the wastewater influent concentrationand C_2 the wastewater effluent concentration.

The behavior of wastewater is a first order kinetic reaction, the degradation rate k was estimated with the following eqn (3).

$$K_o = - \ln (Cn / C_o) / \tau, \tag{3}$$

where τ = retention time for BOD removal, Cn = BOD influent concentration of the reactor "n" (mg/L), Co = influent concentration, K_o = degradation constant.

2.5 Wastewater characterization

In order to study the variables of the wastewater, sampling points were established and sampled throughout the reactor (Table 1, Fig. 1). Three samples were taken by triplicate per day during 5 days. For the kinetic study of the BOD, the influent and effluent of the reactor was monitored, taking a simple daily sample for 7 days, and up to one year of operation in the reactor.

Table 1: Wastewater methods of analysis applied for control parameter determinations.

Parameter	Environmental regulation in Mexico
Temperature	NMX-AA-007-SCFI-2000
Turbidity	NMX-AA-038-SCFI-2001
Electrical conductivity	NMX-AA-093-SCFI-2000
pH	NMX-AA-008-SCFI-2000
Biological oxygen demand	NMX-AA-028-SCFI-2001
Total volatile solids	NMX-AA-034-SCFI-2001

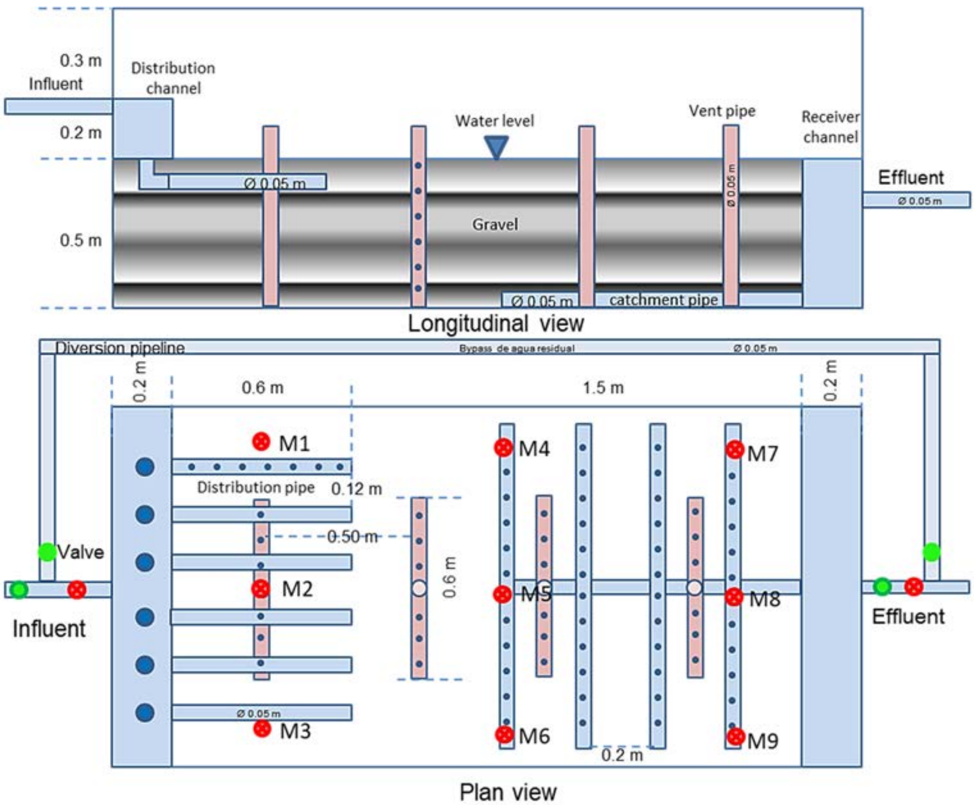

Figure 1: The red points from M1 to M9 indicate the sampling point in the Subsurface Horizontal Flow – Constructed Wetland (HF-CW).

2.6 Biomass on the support medium

The biomass, refering to the quantity of microorganisms on the rocks was determined by gravimetry adapting the total volatile solids (SVT) method to a sample of the support medium at each sampling point (Fig. 1). Each sample considers the density and porosity of the system [21].

2.7 Modeling of the pollutants distribution

To achieve the modeling, the daily average of each sampling point referring to variables such as temperature, turbidity, electric conductivity, pH and biomass were monitored and analyzed. The pollutants distribution inside the reactor was plotted using the software Surfer 8.0 [22], which allows the determination of the spatial distribution within a coordinate system based on a linear interpolation and a quadratic diagram (isoconcentration map).

3 RESULTS AND DISCUSSION

3.1 Hydraulic retention time (HRT)

The HF-CW was designed to operate with 200 L/day. However, when performing the corresponding gaugings and the volumetry of the wastewater flowrate, the HRT was estimated in 4.2 days. This value fulfilled the design criteria for CW established by several authors [8], [13], [15], [19] Fig. 2 shows the HRT at different operating wastewater flowrates. The average operating flow in this period was 204 ± 66 L/day, with 108 L/day and 370 L/day, as minimum and maxium operation wastewater flow values, respectively.

3.2 Degradation rates and kinetic coefficient

When the wastewater flowrate exceeded 200 L/d, the HRT was observed to decrease. Therefore, the wastewater did not have enough time to be in contact with the microorganisms and vegetation, resulting in a low degradation. In order to analyse the influence of design parameters (loading rate, flow and temperature) on the degradation of pollutants in the wastewater, the removal of BOD was measured. It is important to note that facultative microorganisms eliminate part of the BOD through biological and physic process, mainly. The pollution removal rate in wastewater is related to HRT and temperature. The temperature

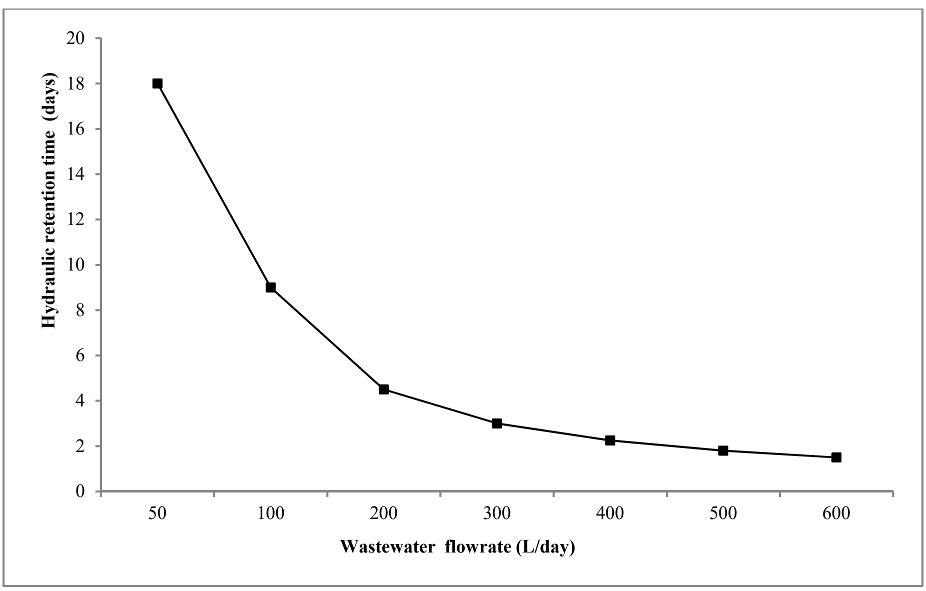

Figure 2: HRT for different operating wastewater flowrates in the HF-CW.

during the evaluation was 27°C on average. The HRT was 4.5 days and *k* was calculated in -0.43 days^{-1}. The Mexican water regulations established a daily discharge average of 75 mg/L for the HRT parameter. This value can be achieved between the fourth and fifth day of operation, as shown in Fig. 3 [23].

The HF-CW (HRT = 6 days) removed nearly 90% of the organic pollutants in the wastewater and comply with the most stringent criteria established by the Mexican regulations for the aquatic life protection. Similar results were reported elsewhere [24], concluding that 8 days of HRT is adequate for the removal of organic matter at temperatures above 25°C. The kinetic degradation behavior of the organic matter in the HF-CW is described as a first order kinetic (Table 3).

3.3 BOD removal efficiency

The maximum BOD removal efficiency was 92.8%. The average removal efficiency was found to be between 80 and 85%. Unlike the operation of other experimental reactors in series [19], [25], the current reactor is setup with a primary and secondary treatment, satisfying the regulations for wastewater discharge to rivers for urban public use (75 mg/L). Furthermore, it complies occasionally to the aquatic life protection limit (30 mg/L) [23].

3.4 Spatial distribution of pollutants in the HF-CW

The basic parameters of pollutants monitored in wastewater for the spatial distribution analysis can be seen in Table 3.

Regarding the pH values, some differences were observed in the spatial distribution, but the values remained in the neutral range of 7.2–7.9 (Fig. 5). This variation may be explained by the type of substrate and biofilm employed in this investigation [25]. The temperature was 29.2 ± 0.8°C on average, with variation from the input to the output of the reactor with 30.5 to 28.1°C, respectively (Fig. 6). This temperature behaviour favors the growth and the stabilization of mesophilic microorganisms [26]. The electrical conductivity values were measured around 3442.7 ± 408.0 µS/cm, in compliance to the regulation for agricultural

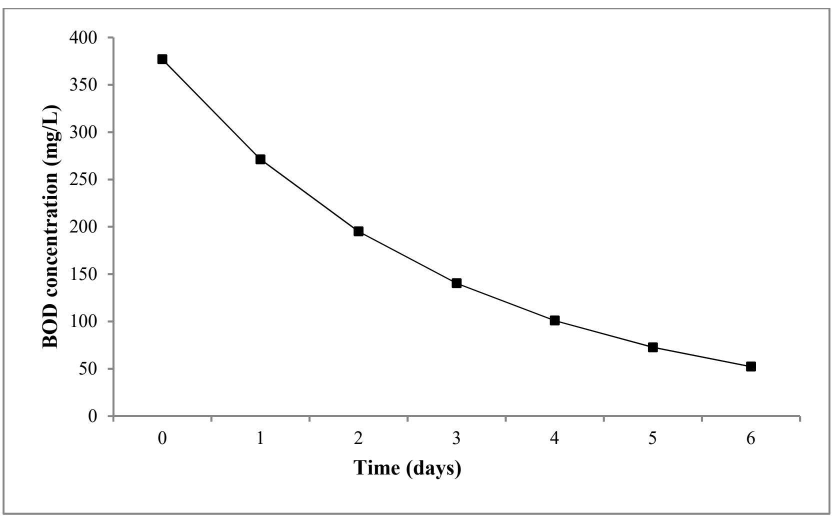

Figure 3: Degradation of organic matter in the HF-CW with *Thalia geniculata* vegetation.

Table 2: Estimation of the kinetic degradation constants evaluated in the HF-CW with *Thalia geniculata* vegetation.

Days	BOD influent (mg/L)	BOD effluent (mg/L)	k (d^{-1})	η (%)
1	375.50	66.20	-0.39	82.4
2	369.40	65.30	-0.39	82.3
3	403.20	66.50	-0.40	82.5
4	437.10	65.90	-0.42	84.9
5	391.70	43.30	-0.49	88.9
6	407.70	29.50	-0.58	92.8
7	254.20	50.50	-0.36	80.1
Average	376.97	55.31	-0.43	85.3

Table 3: Basic pollutant parameters measured in wastewater for the HF-CW.

Days	Parameters	M1	M2	M3	M4	M5	M6	M7	M8	M9
1	pH	7.9	7.7	7.5	7.8	7.6	7.5	7.8	7.4	7.4
	Temperature (°C)	32.3	32.2	31.2	29.4	30.9	31.3	29.6	29.5	29.5
	Conductivity (µS/cm)	4160	4010	3770	3450	3640	3280	2760	2870	2530
	Turbidity (NTU)	226	176	176	165	171	110	23.1	17.6	6.9
2	pH	7.5	7.5	7.5	7.6	7.5	7.5	7.5	7.4	7.2
	Temperature (°C)	30.5	30.8	29.7	27.2	27.5	26.7	26.7	25.1	25.1
	Conductivity (µS/cm)	4010	3680	3890	3070	2370	2950	2300	2240	2520
	Turbidity (NTU)	195	196	178	103	95	90	89	72	68
3	pH	7.4	7.4	7.4	7.6	7.6	7.6	7.6	7.6	7.7
	Temperature (°C)	30.0	29.3	29.6	29.7	29.1	29.1	28.8	28.3	29
	Conductivity (µS/cm)	3950	3920	3920	2710	4020	3840	4000	3740	3640
	Turbidity (NTU)	42.0	42.6	42.6	12.4	12.5	8.71	3.64	2.8	2.1
4	pH	7.3	7.5	7.8	7.4	7.5	7.8	7.4	7.6	7.8
	Temperature (°C)	29.9	29.2	29.5	29.8	29.3	29.4	29.6	29.1	29.4
	Conductivity (µS/cm)	3930	3930	3940	3940	4080	4000	3910	4080	3650
	Turbidity (NTU)	38.7	35.8	39.7	10.7	10.7	11.5	2.95	3.49	3.67
5	pH	7.3	7.3	7.3	7.7	7.7	7.7	7.7	7.6	7.6
	Temperature (°C)	29.5	29.2	30.2	28.7	28.9	29.1	28.8	28.8	28.6
	Conductivity (µS/cm)	4000	4020	3860	3190	3190	3040	2300	2350	2270
	Turbidity (NTU)	220	227	210	81	83	76	3.1	2.57	6.73

irrigation in Mexico. Salinity is considered low, with values from 4000 to 2900 µS/cm (Fig. 7). The HF-CW reduced the salinity of the wastewater; thus water is suitable to be used without restriction for crops irrigation of [27].

Turbidity shows a significant decrease from influent to effluent in the HF-CW. This behavior is associated with *Thalia geniculata* vegetation, which is very effective at removing up to 87% of the sediments (Fig. 8). Finally, the biomass adhered to the support medium presented the highest concentration of microorganisms at the reactor inlet and decreasing significantly at the reactor outlet. The biomass concentration was found to be higher than 33,000 mg/kg (mg of biomass on kg of rock “medium support”) in the influent and reaching values of 3000 mg/kg at the end of the reactor. The support medium has a diameter of 2.8 ± 0.8 cm, porosity n = 56.3 ± 3.5 and density of 1666.7 ± 119.3 kg/m^3. These characteristics allow a water volume of 0.85 ± 0.05 m^3 and 0.66 ± 0.05 m^3 of gravel within the HF-CW.

A HF-CW experimental study reported BOD removals greater than 90% with *Typha* and *Phragmites* vegetation [28]; while a removal efficiency of 79% was observed with *Typha latifolia* for combined HF reactors [29]. Similar removal efficiencies for BOD (80%) were reported with *Typha* and *Phragmites* in the second year of evaluation of an HF-CW [30]. In this study, *Thalia genicualata* vegetation showed a high removal efficiency of BOD (85%). The BOD removal in a CW is generally high because the organic components are degraded aerobically and anaerobically by the bacteria adhered to the roots and rhizomes of the plants with the porous medium [31]. The water treatment in a HF-CW with a HRT of 6 days and temperatures around –28°C is sufficient to obtain removals greater than 90%. Similar results were concluded for 8 days of HRT for the removal of organic matter at temperatures above 15°C [24]. *Thalia geniculata* is not frecuently cited in the literature, however in this study it is demonstratred the great potential for wastewater treatment from this native vegetation in the tropical region of Mexico. The most important effects of emerging macrophytes in wastewater treatment are plant tissue, wind speed reduction that supports the sedimentation of suspended solids, filtering effect or adherence of microorganisms to the roots plants that can be a significant route for the elimination of nutrients, especially under low loading rates [32]. Therefore, *Thalia geniculata* is highly efficient for the removal of basic pollutants in the treatment of domestic wastewater.

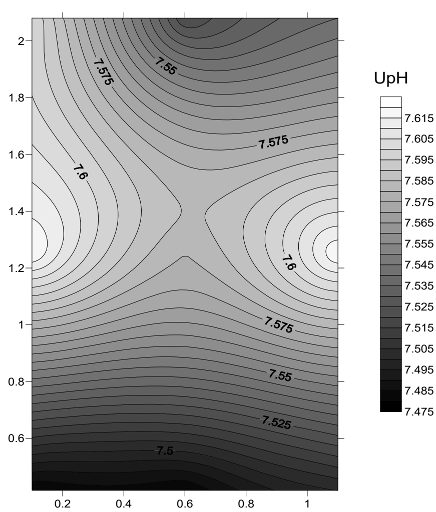

Figure 4: pH spatial distribution.

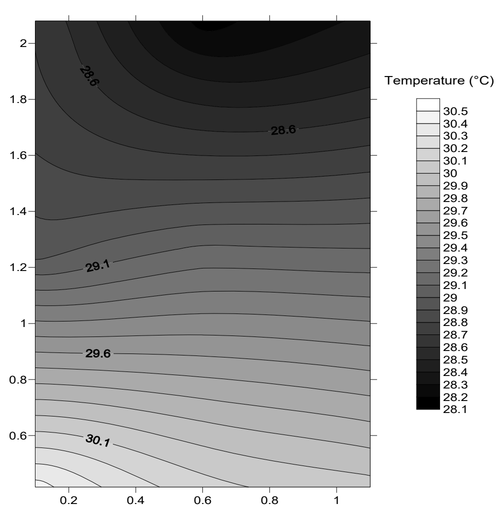

Figure 5: Temperature spatial distribution.

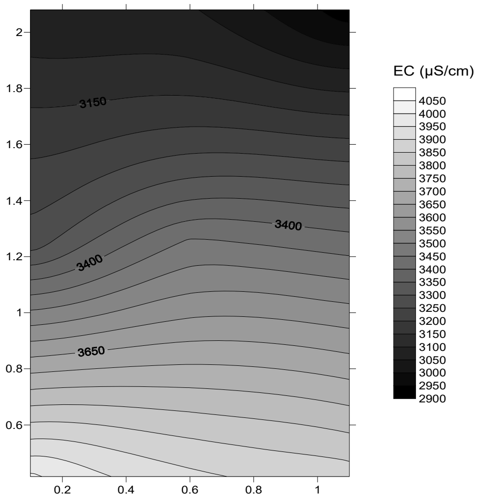

Figure 6: Electrical conductivity spatial distribution.

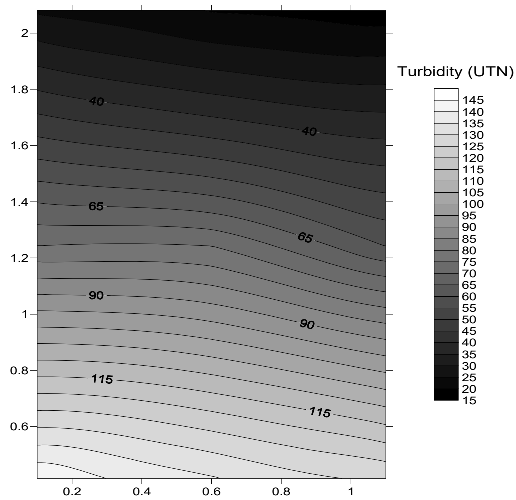

Figure 7: Turbidity spatial distribution.

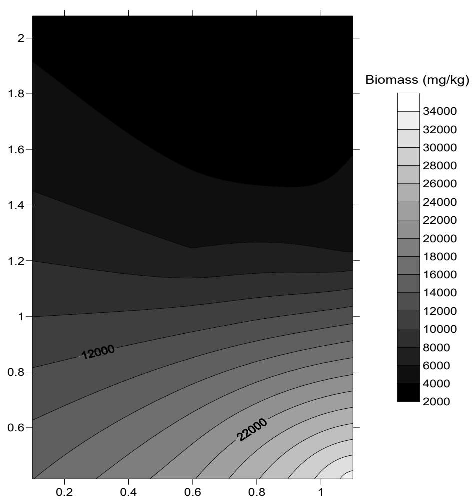

Figure 8: Biomass spatial distribution.

4 CONCLUSIONS

Thalia geniculata vegetation was found to be quite efficient for wastewater treatment in a subsurface flow constructed wetland (HF-CW) with 85% of BOD removal. The daily average temperature (28°C) showed a k of -0.43 days^{-1}, promoting the pollutant removal, similarly as a secondary water treatment. The support medium influenced significantly the microorganism's fixation, meaning that the mixed gravel employed was appropriate for the treatment.

From the operation standpoint, the experimental reactor attained the highest removal efficiencies of basic contaminants when the HRT ranged from 4.2 to 6 days. The experimental design of the HF-CW proposed in this research complied with the environmental regulation for water standards in Mexico (75 mg/L).

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