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Biodegradation of diesel fuel-contaminated wastewater using a three-phase fluidized bed reactor

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Abstract

Aerobic biodegradation of diesel fuel (DF)-contaminated wastewater is carried out in a three-phase fluidized bed reactor under unsteady and steady state conditions. The solid phase lava rock particles, which act as the support for the biomass, are fluidized by the upward flows of influent wastewater, and air. The results show that the reactor under unsteady state operation achieved 100% DF removal from synthetic wastewater loaded with 0.43–1.03 kg/m^3 day of DF. An average of over 97% of the influent chemical oxygen demand (COD) was also removed from the wastewater with COD concentrations in the range, 547–4025 mg/L. For influent COD concentrations up to 1345 mg/L, the removal is greater than 90%. Under steady state operation, the reactor was able to remove 100% of the DF, and an average of 96% of the COD from the wastewater. It had approximately 200 mg/L of DF, and 1237 mg/L of COD at a low hydraulic residence time of 4 h. In general, the results demonstrate that the reactor is very efficient, and requires short residence times to remove both DF and COD from heavily contaminated wastewater.

Keywords: Diesel fuel removal; Wastewater biodegradation; Aerobic fluidized bed reactor

1. Introduction

Accidental release of petroleum products, particularly DFs, into the environment is a widespread problem as two billion tons of petroleum gets refined each year worldwide. The toxicity of DF is due to the presence of aromatic hydrocarbons such as benzene, toluene, ethylbenzene and xylene, which are collectively termed BTEX. The carcinogenic potential of DF is due to the presence of co-carcinogens such as C_{10}–C_{20} alkenes, and alkylated benzenes [1,2]. The development of effective technologies to treat toxic, DF-contaminated water is a pressing issue to be surmounted.

There are several methods for the remediation of petroleum-contaminated sites [3–5]. These methods can be broadly categorized into mechanical (e.g. oil–water separators), chemical (using chemicals dispersants such as surfactants), or biological (based on bioremediation) treatment methods. Over the last few years, much attention has been devoted to fluidized bed bioreactors as an effective technology for treating organic pollutants [6–10]. This technology seems to be more effective than suspended growth technologies because it enables high biomass concentrations, low hydraulic residence times, small footprint requirements, and no mechanical moving parts.

The complete oxidation and metabolic pathways of straight chains and aromatic hydrocarbons have been investigated during the past few years. Aerobic processes by natural populations of microorganisms represent one of the primary mechanisms for the elimination of hydrocarbons in the environment [11]. Margesin and Schinner [4] and Erickson et al. [12] have suggested that aerobic biodegradation by natural populations of microorganisms is a favorable possibility by which DF can be degraded. Yang et al. [5] have also indicated the potential of biological degradation of DF in water using a trickling filter. The present experimental study investigates the performance of an aerobic three-phase fluidized bed bioreactor for the treatment of feed water contaminated with DF.

2. Experimental

Fig. 1 shows the experimental setup in the Laboratory of Water and Wastewater Treatment Technologies at Ryerson

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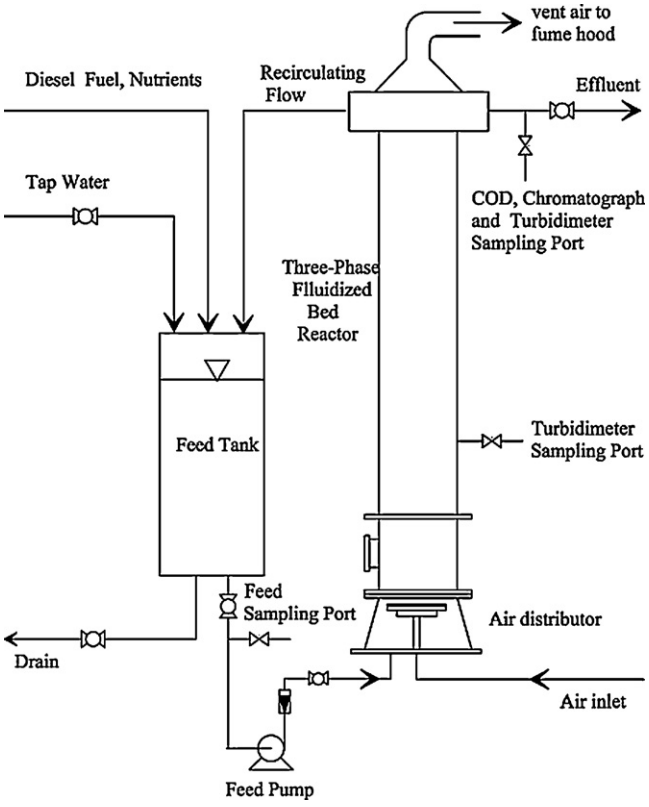

Fig. 1. Schematic diagram of the experimental set-up.

University, Toronto. The setup comprises a cylindrical three-phase fluidized bed reactor of 3 m height, and 17 cm internal diameter. DF-contaminated water from a 200 L feed tank enters at the bottom of the reactor along with an air stream. Inside the reactor, wastewater and air come in contact with each other as well as with the biomass growing as biofilms on the surface of fluidized lava rock particles. Treated wastewater and air are discharged from the top of reactor. A part of the treated wastewater can be recycled if necessary to adjust the residence time of the wastewater inside the reactor.

One kilogram of lava rock particles were used to support biomass inside the reactor (Fig. 2). The properties of lava rock particles are shown in Table 1. Fresh biomass from a Rotating Biological Contactor (RBC) was used to inoculate the reactor. The RBC is located in the same laboratory, and had been operational for a whole year treating wastewater also contaminated with DF. Thus, the microbial population of the biomass was well adapted to the presence of DF in the wastewater. DF-contaminated wastewater (i.e. synthetic wastewater) was prepared by adding DF and nutrients (for the growth of microorganisms) to water in a small feed tank. This tap water was continuously fed from a larger, 5000 L reservoir tank. Table 2 lists the concentration of nutrients in the feed tank. The concentration of DF, and chemical oxygen demand (COD) in the wastewater was in the range, 50–700 and 547–4025 mg/L, respectively. The pH of the wastewater was maintained in the range 6.7–7.8 by adding NaOH on demand. The temperature of water during the experiments was $20 \pm 5\,^{\circ}C$. Sampling was effected through three ports along the column height and at the feed tank. The first port is at the bottom of the feed tank. The second port is located on the reactor at 250 mm above the point of air injection. The third port is at the top of the reactor. While the first port was used to take the influent wastewater samples, the other two ports were used to take samples of both wastewater and bioparticles.

Fig. 2. Lava rock particles used to support biomass.

Table 1
Properties of lava rock particles used to support the biomass in the fluidized bed reactor

Parameter	Value
Average particle diameter (μm)	600
Particle density (kg/m^3)	1790
Minimum fluidization velocity in water (cm/s)	0.3

2.1. Analytical methods

The parameters analyzed in this work were DF concentration, chemical oxygen demand, turbidity, pH, and dissolved oxygen (DO) of the wastewater. The DF concentration in wastewater was determined using liquid–liquid extraction and gas chromatography. The method followed by the Environmental Protection Agency, EPA Method 8041, was used for the liquid–liquid extraction. Dichloromethane was used as a solvent. The calibration curve for DF was established using known concentrations of Diesel Range Organics (DRO) and 5-Alpha Androstane as an internal standard. A wastewater sample was transferred to a

Table 2
Concentrations of bacterial nutrients in the feed tank

Nutrient	Concentration (mg/L)
NH_4Cl	102.6
NaCl	203.3
$CaCl_2{\cdot}6H_2O$	0.4
$MgCl_2{\cdot}4H_2O$	0.8
Na_2HPO_4	406.6
KH_2PO_4	40.6
$MgSO_4{\cdot}7H_2O$	8.0
$FeSO_4{\cdot}7H_2O$	0.4

1 L separatory funnel, and was shaken for 2 min after adding dichloromethane. The lower organic layer was withdrawn from the bottom of the flask. After allowing the organic phase to rest for 15 min, it was passed through a funnel containing a plug of glass wool with 20 g of anhydrous sodium sulfate, and transferred to an auto sampler vial. A gas chromatograph coupled with a mass spectrometer was used to determine the amount of DF in the sample. The total area of peaks between C_{10} and C_{20} was considered to represent the total DRO.

The colorimetric MICRO-COD test method developed by Bioscience Inc. was used to determine the COD of the wastewater samples. In this method, the sample is oxidized using a solution of dichromate ions. Test kit No. 975-62 for the COD range, 100–4500 mg/L, was employed. A 50 mL sample was homogenized for about 2 min after collection, and a representative sample of 0.5 mL was obtained in a twist-cap vial (VWR International) for placement in a COD heater block at $150 \pm 2\,^{\circ}C$ for 2 h. Next, the vial was removed and allowed to cool for 10 min. COD of the sample was then directly read in mg/L using an Orbeco-Hellige 975 MP colorimeter.

The turbidity of the treated wastewater was measured in nephelometric turbidity units (NTU) using a Turbidity Meter Model 800 (VWR International). The meter was calibrated with solutions of 0 and 10 NTU. The measurements were done immediately after sampling in order to avoid particle flocculation and sedimentation. The probe was calibrated with a buffer solution of 4 NTU. A Hanna HI 9025 pH meter was used to measure the acidity/alkalinity of the wastewater. Magnetic stirrers homogenized the samples. The DO of the wastewater was measured using an YSI Model 58 oxygen meter connected to the three-phase fluidized bed reactor. The probe was constantly calibrated with saturated air, and magnetic stirrers were used for homogenous sampling. Finally, the thickness of the biofilms was measured using a Renishaw Raman Imaging Microscope. The total suspended solids (TSS) were determined following the protocol of Test 2540, APHA [13].

2.2. Reactor operation

After the inoculation with the biomass from the rotating biological contactor treating DF, the three-phase fluidized bed reactor was operated for a three month cultivation period with 100% recycle to ensure proper growth and immobilization of the biomass on lava rock particles. The DF concentration in the synthetic wastewater was 50 mg/L at the start-up. The operation was carried out with the minimum liquid fluidization velocity of 0.3 cm/s corresponding to the volumetric liquid flow rate of 5.7 m^3/day. Air was injected at the superficial velocity of 1 cm/s. Wastewater samples of 1 L and 100 mL were taken from the first and second sampling port to monitor DO and pH. The pH was maintained between 6.7 and 7.3. Samples of bioparticles were taken from the second sampling port for microscopy inspection. The makeup wastewater was provided from the 5000 L feed tank. During this operation, the growth of the biomass was visually observed as light and sometimes dark gray coatings or biofilms on the support particles.

After the three-month cultivation period, the reactor was operated under unsteady state for 12 days to assess the upper limit of DF removal by the reactor. Fresh water, nutrients and a variable amount of DF in the range 50–700 mg/L were daily charged to the reactor. The DF loading corresponded to 0.4–4.3 kg/m^3 day with 1.24 kg/m^3 day standard deviation. The influent COD was in the range, 547–4025 mg/L with 1267 mg/L standard deviation. During the 12 day period, the hydraulic residence time of 4 h was maintained. The experiment was repeated three times, and the results reported are averaged with standard deviations within ±5%. Liquid fluidization velocity was lowered to 0.02 cm/s corresponding to the liquid flow rate of 0.43 m^3/day in order to avoid any damage to newly-grown biofilms. Air was injected at the superficial velocity of 1 cm/s. From the first and the third sampling ports, samples of 100 mL, 50 mL and 1 L were taken to determine the respective levels of DF, COD, DO, and pH in the influent and treated wastewater. Samples of 1 L and 100 mL were taken from the second sampling port to monitor DO and pH in the reactor.

After the 12th day of unsteady state operation, the reactor was placed at steady state for five days (from Days 13 to 17). The DF loading corresponded to 1.1–1.3 kg/m^3 day with 0.06 kg/m^3 day standard deviation. The influent COD was in the range, 1156–1390 mg/L with 94 mg/L standard deviation. Liquid fluidization velocity, and air superficial velocity were the same as in the unsteady state operation. As before, the hydraulic residence time of 4 h was maintained. The experiment was repeated three times, and the results reported are averaged with standard deviations within ±5%. The sampling procedures of the unsteady state operation were repeated. Bioparticle samples were extracted for microscopy analysis from the second and third sampling ports. To determine the TSS in the treated wastewater, 100 mL of the wastewater sample was taken from the third sampling port. Turbidity was determined by taking 100 mL of the influent and the treated wastewater samples respectively from the first and the third sampling ports.

3. Results and discussion

As mentioned above, the three-phase fluidized bed reactor was first operated at unsteady state by varying the concentrations of DF and COD in the influent wastewater for 12 days. Fig. 3 shows the percentage of DF removed from influent wastewater as well as its DF loading rate versus time. As shown in the figure, DF removals up to 100% were achieved with the influent loading rates of DF in the range 0.43–1.03 kg/m^3 day corresponding to 70–200 mg/L of DF. For the influent loading rates greater than 1.03 kg/m^3 day after Day 6, the removal of DF decreased with the increase in its loading rates, and was at the minimum of 10% at the loading rate of 4.33 kg/m^3 day. The reason for the decrease in DF removal is that beyond a certain DF load, the biomass and oxygen supply are insufficient to sustain the metabolic activity of the microorganisms. This phenomenon reduces the biomass concentration, thereby slowing down the biological degradation of DF. The maximum DF removal takes place at DF loadings up to 1.03 kg/m^3 day.

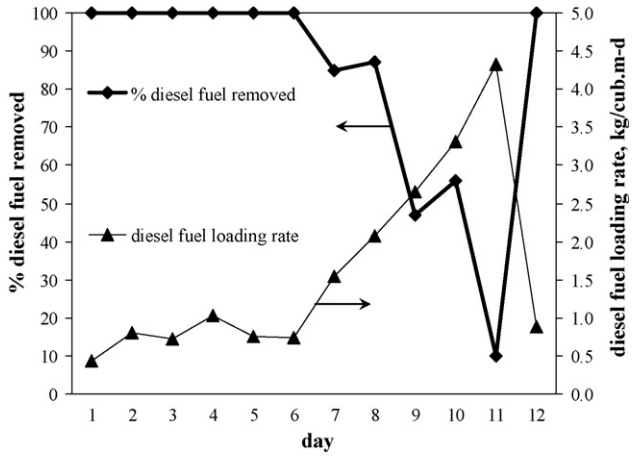

Fig. 3. Percentage of DF removed and DF loading rate of influent wastewater during the unsteady state operation.

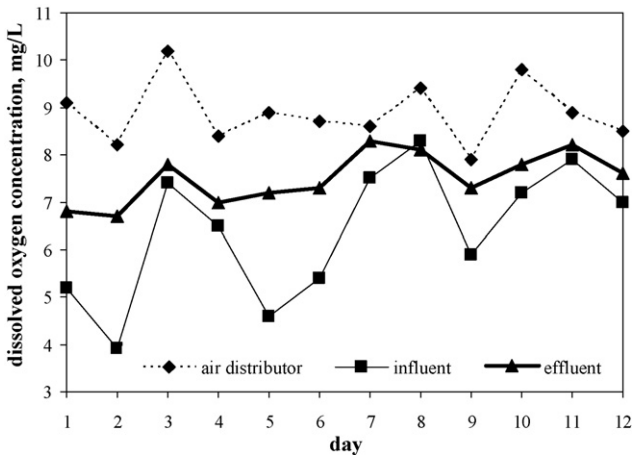

Fig. 5. Concentration of dissolved oxygen in wastewater during the unsteady state operation.

The percentage of COD removed from influent wastewater, and its COD concentration during the unsteady state operation of the reactor is shown in Fig. 4. As shown in this figure, COD removal as high as 97% was achieved. The removal of COD was greater than 90% for COD concentrations up to 1345 mg/L. For COD concentrations higher than 1500 mg/L, the removal of COD generally decreased with the increase in its concentration. The lowest value of COD removal was 29% corresponding to the COD concentration of 4025 mg/L.

The concentration of DO in the influent wastewater, near the distributor of the three-phase fluidized bed reactor, and in the effluent is shown versus time in Fig. 5. It is well known that DO concentration in the range 2–4 mg/L is favorable for aerobic biodegradation processes. In this study, the oxygen concentration of the influent wastewater was kept equal to or greater than 7.9 mg/L. As seen in the figure, the oxygen concentrations near the distributor, and that at the outlet were mostly in phase with the oxygen concentration of the influent. Because of the air jets near the distributor, the oxygen mass transfer at this point is higher [14,15], and the oxygen concentration was always higher than that of the influent. This behavior was expected as the oxygen demand of the synthetic wastewater reduces DO with the column height in the reactor This fact underlines both the importance of the zone near the distributor as a region of enhanced oxygen transfer and the strong oxygen demand as a result of the biological activity. When the concentration of DO in the influent was at the lowest value of 3.9 mg/L (Day 2), the removal of the DF (Fig. 3), and COD (Fig. 4) was 100% and 94.3%, respectively.

An important factor to be considered for the aerobic biological oxidation of wastewater is its pH. Fig. 6 shows the pH of the influent and effluent versus time during the unsteady state operation of the three-phase fluidized bed reactor. The pH of the influent was maintained in the range, 7.4–7.8. The effluent pH was found to be typically lower, and in the range, 6.6–7.6. This range is well within the allowable pH range of 6.5–8.5 for treated effluent discharged to the environment [16]. It is important to point out that complete biological aerobic oxidation tends to decrease the pH in a fluidized biofilm process, and in other processes where biofilm technology is applied. Lower effluent pH values suggest the presence of extra-cellular products like fatty acids, which result from the microbial utilization of straight-chain hydrocarbons [11].

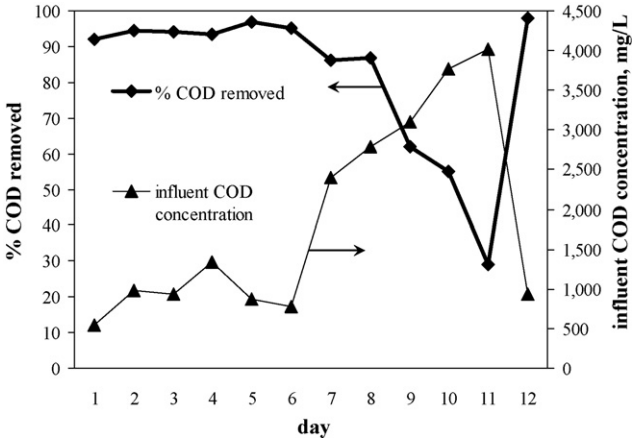

Fig. 4. Percentage of COD removed, and influent COD concentration during the unsteady state operation.

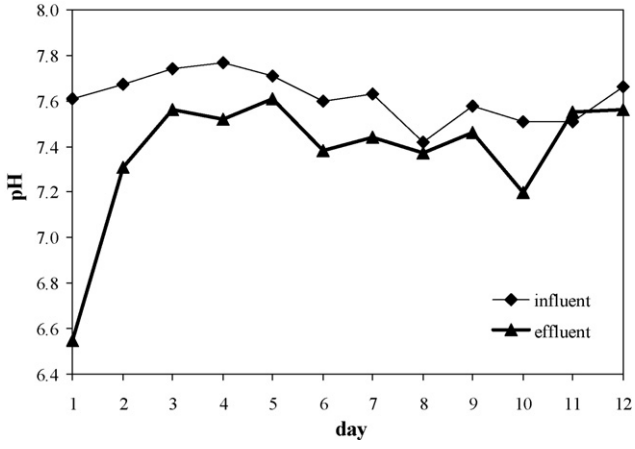

Fig. 6. The pH of influent and effluent wastewater during the unsteady state operation.

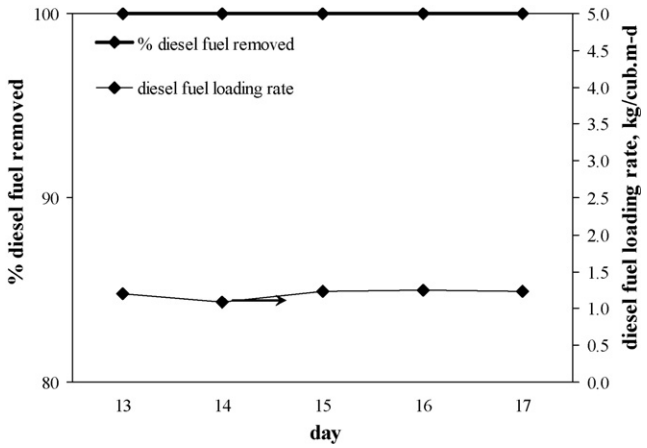

Fig. 7. Percentage of DF removed, and DF loading rate during the steady state operation.

Following the unsteady state operation, the three-phase fluidized bed reactor was operated under steady state for five days (Days 13–17) for the treatment of influent wastewater with approximately 200 mg/L of DF. During that operation, the influent loading of DF varied in the range, 1.09–1.25 kg/m^3 day with a standard deviation of 0.06 kg/m^3 day. On the other hand the influent COD concentration varied in the range, 1156–1389.6 mg/L with the standard deviation of 93.9 mg/L. The hydraulic residence time inside the reactor was about four hours. As seen in Fig. 7, the removal of DF from the influent was 100% throughout the experiment. The removal of COD from the influent varied in the range, 93.6–99%, as seen in Fig. 8.

The reactor significantly reduced the concentration of TSS in the wastewater from 900 mg/L to very low concentrations averaging 0.87 mg/L. It may be noted that a fluidized bed reactor does not behave as an activated sludge process where biomass is suspended in the liquid phase. In the present experiments, biomass was visually observed attached to the solid support of lava rock particles. The reduction of TSS in the treated effluent is a direct evidence of the strong biological degradation taking place in the reactor.

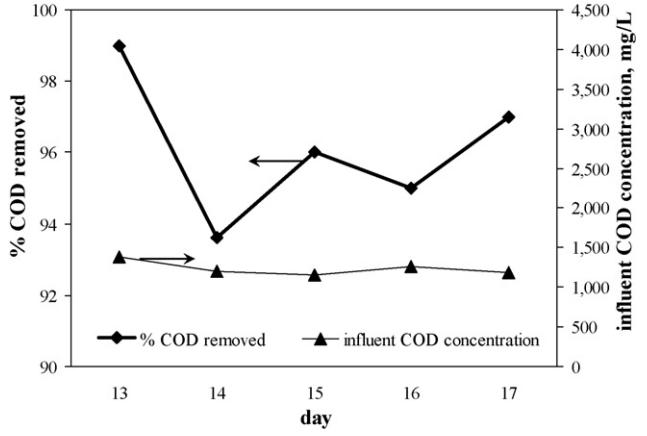

Fig. 8. Percentage of COD removed, and COD concentration of the influent during the steady state operation.

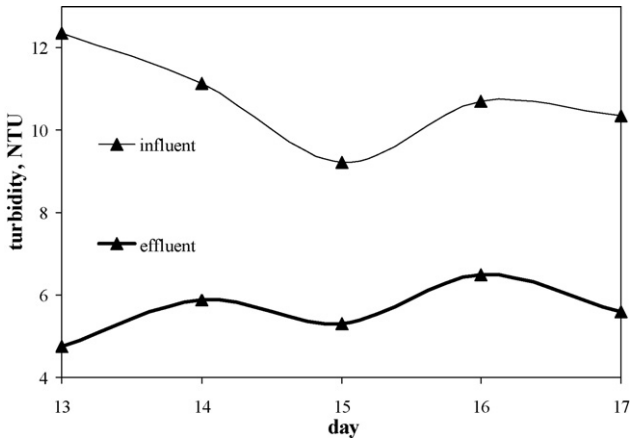

Fig. 9. Turbidity of wastewater during the steady state operation.

The turbidity values as nephelometric turbidity units (NTU) were also measured for the wastewater during the steady state operation of the reactor. As depicted in Fig. 9, the turbidities of influent and effluent were in the range 9.2–12.4 and 4.8–5.9 NTU, respectively. The effluent turbidity was well below the maximum allowable turbidity of 10 NTU for an effluent to be discharged into the environment.

In general, the three-phase fluidized bed reactor did not have particles exhibiting uniform thickness of biofilm. Biofilms with thickness in the approximate range, 50–700 μm, were observed microscopically. Particles at the top of reactor were observed to have thicker biofilms whereas those at the bottom of reactor were observed to have thinner biofilms. The reason for this behavior is that the particles with thicker biofilms have lower densities while those with thinner biofilms have higher densities. The fluidized bed was observed stratified into three visible regions along the height of the reactor. The first region was in the lower part of reactor, and was located next to the distributor. The thickness of biofilm was measured to be around 50 μm in this region (Fig. 10). It appears that higher turbulence, shear stress, greater inter-particle collisions in this region, and lower residence time would have promoted the formation of thin biofilms. The second region of the bed was in the middle of the reactor with noticeably thicker biofilms observed as dark or light coatings on

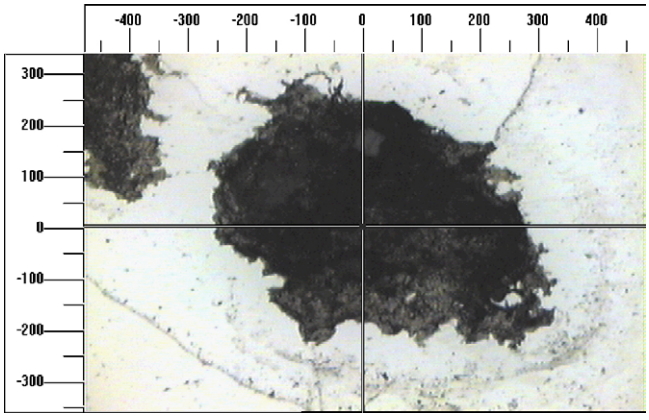

Fig. 10. Microscopy photograph of the active biofilm developed in the lower part of the reactor (scale is in μm).

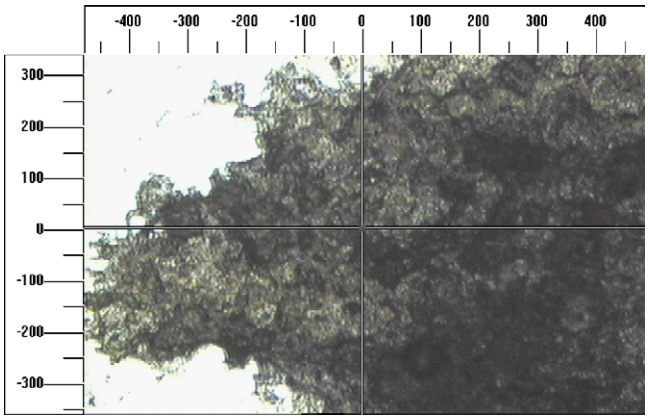

Fig. 11. Microscopy photograph of the active biofilm developed in the upper part of the reactor (scale is in μm).

the lava rock particles. In this region, the turbulence was lower, and would have permitted the formation of thicker biofilms on lava rock particles. The third region of the bed was in the upper part of reactor having particles with biofilms as thick as about 700 μm (Fig. 11). Very low settling velocities were observed in this region. As a result, low values of shear on the particles with longer residence time in this region would have facilitated the formation of thick biofilms.

Table 3 lists the wastewater and hydrodynamic properties during the steady-state operation of the three-phase fluidized bed bioreactor. A good quality effluent with undetectable DF having average concentrations of 47 mg/L COD, 0.87 mg/L TSS, and 5.61 NTU turbidity was obtained from influent wastewater loaded with 1.2 kg/m^3 day of DF.

It may be noted that the lava rock particles are impervious, and the DF removal due to adsorption on the biofilms is negligible. For example, at the maximum DF concentration (4025 mg/L) in the reactor when the DF removal is the lowest (10%; Fig. 3) and biodegradation is negligible, the DF removal due to adsorption alone would be less than 5.5%. This percentage is obtained after accounting for air stripping which could remove 4.5% of the DF (i.e., 45% of the total 10% DF removal). Hence, the DF removal in the aerated three-phase fluidized bed bioreactor was mostly due to biodegradation, and volatilization during which the lighter components of DF volatilized into the air bubbling through the reactor. To find out DF removal solely due to volatilization in presence of air, four different experiments were performed with clean lava rock particles inside the three-phase fluidized bed reactor without any microorganisms [17]. The operating conditions and the DF-contaminated influent were similar to those under the steady state biodegradation experiments. Effluent analyses showed that on an average, approximately 45% of the total DF removal was due to volatilization into air. For a higher DF loading of about 4.3 kg/m^3 day, the DF removal due to the air stripping was about 3.5%, i.e. 35% of the total 10% DF removal as shown in Fig. 3. These results conform to the past findings by other researchers [3,18], and indicate that up to 45% of the total DF removal could be ascribed to air stripping in the overall wastewater treatment carried out in the present study. Therefore, with such reactors, care should be taken to avoid the release of volatilized DF components to atmosphere. The air discharged from the reactors should be stripped of the volatilized components by suitable means such as combustion followed by the removal of particulate matter. While the reactor enables very high efficiency of DF separation, auxiliary equipment is needed to clean the discharged air.

Table 3
Wastewater and hydrodynamic properties during the steady-state operation of three phase fluidized bed bioreactor

Property	Value		% Removal
	Influent	Effluent	
Average DF concentration (mg/L)	200	Not detected	100
Average chemical oxygen demand (mg/L)	1236.92	47.04	96.2
Average total suspended solids (mg/L)	900	0.87	99.9
Average turbidity (NTU)	10.75	5.61	47.8
Biofilm thickness (μm)	50–700		
Average DF loading rate (kg/m^3 day)	1.2		
Hydraulic residence time (h)	4		

4. Conclusions

The biodegradation of the DF contaminated wastewater was investigated in a three-phase aerobic fluidized bed reactor. In the first step, the performance of the reactor was examined under unsteady state condition for 12 days by varying the concentrations of DF and COD in influent wastewater. The reactor with a hydraulic residence time of 4 h was able to remove 100% of DF from the influent wastewater at loading rates in the range, 0.43–1.03 kg/m^3 day corresponding to 70–200 mg/L of DF. The treatment capacity of the reactor was identified using higher influent DF loading rates for which DF removal was lower. Further, the reactor was able to remove up to 97% of COD from influent wastewater having COD concentrations in the range, 547–4025 mg/L. The removal of COD was greater than 90% for influent COD concentrations equal or less than 1345 mg/L. For influent COD concentrations higher than 1500 mg/L, the removal of COD was found to be in the range 30–85%.

In the second step, the reactor was operated under steady state condition for five days to treat influent wastewater having approximately 200 mg/L of DF. The DF loading corresponded to 1.1–1.3 kg/m^3 day, while the influent COD was in the range, 1156–1390 mg/L. The reactor removed 100% of DF, and 96.2% of COD on an average from the influent with a low hydraulic residence time of 4 h. The reduction of the TSS from 900 mg/L to less than 1 mg/L indicates very high biodegradation enabled by the reactor. Thus, in general, the reactor carried out a high removal of both DF, and COD from heavily contaminated wastewater. The treated effluent had acceptable pH values, low turbidity, and very low concentration of suspended solids. Volatilization into air accounted for about 45% of the total DF removal from the wastewater.

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