

Performance of a Packed Reactor with *Opuntia imbricata* for Municipal Wastewater Treatment

F. May-Esquivel, L.J. Rios-González, Y. Garza-García and J. Rodríguez Martínez
 Department of Biotechnology, Faculty of Chemistry, Universidad Autónoma de Coahuila,
 Boulevard V. Carranza and Ing. J. Cárdenas V., Col. República,
 Z.C. 25280. Saltillo, Coahuila, Mexico

Abstract: The oxygen transfer rate and the performance of both a packed reactor with a natural support of *Opuntia imbricata* represented as (R1) and a reactor without support represented as (R2) were compared in the treatment of municipal wastewater. The results demonstrated that the dissolved oxygen (DO) and oxygen transfer rate (K_La) were affected positively by air flow rate when increased from 0-2 lpm, mainly in R1. COD removal efficiency was affected by decreasing hydraulic retention time from 30-10 h in both reactors, but highly significant in R2. The R1 reactor showed higher COD removal rate than R2, when the Organic Loading Rate (OLR) was increased from 0.44-1.32 kg COD m⁻³ day⁻¹. Biomass concentration after 100 day of continuous operation was 2.49 and 12.35 g VSS L⁻¹ for R1 and R2, respectively. The higher oxygen transfer rate, concentration of active biomass and performance of R1 were result of a greater specific surface area of *Opuntia imbricata*.

Key words: Packed reactor, natural support, biofilms, oxygen transfer, COD removal, municipal wastewater

INTRODUCTION

The demanding norms to maintain the quality of water have stimulated development of new technologies for wastewater treatment. Treatment of wastewater in packed reactors using immobilized cells fixed on the surface of natural support is attracting interest due the application of different immobilization methods and a variety of supports (Sorlini *et al.*, 1990; Zhou *et al.*, 2003). Some researches have used materials of vegetal origin for the immobilization of microorganisms as in the case of bamboo (Camargo and Nour, 2001).

Specific surface area, porosity, surface roughness, pore size and orientation of the packing material were found to play an important role in aerobic filter reactor performance (Yang *et al.*, 2004). Picanco *et al.* (2001) reported that the efficiency of removing organic matter in fixed-bed reactors is directly related to the characteristics of the support material used for immobilization.

Biofilm reactors offer the advantage of higher load systems which require less volume and space and less investment in comparison to conventional systems (Karimniae-Hamedani *et al.*, 2003; Liu and Tay, 2002; Mosquera-Corral *et al.*, 2003; Nicolella *et al.*, 2000;

Rodgers *et al.*, 2004). In addition biofilm reactors allowed the separation of Hydraulic Retention Time (HRT) from solids retention time and together with fixed biomass permit higher loading rates and short residence times (Souza *et al.*, 2004; Wäsche *et al.*, 2002). Besides biofilm reactors provide a large surface area per volume unit of reactor, which aids in the enhancement of oxygen transfer through the biofilms and easing biotransformation of COD (Casey *et al.*, 1999; Rosenberger *et al.*, 2002). The oxygen transfer rate and organic loading rate is very important for design control and optimization of the process in any municipal wastewater treatment (DeMoyer *et al.*, 2003; Fonade *et al.*, 2001; Liwarska-Bizukojc *et al.*, 2002; U.S. EPA, 1999). According to Arslan and Ayberk (2003) the municipal wastewater contains a great portion of easily biodegradable organic matter, which is a good parameter that reflects microbial activity and performance of aerobic biological processes. Oxidation of organic matter, biosynthesis of new products and generation of new biomass are the result of oxygen consuming reactions and this point out the importance of transference oxygen in the enhancement of biodegradation of COD by cell systems. The determination of kinetics is of the most common used tool,

Corresponding Author: F. May-Esquivel, Department of Biotechnology, Faculty of Chemistry, Universidad Autónoma de Coahuila, Boulevard V. Carranza and Ing. J. Cárdenas V., Col. República, Z.C. 25280. Saltillo, Coahuila, Mexico

which help to define the aeration mode, air flow rate and the energy needed for the aerobic system (Rodde-Pellegrin *et al.*, 2002).

The objective of this study was to evaluate use of *Opuntia imbricata* as a kind of support for fixed biofilms and to check the influence of the air flow rate on the oxygen transfer rate and efficiency of a packed reactor on municipal wastewater treatment.

MATERIALS AND METHODS

Plant configuration: The reactors were built with two acrylic columns. The principal column (15×61 cm and downflow) was united to another small column by a PVC elbow in the bottom of the principal column (3×50 cm and upflow). The reactor R1 was packed with pieces of *Opuntia imbricata* (1.2 kg of total dry weight) of 10 cm length and external and internal diameters of 3.5 and 3 cm respectively (Fig. 1). The reactor R2 did not contain any support only aerobic sludge. Total working volume of R1 and R2 reactors was 9 L.

Natural support *Opuntia imbricata*: Is a cactus plant that grows throughout over northern Mexico and the southwest of USA and is relatively inexpensive raw material, with a highly rough and porous surface (Fig. 2b). We have been working at the Environmental Biotechnology Lab of the Chemistry Faculty Universidad Autónoma de Coahuila with dried up pieces of stems of *Opuntia imbricata* (Fam. Cactaceae) as a natural support for the growth of biofilms under aerobic and anaerobic conditions (Rodríguez-Martínez and Garza-García, 2002). Our studies showed the potential of this material as a support for biofilms development because of its inherent structural and physical characteristics. The cylindrical skeleton of *Opuntia imbricata* used in this study (previously rinsed and washed with water) is shown in the Fig. 2a. The Fig. 2c illustrates a micrography of biofilms structure obtained by Scanning Electron Microscopy (SEM) at 10.00 kX of R1 reactor after 100 days of continuous operation.

Aeration systems: An air compressor (Mod DOA-P104-AA) fed air to both reactors and was connected to a general filter (Mod F72G-2AN-AL3, NORGREN, USA) and later to two flowmeters (Series FR2000, Mod 2A15, Key Instruments, USA) graduated in liters per minute (lpm). Flow rate aeration for this study was fixed from 0.25-2.0 lpm.

Wastewater and inoculum: Sewage sludge used as inoculum (500 mL active sludge) was obtained directly

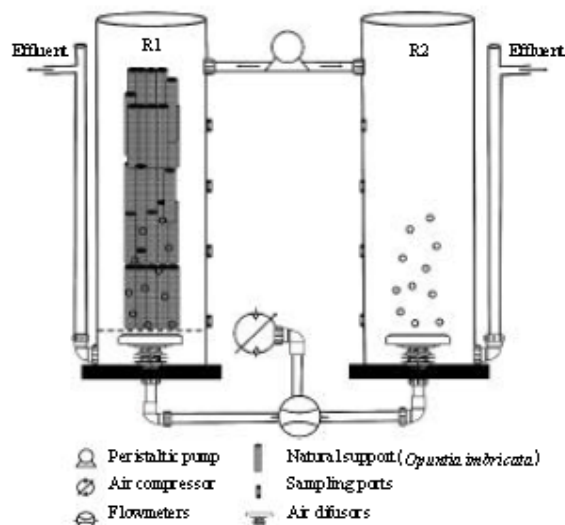


Fig. 1: Schematic diagram of bioreactors

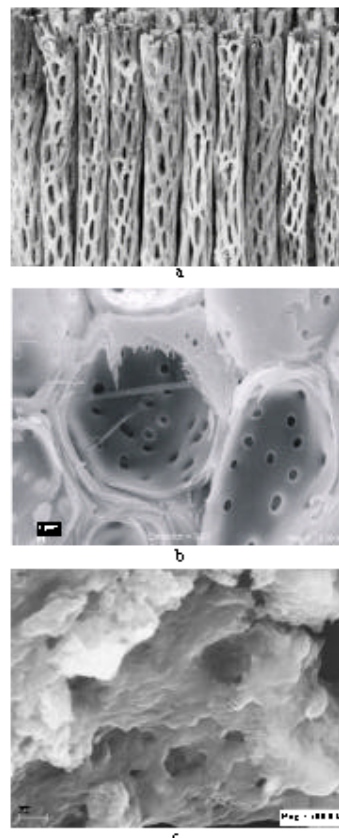


Fig. 2: (a) Cylindrical skeleton of *Opuntia imbricata*, (b) Micrography of *Opuntia imbricata* structure obtained by SEM at 2.00 kX, (c) Micrography of biofilms structure obtained by SEM at 10.00 kX of R1 reactor after 7 days of development

from a full-scale aerobic reactor at the municipal wastewater treatment plant from the Ecology Department of the State Coahuila at Saltillo Mexico. The sewage sludge was kept in recirculation for 7 days together with municipal wastewater to allow the acclimatization and fixing of biomass on the support as biofilm in R1 reactor. The wastewater used was collected from municipal drainage. The characteristics of wastewater were: Average 550 mg L^{-1} COD, pH 7.0. The study was conducted at ambient temperature (20-25°C). The inflow rates were varied to obtain different Hydraulic Retention Times (HRT).

Analysis of methods: Chemical Oxygen Demand (COD) and Volatile Suspended Solids (VSS) were measured according to the standard APHA method (APHA, 1998). The biomass attached to *Opuntia imbricata* was shaken by removes and obtain the concentration in terms of gVSS L^{-1} . Dissolved Oxygen (DO) was measured *in situ* with an Oxygen Meter (Oxygen meter, Traceable®, USA). The oxygen electrode was placed in the principal column at a distance of 40 cm above the diffuser any time that oxygen was measured. During this time the compressor was kept in stop. The volumetric oxygen transfer coefficient ($K_L a$) was measured by the dynamic method (DeMoyer *et al.*, 2003; Fonade *et al.*, 2001; Klein *et al.*, 2002; Rodde-Pellegrin *et al.*, 2002), assuming the mixture of gas and liquid phases. The liquid was deaerated by stripping with nitrogen and then nitrogen was replaced by air. The $K_L a$ was measured in both reactors in the presence of biomass. For Scanning Electron Microscopy (SEM) Leica-Cambridge model Stereoscan 440, were obtained micrographies of the pieces of *Opuntia imbricata* before and after it's colonization by the microbial consortia (Fig. 2b and c).

All data presented represents the means from 3 replications that were kept for each experiment (Steel *et al.*, 1980).

Experimental setup: The system was operated at steady-state at least for five days for each experimental condition of hydraulic retention time, COD removal rate, of oxygen transfer rate determination and dissolved oxygen. Mean of the three principal measurements was used for the interpretation of data.

Statistical analysis: Statistical analyses of the results were performed using SAS (1996). An analysis of variance was carried out using a data set of three replicates. The resultant data were statistically evaluated applying ANOVA at a 5% level of significance. Duncan's critical range tests were used to determine the significant difference between dependent variables.

RESULTS AND DISCUSSION

Start up of the reactors in overall operation time: The start-up period included the preliminary stages needed for stabilization of the system. This was defined as the moment in which the removal rate was constant or spread to stay constant (Rosenberger *et al.*, 2002; Zhou *et al.*, 2003). During the start-up period of both reactors, the maximum COD removal achieved at 20 h of HRT. This was defined as optimum for both R1 and R2 (Fig. 3). In due course of time there was consistent removal of COD and the operation of the reactors was stable. The R1 reactor showed high stability and high COD removal (90 %) from 5th day, while the R2 reactor reached stability shortly after 60th day, with a COD removal of <85%. Kariminiaae-Hamedani *et al.* (2003) reported a 90% COD removal at an HRT of 30.17 h after 79 days of operation. For that research a reactor packed with ceramic as support material was utilized in municipal wastewater treatment.

The COD removal efficiency of the R1 increased the first days to a mean value 90% in a relatively short period. This indicated that a very short start-up period was required for R1 reactor. A similar result was earlier observed by Sarti *et al.* (2001). The R1 reactor maintained a COD removal efficiency >95% until 100th day of continuous operation.

Development of the biomass concentration: In the steady state phase up to 95% of the natural support was covered with biofilms. Data from Zhou *et al.* (2003) showed similar result. Biomass concentration increased from 0 at the beginning of the operation to $2.49 \text{ g VSS L}^{-1}$ on 100th day in the R1 (Fig. 4). Mosquera-Corral *et al.* (2003) reported the degradation of polymers in a biofilm airlift suspension reactor using basalt as carrier material and an air flow rate of 3.0 lpm. Almost 100% of the carrier got completely covered by biomass within 20 days. Biomass concentration increased from 1.5 gVSS L^{-1} at the beginning of the operation to $12.5 \text{ g VSS L}^{-1}$ at the beginning of 63rd day.

Biomass concentration in the R2 reactor was of $12.35 \text{ g VSS L}^{-1}$ at 100th day. Washout of biomass due to application of short HRT could be the reason for the reduced biomass concentration in the reactor R2.

Casey *et al.* (1999) mentioned that excessive growth of biofilm is frequently a problem in the conventionally aerated biofilm reactors used in wastewater treatment. On the contrary the cylindrical and rugged structure of *Opuntia imbricata* and high surface area of the natural support controlled the growth of a thin biofilms. This structure permitted a low air flow rate which provided larger amount of oxygen (Fig. 2a and b). Wäsche *et al.*

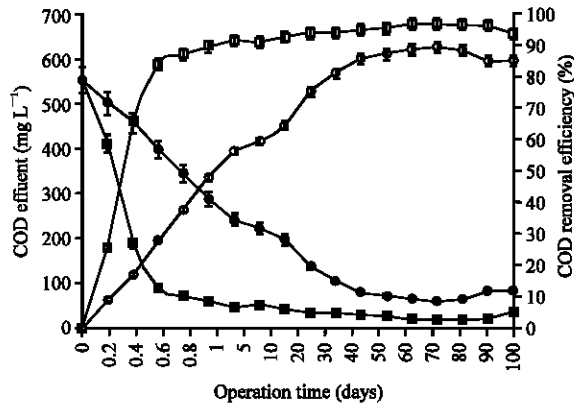


Fig. 3: Effluent COD and COD removal efficiency in R1 (■, □) and R2 (●, ○) reactors at HRT of 20 h, air flow rate of 1 lpm and an influent COD of 550 mg L⁻¹

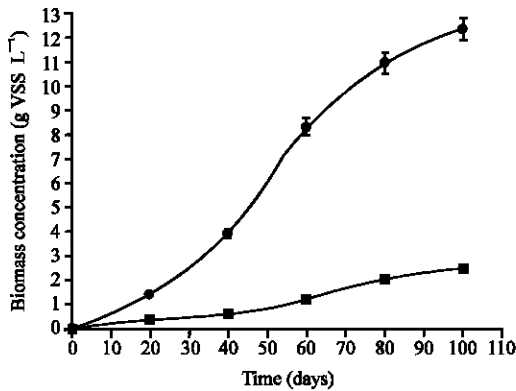


Fig. 4: Biomass concentrations in R1 (■) and R2 (●) reactors at a HRT 20 h and an air flow rate of 1 lpm

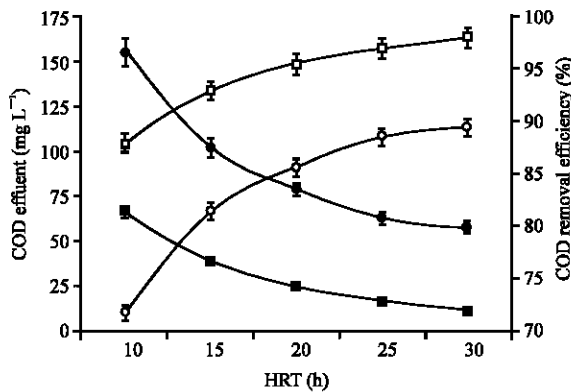


Fig. 5: Effluent COD and COD removal efficiency in the R1 (■, □) and R2 (●, ○) reactors of an air flow rate of 1 lpm and an influent COD of 550 mg L⁻¹

(2002) mentioned that the hydrodynamic conditions and the substrate loading rate during the growth phase of the biofilm are the two key parameters that influence the biofilm growth, particularly the structure, density and thickness of the biofilm system.

Performance of reactors in relation to HRT: As described in Fig. 5, reduction of HRT from 30-10 h produced a shock and decreased the efficiency of the reactors performance. The concentrations of effluent COD increased, which indicated a reduction in COD removal efficiency. In the R2, COD removal efficiency was 89.45 % at 30 h of HRT. This amount decreased significantly to 71.84% at 10 h of HRT.

In R1, the COD removal efficiency was >95% at a HRT of 30, 25 and 20 h and the COD_{effluent} was less than 25 mg L⁻¹. When the HRT was decreased to 10 h, the COD removal efficiency decreased to 87.87% and the COD_{effluent} was 66.7 mg L⁻¹. Tawfik *et al.* (2002) showed similar results. The difference in the efficiency of COD removal in R1 was not significant as in R2. Zita and Hermansson (1997) reported that a low level of cell surface hydrophobicity could be the reason for non-attachment of free-living cells to flocs. These cells escape sedimentation in the treatment system and reduce the quality of the effluent. On the contrary *Opuntia imbricata* had higher hydrophobicity with high fixed concentrations of active biomass. This quality conferred a bigger flexibility and operational stability to R1 as evidenced by Nicolella *et al.* (2000) and Zhou *et al.* (2003). Immobilized biomass is also less susceptible to irreversible damage when shock loads are encountered (Casey *et al.*, 1999).

Rodgers *et al.* (2004) developed a vertically moving biofilm system to treat industrial wastewater, which showed a removal of 93.2% of filtered COD and 97.9% biological oxygen demand (BOD₅). However a decrease in the HRT from 1.1-0.75 days produced a shock to the system. When the HRT was shortened to 0.75 days, the BOD₅ first increased to about 100 mg L⁻¹ and then decreased gradually. The performance of a biofilm reactor is directly associated with the metabolic activity of fixed bacteria under given operational conditions (Liu and Tay, 2002).

Influence of Organic Loading Rate (OLR) on the rate of COD removal: Figure 6 showed the COD removal rate, it increased in R1 from 24-60.425 mg COD L⁻¹ h⁻¹, when OLR was increased from 0.44-1.32 kg COD m⁻³ dya⁻¹. The COD removal rate in R2 was 37.1 mg COD L⁻¹ h⁻¹ at the maximum OLR of 1.32 kg COD m⁻³ day⁻¹. Nicolella *et al.* (2000) mentioned that biofilm reactors are an effective solution to retain biomass in the reactors and to improve the higher removal rate.

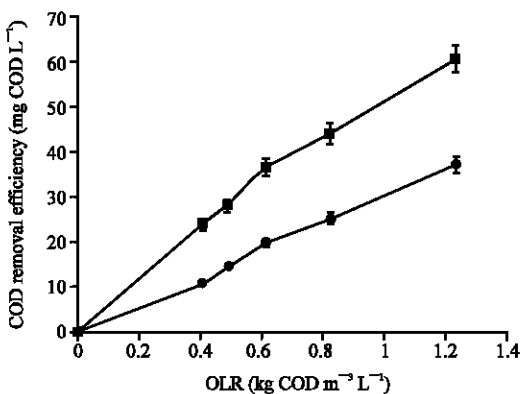


Fig. 6: Effect of OLR on COD removal rate for R1 (■) and R2 (●) reactors with an air flow rate of 1 lpm

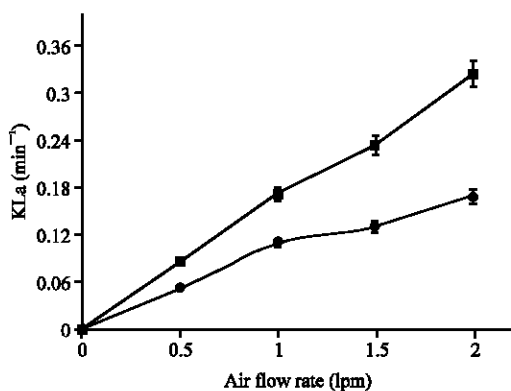


Fig. 7: Behavior of $K_{L,a}$ for R1 (■) and R2 (●) reactors

This capacity of standing the changes in organic load by R1 confirmed its flexibility and operational stability. In Comparison with R2 reactor, R1 showed a higher biomass activity, a longer solid retention time and a larger oxygen transfer coefficient. The results of COD removal rate by R1 indicated that a higher OLR can be applied to obtain good effluent in short period time.

Volumetric oxygen transfer coefficient ($K_{L,a}$): The *Opuntia imbricata* stem used as a kind of support for the packed reactor (R1) showed a positive influence on $K_{L,a}$, which increased from 0.0864 min^{-1} at 0.5 lpm until 0.3208 min^{-1} at 2 lpm of air flow rate as observed in Fig. 7.

In the case of the reactor without support (R2), the maximum $K_{L,a}$ was 0.1671 min^{-1} at 2 lpm. With increased aeration rate the air bubbles could be crushed into fine bubbles by surface of the natural support and this improved high oxygen transfer in the R1 reactor. Small bubbles have a higher specific surface area and could result in high $K_{L,a}$ (Zhou *et al.*, 2003).

A higher gas flow rate produced more bubbles and therefore more interfacial area was available for transfer.

Since a higher rate of oxygen transfer is due to bubbles, the ambient water DO also increased more rapidly at a higher gas flow rate (DeMoyer *et al.*, 2003). $K_{L,a}$ refers to volumetric oxygen transfer coefficient in a unit time. Reactor R1 showed better oxygen transfer than R2. The increased air flow rate supplied larger amount of oxygen, enhanced the velocity of liquid circulation and intensity of mixing, which had a positive effect on the oxygen transfer coefficient (Klein *et al.*, 2002). These results showed that an economical approach to increase the dissolved oxygen in the liquid face is increasing the oxygen transfer coefficient $K_{L,a}$ by high surface area of natural support as in R1.

Effect of air flow rate on Oxygen Transfer Rate (OTR) and Oxygen Uptake Rate (OUR): The air flow rate of 1 lpm for R1 reactor supplied the oxygen required for satisfying their OUR, as described Fig. 8a. Casey *et al.* (1999) reported that an increased oxygen transfer eliminated the problems of oxygen limitation. For the technological choice the scale up of the aeration system is an important part because consumed energy and daily cost of the operation are important parameters. In order to optimize the process, it is necessary to determine the microbial oxygen demand (DeMoyer *et al.*, 2003).

On the contrary, the reactor R2 at air flow rate of 1 lpm achieved an OTR of $0.0704 \text{ mg O}_2 \text{ L}^{-1} \text{ min}^{-1}$ that did not supply the oxygen required for OUR (Fig. 8b). When 2 lpm were provided the OTR needs were met for supplying the oxygen needed for OUR ($0.1483 \text{ mg O}_2 \text{ L}^{-1} \text{ min}^{-1}$). However the oxygen requirements of the microorganisms in suspension in R2 were hardly met with an air flow rate of 2 lpm. Consequently, a greater energy expense is required to overcome the OUR in the reactor without support. About 70% of the total energy consumption of a municipal wastewater treatment plant is used for oxygen supply (Fonade *et al.*, 2001; U.S. EPA., 1999).

Effect of the air flow rate on dissolved oxygen *in situ* in the reactors: The DO concentration in the R1 increased from $0.41\text{-}4.82 \text{ mg L}^{-1}$ with an air flow rate of 0.25-2 lpm (Fig. 9). On the other hand, DO concentration for R2 did not exceed 3.05 mg L^{-1} at 2 lpm. Fluctuations in dissolved oxygen concentration potentially affected the overall productivity of bioreactors. Another paper reported that the perforated membrane diffusers required 30-40% less air than coarse bubble diffusers to maintain constant mixed liquor DO (U. S. EPA., 1999).

For the R1 reactor, an air flow rate of 1 lpm maintained a dissolved oxygen concentration of 1.5 mg L^{-1} which is lower than 2 mg L^{-1} . This is the amount recommended for

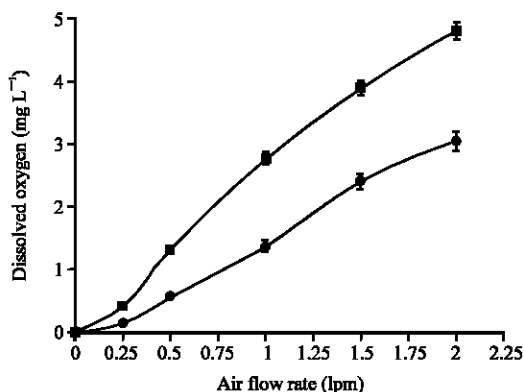


Fig. 8: Effect of air flow rate on OTR (■) and OUR (□) for R1 (a) and R2 (b) reactors

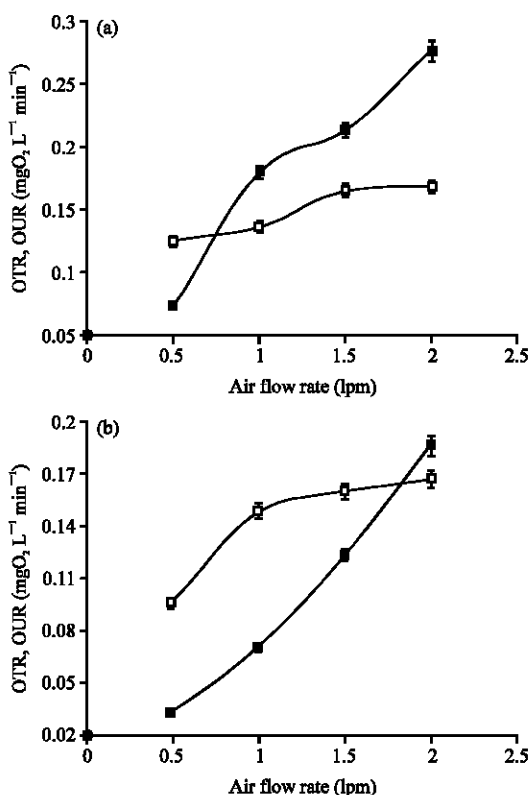


Fig. 9: Effect of the air flow rate on the DO for R1 (■) and R2 (●) reactor with 20 h HRT

aerobic system of wastewater treatment (Hu *et al.*, 2005; Karimniaae-Hamedani *et al.*, 2003; Rodgers *et al.*, 2004; U. S. EPA., 1999). It was necessary to have an air flow rate of 1.5 lpm for the R2 reactor in order to maintain a DO concentration of 2 mg L⁻¹ because oxygen has a low solubility in water. The bubbles generated by the diffusers at the bottom of the principal column packed with the natural support facilitated the oxygen solubility

and mixing of the bulk fluid to make an efficient contact between the wastewater and the biofilm in the case of R1 reactor (Casey *et al.*, 1999). Hu *et al.* (2005) reported the performance of sequence batch reactor by mechanical mixing and treating synthetic wastewater with a COD of 500 mg L⁻¹ under three different air flow rates. Those researches obtained high OD concentration > 4 mg L⁻¹ in the first 400 min of operation at an air flow rate of 2.66 lpm. However an air flow rate of 0.66 and 0.33 lpm recorded a DO less than 0.5 mg L⁻¹.

CONCLUSION

The results obtained for the packed reactor with natural support (R1) showed that the system effectively removed COD of municipal wastewater under certain conditions.

R1 reactor achieved a COD removal efficiency of more than 95% at 20 h of HRT and an air flow rate of 1 lpm. The natural support called *Opuntia imbricata*, dead stems used for fixing aerobic microorganisms as biofilms was responsible of high removal efficiency of COD in wastewater treatment. It offered higher surface area allowing a high DO concentration and a continuous and stable performance of reactor R1 for a longer period. Higher removal of COD was obtained in spite of the low volume of support media and low HRT. Reactor R2 without support showed a removal efficiency of COD less than 85% at similar conditions. Biomass concentration in R1 was of 2.49 g VSS L⁻¹ on 100th day. The skeleton of *Opuntia imbricata* included in the R1 provided large surface areas for the colonization of microorganisms, which increased the biomass concentration and allowed the growth of a thin biofilm. The removal rate of COD in R1 reactor increased from 24-60.425 mg COD L⁻¹ h⁻¹, when the Organic Loading Rate (OLR) was increased from 0.44-1.32 kg COD m⁻³ d⁻¹. R1 reactor showed high COD removal rate and demonstrated good operational stability. R1 reactor achieved a higher oxygen transfer coefficient of 0.3208 min⁻¹ with air flow rate of 2 lpm. The DO concentration in the R1 increased from 0.41-4.82 mg L⁻¹ with an air flow rate of 0.25 at 2 lpm. Increasing air flow rate in the R1 increased the amount of dissolved oxygen and improved the performance of R1 reactor. The main advantages of the natural support used for packed reactor were the lower hydraulic retention time and the small size of reactor. This new bioreactor could be an alternative for municipal wastewater treatment at a low cost.

ACKNOWLEDGEMENT

We thank Dr. Manuel García Hipólito and Dr. José Guzmán Mendoza (Instituto de Investigaciones en

Materiales, Universidad Nacional Autónoma de México) for their collaboration in the preparation of samples and taking of micrographs and CONACYT (Consejo Nacional de Ciencia y Tecnología) for financial support.

REFERENCES

- APHA, 1998. Standard Methods for the Examination of Water and Wastewater. 20th Edn. Washington, DC, USA
- Arslan, A. and S. Ayberk, 2003. Characterization and biological treatability of "Izmit industrial and municipal wastewater treatment plant" wastewaters. *Water SA.*, 29: 451-456.
- Camargo, S.A.R. and E.A.A. Nour, 2001. Bamboo as an anaerobic medium: effect of filter column height. *Wat. Sci. Technol.*, 44 (4): 63-70.
- Casey, E., B. Glennon and G. Hamer, 1999. Oxygen mass transfer characteristics in a membrane-aerated biofilm reactor. *Biotechnol. Bioeng.*, 62: 183-192.
- DeMoyer, C.D., E.L. Schierholz, J.S. Gulliver and S.C. Wilhelms, 2003. Impact of bubble and free surface oxygen transfer on diffused aeration systems. *Water Res.* 37: 1890-1904.
- Fonade, C., N. Doubrovine, C. Maranges and J. Morchain, 2001. Influence of a transverse flowrate on the oxygen transfer performance in heterogeneous aeration: Case of hydro-ejectors. *Water Res.*, 35: 3429-3435.
- Hu, L., J. Wang, X. Wen and Y. Qian, 2005. Study on performance characteristics of SBR under limited dissolved oxygen. *Process Biochem.*, 40: 293-296.
- Karimniaae-Hamedani, H.R., K. Kanda and F. Kato, 2003. Wastewater treatment with bacteria immobilized onto a ceramic carrier in an aerated system. *J. Biosci. Bioeng.*, 95: 128-132.
- Klein, J., M. Rosenberg, J. Markos, O. Dolgos, M. Krosiak and Lu. Kristofikova, 2002. Biotransformation of glucose to gluconic acid by *Aspergillus Niger*, study of mass transfer in an airlift bioreactor. *Biochem. Eng. J.*, 10: 197-205.
- Liu, Y. and H. Tay, 2002. The essential role of hydrodynamic shear force in the formation of biofilm and granular sludge. *Water Res.*, 36: 1653-1665.
- Liawska-Bizukojc, E., M. Bizukojc and S. Ledakowicz, 2002. Kinetics of the aerobic biological degradation of shredded municipal solid waste in liquid phase. *Water Res.*, 36: 2124-2132.
- Mosquera-Corral, A., A. Montras, J.J. Heijnen and M.C.M. Van Loosdrecht, 2003. Degradation of polymers in a biofilm airlift suspension reactor. *Water Res.*, 37: 485-492.
- Nicolella, C., M.C.M. Van Loosdrecht and S.J. Heijnen, 2000. Particle-based biofilm reactor technology. *Trends Biotechnol.*, 18: 312-320.
- Picanco, A.P., M.V.G. Vallero, E.P. Gianotti, M. Zaiat and C.E. Blundi, 2001. Influence of porosity and composition of supports on the methanogenic biofilm characteristics developed in a fixed bed anaerobic reactor. *Water Sci. Technol.*, 44: 197-204.
- Rodde-Pellegrin, L., C. Wisniewski, A. Grasmick, A. Tazi-pain and H. Buisson, 2002. Respirometric needs of heterotrophic populations developed in an immersed membrane bioreactor working in sequenced aeration. *Biochem. Eng. J.*, 11: 2-12.
- Rodgers, M., X. Zhan and A. Casey, 2004. Oxygen transfer and industrial wastewater treatment efficiency of a vertically moving biofilm system. *Water Air Soil Pollut.*, 151: 165-178.
- Rodriguez-Martínez, J. and Y. Garza-García, 2002. Application of *Opuntia imbricata* (teasel), as support for the immobilization of microbial consortiums and the removal of different organic and inorganic contaminants contained in residual waters. PN: MXNL02000043 A.
- Rosenberger, S., U. Kruger, R. Witzig, W. Manz, U. Szwczyk and M. Kraume, 2002. Performance of a bioreactor with submerged membranes for aerobic treatment of municipal wastewater. *Water Res.*, 36: 413-420.
- Sarti, A., L. Tavares, E. Foresti and M. Zaiat, 2001. Influence of the liquid-phase mass transfer on the performance of a packed-bed bioreactor for wastewater treatment. *Bioresour. Technol.*, 78: 231-238.
- SAS®, 1996. Institute Inc., SAS® User's Guide: Statistics. Versión 6.12. Cary, NY, USA.
- Sorlini, C., G. Ranalli and S. Merlo, 1990. Microbiological aspects of anaerobic digestion of swine slurry in upflow fixed-bed digesters with different packing materials. *Biol. Wast.*, 31: 231-239.
- Souza, R.R., I.T.L. Bresolin, T.L. Bioni, M.L. Gimenes and B.P. Dias-Filho, 2004. The performance of a three-phase fluidized bed reactor in treatment of wastewater with high organic load. *Braz. J. Chem. Eng.*, 21: 219-227.
- Steel, R.G.D. and J.H. Torrie, 1980. *Bioestadística. Principios y procedimientos.* McGraw-Hill de México, México, D.F.
- Tawfik, A., B. Klapwijk, F. el-Gohary and G. Lettinga, 2002. Treatment of anaerobically pre-treated municipal sewage by a rotating biological contactor. *Water Res.*, 36: 147-155.
- U.S. EPA., 1999. Wastewater technology Fact sheet: *Fine bubble aeration.* United States Environmental Protection Agency. Office of Water. Washington, D.C. EPA 832-F-99-065.

- Wäsche, S., H. Horn and D.C. Hempel, 2002. Influence of growth conditions on biofilm development and mass transfer at the bulk/biofilm interface. *Water Res.*, 36: 4775-4784.
- Yang, Y., C. Tada, K. Tsukahara and S. Sawayama, 2004. Methanogenic community and performance of fixed- and fluidized-bed reactors with reticular polyurethane foam with different pore sizes. *Mater. Sci. Eng.*, 24 (6-8): 803-813.
- Zhou, P., J. He and Y. Qian, 2003. Biofilm airlift suspension reactor in the treatment of municipal wastewater. *Water Air Soil Pollut.*, 144: 81-100.
- Zita, A. and M. Hermansson, 1997. Effects of bacterial cell surface structures and hydrophobicity on attachment to activated sludge flocs. *Applied Environ. Microbiol.*, 63: 1168-1170.