Technical Article

Evaluation of the phytotoxicity of landfill leachate treated with a Rotating Biological Reactor

Avaliação da fitotoxicidade de lixiviado de aterro tratado com um Reator Biológico Rotativo

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ABSTRACT

One of the major drawbacks of this final disposal technique is leachate production, which occurs due to the degradation of organic matter and rainwater percolation. The leachate composition is guite varied, with high organic load and various compounds that may be toxic to the environment. To reduce the polluting potential of the leachate, it must be subjected to an appropriate treatment. Biological treatments are widely used in the treatment of leachate, although these technologies have some drawbacks. As an alternative to conventional biological treatments, rotating biological reactors are used. The objective of this study was to determine the efficiency of a pilot scale rotating biological reactors in the removal of biochemical oxygen demand, chemical oxygen demand, and phosphorus from the landfill leachate. Effluent toxicity and rotating biological reactors's ability to remove this toxicity were also verified by calculating the germination index of lettuce (Lactuca sativa) and cucumber (Cucumis sativus) seeds. The reactor showed an average efficiency of 75.99% of biochemical oxygen demand removal, 33.53% of chemical oxygen demand removal, and 16.04% of phosphorus removal. In fact, the cucumber and lettuce seeds proved sensitive to the toxicity of the effluent. For crude leachate, the germination index values of lettuce and cucumber were 13.28 and 49.61%, respectively. In this study, rotating biological reactors obtained a good efficiency in the removal of toxicity from leachate. The germination index of the seeds for the treated effluent was 60.9% for lettuce and 82.49% for cucumber.

Keywords: biological treatment; germination index; toxicity; leachate.

RESUMO

Diante do crescimento populacional acelerado e do desenvolvimento econômico, grande quantidade de resíduos tem sido gerada nos grandes centros urbanos. Esses resíduos devem ser coletados e dispostos adequadamente, sendo os aterros sanitários os locais mais adequados para a sua disposição. Um dos grandes inconvenientes dessa técnica de disposição final é a produção de um líquido lixiviado, proveniente da degradação da matéria orgânica e da percolação da água da chuva. A composição do lixiviado é bastante variada, com elevada carga orgânica e diversos compostos que podem ser tóxicos ao meio ambiente. Para determinar a toxicidade do lixiviado são amplamente utilizados testes fitotoxicológicos, como o índice de germinação de sementes. Com o intuito de reduzir o potencial poluidor do lixiviado, este deve ser submetido a um tratamento adequado. Tratamentos biológicos são bastante utilizados, mas possuem algumas desvantagens. Como alternativa aos tratamentos biológicos convencionais, surgem os reatores biológicos rotatórios. O objetivo deste estudo foi determinar a eficiência de uns reatores biológicos rotatórios em escala piloto na remoção da demanda bioquímica de oxigênio, da demanda química de oxigênio e de fósforo do lixiviado do aterro. Também foi verificada a toxicidade do efluente e a capacidade dos reatores biológicos rotatórios para remover essa toxicidade por meio do cálculo do índice de germinação de sementes de alface (Lactuca sativa) e pepino (Cucumissativus). O reator utilizado neste trabalho obteve eficiência média de 75,99% de remoção de demanda bioquímica de oxigênio, 33,53% de remoção de demanda química de oxigênio e 16,04% de remoção de fósforo. As sementes de alface e pepino mostraram-se bastantes sensíveis à toxicidade do efluente. Para o lixiviado bruto, os índices de germinação da alface e do pepino foram, respectivamente, 13,28 e 49,61%. Neste estudo, os reatores biológicos rotatórios obtiveram boa eficiência na remoção da toxicidade do lixiviado. O índice de germinação das sementes para o efluente tratado foi de 60,9% para a alface e de 82,49% para o pepino.

 $\textbf{Palavras-chave:} \ tratamento \ biológico; \'indice de germina \\ \varsigma\~ao; toxicidade; lixiviado.$

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INTRODUCTION

The fast economic development in recent years has resulted in a huge generation of municipal and industrial solid waste (SRIVASTAVA *et al.*, 2015). It is necessary to dispose the waste generated in the cities in suitable places. According to the study by Zhang *et al.* (2016), landfills are the most common final disposal techniques for urban solid waste and are found in several locations around the world. Other alternatives are needed for sustainable disposal of the industrial wastewater (Carlos *et al.*, 2017a).

One of the T drawbacks of waste disposal in landfills is the production of leachate. This leachate is produced by the percolation of rainwater during the process of decomposition of the organic matter present in the residues (TIGINI et al., 2014). The composition of this type of effluent is quite varied and may contain high concentrations of dissolved organic compounds, xenobiotic compounds, inorganic salts, ammonia, heavy metals, and other toxic compounds (CHRISTENSEN et al., 2001; PIVATO et al., 2006; WANG et al., 2010).

The pollutants' toxicity can be measured through standard toxicity tests using bacteria, crustaceans, algae, and seed germination tests (MALACHOVA *et al.*, 2013). In phytoindicative methods, observation of plant performance gives us information about environmental conditions (BIAŁOWIEC, 2015).

Phytotoxicity is the toxic action induced in plants, by one or more substances, which impairs or inhibits its development and/or germination, i.e., the toxic action of heavy metals. It is the most used bioindicator in the evaluation of the animal waste toxicity, to assess its polluting potential (MENDES, 2011).

When physicochemical parameters are not reliable, i.e., to assure that residues are environmentally safe for agricultural use (JURADO *et al.*, 2014), it is recommended to use phytotoxicity tests as supplementary methods (ZUCCONI *et al.*, 1981; TIQUIA *et al.*, 1998; OZORES-HAMPTON *et al.*, 2013; DELGADO *et al.*, 2010; GALENDE *et al.*, 2014). There are several advantages of using plants, such as *Lactuca sativa*, in phytotoxicity tests;. The tests are reliable, quick, and simple, as well as of low cost and do not require a lot of equipment (CHARLES *et al.*, 2011).

The germination index (GI) is a usual phytotoxicity indicator (VENEGAS *et al.*, 2018). The GI is an index that takes into consideration both seed germination and root growth (OLESZCZUK, 2010; RODRIGUEZ-RODRIGUEZ *et al.* 2011).

The presence of refractory compounds in the landfill leachate can become a source of soil and water contamination (HAN *et al.*, 2014), as well as hinder the biological treatment system, reducing its efficiency. Recalcitrant compounds are hardly identifiable, but their impact may be potentially devastating due to their toxicity (TIGINI *et al.*, 2014).

The landfill leachate is characterized as a complex of different mixtures of recalcitrant organic and inorganic contaminants, such as humic and fulvic acids (FAN *et al.*, 2006), polycyclic aromatic hydrocarbons (JONSSON *et al.*, 2003; MARTTINEN *et al.*, 2003), pesticides (ÖMAN *et al.*, 2008), trace elements (JENSEN *et al.*,1999), and high levels of ammoniacal nitrogen (AZIZ *et al.*, 2010).

To reduce the polluting potential of the leachate and its toxicity to the environment, the effluent must be treated properly prior its disposal in nature. Hilles *et al.* (2015) highlighted the biological, chemical, and physical processes as the most used for the treatment of leachates. Zhao *et al.* (2013) mentioned that the main difficulties found in the treatment of leachate by conventional techniques are associated with the high concentration of organic matter in the slurry and the presence of toxic organic compounds and heavy metals.

Biological treatment of landfill leachate encloses suspended growth biomass systems, such as activated sludge, sequencing batch reactors, and lagooning (CORTEZ, 2010). The treatment also encloses attached growth biomass systems, i.e., rotating biological contactors, trickling filters, and suspended carrier biofilm reactors (CORTEZ, 2010).

According to the study by Tigini *et al.* (2014), innovative techniques for the treatment of leachates are necessary for the removal of macropollutants and micropollutants. As an alternative to conventional biological treatments, rotating biological reactors (RBRs) are used. The RBR consists of a series of disks regularly arranged on a rotating axis, which rotate at low speed under aerobic and anaerobic conditions, facilitating the formation of a biofilm, responsible for the effluent treatment (FONSECA *et al.*, 2010).

Biofilm reactors, such as RBR, offer some advantages over suspended biomass reactors, such as increased resistance to toxic compounds, organic charge, and temperatures shock (ARVIN *et al.*, 1990), due to the complexity of the biofilm, which are a heterogeneous combination of cells that are influenced by the environment and conditions they are subjected (BUTLER *et al.*, 2014). The RBRs can operate in conditions of BOD removal, which is associated with nitrification, and also in nitrification/denitrification processes (SCHWERZ, 2012).

Other advantages of this type of reactor include a high contact area of the disks with the effluent, simple design and operation, reduced area requirements, low operating and maintenance costs, and high treatment efficiency (CORTEZ et al., 2008; GUIMARÃES et al., 2005). Therefore, the objective of this work was to analyze the phytotoxicity of leachate from a landfill and to evaluate the efficiency of an RBR in the reduction of organic load and decrease in leachate phytotoxicity.

METHOD

The leachate used came from a landfill implemented in 2010, which received up to 170 tons of solid waste per day. The leachate was collected weekly, before the treatment system, stored in gallons of 25 L, and taken to the Laboratory of Water and Effluent Treatment of the Federal University of Pelotas to conduct the experiment.

Effluent treatment

The RBR was constructed from fiberglass and resin (Figure 1), with a useful volume of $24\,\mathrm{L}$ and a metal axis containing 30 disks evenly arranged in 2 groups of 15 disks. Each disk had 0.3 m diameter, spaced apart by 1 cm, and the phases were covered by plastic polymer foam. The specific area was $0.14\,\mathrm{m}^2$, totaling a surface area of $4.24\,\mathrm{m}^2$. The system had 40% of its area submerged. The rotation was performed using an electric motor with speed variation, operating at a speed of 1 rpm.

The feeding of the RBR was performed through a peristaltic pump with a flow rate of 0.33 $\rm L.h^{-1}$, determined to obtain a hydraulic retention time (HRT) of 72 h.

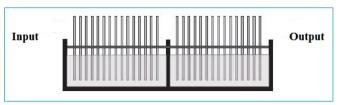


Figure 1 - Schematic drawing of the rotating biological reactors.

The RBR was inoculated with biomass from an activated sludge reactor operating in a parboiled rice industry with a volatile suspended solid concentration of 4200 mg.L⁻¹. The adaptation period was 2 weeks, and the data collection period was 4 weeks, divided into 8 collections. Samples were taken two times per week at the inlet and outlet of the reactor. The parameters of biochemical oxygen demand (BOD), chemical oxygen demand (COD), total phosphorus, and pH were evaluated in duplicates, according to the methodology described in Standard Methods for the Examination of Water and Wastewater (EATON *et al.*, 2005).

Phytotoxicity of the effluent

Phytotoxicity analysis was performed with lettuce (*L, sativa*) and cucumber (*Cucumis sativus*) seeds. The seeds were purchased from an agricultural store. The seed germination tests were performed in triplicate and followed the methodology developed by the Environmental Protection Agency (EPA, 2012). Ten cucumber seeds and ten lettuce seeds were placed in Petri dishes of 9 cm diameter each, containing one layer of filter paper and 5 mL of effluent. For the control sample, distilled water was used.

After the plates were prepared, they were incubated in the dark at a temperature of $25\,^{\circ}\text{C}$ for a period of $72\,\text{h}$. At the end of this period, the germinated seeds were counted, and the root size was measured. With the values found, the GI was calculated by using the following Equation 1:

$$GI = \frac{G * Ls}{Gc * Lc} \tag{1}$$

where:

GI = germination index (%).

 G_s and L_s = seed germination (%) and root length (mm) for the samples, respectively. G_c and L_c = seed germination (%) and root length (mm) for the control sample, respectively.

To facilitate the comparison between the different tests, the GI was expressed as a percentage of GI of the control sample, which was stipulated as 100.

Statistical analysis

All the data and experiments were done in duplicate. Statistical analysis of the data was done using the Statistical Analysis Software (WINSTAT) (MACHADO,

2001) and the analysis of variance (F test), according to the recommendations by Silva (1997). The significant differences were determined by Tukey's multiple comparison tests, and the comparison of the averages was analyzed using the Duncan's test.

RESULTS AND DISCUSSION

The mean values of pH, BOD, COD, and P-PO $_4$ contents found for the crude leachate (CL) and for the effluent treated with RBR (treated leachate [TL]), and the efficiency of reduction of the parameters evaluated after treatment in the RBR are presented in Table 1. The pH of the CL ranged from 7.8-8.7; BOD ranged from 701.4-2124.3 mg.L $^{-1}$; COD ranged from 2788.1-5863.9 mg.L $^{-1}$, and phosphorus ranged from 24.7-38.3 mg.L $^{-1}$. At the exit of the RBR, the pH ranged from 8.5-9.4; BOD ranged from 93.1-511.8 mg.L $^{-1}$; COD ranged from 1301.1-4607.3 mg.L $^{-1}$, and phosphorus ranged from 18.2-36.6 mg.L $^{-1}$.

Significant differences were observed by the Duncan's test at 1% probability, for the pH at the RBR input and output, except for the third collection. The RBR provided an increase in the pH of the effluent. This behavior corroborates the work by Cortez *et al.* (2010), who treating leachate from landfill in contact reactor found the values between 6.4 and 7.3 for the CL and from 7.8 – 9.4 for the TL. This behavior, according to Cortez *et al.* (2011), is due to the consumption of hydrogen ions resulting from the conversion of N-NO₃⁻ to nitrous oxides in the reactor due to denitrification process. However, since no analysis of nitrogen was carried out, it is not possible to allege that this behavior is due to the denitrification process.

The RBR provided a significant reduction in COD and BOD contents in all samples (Table 1). Removal efficiencies were obtained between 54% and 88% for BOD and 21% and 53% for COD (Table 1). The RBR obtained average BOD and COD removal values of 76.0% and 33.5%, respectively. The average BOD removal is in compliance with the standard established by CONAMA Resolution n°. 430/2011, which determines that at least 60% of BOD must be removed. Figure 2 presents the average BOD values for both CL and TL.

Table 1 - Analysis of the average of the parameters, such as biochemical oxygen demand, chemical oxygen demand, and phosphorus, during rotating biological reactors operation and the efficiency of reduction of the parameters evaluated after treatment in the rotating biological reactors.

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Collect	pН		BOD (mg L ⁻¹)		E%	COD (mg.L ⁻¹)		E%	P-PO4 (mg.L ⁻¹)		E%
	CL	TL	CL	TL	BOD	CL	TL	COD	CL	TL	P-PO ₄
1	8.3b*	8.9a	701.4a	93.1b	86.7	2788.1a	1301.1b	53.1	24.7	22.1	10.3
2	8.5b	9.4a	644.6a	77.0b	88.0	4271.8a	2091.1b	50.7	27:1b	36.6a	-36.8
3	8.7	8.5	1135.1a	511.8b	54.7	3274.1a	2354.8b	27.8	37.5a	18.2b	50.9
4	7.8b	8.8a	1524.3a	219.8b	85.6	3366.0a	2560.0b	23.6	34.3a	24.7b	28.1
5	8.4b	9.2a	1333.8a	373.0b	72.0	5358.3a	3493.3b	34.8	34.3a	23.9b	30.6
6	8.2b	9.2a	1350.0a	391.4b	71.0	5695.8a	3843.6b	32.5	38.3a	30.3b	20.4
7	8.1b	9.3a	1645.9a	429.9b	73.8	4638.8a	3495.2b	24.7	25.5	26.6	-4.8
8	8.2b	9.1a	2124.3a	508.9b	76.1	5863.9a	4607.3b	21.3	31.9a	22.2b	29.8
Average	8.3 ± 0.33	9.1 ± 0.28	1307.4 ± 434.96	325.6 ± 168.86	76.0	4407.1 ± 1195.81	2968.3 ± 914.34	33.6	31.7 ± 7.99	25.6 ± 7.18	16.0

(E%): efficiency of reduction; BOD: biochemical oxygen demand; COD: chemical oxygen demand; CL: crude leachate; TL: treated leachate.

^{*}Average followed by the same lowercase letter, within the same line, for the same parameter, does not present significant difference by Duncan's test 1% of probability.

Pérez (2010), using an RBR for landfill leachate treatment, obtained removal efficiencies of 95% for BOD and 68.3% for COD. Jemec *et al.* (2012), treating landfill leachate in an existing biological wastewater treatment plant, obtained 71% efficiency in the removal of BOD and 54% in the removal of COD. However, Cassano *et al.* (2011), using a biological system to treat landfill leachate, obtained efficiencies of 98% for BOD and 54% for COD.

The average removal of P-PO $_4$ obtained in the RBR was 16%. Pérez (2010), working with RBR, obtained phosphorus removal of 38%. The RBR was not stable

for the removal of phosphorus, due to the variability of the removal efficiency. Increases in phosphorus levels were observed in two collections (collections 2 and 7). This may have been due to problems during the analysis of phosphorus and contamination of glassware. Considering only the collections where phosphorous removal was verified, the removal efficiencies varied between 10.3 and 50.8%. Schwerz (2012) obtained similar results, where nitrogen analyses were compromised by the sludge detachment of the disks. Figure 3 presents the average P-PO₄ values for both CL and TL.



Figure 2 - Average biochemical oxygen demand values of crude and treated leachate.

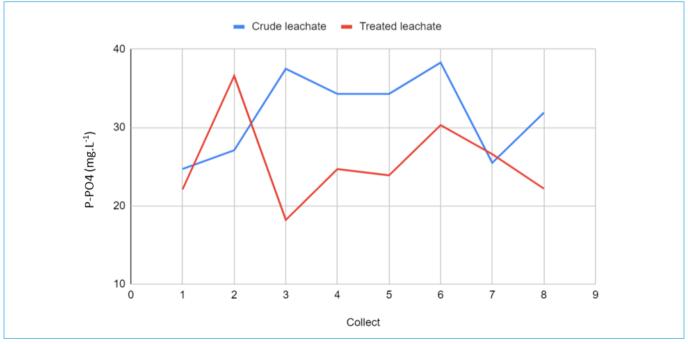


Figure 3 - Average P-PO₄ values of crude and treated leachate.

For the GIs of lettuce and cucumber seeds, presented in Table 2, no significant differences were observed between the treatment interactions and the date of collection, allowing to conclude separately about the factors.

The average GI of the lettuce before treatment was 13.28%. After treatment with the RBR system, the phytotoxicity decreased and the GI of the lettuce increased significantly to 60.9%. The cucumber was less sensitive to the toxicity of landfill leachate. The average GI of the samples with crude effluent was 49.61%, a higher value when compared to the average GI of the lettuce. As occurred with the lettuce seeds, the TL had lower toxicity to cucumber seeds. The average GI of cucumber seeds in the treated effluent was 82.49%. Roig *et al.* (2012) considered that a GI below 50% indicates high toxicity effects.

Table 2 - Evaluation of the germination index average (%) of lettuce and cucumber seeds.

Collection	Lett	uce	Cucumber			
Collection	CL	TL	CL	TL		
1	14.26b*	132.14a	26.32b	113.34a		
2	34.32	46.90	67.63	70.07		
3	34.97b	152.05a	73.89	81.68		
4	22.67b	53.76a	38.62b	64.78a		
5	0.00b	46.14a	47.67b	60.00a		
6	0.00b	16.61a	43.45b	97.12a		
7	0.00b	18.81a	23.89b	73.47a		
8	0.00b	20.79a	75.41b	99.43a		
Average	13.28	60.90	49.61	82.49		

*Average followed by the same lowercase letter, within the same line;, for the same parameter, it does not present significant difference by Duncan's test 1% of probability. CL: crude leachate; TL: treated leachate.

Kalčíková *et al.* (2012), using lentil, wheat, and mustard seeds, found that the treated landfill leachate had lower toxicity to the seeds, with mustard seed being the most sensible to the toxicity of the landfill leachate.

CONCLUSIONS

The RBR had a satisfactory efficiency for reducing BOD, greater than the required by the Brazilian legislation; however, the same efficiency cannot be pronounced in relation to COD and phosphorus parameters. Nevertheless, it can be considered a satisfactory performance due to its advantages regarding conventional biological treatments, such as the low cost of operation and maintenance, low area requirements, and easy operation. Therefore, this type of reactor is indicated for the treatment of landfill leachate.

The RBR used in the treatment of leachate presented great effluent toxicity removal capacity. After the analysis of the phytotoxicological tests, we can conclude that the seeds used in the study (i.e., cucumber and lettuce) presented good sensitivity to the toxicity of the landfill leachate. The lettuce showed to be more sensitive to the toxicity of the leachate used in this study, showing a better indicator than the seed of cucumber.

AUTHORS' CONTRIBUTIONS

HOSS, L.: Writing – original draft; Writing – review & editing. ZANATTA, R.: Conceptualization, Data curation, Formal Analysis. QUADRO, M.: Writing – Conceptualization, original draft Writing – review & editing. CORRÊA, E.K: Formal Analysis. ANDREAZZA, R.: Writing – original draft; Writing – review & editing.

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