frontiers | Frontiers in Energy Research

TYPE Original Research
PUBLISHED 19 March 2025
DOI 10.3389/fenrg.2025.1549247

Check for updates

OPEN ACCESS

EDITED BY
Tingting You,
Beijing Forestry University, China

REVIEWED BY
Koray Alper,
Bülent Ecevit University, Türkiye
Sikandar I. Mulla,
REVA University, India

*CORRESPONDENCE
Maitreyie Narayan,
maitreyie25apr@gmail.com
Amit Mittal,
amitforestry26@gmail.com
Nandan Singh,
nandanforestry@gmail.com

RECEIVED 20 December 2024
ACCEPTED 24 February 2025
PUBLISHED 19 March 2025

CITATION
Narayan M, Solanki P, Srivastava RK, Mittal A, Singh N, Akhter F and Siddiqui S (2025) Optimizing COD removal, lignin degradation and electricity generation from pulp and paper industry wastewater by CW-MFC using box-behnken design.
Front. Energy Res. 13:1549247.
doi: 10.3389/fenrg.2025.1549247

Optimizing COD removal, lignin degradation and electricity generation from pulp and paper industry wastewater by CW-MFC using box-behnken design

Maitreyie Narayan[1]*, Praveen Solanki[2], Rajeev Kumar Srivastava[3], Amit Mittal[4]*, Nandan Singh[1]*, Fareeda Akhter[5] and Sazada Siddiqui[6]

[1]Department of Forestry and Environmental Science, Kumaun University, Nainital, Uttarakhand, India, [2]Krishi Vigyan Kendra, Narsinghpur, MP, India, [3]Department of Environmental Science, GBPUA&T, Pantnagar, Uttarakhand, India, [4]School of Allied Sciences, Graphic Era Hill University Bhimtal Campus, India, [5]Department for Molecular Biology, Central University of Jammu, Jammu Kashmir, India, [6]Department of Biology, College of Science, King Khalid University, Abha, Saudi Arabia

The novel and distinctive microbial fuel cell coupled with a built wetland (CW-MFC) is a technology that treats different kinds of wastewater and produces electricity. In comparison to traditional MFCs, it is also easier to maintain and has better wastewater treatment efficiency. This study looked into the impact of wetland plants and hydraulic residence time (HRT) on treatment and energy production. There was also investigation into the features of wastewater deterioration. The lignin-degrading microcosm uses immobilized peroxidase enzyme that has been synthesized on sodium alginate beads. Using the Response Surface Methodology model, for optimization of lignin degradation, power production, and COD removal by peroxidase enzyme was mathematically investigated. Following optimization, the highest treatment given to the wastewater was 74.99%, 3.08 days of HRT, and 10 mg/L for peroxidase concentration andthe results we got were: lignin degradation was 751.1 mg/L, voltage generation was 32.09 mV and COD removal was 245.07 mg/L. The desirability of model was 0.809.

KEYWORDS

constructed wetland-microbial fuel cellWastewater treatment, electricity generation, response surface methodology, box-behnken design, lignin degradation

1 Introduction

Indian paper industry highly dispersed according to Indian geography and is also one of the most fragmented industries, with production capacities ranging from 5 tons per day to 1,650 tons per day. During the decade from 2010 to 2020, the Indian paper industry had significant expansion, resulting in a twofold rise in output from 10.99 million tonnes in 2010 to 21.36 million tonnes in 2020 (CPPRI Annual Report 2020–21). Today, there are around 900 paper mills located throughout the country. The entire installed capacity in the sector is 23.99 million tonnes per year, and the production is 21.36 million tonnes

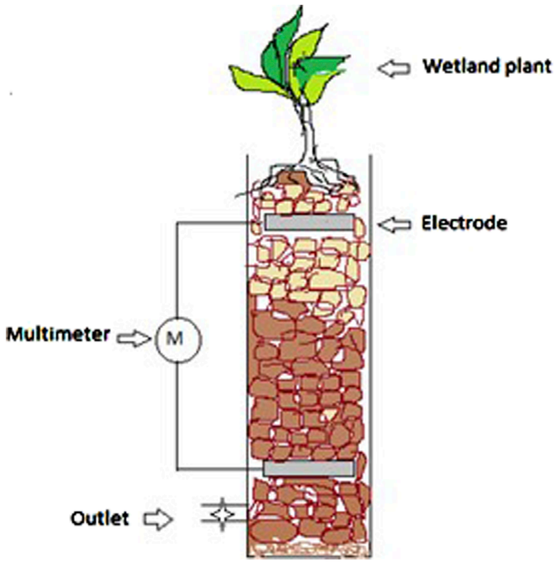

FIGURE 1
Schematic of novel constructed wetlands cum microbial fuel cell.

per year. Additionally, due to population growth, India's consumption of paper has climbed from 16.91 million tonnes in 2016–17 to 22.83 million tonnes in 2019–20. During this time, India's paper consumption grew at a CAGR of 6%, compared to a global growth rate of 3%, making India one of the world's fastest growing paper markets (CPPRI Annual Report 2020–21). Due to the increase in growth of pulp and aper industry there is subsequent increase of demand of water and raw material for its operation and production.

Pulp and paper industry is the third largest water consuming industrialized sector in the country. Fresh water consumption in wood based, agro-based and waste paper based mills is 125–200, 125–225, 75–100 m^3 per tonne of paper respectively (Chakrabarti 2006). With average water consumption was 151 m^3 per tonne of paper (Soloman et al., 2009).

Based on the raw materials used and the manufacturing process adopted, pulp and paper industry produces relatively large amounts of both wastewater and solid wastes (Narayan et al., 2018).

Annually, pulp and paper industry generates millions of tons of toxic and hazardous wastewater which directly affect the aquatic biodiversity (Sharma et al., 2022). High pH, turbidity, color, temperature, BOD, COD, nutrients and the addition of toxic and persistent pollutants contaminate the vicinity aquatic water bodies (Sharma et al., 2021). On-site, reuse and recycling, and also modifications in the technology are among the most efficient economic and environmental options dealing with the pollutants that are produced (Kamali et al., 2016). In this regard, measures for minimizing the produced wastes, and recovery of energy and unavoidable wastes have been introduced and adopted by pulp and paper industry (Ericsson et al., 2011). However, the outdoor wastewater treatment plants are still the main ways to deal with the pollutants from pulp and paper industries (Tiku et al., 2007). So far, various types of treatments, i.e., primary, secondary, and tertiary have been developed and applied in order to enhance the treatment efficiency of both pulp and paper industry wastewater (MG/LW) and sludge (MG/LS) with the intend of dipping the amount of the produced ultimate wastes, and also to put off the probable successive toxic effects induced by the presence of harmful compounds when released into the receiving environment (Waye et al., 2014).

Microbial fuel cell (MFC) is a novel bioelectrochemical device that can utilize various organic substrates under anaerobic conditions and directly transform chemical energy to electricity via a series of electrochemical reactions catalyzed by microorganisms (Logan et al., 2006). There is currently a movement in expanding the use of MFCs to new application-treatment of recalcitrant environmental pollutants coupled with bioelectricity generation (Luo et al., 2009; Morris et al., 2009; Li et al., 2010; Liu et al., 2013). Recent years have seen a significant increase in interest in microbial fuel cells (MFCs), which may recover energy from waste organic sources and transform chemical energy into electrical energy during wastewater treatment (Logan 2008; Puig et al., 2012). MFCs have recently showed the potential to produce energy while also degrading several biorefractory pollutants (Luo et al., 2009; Morris et al., 2009). Dual chamber MFC (composed of an anode chamber and a cathode chamber separated by a proton exchange membrane) and membrane-less single chamber MFC are both frequently utilized in wastewater treatment (Kiely et al., 2011; Sun et al., 2009). According to several studies, membrane-less single chamber MFCs have more advantages than dual-chamber MFCs, such as low cost, simple design, and high power density (Liu and Logan 2004). In particular, this study aims to demonstrate the optimization of lignin degradation, with the help of immobilized peroxidase enzyme, (Bankole et al., 2022) revealed that laccase and lignin peroxidase played vital role in the lignin degradation. COD removal and electricity generation in terms of voltage and current using response surface methodology. Response surface methodology (RSM) is used to investigate the interaction between the independent process variables and estimate the best conditions that will result in the highest yield. The study demonstrates the use of modeling techniques for identifying optimal conditions and capturing inherent characteristics for better yield predictions, which could reduce the time-consuming and energy-intensive scale-up experiments before moving on to real-time applications. The main findings of this research study may lead to the development of sustainable voltage and current along with the treatment of wastewater.

2 Materials and methods

2.1 Wastewater collection

For the present study, samples were collected from the Century Pulp and Paper Mill, Lalkua, Nainital (Uttarakhand) situated at about 7 km from G.B. Pant University of Agriculture and Technology, Pantnagar on the Bareilly-Nainital highway. This industry was established in June 1984 and is engaged in the manufacture of some finest varieties of writing and printing paper. The mill has three drains, which combine to form one main effluent channel (combined effluent). The major sampling source for pulp and paper industry wastewater were from the black liquor area. In most pulp and paper mills, raw materials go through a series of unit activities such as raw material preparation, pulp digestion, washing,

10.3389/fenrg.2025.1549247

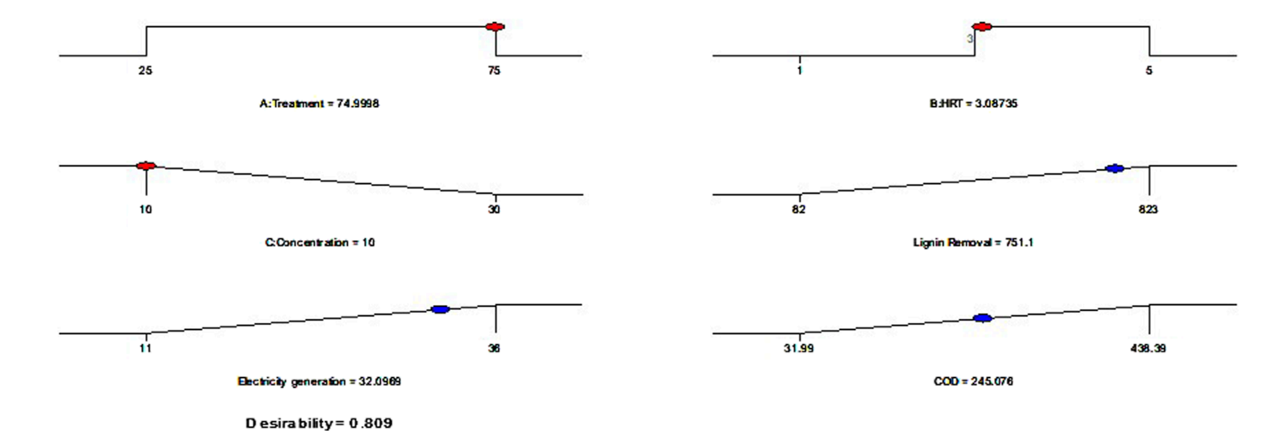

FIGURE 2
Copper and zinc electrodes.

TABLE 1 Characteristics of paper and pulp industry effluent at different months.

Months→Parameters↓	March (2017)	May (2017)	July (2017)	September (2017)	November (2017)	January (2018)
pH	8.0	8.1	7.6	7.6	7.9	8.2
EC (ds/m)	1.94	1.60	1.72	1.89	2.01	2.03
TS (mg/L)	1,408	1,462	1,389	1,316	1,525	1,558
TSS (mg/L)	208	211	163	176	316	335
TDS (mg/L)	1,200	1,251	1,226	943	1,209	1,123
COD (mg/L)	1958.3	2014.6	1855.2	1909.3	2057.1	2,123.7
BOD (mg/L)	621.5	609.1	614.2	662.3	714	676
TN (mg/L)	86.34	70.15	68.37	63.52	71.05	73.14
Chloride *(mg/L)*	731.6	726.1	719.2	684.5	693.7	701.9
Sodium (mg/L)	624.1	613.2	539.1	562.7	608.3	581.6
Potassium (mg/L)	133.5	101.7	168	154.3	128.6	117.2
Sulphate (mg/L)	175.1	163.8	151.7	153.3	149.1	137.8
Chromium (mg/L)	3.32	3.51	3.90	3.13	3.60	3.36
Iron (mg/L)	7.55	8.10	7.45	7.68	8.06	7.33
Lead (mg/L)	2.04	2.01	2.09	2.14	2.19	2.05
Cadmium (mg/L)	0.74	0.89	0.71	0.85	0.81	0.70

bleaching, and eventually papermaking. The pulping and bleaching operations use a lot of freshwater and produce a lot of contaminated wastewater. The features of wastewater are usually determined by the process, and hence the wastewater collected from the black liquor area. Tap water sample were taken from the university premises. To maintain the integrity of the sample, appropriate selection of container, pre-treatment of container is necessary. To minimize the potential for volatilization or biodegradation between sampling and analysis, the samples were kept at cool place without freezing. Therefore, the collected samples were kept at refrigeration system set at 4°C. The wastewater was characterized in different months by using the standard methods (APHA 2012).

TABLE 2 The lignin content of pulp and paper industry.

S. No.	Month/Year	Lignin (mg/L)
1	March/2017	1,319
2	May/2017	1,304
3	July/2017	1,172
4	September/2017	1,140
5	November/2017	1,263
6	January/2018	1,296

2.2 CW-MFC construction

A vertical flow constructed wetland microcosm was fabricated in the laboratory. CW-MFC was designed and fabricated using PVC pipes with diameter of 18 cm and height of 100 cm. The bottom layer is filled with glass beads with diameter of 10 mm up to 10 cm high from the bottom to ensure even distribution of influent. After that from bottom to upward, there were four layers, i.e., of gravels layer up to 20cm, pebbles layer up to 20cm, sand layer up to 20 cm and finally clay layer up to 20 cm in which wetland plant is planted.

Glass wool was employed as a separator in CW-MFC by Yadav et al. (2012) and Zhao et al. (2013). The microcosm's upper end was left open, while the bottom end was sealed with epoxy materials. A sampling port was supplied in this microcosm to gather samples. To close the open ends of the sample ports, stoppers/taps were utilized. A setup comparable to a regular MFC was created inside the built marsh microcosm to measure electricity. A cathode electrode was put near the rhizospheric zone with the expectation that it would be more aerobic than the deeper zones due to air diffusion from the nearby outside atmosphere and air release from the rhizosphere. The anode was placed near the bottom of the microcosm with the hope that this zone would be relatively anaerobic and suited for MFC anodic reaction. To link the electrodes, an insulated copper wire was employed. Figure 1 demonstrates a schematic diagram of the constructed wetland-microbial fuel cell.

2.3 Inoculation

Each CW-MFC was inoculated with activated sludge, collected from a pulp and paper industry, wastewater treatment plant in Uttarakhand, India and was used as an inoculums source. Glucose (600 mg/L) was used as a carbon source and was added directly to the medium solution (NH_4Cl: 0.31 g/L, NaH_2PO_4: 4.97 g/L, Na_2HPO_4: 2.75 g/L, KCl: 0.13 g/L, $NaHCO_3$: 3.13 g/L (Liu et al., 2009). The systems were operated for several days until the reproducible maximum voltages were observed.

2.4 Wetland plants

The plants that grow in constructed wetlands have several properties related to the water treatment process that make them an essential component of the design. Macrophytes are the main source of oxygen in CWs through a process that occurs in the root zone, called radial oxygen loss [ROL] (Wang et al., 2018). The ROL contributes to the removal of pollutants because it favors an aerobic micro-environment, and waste removal is therefore accelerated as compared to anaerobic microbes. In the present research work, phytoremediation of industrial wastewater was done using Canna plants.

2.5 Electrode

Copper and zinc electrodes were used throughout the study. The surface area of copper and zinc electrode was 174 cm^2. Shown in Figure 2.

2.6 Biodegradation of lignin by immobilized peroxidase enzyme

The lignin component of wood is highly susceptible to becoming strongly colored due to its high content of aromatic rings, in addition to other unsaturated structures (Sarkanen 1971), Biodegradation of lignin was tested using immobilized peroxidase enzyme in sodium alginate beads. Sodium alginate beads were prepared by using 4% sodium alginate mixed with peroxidase enzyme. Mixture was pour drop by drop into pre-chilled autoclaved $CaCl_2$ (0.4 M) through a syringe without needle to makes fine beads of 2 mm in the laminar air flow. Beads were stored at 4°C for overnight.

For biodegradation experiment, sodium alginate beads were added in the microcosm according to the seventeen experiments proposed by the design expert 10.0 version software. Quantification of lignin was done by UV-VIS Spectrophotometer (Chen et al., 2011; Xiao et al., 2015).

2.7 Analytical methods

The response surface methodology (RSM) was investigated in order to optimize electricity generation in terms of millivolt and milliampere, lignin degradation and COD removal using the pulp and paper industry wastewater. The Box-Behnken design, which included 17 experimental runs with three replicates at the centre point, was utilized to optimize the dependent variables that significantly influenced biodegradation. Three key variables and their ideal values selected, i.e., electricity generation in terms of millivolt and milliampere, lignin degradation and COD removal for this experiment were: treatment (25%, 50%, and 75%), HRT (1 Day, 3 Days and 5 days) and concentration of perxoidase (10 mg/L, 20 mg/L and 30 mg/L). Seventeen experiments as proposed by the design expert 10.0 version software were proceeded in the laboratory accordingly.

The voltage (V) and current (I) have been monitored with a Haoyue (DT830D) multimeter that is digital and transformed to power (P = IV). The power density (mW cm^{-2}) and the density of current (mA cm^{-2}) were computed by dividing the power and current by the anode's surface area (cm^2).

TABLE 3 (a) Levels of different process variables in coded and ciphered form for lignin degradation, COD removal and electricity generation in terms of voltage and current (b). Box-Behnken experimental design and response of dependent variable for lignin degradation, COD removal and electricity generation in terms of voltage and current.

a

Variables	Code	Levels −1 0 + 1		
Treatment (mg/L)	A	25	50	75
HRT (Days)	B	1	3	5
Concentration (mg/L)	C	10	20	30

b

Std	Run	Factor 1 A:Treatment (%)	Factor 2 B:HRT (days)	Factor 3 C: Concentration (mg/L)	Response 1 lignin degradation (mg/L)	Response 2 electricity generation in terms of voltage and current (mV)	Response 3 COD removal (mg/L)
4	1	75	5	20	217	26	152.36
8	2	75	3	30	431	36	226.07
13	3	50	3	20	459	26	272.21
3	4	25	5	20	96	13	31.99
9	5	50	1	10	564	19	285.8
16	6	50	3	20	493	23	236.82
10	7	50	5	10	374	18	85.34
11	8	50	1	30	501	18	343.8
12	9	50	5	30	120	16	74.16
17	10	50	3	20	411	23	245.21
14	11	50	3	20	473	25	263.48
2	12	75	1	20	739	26	438.39
1	13	25	1	20	197	11	145.68
7	14	25	3	30	82	17	77.21
6	15	75	3	10	823	32	247.86
5	16	25	3	10	110	15	96.37
15	17	50	3	20	494	23	269.75

2.8 Statistical analysis and modeling

In the setups the lignin content was checked and for degradation of lignin immobilized horseradish peroxidase enzyme was used. According to Wang et al. (2018) peroxidase is able to oxidize the non-phenolic part of lignin (which forms 80%–90% of the lignin composition), it is absent from many lignin-degrading fungi.

TABLE 4 (a) ANOVA for response surface quadratic model of lignin degradation. Response 1 Lignin Degradation. (b) Estimated regression coefficients for lignin degradation

a

ANOVA for response surface quadratic model

Analysis of variance table [partial sum of squares - type III]

Source	Sum of squares	df	Mean square	F value	p-value prob > F	
Model	7.608 E+005	9	84,532.59	22.58	0.0002	significant
A-Treatment	3.720 E+005	1	3.720 E+005	99.38	<0.0001	
B-HRT	1.782 E+005	1	1.782 E+005	47.61	0.0002	
C-Concentration	67,896.13	1	67,896.13	18.14	0.0038	
AB	44,310.25	1	44,310.25	11.84	0.0108	
AC	33,124.00	1	33,124.00	8.85	0.0207	
BC	9,120.25	1	9,120.25	2.44	0.1625	
A^2	34,867.37	1	34,867.37	9.32	0.0185	
B^2	16,579.21	1	16,579.21	4.43	0.0734	
C^2	767.37	1	767.37	0.21	0.6644	
Residual	26,200.25	7	3,742.89			
Lack of Fit	21,564.25	3	7,188.08	6.20	0.0551	not significant
Pure Error	4,636.00	4	1,159.00			
Cor Total	7.870 E+005	16				

b

Std. Dev	61.18	R^2	0.9667
Mean	387.29	Adjusted R^2	0.9239
C.V. (%)	15.80	Predicted R^2	0.5524
		Adeq Precision	15.552

• Fcal: Fisher's ratio, df, Degree of freedoms.

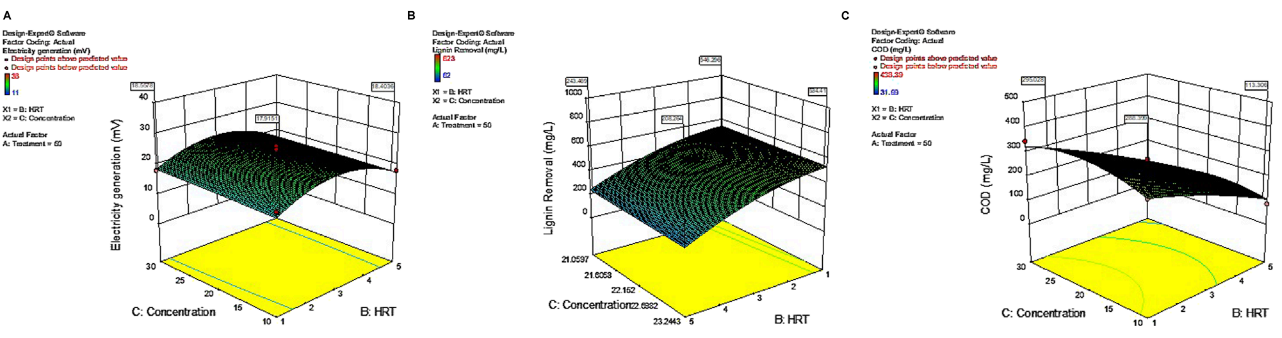

FIGURE 3
(A) Interactive effect of treatment **(A)** and HRT **(B)** on lignin degradation. **(B)** Interactive effect of peroxidase concentration **(C)** and HRT **(B)** on lignin degradation. **(C)** Interactive effect of peroxidase concentration **(C)** and treatment **(A)** on lignin degradation.

TABLE 5 (a) ANOVA for response surface quadratic model of electricity generation in terms of voltage and current. Response 2 Electricity generation in terms of voltage and current. (b) Estimated regression coefficients electricity generation in terms of voltage and current.

a						
ANOVA for response surface quadratic model						
Analysis of variance table [partial sum of squares - type III]						
Source	Sum of squares	df	Mean square	F value	p-value prob > F	
Model	676.87	9	75.21	18.00	0.0005	significant
A-Treatment	512.00	1	512.00	122.53	<0.0001	
B-HRT	0.13	1	0.13	9.030	0.8676	
C-Concentration	1.13	1	1.13	8.27	0.6198	
AB	1.00	1	1.00	0.24	0.6397	
AC	1.00	1	1.00	0.24	0.6397	
BC	0.25	1	0.25	0.060	0.8138	
A^2	5.33	1	5.33	1.28	0.2960	
B^2	157.96	1	157.96	37.80	0.0005	
C^2	0.066	1	0.066	0.016	0.9037	
Residual	29.25	7	4.18			
Lack of Fit	21.25	3	7.08	3.54	0.1267	not significant
Pure Error	8.00	4	2.00			
Cor Total	706.12	16				

b			
Std. Dev	2.04	R^2	0.9586
Mean	21.59	Adjusted R^2	0.9053
C.V. (%)	9.47	Predicted R^2	0.5008
		Adeq Precision	14.830

• Fcal: Fisher's ratio, df, Degree of fReedoms.

3 Results and discussion

3.1 Optimization of conditions for lignin degradation, COD removal and electricity generation in terms of voltage and current using response surface methodology

In the optimization study, Box–Behnken Factorial Design (BBD) was employed to determine the three-level-three-factor test. Treatment (A), HRT (B) and concentration (C) were selected as independent variables for the optimization studies of lignin degradation, COD removal, and electricity generation in terms of voltage and current which are depended on variables. Low and high levels of each variable were coded as −1 and+1 by keeping 0 as mid-point. The characteristics of the Paper and Pulp Industry effluent are shown in Tables 1, 2 shows the lignin content of the pulp and paper industry.

For optimization of different parameters for lignin degradation, COD removal, and electricity generation in terms of voltage and current, three independent (treatment, HRT and concentration) and three dependent variables (lignin degradation, COD removal, and electricity generation in terms of voltage and current) were optimized. Three different treatments of wastewater 25 mg/L, 50 mg/L, and 75 mg/L were taken along with 1, 3 and 5 HRT in days and concentration of immobilized horseradish peroxidase enzyme 10 mg/L, 20 mg/L, and 30 mg/L were used, given in Table 3A. To optimize these parameters, a total of seventeen tests were carried out with the use of Box-Behnken Design; all the values are given in Table 3B. Each combination given by Box-Behnken

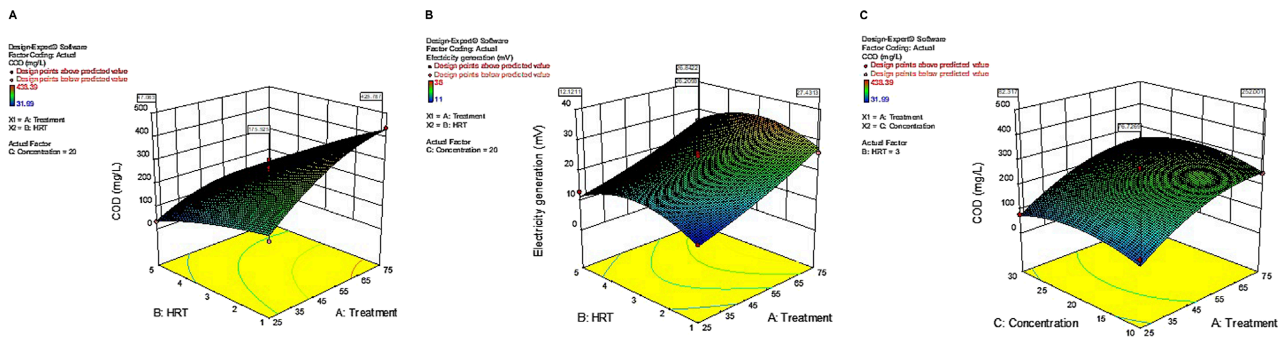

FIGURE 4
(A) Interactive effect of Treatment **(A)** and HRT **(B)** on electricity generation in terms of voltage and current. Interactive effect of peroxidase concentration **(C)** and HRT **(B)** on electricity generation in terms of voltage and current. Interactive effect of peroxidase concentration **(C)** and treatment **(A)** on electricity generation in terms of voltage and current.

Design was applied to CW-MFC. The optimized value was applied to CW-MFC for further study.

3.2 Lignin degradation

The ANOVA test of the data was performed to observe the effect of independent finishing variables on lignin degradation. The model's p-value was discovered to be 0.0002, indicating that the model was significant because the p-value was below 0.05. Furthermore, as the p-value was greater than 0.07, or 0.05, the lack of fit was not statistically significant. This demonstrated the validity of the model.

It is clearly presented in Table 4A that the overall model (Fcal = 22.58 > Ftab = 14.66), was significant confirming the significant effect of all the independent factors on lignin degradation. At the 1% level of significance, treatment (Fcal = 99.38 > Ftab = 21.20) and HRT (Fcal = 47.61 > Ftab = 21.20) had a substantial impact on the lignin degradation of wastewater samples. Whereas the concentration of peroxidase (Fcal = 18.14 > Ftab = 7.71) had a significant effect on lignin degradation from pulp and paper industry wastewater samples at a 5% level of significance. It implies that lignin degradation from wastewater samples was affected by changing the parameters for treatment, HRT and peroxidase concentration on the wastewater samples in the CW-MFC.

As can be seen from Table 4B, the total influence of all factors at the linear, interactive, and quadratic levels produced the coefficient of determination (R^2) for the regression model of lignin degradation, which was 0.96, adjusted R^2, and predicted R^2, which was 0.55. Regression analysis was used to estimate the regression equation for lignin degradation, and the model's sufficient precision value was 15.55, which is greater than 4 (signals noise ratio).

3.3 Graphical representation of lignin degradation

The interactive effect of independent variables, i.e., treatment, HRT and peroxidase concentration on lignin degradation in pulp and paper industry wastewater was studied using 3D graphs. The graphs showing the interaction among the factors are shown in Figures 3A–C.

3.4 Electricity generation in terms of voltage and current

To see how independent finishing variables affected the voltage and current generated during power generation, an ANOVA test was run on the data. The model's p-value, which was less than 0.05, was determined to be 0.0005, suggesting that the model was significant. Furthermore, as the p-value was greater than 0.07, or 0.126, the lack of fit was not statistically significant. This demonstrated the validity of the model.

It is clearly presented in Table 5A that the overall model (Fcal = 18 > Ftab = 14.66), was significant confirming the significant effect of all the independent factors on electricity generation in terms of voltage and current. Treatment (Fcal = 122.53 > Ftab = 21.20) had significant effect on electricity generation in terms of voltage and current of wastewater samples at 1% level of significance. Whereas concentration of peroxidase (Fcal = 8.27 > Ftab = 7.71) and HRT (Fcal = 9.03 > Ftab = 7.71) had a significant effect on electricity generation in terms of voltage and current from pulp and paper industry wastewater samples at 5% level of significance. It implies that electricity generation in terms of voltage and current from wastewater samples was affected by changing the parameters for treatment, HRT and peroxidase concentration on the wastewater samples in the CW-MFC.

Table 5B makes this clear: the total effect of all factors at the linear, interaction, and quadratic levels produced an adjusted R^2 of 0.90, a projected R^2 of 0.50, and a coefficient of determination (R^2) of 0.95 for the regression model of lignin degradation. Regression analysis was used to forecast the regression equation for electricity generation in terms of voltage and current, and the model's sufficient precision value was 14.83, which is greater than 4 (signals noise ratio).

3.5 graphical representation of electricity generation in terms of voltage and current

Using 3D graphics, the interaction effects of independent variables—treatment, HRT, and peroxidase concentration—on the

TABLE 6 (a) ANOVA for response surface quadratic model of COD removal Response 3 COD removal. (b) Estimated regression coefficients COD removal

a						
ANOVA for response surface quadratic model						
Analysis of variance table [partial sum of squares - type III]						
Source	Sum of squares	df	Mean square	F value	p-value prob > F	
Model	1.889 E+005	9	20,993.42	35.40	<0.0001	significant
A-Treatment	63,622.80	1	63,622.80	107.28	<0.0001	
B-HRT	94,573.35	1	94,573.35	159.46	<0.0001	
C-Concentration	4.31	1	4.31	7.862	0.9345	
AB	7,425.27	1	7,425.27	12.52	0.0095	
AC	1.73	1	1.73	2.916E-003	0.9584	
BC	1,196.47	1	1,196.47	2.02	0.1985	
A^2	10,692.55	1	10,692.55	18.03	0.0038	
B^2	946.83	1	946.83	1.60	0.2469	
C^2	8,611.13	1	8,611.13	14.52	0.0066	
Residual	4,151.53	7	593.08			
Lack of Fit	3,170.62	3	1,056.87	4.31	0.0960	not significant
Pure Error	980.91	4	245.23			
Cor Total	1.931 E+005	16				

b			
Std. Dev	24.35	R^2	0.9785
Mean	205.44	Adjusted R^2	0.9509
C.V. (%)	11.85	Predicted R^2	0.7293
		Adeq Precision	21.191

• Fcal: Fisher's ratio, df: Degree of fReedoms.

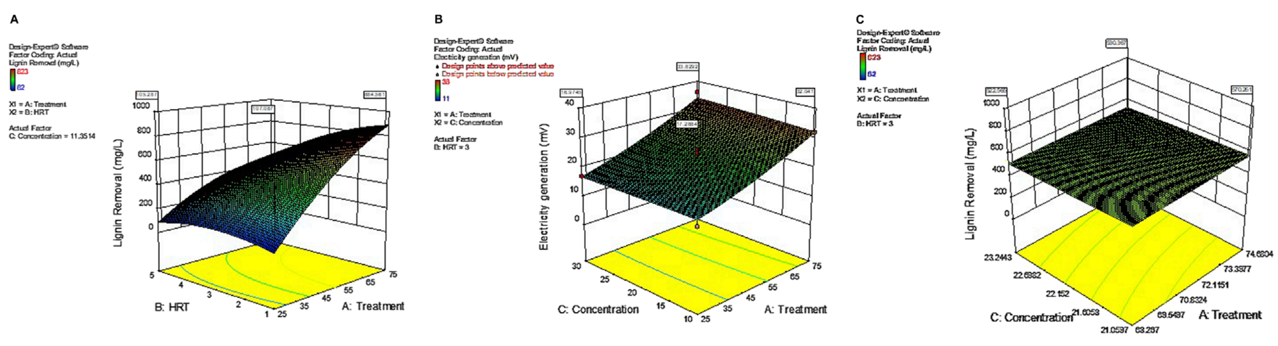

FIGURE 5
(A) Interactive effect of Treatment **(A)** and HRT **(B)** on COD removal. **(B)** Interactive effect of peroxidase concentration **(C)** and HRT **(B)** on COD removal. **(C)** Interactive effect of peroxidase concentration **(C)** and treatment **(A)** on COD removal.

TABLE 7 Constraints for optimization of variables for lignin degradation, electricity generation and COD removal with pulp and paper industry wastewater.

S.No.	Printing variables	Goals	Lower limit	Upper limit	Importance
I	Independent variables				
1	Treatment (A)	is in range	25	75	++++
2	HRT (B)	is in range	3	5	++++
3	Concentration (C)	Minimum	10	30	++++
II	Dependent variables				
1	Lignin degradation	Maximize	82	823	++++
2	Electricity generation in terms of voltage and current	Maximize	11	36	++++
3	COD removal	Maximize	31.99	438.39	++++

FIGURE 6
Optimized conditions obtained for treatment, HRT and concentration.

voltage and current produced by wastewater from the pulp and paper industry were investigated. The graphs in Figures 4A–C illustrate how the factors interact with one another.

3.6 COD removal

To investigate the impact of independent finishing variables COD removal, an ANOVA test was run on the data. The model's p-value was discovered to be 0.0001, indicating that the model was significant because the p-value was below 0.05. Further, the lack of fit was non-significant as the p-value was more than 0.07 that is 0.096. This demonstrated the validity of the model.

It is clearly represented in Table 6A that the overall model (Fcal = 35.40 > Ftab = 14.66), was significant confirming the significant effect of all the independent factors on electricity generation in terms of voltage and current. Treatment (Fcal = 107.28 > Ftab = 21.20) and HRT (Fcal = 159.46 > Ftab = 21.20) had significant effect on COD removal from pulp and paper industry wastewater samples at 1% level of significance. Whereas the concentration of peroxidase concentration (Fcal = 7.86 > Ftab = 7.71) had a significant effect on COD removal of wastewater samples at a 5% level of significance. It implies that COD removal from wastewater samples was affected by changing the parameters for treatment, HRT and peroxidase concentration on the wastewater samples in the CW-MFC.

Table 6B makes this clear: the overall effect of all factors at the linear, interaction, and quadratic levels produced the expected R^2 of 0.72, adjusted R^2 of 0.95, and coefficient of determination (R^2) of 0.97 for the regression model of lignin degradation. Regression analysis was used to forecast the regression equation for electricity generation in terms of voltage and current. The model's sufficient precision value was 21.19, which are greater than 4 (signals noise ratio).

3.7 Graphical representation of COD removal

The interactive effect of independent variables, i.e., treatment, HRT and peroxidase concentration on electricity generation in terms of voltage and current from pulp and paper industry wastewater was studied using 3D graphs. The graphs showing the interaction among the factors are shown in Figures 5A–C.

3.8 Constraints set for independent and dependent variables

The restrictions established for the optimization of responses and independent variables are shown in Table 7. To find the greatest possible combination of the independent variables, namely, therapy (A), HRT (B), and concentration (C) that may produce the best overall results with desirability close to 1, the aims and importance for the independent and dependent variables were fixed. An independent variable, such as the concentration

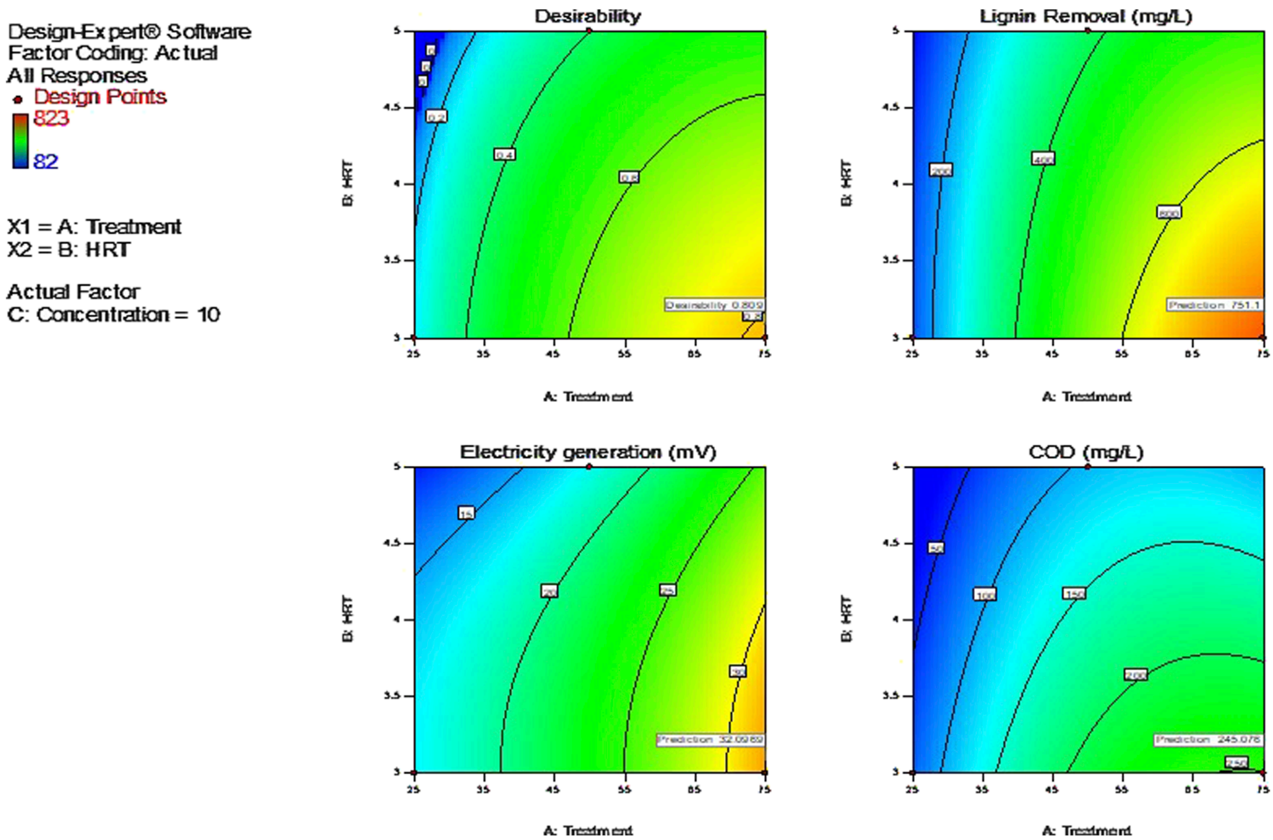

FIGURE 7
Optimized conditions obtained for treatment, HRT and concentration.

of peroxidase, was set to "minimize" with the intention of achieving an optimized value that would ultimately lead to reasonable treatment and cost considerations, and HRT was set within the range.

The goal was to produce an optimized value that liberates as little pollutant and lignin in the wastewater as possible. Other dependent variables, such as lignin content, electricity generation in terms of voltage and current, and COD removal from the wastewater, were set at "maximize."

3.9 Results of the experiments under optimal conditions

In order to improve responses, the BBD model was used to predict an ideal condition for each independent variable, including treatment, HRT, and concentration. Figures 6, 7 depict the anticipated results of running the experiment under the independent variable optimization settings. This figure reveals that the optimum treatment, HRT and peroxidase concentration should be 74.99 mg/L, 3.08 days and 10 mg/L respectively. Therefore the experiment carried out gives better results, i.e., lignin degradation was 751.1 mg/L, voltage generation was 32.09 mV and COD removal was 245.07 mg/L. The desirability of model was 0.809 which indicates model is best suited up to 80%.

A quadratic model was built to predict lignin degradation efficiency, electricity generation in terms of voltage and current and COD removal efficiency. ANOVA was performed to examine the significance of the variables and their interactions. Schenone et al. (2015) used ferrioxalate complex and response surface methodology (RSM) to model and optimize photo-Fenton degradation of 2, 4-D.

A three-level factorial experimental design combined with response surface methodology (RSM) was used to optimize the biodegradation of 2, 4-D. The analysis of variance (ANOVA) was used to evaluate significance models and their coefficients. All results of this study shown similarity with other studies for optimization using RSM (Mansouriieh et al., 2015; Uniyal et al., 2016; Jabeen et al., 2015; Sohrabi et al., 2016). The major findings of this research study could lead to the generation of sustainable bioelectricity from wastewater while also ensuring greener waste-to-wealth technologies in the pulp and paper industries.

4 Conclusion

The goal of this research is to investigate how combined microbial fuel cells (MFCs) and built wetlands (CWs) can be used to treat wastewater more effectively and generate power in the form of voltage and current while requiring less money up front and over time by using Response Surface Methodology.

The effects of hydraulic residence time (HRT) were investigated in this study; various concentrations of wastewater as well as a concentration of immobilized peroxidase enzyme were also studied for electricity generation, COD removal and the lignin degradation from pulp and paper industry wastewater. The CW-MFC is a promising technology in the fields of wastewater treatment and bioenergy.

Wetland plants has also shown good efficacy in eliminating pollutants from the wastewater. An enzyme called peroxidase that had been immobilized on sodium alginate beads was developed and used in the microcosm to break down lignin. Optimization of treatment, HRT and concentration for maximum biodegradation of the lignin, electricity generation in terms of voltage and current and Response Surface Methodology was also used to achieve COD elimination by peroxidase enzyme. The optimized parameters were treatment of wastewater-74.99%, HRT-3.08 days, and concentration-10 mg/L. The experiment was designed using the Box-Behnken Experimental design (BBE).

These days technology involved in wastewater treatment uses innovative, efficient and advanced methods. Based on the above empirical models, this study recommends the RSM-BBD design as a practical and applicable technology for sustainable power generation source suitable for COD removal and lignin degradation. This can be a current method for efficient and accurate wastewater treatment technology. However, these need to be economically viable, especially in the industrial areas, municipal, domestic as well as in the rural part of India in order to support wastewater treatment and its reuses.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

MN: Conceptualization, Methodology, Validation, Visualization, Writing–original draft. PS: Conceptualization, Data curation, Formal Analysis, Methodology, Visualization, Writing–original draft. RS: Conceptualization, Supervision, Validation, Writing–review and editing. AM: Funding acquisition, Writing–review and editing. NS: Writing–review and editing, Supervision. FA: Writing–review and editing. SS: Funding acquisition, Writing–review and editing.

Funding

The author(s) declare that financial support was received for the research, authorship, and/or publication of this article. The authors extend their appreciation to the Deanship of Research and Graduate Studies at King Khalid University for funding this work through Large Group Project under grant number RGP2/89/46.

Acknowledgments

The authors are thankful to the Department of Environmental Science, G. B. Pant University of Agriculture & Technology, Pantnagar, India for all the facilities to conduct this experiment.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Generative AI statement

The author(s) declare that no Generative AI was used in the creation of this manuscript.

Publisher's note

References

APHA (2012). "Standard methods for the examination of water and wastewater," in *American public health association (APHA). American water works association (AWWA) and water environment federation (WEF).* 22nd ed. (Washington DC, USA).

Bankole, P. O., Omoni, V. T., Mulla, S. I., Adebajo, S. O., and Adekunle, A. A. (2022). Co-biomass degradation of fluoranthene by marine-derived fungi; Aspergillus aculeatus and Mucor irregularis: comprehensive process optimization, enzyme induction and metabolic analyses. *Arabian J. Chem.* 15 (9), 104036. doi:10.1016/j.arabjc.2022.104036

Chakrabarti, S. K. (2006). Environment management in pulp and paper industry beyond current legislations and CREP commitments. *IPPTA* 18 (4), 143.

Chen, Y. H., Sheu, S. C., Mau, J. I., and Hsieh, P. C. (2011). Isolation and characterization of a strain of *Klebsiella pneumoniae* with citrinin-degrading activity. *Microbiol. Biotechnol.* 27, 487–493. doi:10.1007/s11274-010-0478-4

Ericsson, K., Nilsson, L. J., and Nilsson, M. (2011). New energy strategies in the Swedish pulp and paper industry-The role of national and EU climate and energy policies. *Energy Policy* 39, 1439–1449. doi:10.1016/j.enpol.2010.12.016

Jabeen, H., Iqbal, S., Anwar, S., and Parales, R. E. (2015). Optimization of profenofos degradation by a novel bacterial consortium PBAC using response surface methodology. *Biodegrad* 100, 89–97. doi:10.1016/j.ibiod.2015.02.022

Kamali, M., Gameiro, T., Costa, M. E. V., and Capela, I. (2016). Anaerobic digestion of pulp and paper mill wastes-An overview of the developments and improvement opportunities. *Chem. Eng. J.* 298, 162–182. doi:10.1016/j.cej.2016.03.119

Kiely, P. D., Rader, G., Regan, J. M., and Logan, B. E. (2011). Long-term cathode performance and the microbial communities that develop in microbial fuel cells fed different fermentation end products. *Bioresour. Technol.* 102, 361–366. doi:10.1016/j.biortech.2010.05.017

Li, Z., Zhang, X., Lin, J., Han, S., and Lei, L. (2010). Azo dye treatment with simultaneous electricity production in an anaerobic–aerobic sequential reactor and microbial fuel cell coupled system. *Bioresour. Technol.* 101, 4440–4445. doi:10.1016/j.biortech.2010.01.114

Liu, H., and Logan, B. E. (2004). Electricity generation using an air-cathode single chamber microbial fuel cell in the presence and absence of a proton exchange membrane. *Environ. Sci. Technol.* 38, 4040–4046. doi:10.1021/es0499344

Liu, S., Song, H., Li, X., and Yang, F. (2013). Power generation enhancement by utilizing plant photosynthate in microbial fuel cell coupled constructed wetland system. *Int. J. Photoenergy* 2013, 1–10. doi:10.1155/2013/172010

Liu, Z., Liu, J., Zhang, S., and Su, Z. (2009). Study of operational performance and electrical response on mediator-less microbial fuel cells fed with carbon- and protein-rich substrates. *Biochem. Eng. J.* 45, 185–191. doi:10.1016/j.bej.2009.03.011

Logan, B. E. (2008). *Microbial fuel cell. John wiley and sons*. New York.

Logan, B. E., Hamelers, B., Rozendal, R., Schröder, U., Keller, J., Freguia, S., et al. (2006). Microbial fuel cells: methodology and technology. *Environ. Sci. and Technol.* 40 (17), 5181–5192. doi:10.1021/es0605016

Luo, H., Liu, G., Zhang, R., and Jin, R. (2009). Phenol degradation in microbial fuel cells. *Chem. Eng. J.* 147, 259–264. doi:10.1016/j.cej.2008.07.011

Mansouriieh, N., Sohrabi, M. R., and Khosrav, M. (2015). Optimization of profenofos organophosphorus pesticide degradation by zero-valent bimetallic nanoparticles using response surface methodology. *Arab. J. Chem.* doi:10.1016/j.arabjc.2015.04.009

Morris, J. M., Jin, S., Crimi, B., and Pruden, A. (2009). Microbial fuel cell inenhancing anaerobic biodegradation of diesel. *Chem. Eng. J.* 146, 161–167. doi:10.1016/j.cej.2008.05.028

Narayan, M., Solanki, P., Rabha, A. K., and Srivastava, R. K. (2018). Treatment of pulp and paper industry effluent and electricity generation by constructed wetland coupled with microbial fuel cell (CW-MFC). *J. Pharmacogn. Phytochemistry* 7 (6), 493–498.

Puig, S., Coma, M., Desloover, J., Boon, N., and Balaguer, M. D. (2012). Autotrophic denitrification in microbial fuel cells treating low ionic strength waters. *Environ. Sci. Technol.* 46, 2309–2315. doi:10.1021/es2030609

Sarkanen, K. V. (1971). *Lignins: occurrence, formation, structure and reactions*. New York 916: Wiley-Interscience.

Sharma, P., Iqbal, H. M., and Chandra, R. (2022). Evaluation of pollution parameters and toxic elements in wastewater of pulp and paper industries in India: a case study. *Case Stud. Chem. Environ. Eng.* 5, 100163. doi:10.1016/j.cscee.2021.100163

Sharma, P., Ngo, H. H., Khanal, S., Larroche, C., Kim, S. H., and Pandey, A. (2021). Efficiency of transporter genes and proteins in hyperaccumulator plants for metals tolerance in wastewater treatment: sustainable technique for metal detoxification. *Environ. Technol. and Innovation* 23, 101725. doi:10.1016/j.eti.2021.101725

Sohrabi, M. R., Moghri, M., Masoumi, H. R. F., Amiri, S., and Moosavi, N. (2016). Optimization of reactive blue 21 removal by nanoscale zero-valent iron using response surface methodology. *Arabian J. Chem.* 9 (4), 518–525. doi:10.1016/j.arabjc.2014.11.060

Soloman, P. A., Basha, C. A., Velan, M., Balasubramanian, N., and Marimuthu, P. (2009). Augmentation of biodegradability of pulp and paper industry wastewater by electrochemical pre-treatment and optimization by RSM. *Sep. Purif. Technol.* 69 (1), 109–117. doi:10.1016/j.seppur.2009.07.002

Schenone, A. V., Conte, L. O., Botta, M. A., and Alfano, O. M. (2015). Modeling and optimization of photo-Fenton degradation of 2, 4-D using ferroxalate complex and response surface methodology (RSM). *J. Environ. Manage.*, 177–183.

Sun, J., Hua, Y. Y., Bi, Z., and Cao, Y. Q. (2009). Simultaneous decolorization of Azo dye and bioelectricity generation using a microfiltration membrane air-cathode singlechamber microbial fuel cell. *Bioresour. Technol.* 100, 3185–3192. doi:10.1016/j.biortech.2009.02.002

Tiku, D. K., Kumar, A., Sawhney, S., Singh, V. P., and Kumar, R. (2007). Effectiveness of treatment technologies for wastewater pollution generated by Indian pulp mills. *Env. Monit. Assess.* 132, 453–466. doi:10.1007/s10661-006-9548-3

Uniyal, S., Paliwal, R., Sharma, R. K., and Rai, J. P. N. (2016). Degradation of fipronil by Stenotrophomonas acidaminiphila isolated from rhizospheric soil of Zea mays. *3 Biotech.* 6, 1–10.

Wang, Q., Hu, Y., Xie, H., and Yang, Z. (2018). Constructed wetlands: a review on the role of radial oxygen loss in the rhizosphere by macrophytes. *Water* 10, 678. doi:10.3390/w10060678

Waye, A., Annal, M., Tang, A., Picard, G., Harnois, F., Guerrero-Analco, J. A., et al. (2014). Canadian boreal pulp and paper feedstocks contain neuroactive substances that interact *in vitro* with GABA and dopaminergic systems in the brain. *Sci. total Environ.* 468, 315–325. doi:10.1016/j.scitotenv.2013.08.040

Xiao, Y., Chen, S., Gao, Y., Hu, W., Hu, M., and Zhong, G. (2015). Isolation of a novel beta cypermethrin degrading strain Bacillus subtilis BSF01 and its biodegradation pathway. *Appl. Microbiol. Biotechnol.* 99, 2849–2859. doi:10.1007/s00253-014-6164-y

Yadav, A. K., Dash, P., Mohanty, A., Abbassi, R., and Mishra, B. K. (2012). Performance assessment of innovative constructed wetland-microbial fuel cell for electricity production and dye removal. *Ecol. Eng.* 47, 126–131. doi:10.1016/j.ecoleng.2012.06.029

Zhao, Y., Collum, S., Phelan, M., Goodbody, T., Doherty, L., and Hu, Y. (2013). Preliminary investigation of constructed wetland incorporating microbial fuel cell: batch and continuous flow trials. *Chem. Eng. J.* 229, 364–370. doi:10.1016/j.cej.2013.06.023