

TREATMENT OF ENGINE OIL EFFLUENT BY ELECTROOXIDATION AND AEROBIC BIOLOGICAL DEGRADATION

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ABSTRACT

Electrooxidation (EO) and biochemical oxidation (BO) experiments were conducted to reduce the chemical oxygen demand (COD) and biological oxygen demand (BOD) of the engine oil effluent (EOE). BO process was carried out for the effluent containing 5, 10 and 15 mL of inoculum (two different culture concentrations) in aerobic conditions. COD and BOD removal efficiencies of 86 and 89% for *Bacillus* sp. and 82 and 85% for *Alcaligenes* sp. were achieved at the end of sequential integrated oxidation process.

The experimental results showed that the integrated EO with BO is a viable option to reduce COD and BOD of the EOE containing the bio-recalcitrant phenol, anthracene, naphthalene and benzo(a)pyrene using the *Bacillus* sp. and *Alcaligenes* sp. The end products were analyzed by Fourier Transform Infrared spectrophotometer, ¹H nuclear magnetic resonance, high performance liquid chromatography and gas chromatography-mass spectrometry. The treated water can be reused in automobile industries for general purpose and gardening.

INTRODUCTION

Environmental pollution with petroleum and petrochemical products has been recognized as one of the most serious problems. Automobile workshops are an important component of the service sector industry. With rapid socioeconomic and infrastructure development, the number of vehicles in the world, especially in the urban areas, has increased significantly.

The most significant environmental impact associated with the existing workshops is the seepage of used engine oil into the soil. 1.7, 3.5 Mt of engine oil effluent is collected in the European Union and USA each year, respectively. Millions of tone of used oils is disposed through dumping on the ground or in water, land filling, or nonenergy recovery [1]. Contamination of soil by oil causes it to lose its useful properties such as fertility, water-holding capacity, permeability and binding capacity. Used motor oil is a very dangerous polluting product due to its chemical composition [2]. Prolonged exposure and high oil concentration may cause the development of liver or kidney disease, possible damage to the bone marrow and an increased

risk of cancer [3].

Engine oil effluent (EOE) normally consists of high content of polycyclic aromatic hydrocarbons (PAHs, e.g., naphthalene, anthracene, phenols and these derivatives) and other organic compounds, including some organometallic constituents which are toxic to the environment [4], generate carcinogens and endocrine disruptors [5,6]. Therefore, in order to prevent the environmental pollution, the EOE was treated to reduce the toxic level. A number of EOE treatment methods have been employed, namely supercritical fluid extraction and liquid-liquid extraction methods for the removal of PAHs [7]. The oil and oil based effluent was treated using advanced oxidation processes, sorption and coalescence method [8], chemical oxidation process [9], ozonation and cavitation processes [10]. It is important to select proper pretreatment technique to improve the overall efficiency of the wastewater treatment unit [11].

In recent years, electrochemical techniques in combination with biological processes have received greater attention due to the distinctive advantages of electrochemical methods such as, environmental com-

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patibility; the main reactant is the electron which is a clean reagent. Versatility, a plethora of reactors and electrode materials, shapes, and configurations can be utilized. It is noteworthy that the same reactor can be used frequently for different electrochemical reactions with minor changes only and the electrolytic processes can be scaled easily from the laboratory to the plant. Also, electrochemical methods are generally safe because of the mild conditions usually employed and the small amount and innocuous nature of the added chemicals. Electrodes and cells can also be designed to minimize power losses due to poor current distribution and voltage drops.

In batch electrochemical reactor, electrochemical oxidation (EO) has been carried out as a pre treatment before the biochemical oxidation (BO) to increase biodegradability and at the end as a post treatment to meet the required standards. In this work, integrated EO and BO methods have been attempted to degrade the EOE containing the bio-recalcitrant phenol, anthracene, naphthalene and benzo(a)pyrene. *Bacillus* sp. and *Alcaligenes* sp. were used in BO. The treated water has been recommended for reuse which has standard discharge characteristics as per the pollution control norms. It can be transferred the efficient laboratory scale technique to large-scale operations.

Biodegradation of oil contaminated water has been established as one of the efficient, economic, versatile and environmentally sound treatment [12]. It makes use of indigenous oil-consuming microorganisms, called petrophiles that can naturally degrade large hydrocarbons and utilize them as a food source. It converts the hydrocarbon into CO₂ and water [13,14]. Aerobic treatment has a great efficiency in degrading polyphenolic compounds [15-18]. Basha et al. [19] reported that the coupling of EO with BO for non-biodegradable organics was one of the best treatment methods.

MATERIALS AND METHODS

1. EOE

The EOE was collected from an automobile

work shop (Chennai, India) and its characteristic features were analyzed such as pH, color, total nitrides (TN), total dissolved solids (TDS), total organic carbon (TOC), biological oxygen demand (BOD) and chemical oxygen demand (COD). The analyzed results are tabulated in Table 1.

2. Pre-treatment of EOE by EO

A batch electrolytic cell was used for the EO process. A laboratory model DC power supply (APlab L-3210) was used for electrolysis. Electrolysis was carried out at room temperature (28 °C) in a cylindrical open glass cell of 1 L capacity equipped with two electrodes by vigorously stirred with a magnetic bar (500 rpm). The working electrodes were Ti/RuO₂ (anode) and stainless steel (cathode). The schematic diagram of experimental setup is shown in Fig. 1.

The EO experiments were carried out at 5 A dm⁻² and NaCl was added to the influent prior to electrolysis as a supporting electrolyte to enhance the reactivity. The overall time of operation was 150 min. At the end of the process, COD was measured.

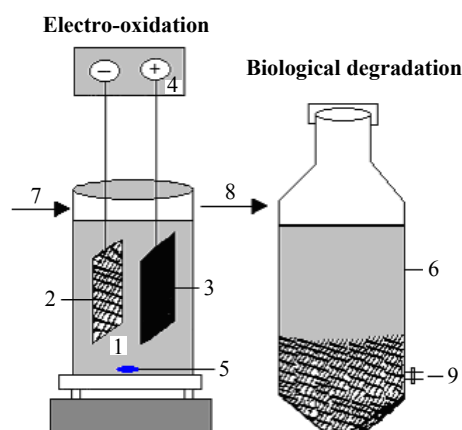


Fig. 1. Schematic representation of engine oil effluent treatment process. (1) EOE, (2) anode, (3) cathode, (4) DC power supply, (5) magnetic stirrer, (6) bio setup, (7) crude EOE, (8) pretreated EOE and (9) Outlet of treated water.

Table 1. Characteristic of the engine oil effluent

Parameter	Before treatment	After treatment (Electro-oxidation)	After treatment (Biological oxidation) at pH 6.8	
			<i>Bacillus</i>	<i>Alcaligenes</i>
Color	Dark Brown	Light yellow	Colorless	Colorless
pH	7.8	6.6	8.8	8.2
COD (mg L ⁻¹)	3280	1072	470	585
BOD (mg L ⁻¹)	1975	868	226	297
TDS (mg L ⁻¹)	140	112	88	102
TN (mg L ⁻¹)	32	19	10	14
TOC (mg L ⁻¹)	473	155	63	87

3. BO Treatment

By performing biochemical tests, it was found that *Bacillus* sp. and *Alcaligenes* sp. have the ability to degrade EOE and used in this study. The pure culture of microorganisms was inoculated in a flask, containing 100 mL nutrient broth [Composition (g L⁻¹): peptone-10, NaCl-5, yeast extract-2 and beef extract-1] sterilized by autoclaving at 20 °C for 20 min and incubated at 30 ± 2 °C for 24 h under static condition. Both these cultures utilize the organic carbon as a sole source of energy. The successive transfers of culture into EOE were done at 30 °C in static condition. This acclimatized culture was used for BO studies.

BO experiments were carried out at room temperature (28 °C), by batch mode in 250 mL Erlenmeyer flasks for 5 d in shaking conditions (140 rpm) with the help of Orbital Shaker. The pre-treated EOE by EO was further treated by BO under different inoculum concentrations (5, 10, 15 and 100 mL⁻¹).

4. Analysis of COD and BOD

COD and BOD were measured after centrifugation with 1 d interval. COD of all samples were determined by the dichromate closed reflux method using thermo reactor TR620-Merck. Analysis of BOD was done by Winkler's method, strictly following the APHA procedures [20]. Experiments were repeated until the error was less than 3%.

The treated and untreated effluent samples were collected and subjected to Fourier Transform Infrared spectrophotometer (FTIR, Tensor 27 model), ¹H-Nuclear Magnetic Resonance (NMR, Bruker 400 MHz NMR spectrometer), High performance Liquid Chromatography (HPLC, Shimadzu) and Gas chromatography-mass spectrometry (GC-MS).

5. FTIR Analysis

To determine the presence of different molecules in the sample FTIR spectrometer was used. The source setting of FTIR was middle-infrared light. DLaTGS was used as a detector. The disc used for FTIR analysis was cleaned using dichloromethane and a few drops of sample to be analyzed was placed in between the discs. The data was scanned from 4000 to around 400 cm⁻¹ with 64-scan speed and the spectrum was recorded.

6. ¹H-NMR Analysis

The ¹H NMR used was Bruker 400 MHz ultra-shield FT-NMR. A few drops of the sample were taken in 17.5 cm NMR tube. About 0.5 mL of NMR solvent (chloroform) was added to the NMR tube. The NMR tube should be free from vacuum and the sample should be completely dissolved in the NMR sol-

vent.

7. HPLC Analysis

HPLC analysis was carried out on a Shimadzu model LC-8A chromatograph (Shimadzu, Kyoto, Japan) equipped with Shimadzu model UV Spectrophotometric detector and CLC-ODS-C18 (column with 4.6 × 150 mm inside diameter, Senshu Scientific, Tokyo, Japan). A stationary phase consists of silica gel and a mobile phase composed of methanol, methanol-water, acetonitrile, and acetonitrile-water. The software used was Spinchrome CFR Single Channel. In HPLC analysis the effluent was dissolved in small volume of methanol and 20 µL aliquots were injected into the chromatograph. They were monitored by UV absorption at 275 nm.

8. GC-MS Analysis

GC-MS analysis was used to identify the components present in the EOE. An Agilent 7890A Series GC equipped with Electronic pneumatics controls and New Nitrogen Phosphorous Detector were used. Samples for GC/MS analysis should be fully dissolved in an organic solvent at a concentration between 10 and 1000 ppm (w/v). The column temperature was programmed from an initial temperature at 35 °C for 0.5 min to 325 °C with a rate of 25 °C min⁻¹. After this, the temperature was held constant at 325 °C for 8 min. The injection port temperature was 275 °C. The total ion chromatogram was obtained using a scan range of 30-400 amu.

RESULTS AND DISCUSSION

1. Pre-treatment by EO Process

Electrochemical treatment is undoubtedly an energy-intensive process and its efficiency is usually assessed in terms of specific energy consumption (SEC). This was defined as the amount of energy consumed per unit mass of organic load (e.g. COD) removed. Energy consumption was directly affected by the current density applied to the system. The SEC is expressed in kWh kg⁻¹ of COD removal and was given as:

$$\text{Energy Consumption} = \frac{\left(\frac{IVt}{vol} \right)}{\Delta_{COD}} \quad (1)$$

Where Δ_{COD} was difference in COD between initial and final in g L⁻¹, *vol* was the volume of reactor in L, *I* the current passed in A, *t* the retention time in h, *V* the cell voltage in V.

The EO pre-treatment was carried out at optimized current density of 5 A dm⁻² for 150 min at

which the maximum reduction of COD was observed from 3280 to 1072 mg L⁻¹ (67% efficiency) as shown in Fig. 2 at the end of 150 min. The energy density was determined as 2.8 kWh kg⁻¹ of COD removal. Hence all the experiments were carried out at the same current density. The slow rate of COD reduction at the initial stage of operation was observed and that could be attributed to the occurrence of secondary reactions (involving Cl₂, O₂ and H₂ evolution), perhaps favoured by the higher cathode-anode potential gap employed.

The overall reduction in levels of COD observed during electrolysis may be a consequence of: (i) anodic oxidation of phenol, naphthalene, anthracene and benzo(a)pyrene at the Ti/RuO₂ electrode (electrodegradation), and (ii) aggregation of suspended oil droplets drawn to the surface by gases formed at the cathode (H₂) and the anode (O₂ and Cl₂) (electroflotation). Electrochemical processes are typically characterized by the evolution of small bubbles of gases, the sizes of which (8-15 µm) are appropriate for the flotation process to occur.

Results clearly indicated that the optimized current density strongly influences the maximum reduction of COD. The conversion of phenol, naphthalene, anthracene and benzo(a)pyrene to acetaldehyde and pyruvate is exclusively attributed to EO. The end products such as acetaldehyde and pyruvate were biodegradable in nature and were analysed by FTIR, ¹H NMR, HPLC and GC-MS.

2. Biological Oxidation

A batch bioreactor without mechanical agitation such as a bubble column, proved to be an adequate bioreactor for *Bacillus* sp. and *Alcaligenes* sp. growth on EOE. A moderate mixing was obtained by aeration for the growth of *Bacillus* sp. and *Alcaligenes* sp. on EOE. The biodegradation rate of strains of *Alcaligenes* sp. and *Bacillus* sp. in degrading pre-treated EOE was tested for 5 d with different concentra-

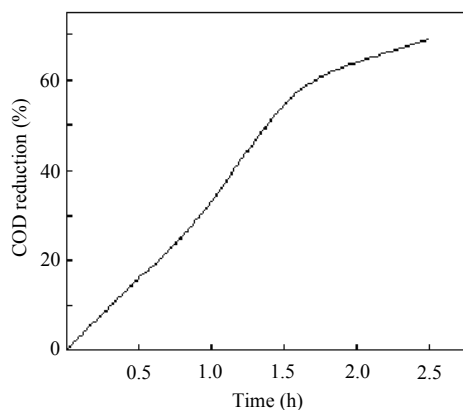


Fig. 2. Percentage of COD reduction after electrooxidation.

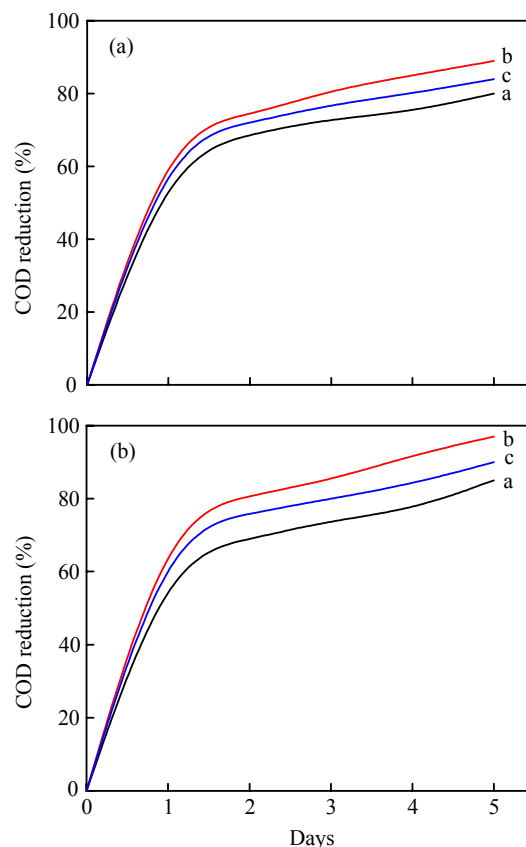


Fig. 3. Percentage of COD reduction at different concentrations by (a) *Alcaligenes* sp. and (b) *Bacillus* sp. at addition of a, 5 mL; b, 10 mL; c, 15 mL.

tion (5, 10 and 15 mL). The maximum degradation was obtained at 10 mL inoculum concentration (*Alcaligenes* sp. and *Bacillus* sp.) as shown in Figs. 3a and 3b. However, *Bacillus* sp. successfully reduced COD about 86% while *Alcaligenes* sp. correspondingly removed COD about 82% under the same operating conditions. Better performance of *Bacillus* sp. than *Alcaligenes* sp. in the biological degradation can be explained as follows: *Bacillus* sp. had higher ability in utilizing hydrocarbon as the sole source of carbon and energy when inoculated directly into the EOE. This may be due to the presence of efficient hydrocarbon degradative enzyme systems and the presence of catabolic genes involved in hydrocarbon degradation in the bacterial species.

From the results, the two organisms were found to degrade the organics present in EOE. It was noted that microorganisms utilize the organic substances present in EOE as nutrients and finally breakdown the organic matter into carbon dioxide and water [21,22].

Table 1 shows the characteristics of EOE before and after EO and BO. From the table, the pH increased considerably at the end of the aerobic treatment, showing values in the range of pH 6.8 to 8.8 after 5 d. Simultaneously, the total alkalinity increased

in parallel with the rise in the pH values. The rise in alkalinity with the time was due to the increase in the bicarbonate concentration as a consequence of the increase in the CO_2 concentration derived from the phenol, naphthalene, anthracene and benzo(a)pyrene degradation and the soluble substrate was later oxidized to CO_2 .

3. FTIR Spectral Studies

The FTIR analysis of real EOE, pretreated effluent by EO and final treatment by BO is present in Fig. 4. Table 2 presents the absorption of the peaks with respect to different functional groups of organics during the degradation. Similar results were also reported by El Hajjouji et al. [23]. It can be seen that some structural changes might have occurred during the integrated EO and BO treatment. The peaks of real effluent at 3436 and 1638 cm^{-1} indicated the presence of functional groups $-\text{OH}$ and $\text{C}=\text{O}$ respectively. Further it indicated increased levels of aliphatic structures (2922 and 1457 cm^{-1}) and polysaccharides (1374 – 1159 cm^{-1}), as well as of nitrogenous and aromatic compounds (1638 cm^{-1}).

The IR spectra of the fraction showed strong peaks at about 2922 – 2856 cm^{-1} , while other peaks at around 1374 – 1457 cm^{-1} decrease in intensity compared to the retained fraction. These retained fractions show an increase in the peaks at 1159 and 1035 cm^{-1} due to COOH and characteristic of the $\text{C}=\text{N}$ of secondary amides or of aromatic $\text{C}=\text{C}$. So, during the treatment, there was a decrease in the relative intensity of the aliphatic chains (2922 and 1457 cm^{-1}), and a new peak mostly occurred at 1636 cm^{-1} which can be identified as $\text{C}=\text{C}$ of the simple aliphatic chain of the low molecular weight fatty acid.

In BO, the appearance of a new peak at 2071 cm^{-1} corresponds to the characteristic peak of CO_2 . This indicates that acetaldehyde and pyruvate present in the

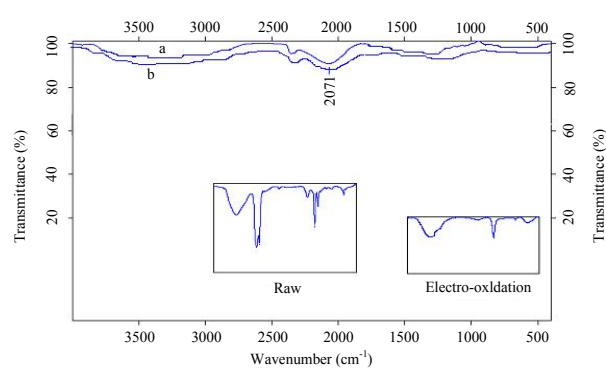


Fig. 4. FTIR spectra of real EOE (raw), treated effluent by electrooxidation, treated effluent by biological oxidation (a) *Bacillus* sp. and (b) *Alcaligenes* sp.

EOE may be converted into CO_2 and water.

4. ^1H -NMR Studies

^1H -NMR was chosen as the primary analytical method for determining PAHs present in the EOE. From the Fig. 5, ^1H -NMR of real EOE showed major peaks at 1–2 ppm chemical shifts. It indicated the presence of aromatic protons. These shifts were reduced drastically by EO and BO as shown in spectrums b, c, and d. It showed that the PAHs present in real EOE completely changed into acetaldehyde and pyruvate. It can also be explained that the longer aromatic and aliphatic chains may break down into smaller one which are biodegradable in nature. Peak at 0.91 ppm corresponded to the terminal $-\text{CH}_3$ group of organics present in EOE. Peak at 2.9 ppm was due to the presence of water. The formation of H_2O was predominant in spectrums c and d which clearly indicated that the BO was more effective method of secondary treatment of EOE. From the NMR studies, it can be concluded that *Alcaligenes* sp. and *Bacillus* sp. consumed carbon and hydrogen from EOE for its growth

Table 2. FT-IR peak assignments

S. no.	Peak (cm^{-1})	Real EOE Assignment	EO treated EOE Assignment	Biodegradation of EO treated EOE Assignment
1.	3436	$-\text{OH}$ (phenols, naphthalene, anthracene, benzo(a)pyrene, and aromatic alcohols.	-----	-----
2.	2922	Unsaturated C-H	-----	-----
3.	2856	-----	Saturated C-H	-----
4.	2071	-----	-----	Characteristic of CO_2
5.	1638	$\text{C}=\text{O}$ stretching in amides.	-----	-----
6.	1635	-----	$\text{C}=\text{C}$ of the simple aliphatic chain	-----
7.	1457	C-H stretching in aliphatic structures	-----	-----
8.	1374	Methyl group	-----	-----
9.	1035	Vibration of aromatic ethers	-----	-----
10.	725	C-H out of plane	C-H out of plane	-----

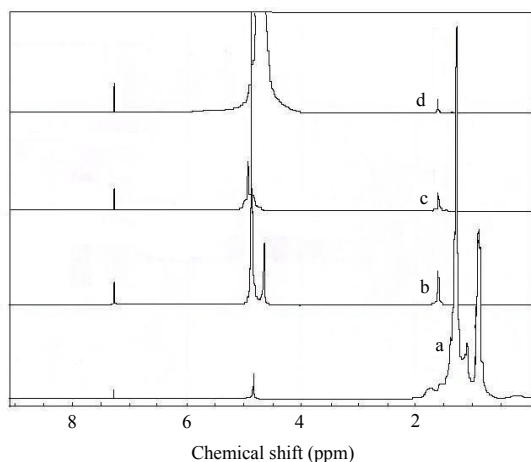


Fig. 5. ^1H -NMR spectra of real EOE (a), treated effluent by electrooxidation (b), treated effluent by biological oxidation of (c) *Alcaligenes* sp. and (d) *Bacillus* sp.

which indirectly favours the BO.

5. HPLC Studies

HPLC analysis using CLC-ODS-C18 column equipped with UV-Visible spectrophotometer was used to identify the degradation compounds of EOE. HPLC of real EOE is shown in Fig. 6. Major peaks were observed between 2-4 min and it indicated the presence of naphthalene, benzo(a)pyrene, phenol and anthracene at 2.2, 3, 3.4 and 4 min respectively. The peak intensity was reduced by electrooxidation and further reduced by *Alcaligenes* sp. and *Bacillus* sp. HPLC studies revealed that the combined treatment of EOE was very effective for the organics present in the EOE.

6. GC-MS Studies

GC-MS analysis was carried out to determine the nature of organic compounds present in EOE in each step during the combined treatment by electrooxidation and biodegradation. A wide variety of PAHs were found in the real samples namely naphthalene, phenol, anthracene and benzo(a)pyrene. The GC-MS spectra indicated that the components present in the initial EOE (Fig. 7) have undergone degradation step by step by electrooxidation (b) and biological degradation by *Alcaligenes* sp. (c) and *Bacillus* sp. (d). GC-MS results concurred with those of FTIR, HPLC and ^1H NMR.

CONCLUSIONS

In the present study, the feasibility of combined treatment of EOE by electrooxidation and aerobic degradation was successfully investigated. The experimental results of the lab-scale setup indicated that

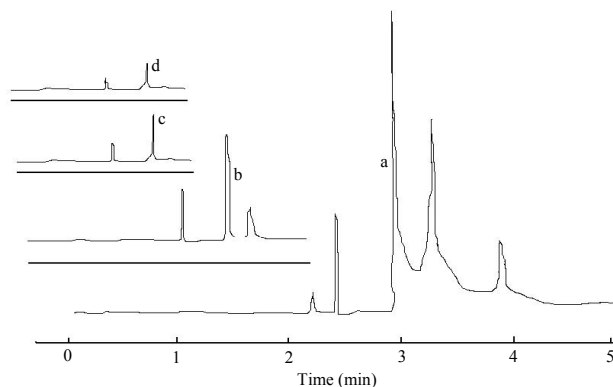


Fig. 6. HPLC spectra of (a) real EOE, (b) treated effluent by electrooxidation, treated effluent by biological oxidation of (c) *Alcaligenes* sp. and (d) *Bacillus* sp.

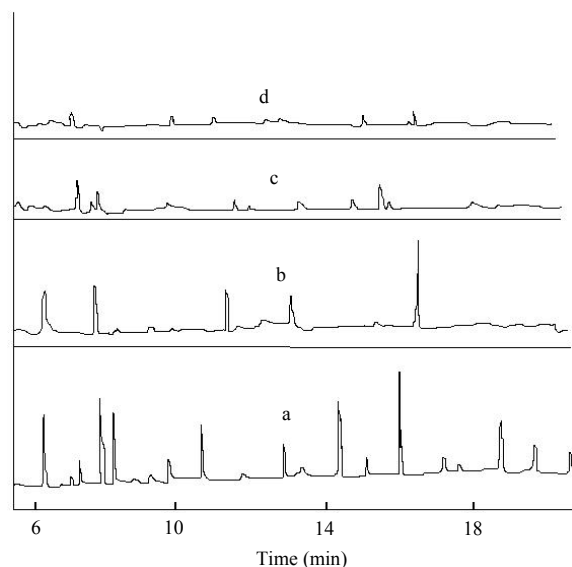


Fig. 7. GC-MS spectra of (a) real EOE, (b) treated effluent by electrooxidation, treated effluent by biological oxidation of (c) *Alcaligenes* sp. and (d) *Bacillus* sp.

the stirred tank reactor was effective and reliable for treating automotive effluent. The combined degradation method of EOE may be effective and economical. Low molecular weight organics were obtained at the end of EO pre-treatment and it was further degraded to CO_2 by biological oxidation. Pretreatment of EOE by EO enhances the biological degradation. Both the combined methods removed COD and BOD to a larger extent. EO removed 67% of the COD and BO reduced COD about 86% (*Bacillus* sp.) which gave better result than *Alcaligenes* sp. FT-IR, ^1H -NMR, HPLC and GC-MS studies confirmed the structural changes occurring in the organics present in EOE during the treatments. It can be utilized for the treatment of non biodegradable organic pollutants from industrial efflu-

ents and help to meet the requirements of pollution control boards. This method does not produce any secondary pollutant and hence it is an eco-friendly method. The treated water can be reused for general purpose and gardening in the industry.

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REFERENCES

1. Rauckytea, T., D.J. Hargreaves and Z. Pawlaka, Determination of heavy metals and volatile aromatic compounds in used engine oils and sludges. *Fuel*, 85(4), 481-485 (2006).
2. Vazquez-Duhalt, R., Environmental impact of used motor oil. *Sci. Total Environ.*, 79(1), 1-23 (1989).
3. Mandri, T. and J. Lin, Isolation and characterization of engine oil degrading indigenous microorganisms in Kwazulu-Natal, South Africa. *Afr. Biotechnol.*, 6(1), 23-27 (2007).
4. Perez, M., R. Rodriguez-Cano, L.I. Romero and D. Sales, Anaerobic thermophilic digestion of cutting oil wastewater: Effect of co-substrate. *Biochem. Eng. J.*, 29(3), 250-257 (2006).
5. USEPA, Recycling Used Oil: What Can You Do? Cooperation Extension Services for University of Kentucky, ENRI 317, Washington, D.C. (1996).
6. Agency for Toxic Substances and Disease Registry Division of Toxicology and Environmental Medicine, Toxicological Profile for Used Mineral-based Crankcase Oil. CAS# 8002-05-9, Atlanta, GA (1997).
7. Wong, P.K. and J. Wang, The accumulation of polycyclic aromatic hydrocarbons in lubricating oil over time-a comparison of supercritical fluid and liquid-liquid extraction methods. *Environ. Pollut.*, 112(3), 407-415 (2001).
8. Ji, F., C.L. Li, X.Q. Dong, Y. Li and D.D. Wang, Separation of oil from oily wastewater by sorption and coalescence technique using ethanol grafted polyacrylonitrile. *J. Hazard. Mater.*, 164(2-3), 1346-1351 (2009).
9. Balcioglu, I.A. and I. Arslan, Partial oxidation of reactive dyestuffs and synthetic textile dye-bath by the O_3 and O_3/H_2O_2 processes. *Water Sci. Technol.*, 43(2), 221-228 (2001).
10. Commenges, N., E. Gonze, Y. Gonthier and A. Bernis, Ultrasonic treatment applied to industrial wastewater: A difficult undertaking. 7th Meeting of the European Society of Sonochemistry, Biarritz-Guéthary, France, May 14-18 (2000).
11. Rajeshwar, K. and J.G. Ibanez, *Environmental Electrochemistry: Fundamentals and Applications in Pollution Abatement*. Academic Press, San Diego, CA (1997).
12. Newman, A.P., C.J. Pratt, S.J. Coupe and N. Cresswell, Oil bio-degradation in permeable pavements by microbial communities. *Water Sci. Technol.*, 45(7), 51-56 (2002).
13. Harder, E., *Bioremediation of Engine Oil*. Little Flower Academy, Dallas, TX (2004).
14. Borja, R., A. Martin, V. Alonso, I. Garcia and C.J. Banks, Influence of different aerobic pretreatment on the kinetics of anaerobic digestion of olive mill wastewater. *Water Res.*, 29(2), 489-495 (1995).
15. Gioia, D., F. Fava, L. Bertin and L. Marchetti, Biodegradation of synthetic and naturally occurring mixtures of mono-cyclic aromatic compounds present in olive mill wastewaters by two aerobic bacteria. *App. Microbiol. Biot.*, 55(5), 619-626 (2001).
16. Zaim, H., J.R. Bailly and L. Codomier, Influence d'une argile sur la formation des substances parahumiques d'origine microbienne. *Agrochimica*, 39(1), 18-25 (1995) (in French).
17. Hafidi, M., G.A. Baddi, A. Chetoui and I. El Hadrami, Traitements des effluents liquides d'huileries d'olive par des micro-organismes aerobies. *Agrochimica*, 48(1), 1-12 (2004) (in French).
18. Armenante, P.M., D. Kafkewitz, G.A. Lewandowski and C.J. Jou, Anaerobic-aerobic treatment of halogenated phenolic compounds. *Water Res.*, 33 (3), 681-692 (1999).
19. Basha, C.A., E. Chithra and N.K. Sripriyalakshmi, Electro-degradation and biological oxidation of non-biodegradable organic contaminants. *Chem. Eng. J.*, 149(1-3), 25-34 (2009).
20. Clescent, L.S., A.E. Greenberg and A.D. Eaton (Eds.), Method 5210 B, In: *Standard Methods for the Examination of Water and Wastewater*. American Public Health Association and American Water Works Association and Water Environment Federation, Washington, D.C., pp. 5_2-5_6 (1998).
21. Ballesteros, M.M.B., J.A.S. Perez, F.G.A. Fernandez, J.L.G. Sanchez, J.L.C. Lopez and S.M.

- Rodriguez, A kinetics study on the biodegradation of synthetic wastewater simulating effluent from an advanced oxidation process using *Pseudomonas putida* CECT 324. J. Hazard. Mater., 151(2-3), 780-788 (2008).
22. Kamath, A.V. and C.S. Vaidyanathan, New pathway for the biodegradation of indole in *Aspergillus niger*. Appl. Environ. Microb., 56(1), 275-280 (1990).
23. El Hajjouji, H., Fakharedine, G.A. Baddi, P. Winterton, J.R. Bailly, J.C. Revel and M. Hafidi, Treatment of olive mill waste-water by aerobic biodegradation: An analytical study using gel permeation chromatography, ultraviolet-visible and Fourier transform infrared spectroscopy. Bioresource Technol., 98(18), 3513-3520 (2007).
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