



National
Technical
University
of Athens



Municipality of Ancient Olympia



PAVE
THE
WASTE
c.c.



NAXOS2018

13-16 June 2018

6th International
Conference

on

Sustainable Solid Waste
Management

<http://www.naxos2018.uest.gr>

NAXOS2018

6th International Conference on
Sustainable Solid Waste
Management

13-16 June 2018



Demonstrating resource efficiency through innovative,
integrated waste recycling schemes for remote areas
LIFE14 ENV/GR/000722



National
Technical
University
of Athens



Municipality of Ancient Olympia

Committees

Scientific Committee

Maria Loizidou, National Technical University of Athens, Chair

Abdul-Sattar Nizami, King Abdulaziz University
Adam Smoliński, Central Mining Institute
Adela Galvin, University of Cordoba
Ahmed El-Gendy, American University in Cairo
Aleksandra Djukic-Vukovic, University of Belgrade
Alexandra Ribeiro, Universidade Nova de Lisboa
Amane Jada, IS2M-CNRS
Ana Jiménez-Rivero, Universidad Politécnica de Madrid
Anastasia Zabaniotou, Aristotle University of Thessaloniki
Anastasios Zouboulis, Aristotle University of Thessaloniki
Andrea Capodaglio, University of Pavia
Andres Illanes, Universidad Católica de Valparaíso
Ange Nzihou, Ecole des Mines d'Albi
Antonis Mavropoulos, International Solid Waste Association
Antonis Zorpas, Open University of Cyprus
Apostolos Koutinas, Agricultural University of Athens
Apostolos Vlyssides, National Technical University of Athens
Barbara Ruffino, Politécnico di Torino
Carlos Ariel Cardona Alzate, Universidad Nacional de Colombia sede Manizales
Carol S.K. Lin, City University of Hong Kong
Celia Dias-Ferreira, Polytechnic of Coimbra
Chew-Tin Lee, University Technology of Malaysia
Christopher Cheeseman, Imperial College
Christopher Koroneos, National Technical University of Athens
Costas Costa, Cyprus University of Technology
Costas Velis, University of Leeds
David Bolzonella, University of Verona
David Newman, World Biogas Association
Dimitrios Kaliampakos, National Technical University of Athens
Dimitrios Komilis, Democritus University of Thrace
Dimitrios Malamis, National Technical University of Athens
Dolores Hidalgo Barrio, CARTIF Technology Centre
Evina Katsou, Brunel
Filiz Dilek, Middle East Technical University
Francesco Fatone, Marche Polytechnic University
Francisco Agrela, University of Cordoba
Francisco Omil, Universidad de Santiago de Compostela
Florian Amlinger, European Compost Network
Georgia Labuto, Universidade Federal de Sao Paulo
Gerasimos Lyberatos, National Technical University of Athens
Giuseppe Mancini, University of Catania
Gregorio Antolín, University of Valladolid
Grigorios Itskos, Purdue University
Hussam Jouhara, Brunel University
Ioannis Skiadas, Technical University of Denmark
Isam Janajreh, Masdar Institute
Jader Busato, Universidade de Brasilia
Jale Yanik, Ege University
Jiri Hrebíček, Masaryk University
Joan Dosta, University of Barcelona
Joseph Patrick Hettiaratchi, University of Calgary
Joseph Zeaiter, American University of Beirut
Juan Antonio Baeza Labat, Universitat Autònoma de Barcelona
Justo García-Navarro, Universidad Politécnica de Madrid
Katarzyna Chojnacka, Wrocław University of Technology
Katherine-Joanne Haralambous, National Technical University of Athens
Katia Lasaridi, Harokopio University
Kiran Thakur, Hefei University of Technology
Konstantinos Aravosis, National Technical University of Athens
Konstantinos Moustakas, National Technical University of Athens
Laurent Lemeé, Université de Poitiers
Lidia Lombardi, Università degli Studi Niccolò Cusano - Telematica Roma
Lionel Limousy, Université de Haute-Alsace
Lorna Anguilano, Brunel University
Luben Tzankov, Technical University of Sofia
Luciano Matos Queiroz, University of Bahia
Marc Deshusses, Duke University
Marco Baratieri, Free University of Bolzano
Maria Zachariou-Dodou, Institute of Environment & Sustainable Development
Mauro Majone, Sapienza University of Rome

Committees

Mejdi Jeguirim, Université de Haute-Alsace
Michalis Kornaros, University of Patras
Nathalie Gontard, French National Institute for Agricultural Research
Nick Voulvoulis, Imperial College
Nickolas Themelis, Columbia University
Nikolaos Mousiopoulos, Aristotle University of Thessaloniki
Nikolaos Tzortzakis, Cyprus University of Technology
Omar Assobhei, University of Fez
Pascale Champagne, Queen's University
Paul Chen, National University of Singapore
Paul Christakopoulos, Lulea University of Technology
Petros Gikas, Technical University of Crete
Rafael Luque, University of Cordoba
Rémy Bayard, DEEP - INSA Lyon - Université de Lyon
Ronghou Liu, Shanghai Jiao Tong University
Rui Cunha Marques, University of Lisbon
Siegfried Vlaeminck, University of Antwerp
Simos Malamis, National Technical University of Athens
Spyros Pavlostathis, Georgia Tech
Stephen Smith, Imperial College
Stijn Van Hulle, Ghent University
Ta Yeong Wu, Monash University
Thierry Ribeiro, UniLaSalle
Thomas Pretz, RWTH Aachen University
Ulku Yetis, Middle East Technical University
Vassilis Inglezakis, Nazarbayev University
Weiguo Shen, Wuhan University of Technology
Zhao Jun Wei, Hefei University of Technology
Zorana Naunovic, University of Belgrade

Organizing Committee

Konstantinos Moustakas, National Technical University of Athens, Head

Christina Papadaskalopoulou, National Technical University of Athens
Despoina Bakogianni, National Technical University of Athens
Dolores Hidalgo Barrio, CARTIF Technology Centre
Dimitrios Lianos, Municipality of Naxos and Small Islands
Dimitrios Malamis, National Technical University of Athens
Evangelia Ploumistou, National Technical University of Athens
Francisco Corona Encinas, CARTIF Technology Centre
Jesús M^a Martín, CARTIF Technology Centre
Nikolaos Kondylis, Municipality of Naxos and Small Islands
Nikolaos Lolos, Municipality of Ancient Olympia
Stergios Vakalis, National Technical University of Athens
Varvara Lavouta, National Technical University of Athens
Vasiliki Panaretou, National Technical University of Athens



Poster session

- | | |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>75. E. Papaefstathiou, M. Stylianou, A. Agapiou, Exploring the side effects of vaping (electronic cigarette)</p> | <p>76. I. Erotokritou, M.A. Stylianou, A. Agapiou, S. Giannopoulos
The potential resource value of coffee residues</p> |
| <p>77. Yan Rosen
Enhancing the feasibility of ozonation pretreatment of lignocellulosic waste by reducing ozonation time</p> | <p>78. B. A. Karamanova, A. E. Stoyanova, M. Schipochka, R. K. Stoyanova
Sustainable porous carbons on the basis of biomaterials for supercapacitors</p> |
| <p>79. P. Yang, G.-B. Shao, W.-X. Jiang
Research and preparation of a degradable material</p> | <p>80. F.R. Beltrán, M.U. de la Orden, E. Climent, J. Martínez Urreaga
Effects of process conditions on the effectiveness of solid-state polymerization in the recycling of poly(lactic acid) waste.</p> |
| <p>81. J. Milić, M. Curčić, Z. Brnjac, H. Carapina, J. Randjelović, K. Krinulović, A. Jovović
The socio-economic impact timeline in Serbia for persistent organic pollutants (POPs)</p> | <p>82. J. Milić, G. Gojgić-Cvijović, T. Šolević Knudsen, M. Ilić, J. Avdalović, M. Lješević, M. M. M. Vrvic
Assessment of screening assays as tool for evaluation of bacterial potential for biodegradation of polycyclic aromatic hydrocarbons</p> |
| <p>83. J. Avdalović, A. Žeradžanin, S. Miletić, O. Božović, T. Šolević Knudsen, D. Stanković, J. Milić, M.M. Vrvic
Investigation of the humic acids evolution during bioremediation of heavy residual fuel oil</p> | <p>84. C. L. B. M da Cruz, A.S. P. Santos, E. Ritter
Study on the viability of incorporation of the water treatment plant sludge in the substrate for the production of native specie the Atlantic Forest (Brazil)</p> |
| <p>85. Kai-Chee Loh, Jingxin Zhang, Yen Wah Tong
Fate of bacterial pathogens and antibiotic resistance genes during anaerobic digestion of food waste by incorporating powdered activated carbon</p> | <p>86. T. Klammsteiner, A. Walter, C. Heussler, B. C. Schlick-Steiner, F. M. Steiner, H. Insam
Hermetia illucens (Diptera: Stratiomyidae) larvae in waste valorization and diet-based shifts in their gut microbiome</p> |
| <p>87. M. Nagler, A. Jäger, B. Hupfau, H. Insam, S. M. Podmirseg
Defining the best pretreatment method for biomethanisation of sewage sludge and corn straw.</p> | <p>88. K. Triantafyllidis, P. Samaras, G. Palantzas, E. Tzamos, A. Zouboulis
Application of the pyrolysis technology for industrial and hazardous waste management</p> |
| <p>89. S. Brivio, D. Papadrossou, M. Kondis
Sustainable FGT solutions for WtE</p> | <p>90. A. Uruña, J.A. Conesa, D. Díez
The influence of temperature and dolomite catalyst on the pyrolysis of corn stover</p> |
| <p>91. A. Uruña, D. Díez, G. Antolín
New biofuel production technology to recover used frying oils and power the Seville's urban bus fleet</p> | <p>92. Yen Wah Tong, Jingxin Zhang, Kai-Chee Loh
Enhancing methane production in anaerobic co-digestion process by anaerobic co-pretreatment</p> |
| <p>93. M. Omirou, I. Anastopoulos, D. Fasoula, I. M. Ioannides
Soil nitrous oxide emissions in agricultural systems fertilized with livestock manure, compost and inorganic fertilizers</p> | <p>94. E. Ell Bestawy
Stimulatory and / or inhibitory effects of heavy metals-contaminated effluents on biochemical characteristics of some selected cyanobacteria</p> |

Investigation of the humic acids evolution during bioremediation of heavy residual fuel oil

J. Avdalović¹, A. Žerađjanin¹, S. Miletic¹, O. Božović², T. Šolević Knudsen¹, D. Stanković^{3,4}, J. Milić¹, M.M. Vrvic⁵

¹Institute of Chemistry, Technology and Metallurgy, University of Belgrade, 11000 Belgrade, Serbia

²Institute of Physical Chemistry, University of Zurich, Zurich, Switzerland

³Vinča Institute of Nuclear Sciences, University of Belgrade, 11000 Belgrade, Serbia

⁴Innovation Centre Faculty of Chemistry, University of Belgrade, 11000 Belgrade, Serbia

⁵BREM GROUP, 11090 Belgrade-Kneževac, Serbia

Keywords: heavy residual fuel oil, bioremediation, humic acids, humification.

Presenting author email: adjunic@chem.bg.ac.rs

The advance of technology and industry in the last hundred years, population growth and the development of big cities have led to the production of waste materials in quantities which exceed self-purification capacity of nature. The accumulation of waste materials of various origin causes increased pollution of the environment. Petroleum and its derivatives such as heavy residual fuel oils are one of the major and most dangerous pollutants of soil. Bioremediation is a method of reducing petroleum pollution which has been widely used in the last years. Bioremediation is a process which is based on the natural capacity of microorganisms to degrade or transform toxic substances from the environment into harmless products (Gomez and Sartaj, 2014.). Bioremediation technologies are in harmony with the principles of sustainable development since waste materials are not generated (Avdalović et al. 2016, Jednak et al. 2017).

The aim of this paper is to study the use of different microorganisms as “biological agents” for *ex situ* bioremediation of a petroleum contaminant (heavy residual fuel oil), along with simultaneous monitoring of the humification process, because there are indications that during biodegradation of polycyclic aromatic hydrocarbons (PAH), substances similar to humic substances are creating (Henner et al. 1997, Ressler et al. 1999).

The content of total petroleum hydrocarbon (TPH) in the sample was extracted as per method ISO 16703 (ISO 16703, 2004) and determined gravimetrically in accordance with DIN (DIN EN 14345, 2004). The results were calculated according to dry matter. For the analysis of humic acid, 40 g of each air-dried soil sample was mixed with 200 ml of NaOH/Na₄P₂O₇ solution (ISO 5073, 1999). The mixture was heated in a boiling water bath for 2 h, with frequently shaking to ensure precipitation of insoluble material. Then, the flask was removed from the water bath, cooled to room temperature, and supernatant was separated from residue by centrifugation at 3000 rpm (residue was discarded). Humic acids were precipitated from supernatant by acidifying with 6 M HCl to pH 1. After that, the suspension was centrifuged. The supernatant was decanted and discarded. The HA was purified according to the method of the International Humic Substance Society (IHSS). The effects of humification on the structure of humic acids were evaluated by Fourier Transform Infrared Spectroscopy (FT-IR), potentiometric titrations, the ratio of absorbances at 465 and 665 nm (E4/E6 ratio), the C/H ratio and gel chromatography.

In the pilot experiment, which lasted for 170 days, bioremediation of a petroleum pollutant was carried out in the layer of artificial soil substrate, with river sand as mother substrate. One container was a control biopile and it contained a mixture of sawdust, sand and the pollutant. The second container was an inoculated biopile and it was prepared in the same way as the control biopile but, in addition to sawdust, sand and the pollutant, it also contained biostimulants, the sources of nitrogen and phosphorus, and biomass. The initial concentration of petroleum hydrocarbons in the pilot experiment was 23,05 mg/kg dry weight. At the end of the experiment, the total hydrocarbons were reduced to 8,15 mg/kg dry weight in the inoculated biopile, while the concentration was 21,35 mg/kg dry weight in the control biopile. It was observed that the content of humic acids increased from 0,32% to 0,42% in the inoculated biopile during the bioremediation process, while the increase in the content of humic acids in the control biopile was insignificant. Additional analyses of humic acids from the inoculated biopile showed that they were transformed during the bioremediation process and that they had a higher degree of aromaticity as well as greater redox and buffering capacity at the end of the experiment. In addition to this, gel chromatography was performed on the samples of humic acids isolated from the inoculated biopile during the bioremediation process. The results showed that on day 0 the share of fraction of great molecular mass (greater than 100 kDa) was 72,67%, while the share of the above mentioned fraction was 76,17%, 170 days after the beginning of the process, which was probably due to the fact that undegraded organic compounds were transformed and included in complex polymer structures which are analogues of humic substances.

References

- Avdalović J., Đurić A., Miletić S., Ilić M., Milić J., Vrijić M.M. (2016) Treatment of a mud pit by bioremediation. *Waste Manage Res* 34:734–739.
- DIN EN 14345 (2004) Characterization of Waste. Determination of hydrocarbon content by gravimetry. DIN, Berlin.
- Gomez F., Sartaj M. (2014) Optimization of field scale biopiles for bioremediation of petroleum hydrocarbon contaminated soil at low temperature conditions by response surface methodology (RSM). *Int. Biodeter. Biodegr.* 89:103-109.
- Henner P., Schiavon M., Morel J.L., Lichtfouse E. (1997) Polycyclic aromatic hydrocarbon (PAH) occurrence and remediation methods. *Analisis* 25:9-10.
- International Humic Substance Society (IHSS) Available at URL: <http://humic-substances.org/> (2013).
- ISO 5073 (1999) Brown coals and lignites — Determination of humic acids. Geneva.
- ISO 16703 (2004) Soil quality – Determination of content of hydrocarbon in the range C10 to C40 by gas chromatography. Geneva.
- Jednak T., Avdalović J., Miletić S., Slavković-Beškoski L., Stanković D., Milić J., Ilić M., Beškoski V., Gojčić-Cvijović G., Vrijić M.M. (2017) Transformation and synthesis of humic substances during bioremediation of petroleum hydrocarbons. *Int Biodeterior. Biodegrad* 122: 47-52.
- Ressler B.P., Kneifel H., Winter J. (1999) Bioavailability of polycyclic aromatic hydrocarbons and formation of humic acid-like residues during bacterial PAH degradation. *Appl. Microbiol. Biot.* 53:85-91.

Acknowledgements

This research was partially financed by the Ministry of Education, Science and Technological Development of the Republic of Serbia as a part of the Projects III 43004.

Investigation of the humic acids evolution during bioremediation of heavy residual fuel oil

J. Avdalović^{1a}, A. Žeradjanić^{1b}, S. Miletić¹, O. Božović², T. Šolević Knudsen¹, D. Stanković^{3,4}, J. Milić¹, M.M. Vrvčić⁵

¹Institute of Chemistry, Technology and Metallurgy, University of Belgrade, 11000 Belgrade, Serbia

²Institute of Physical Chemistry, University of Zurich, Zurich, Switzerland

³Vinča Institute of Nuclear Sciences, University of Belgrade, 11000 Belgrade, Serbia

⁴Innovation Centre Faculty of Chemistry, University of Belgrade, 11000 Belgrade, Serbia

⁵BREM GROUP, 11090 Belgrade- Knezevac, Serbia

^ajavidalovic@chem.bg.ac.rs; ^badjuric@chem.bg.ac.rs



Aim

The aim of this paper is to study the use of different microorganisms as “biological agents” for *ex situ* bioremediation of a petroleum contaminant (heavy residual fuel oil), along with simultaneous monitoring of the humification process, because there are indications that during biodegradation of polycyclic aromatic hydrocarbons (PAH), substances similar to humic substances are creating.

Introduction

The advance of technology and industry in the last hundred years, population growth and the development of big cities have led to the production of waste materials in quantities which exceed self-purification capacity of nature. The accumulation of waste materials of various origin causes increased pollution of the environment. Petroleum and its derivatives such as heavy residual fuel oils are one of the major and most dangerous pollutants of soil. Bioremediation is a method of reducing petroleum pollution which has been widely used in the last years. Bioremediation is a process which is based on the natural capacity of microorganisms to degrade or transform toxic substances from the environment into harmless products. Bioremediation technologies are in harmony with the principles of sustainable development since waste materials are not generated.

Material and Methods

The content of total petroleum hydrocarbon (TPH) in the sample was extracted as per method ISO 16703 (ISO 16703, 2004) and determined gravimetrically in accordance with DIN (DIN EN 14345, 2004). The results were calculated according to dry matter. For the analysis of humic acid, 40 g of each air-dried soil sample was mixed with 200 ml of NaOH/Na₄P₂O₇ solution (ISO 5073, 1999). The mixture was heated in a boiling water bath for 2 h, with frequently shaking to ensure precipitation of insoluble material. Then, the flask was removed from the water bath, cooled to room temperature, and supernatant was separated from residue by centrifugation at 3000 rpm (residue was discarded). Humic acids were precipitated from supernatant by acidifying with 6 M HCl to pH 1. After that, the suspension was centrifuged. The supernatant was decanted and discarded. The HA was purified according to the method of the International Humic Substance Society (IHSS). The effects of humification on the structure of humic acids were evaluated by Fourier Transform Infrared Spectroscopy (FT-IR), potentiometric titrations, the ratio of absorbances at 465 and 665 nm (E4/E6 ratio), the C/H ratio and gel chromatography.

Results & Discussion

In the pilot experiment, which lasted for 170 days, bioremediation of a petroleum pollutant was carried out in the layer of artificial soil substrate, with river sand as mother substrate. One container was a control biopile and it contained a mixture of sawdust, sand and the pollutant. The second container was an inoculated biopile and it was prepared in the same way as the control biopile but, in addition to sawdust, sand and the pollutant, it also contained biostimulants, the sources of nitrogen and phosphorus, and biomass. The initial concentration of petroleum hydrocarbons in the pilot experiment was 23,05 mg/kg dry weight. At the end of the experiment, the total hydrocarbons were reduced to 8,15 mg/kg dry weight in the inoculated biopile, while the concentration was 21,35 mg/kg dry weight in the control biopile.

It was observed that the content of humic acids increased from 0,32% to 0,42% in the inoculated biopile during the bioremediation process, while the increase in the content of humic acids in the control biopile was insignificant.

Additional analyses of humic acids from the inoculated biopile showed that they were transformed during the bioremediation process and that they had a higher degree of aromaticity as well as greater redox and buffering capacity at the end of the experiment.

In addition to this, gel chromatography was performed on the samples of humic acids isolated from the inoculated biopile during the bioremediation process. The results showed that on day 0 the share of fraction of great molecular mass (greater than 100 kDa) was 72,67%, while the share of the above mentioned fraction was 76,17%, 170 days after the beginning of the process, which was probably due to the fact that undegraded organic compounds were transformed and included in complex polymer structures which are analogues of humic substances.

Conclusions

We concluded that in parallel with biodegradation of petroleum contamination there is an increase in the content of humic substances and the structure of these substances undergoes changes as well.

This research was partially financed by the Ministry of Education, Science and Technological Development of the Republic of Serbia as a part of the Projects III 43004.