Reportes Científicos de la FACEN

Vol. 8, Nº 1 (2017): 79-88

Artículo Original

http://dx.doi.org/10.18004/rcfacen2017080179-88

BIODEGRADATION OF THE BENZOPHENONE-3 IN AGRICULTURAL SOILS AMENDED WITH COMPOST FROM URBAN WASTEWATER TREATMENT PLANTS

BIODEGRADACIÓN DE LA BENZOFENONA-3 EN SUELOS AGRÍCOLAS ENMENDADOS CON COMPOST PROCEDENTES DE ESTACIONES DEPURADORAS DE AGUAS RESIDUALES URBANAS

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Abstract: Endocrine disruptors are chemical substances capable of altering the hormonal system in living beings; which they are present in urban or industrial wastewater and can not be completely degraded by treatment in wastewater treatment plants (EDAR). Within this group of contaminants, it includes a wide variety of chemical compounds such as benzophenone-3 (BP-3) with activity as an endocrine disruptor whose biodegradation was studied in amended and unamended agricultural soils with compost from urban sewage sludge EDAR. For this, an analytical methodology has been developed and validated that has allowed the identification and quantification of this pollutant in the selected matrices (agricultural soil and agricultural soil contaminated with compost). A study of the behavior of the microbiota was performed on the treated soils. Firstly, the count of cultivable microbiota was performed on these soils, using the technique of plating. The selected microorganisms were characterized biochemically from the treated soils using kits, where it was concluded that each microorganism metabolized them differently and allowed to discriminate the microorganisms studied and to select those that were different from the metabolic aspect. Subsequently the growth kinetics of selected microorganisms and study of degradation of BP-3 in the presence of these microorganisms was studied. Later the growth kinetics of selected microorganisms and study of degradation of BP-3 in the presence of these microorganisms was studied, demonstrating the ability to grow them in the presence of this compound as a source of carbon and energy (C/E).

Keywords: *compost, soil amended, Benzophenone-3, bioremediation, biodegradation, sewage sludge.*

Resumen: Los disruptures endocrinos son sustancias químicas capaces de alterar el sistema hormonal en seres vivos; los cuales se encuentran presentes en aguas residuales urbanas o industriales y que pueden no ser completamente degradadas por tratamientos en estaciones depuradoras de aguas residuales (EDAR) la Benzofenona-3 (BP-3) se encuentra dentro de estegrupo, con actividad como disruptor endocrino y cuya biodegradación fue estudiada en suelos agrícolas enmendados y no enmendados con compost procedentes de lodos de depuradoras urbanas EDAR. Para ello se ha desarrollado y validado una metodología analítica que ha permitido la identificación y cuantificación de este contaminante en las matrices seleccionadas (suelo agrícola y suelo agrícola contaminado con compost). Se realizó un estudio del comportamiento de la microbiota en los suelos tratados. Primeramente se realizó el recuento de la microbiota cultivable en dichos suelos, mediante la técnica de siembra en placa. Se caracterizaron bioquímicamente los microorganismos seleccionados a partir de los suelos tratados empleando kits, donde se concluyó que cada microorganismo los metabolizaba de forma diferente y permitió discriminar y seleccionar a los que eran diferentes desde el punto de vista metabólico. Posteriormente se estudió a los microorganismos estudiados la cinética de crecimiento de los microorganismos seleccionados y el estudio de degradación de la BP-3 en presencia de estos microorganismos; demostrando la habilidad de crecimiento de los mismos en presencia de este compuesto como fuente de carbono y energía (C/E).

Palabras clave: *compost, suelos enmendados, Benzofenona-3, biorremediación.*

INTRODUCTION

Today's society depends largely on many chemicals, used in the many activities of daily life. Nevertheless, its massive and sometimes uncontrolled use is becoming a major concern for the different social strata, due to its possible environmental impact, since at present, there is no clear and specific legislation for many of these compounds, especially in relation to existing concentrations in the environment (Cunningham, V.L., et al 2006; Calamari, D. 1995; Richardson, M.L., et al 1985; Doughton, C.G., et al 1999). Within this group of contaminants, which are now known as "emerging", includes a wide variety of pharmaceuticals, antimicrobial and numerous chemical compounds with activity as endocrine disruptors (Cunningham, V.L., et al 2006; Calamari, D. 1995; Richardson, M.L., et al 1985. Doughton, C.G., et al 1999).

Recibido: 12/2/2016

Aceptado: 27/7/2017

Endocrine disruptors are chemicals capable of altering the hormonal system, both in humans and animals, responsible for multiple vital functions such as growth or hormonal development (Frang, H., et al 2001; King, K.A., et al 2003). Many of these compounds can have as final destination urban and industrial wastewater, being able to be degraded and removed during the treatment of them in the EDAR. However, it is demonstrated that the purification procedures are not completely effective, since these substances remain in the treated effluentes, being able to reenter the environment becoming a serious danger to the ecosystems (Petrovic, M., et al 2005; Díaz-Cruz, M.S., et al 2006; Temes, T., et al 2006).

At present time, it is thought, although there is still insufficient evidence, that the presence of these substances in this kind of environment may represent a risk to human health and ecosystems, since when they are adsorbed, soil microorganisms can not biodegrade them satisfactorily, becoming bioaccumulable, persistent and toxic substances; With the possibility of being transferred to the harvets, and to enter the food chain of living beings (Drewes, J.L., et al 2003; Kummerer, K. 2003).

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Generally, sewage sludge is subjected to composting treatments, since it is a low-cost treatment that has many advantages such as disinfection of sludge, microbial removal of some contaminants, and obtaining a substrate rich in organic matter and nutrients, suitable for the amendment of agricultural soils. In addition, it is an environmentally more favorable process compared to others such as incineration or landfill. Currently, very little

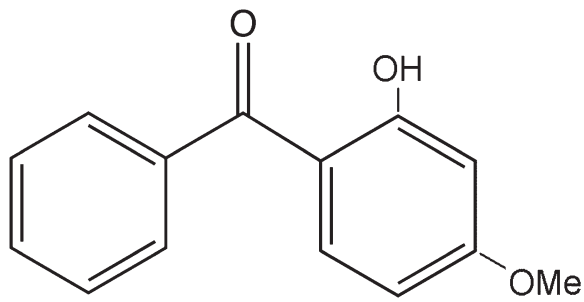

Figure 1. Chemical Structure of the benzophenone-3.

is known about the ability of this process to the definitive removal of these emerging pollutants. Ther is not any evidence of what parameters or factors contribute to the degradation of these substances during this process. On the other hand, it is not exactly known the effect of the amendment of soils with sluges or compost on the pollutants present, as well as the real risk to which humans are exposed, since these substances can be transferred, for example, to groundwater (Spellman, F.R. 2009; Cheremisinoff, N.P. 2002).

The compound selected for this study is Benzophenone-3 (BP-3, Figure 1), a white crystalline substance, insoluble in water. It acts as a filter for UV radiation as it is able to absorb it (promoting its electrons to an excited state) and dissipate it as heat. This is possible because the benzophenone has its energetic states of singlet and triplet very close together. The BP-3 is used in products such as perfumes and soaps to prevent ultraviolet light from degrading the odor and color of these products. It is also used as a component of sunscreens and can be added in the packaging plastics so that they block the UV rays protecting the product (Jeon, H.K., et al 2006; Sánchez-Brunete C., et al 2011; Zhang, Z., et al 2011).

In this work, it has been carried out a study of biodegradation of benzophenone-3 (BP-3) in the presence of microorganisms isolated from agricultural soils amended with compost from wastewater treatment plant EDAR. The microorganisms isolated from the soils were characterized and their kinetic growth was studied in the presence of the compound and in addition a chemical study of the degradation of this substance was carried out. This

allowed to study and evaluate this contaminant in agricultural soils amended and not amended with compost from sludge from urban sewage treatment plants.

MATERIALS AND METHODS

Chemicals and Reagents

Analytical grade reagents were used. The pattern of Benzophenone-3 and the internal standard Benzophenone-d_{10} were supplied by Sigma-Aldrich (St. Louis, MO). The BP-3 solution (200 mg mL^{-1}) was prepared in methanol monthly and stored at -20°C. The standard solution mixture of this compound together with the internal standard was prepared in methanol or in the mobile phase immediately prior to use. This standard was stored at 4°C and was prepared weekly. All solutions were stored in dark glass bottles. The water and methanol used for the preparation of the mobile phase were LC-MS grade supplied by Fluka (St. Louis, MO, USA). The ammonia (> 25%), acetonitrile, and ethyl acetate were purchased from Merck (Darmstadt, Germany). The water (18.2 MΩcm) was purified by a Milli-Q of Millipor system (Bedford, MA, USA). For the study of the growth kinetics of microorganisms, the base culture medium diluted 1/10 with 5 mg L^{-1} of the compound was used.

Instrumentation and software

For the collection of compost samples, conventional shovels were used and for soil samples a draft sampler (surface) and a helical auger for greater depths. The soil temperature and humidity were controlled to the different depths of the studied soil, continuously, by means of an AquaCheck probe. The experimental work was developed using a digital ultrasonic probe Sonifier S450D (BRANSON) equipped with: Type 102 converter, standard resonator 12.7 mm diameter, screw tip of 12.7 mm in diameter, temperature probe, 3 mm threaded end screwdriver, temperature probe, Micro Threaded tip with final diameter 3 mm. A Waters Acquity UPLC™ chromatograph coupled to triple quadrupole mass spectrometer Waters H-Class-Xevo TQS™, equipped with: Pump: Quaternary Solvent Manager. Acquity UPLC H Class. (Waters); Inyector: Sample Manager-FTN. Acquity UPLC. Waters; Detector: Xevo TQ-S. (Waters). The separation of the compound was obtained with a column Acquity UPLC BEH ™ C_{18} (1.7 µm; 2.1 mm × 50 mm) (Waters). An ionization source by electrodepray Z-spray™ ESI for analyte detection. The software used for the detection and integration of the peaks was the MassLynx V.4.1 SCN.803 program for the management and treatment of the data obtained by the chromatograph Waters Acquity UPLC™ H-Class - Xevo TQS™. A Vortex (Yellow line, Wilmington, NC, USA), a centrifuge Hettich Zentrifugen, Universal 32 (Tuttlingen, Germany). On the growth kinetics, the results were expressed according to the optical density relationship/count CFU mL^{-1} through a spectrophotometer (UV/VIS UNICAM 5822) to 600 nm. Finally, the equipments were used (scales, stoves, stirrers, incubators, etc.) and the usual laboratory glassware of analytical chemistry and microbiology. For the statistical treatment, a Statgraphics Plus version 5.0 (Manugistics, Rockville, MD, USA, 2000) software was used. Packages Microsoft® Office: Word®, Excel® and PowerPoint® 2007.

Experimental Development

Field study BP3

The field study was carried out on an experimental parcel which is located in the Huerta de Santa María in the Vega de Granada - Spain. In the agricultural soil, has not used any type of pesticide, insecticide or herbicide in the last ten years with the aim of not altering the soil microbiota (Araujo, A., et al 2003; Haney, R., et al 2000). The samples of compost made from sewage sludge that were used for this assay, were collected at the company "Biomass del Guadalquivir" located in Santa Fe (Granada).

In order to determine the influence of this chemical pollutant under study on the cultivable microbiota present in the agricultural soil, this compound was added to the soil of the experimental parcel, divided in this case into subparcels of 1 m^2 being the amount of compound added 1 gL^{-1} in a volume of well water of 120 L m^{-2} so that the provision of the

compound was homogeneous in the zone of application. Each subparcels was separated by tasquibas of 30 centimeters of thickness. Previously a cleaning of the existing vegetation was carried out to avoid possible interferences of the adsorption-desorption mechanisms, degradation of the compound under study, or interference with some general variable such as the evaporation index of water.

Two different conditions were set: Parcel 1 (P1) soil contaminated with BP-3 applied directly to the soil, Parcel 2 (P2) soil amended with compost and contaminated with BP-3. For this study, three samples of each subparcels were taken along time (0, 15 and 30 days) and at three depths (surface, 30 and 60 cm). The samples were transferred to the laboratory and dried at room temperature in a desiccator for 24 h, in order to remove remaining moisture. Them they were homogenized and sieved using a sterile 1 mm diameter pore sieve.

Count of the cultivable microorganisms

Through the technique of planting in plate. All plantings, as well as processing of the samples (preparation of dilutions), were carried out in a laminar flow hood. Serial dilutions (1/10) in saline solution (NaCl, 0.9%, w/v) were made from 0.1 g of each dilution in TSA medium (trypticase soy agar, Oxoid), by means of a method of sowing the drop. From each solution, three replicates were made. Subsequently, the plates were incubated in aerobiosis for 24/48 h in the oven at 28-30°C.

The results were expressed as logarithm of Colony Forming Units of cultivable heterotrophic microorganisms per gram of soil (log UFC g $soil^{-1}$).

Morphological characterization of the isolated microorganisms and selected from the different plots and studiadas

After the counting of the cultivable microbiota in the different parcel treated with the BP-3 under study, the isolates and selection of the culture-dependent microorganisms present in the different soils were carried out in solid plate medium (TSA). From each soil type, 20-25 isolates were made. The selection of microorganisms was performed according to morphological criteria of the colonies (shape, elevation, edge, size and coloration) as well as microscopic criteria (Gram, morphology, and clustering). In order to obtain an initial information about the microorganisms present in the study soils, Gram staining of the selected microorganisms was performed. This staining generated information about the type of cell wall as well as the morphology and type of cell grouping. Subsequent observation under the microscope allowed to ascertain the basic morphological characteristics of the working microorganisms.

Kinetics of growth of microorganisms and degradation studies of the compounds

A battery of microorganisms from the soil samples treated with the different compounds and composts (coded with the initials of the compound added to that parcel and a consecutive number). The selection was made based on morphological criteria of the colonies. In addition, 10 of the microorganisms were used for the growth and degradation kinetics assays. For the kinetics, a base culture medium diluted 1/10 with 5 mg L^{-1} of the test compound was used and incubated at 30°C for five days.

After the study of microbial counts obtained from the samples of the parcels treated and not treated with compost and the microcontaminant object of study, isolates were made in order to select those microorganisms capable of growing and degrading the different compounds.

Isolates were plated with TSA culture medium. The selection of microorganisms was performed based on morphological criteria of isolated colonies. Of all the microorganisms isolated, a selection of 10 microorganisms was finally carried out for the kinetic growth and degradation assays of each analyte.

The tests were carried out in sterile centrifuge conical bottom tube according to Table 1. With the analyte used as a source of C / E, 20 cultures were carried out + 1 flasks without microorganism subjected to the same incubation process (chemical control).

Table 1. Description of tests (10 microorganisms by analyte).

Test	Samples	Analysis Type
Half TSB1/10 + microorganism + analyte	10	Microbiological / Chemical
Half TSB1/10 + microorganism	10	Microbiological (Controls)
Half TSB1/10 + analyte	1	Chemical (Control)

Culture medium and inoculum. 15 mL TSB diluted 1/10 supplemented with 50 mg L^{-1} of the test compound. Diluted TSB was used according to the Valkova assays(Valkova, N., et al 2001). The inocula were prepared in TSB medium diluted 1/10. Initial inoculation / assay = 105 CFU mL^{-1}.

Incubation, sampling and reading results. All tubes were incubated on an orbital shaker (New Brunswick E25R) at 100 r.p.m. At 30°C for 5 days.

Microbiological analysis. The optical density measurement was performed on a spectrophotometer (UNICAM UV / VIS 5821) at 600 nm. The readings were performed at 0 h, 24 h, 48 h, 72 h and 96 h. The results of UFC mL^{-1} were obtained according to the different calibration curves (relationship optical density D.O. / count UFC mL^{-1}).

Chemical analysis using UPLC-MS/MS. The amount of degraded compound was determined by the UPLC-MS/MS technique using the conditions described in Table 2. After the sample was centrifuged at 13,000 r.p.m. for the removal of cells. Measurements were performed at initial and final test time (0 hours and 96 hours).

Table 2. Optimal values of the chromatographic conditions.

Stationary Phase	ACQUITY UPLC® BEH C18 (2.1 x 100 mm) 1.7µm		
Mobile Phase	A: Water (0.025 % ammonia) B: methanol (0.025 % ammonia)		
Modality: Gradient	Time (min)	A %	B %
	0.0	40	60
	6.0	0	100
	6.5	0	100
	6.6	40	60
	10.0	40	60
Flow	0.3 mL min^{-1}		
Injection volume	4 µL		
Oven temperature	40 °C		
Detection	ESI-MS/MS		

Table 3 shows optimized parameters for chromatographic separation and mass spectrometric detection. Once these variables were optimized, the two transitions were selected in the spectrometer (quantification and identification), optimal for the unequivocal determination of the molecule.

Biochemical characterization of microorganisms

In order to determine the behavior of the microorganisms in relation to their ability to use different substrates as a source of carbon and energy (C/E), as well as their enzymatic capacity, tests were performed for

Table 3. Parameters of the mass spectrometer for the BP-3 (**VC:** Voltage of cone; **EC:** Energy of collision).

Analyte	Transition	VC / EC	Transition	VC / EC	Mode
BP-3	229.0 → 150.8	4/20	229.0 → 104.9	4/18	ESI(+)
BP-d_{10}	193.1 → 109.8	18/16	193.1 → 81.8	18/30	ESI(+)

API 50 CH (study of carbohydrate metabolism) and ZYM API (study of enzymatic activity).

The API 50 CH system is a standardized system consisting of 50 biochemical tests for the study of carbohydrate (CH) metabolism in microorganisms. The API 50 CH is used in combination with the API 50 CHB/E medium for the identification of: Lactobacillus and nearby microorganisms of Bacillus and nearby microorganisms and *Enterobacteriaceae* and *Vibronaceae.*

The gallery API 50 CH is composed of microtubes and allows the study of the use of substrates belonging to the family of carbohydrates and derivatives (heterosidos, polysaccharides, uronic acid). Fermentation assays were inoculated into API 50 CHB/E medium which rehydrates the substrates. During the incubation period, the use of HC results in a color change in the microtube, due to acid production in anaerobiosis (fermentation) and in aerobiosis (oxidation) revealed by the pH indicator of the chosen medium. The first microtube, without active principle, serves as a negative control.

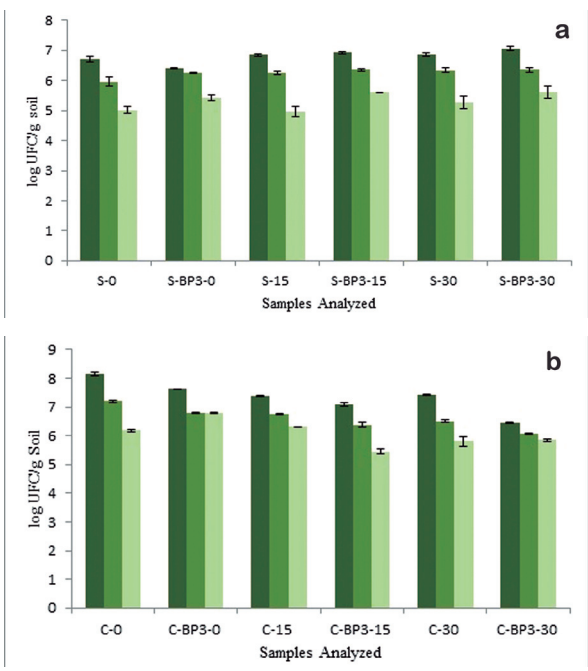

Figure 2. Example of counting of microorganisms (Log CFU/g soil) in samples of soil treated and non treated with BP-3 at different depths, at initial test time (0 days), 15 and 30 days. (a) ground control without compost (S) and contaminated with the compound (S-BP-3). (b) Control of soil + compost (C) and soil + compost contaminated (C-BP-3). Surface (dark tone); 30 cm (medium tone); 60 cm (clear tone).

Of the microorganisms selected, studies were carried out on both oxidation and fermentation, since this gallery allows studying both pathways. The oxidation was investigated in unsealed microtubes, and the fermentation in microtubes sealed with liquid paraffin. The test container consists of 10 galleries API 50 CH, 10 inoculation chambers, 10 results sheets and a technical sheet.

For the test were used inoculation medium (API 50 CHL Medium), paraffin oil, McFarland Standard scale and distilled water. The galleries were kept at 2-8°C until the date of their use.

The microorganisms to perform the carbohydrate test was the one capable of degrading high percentages of the contaminant, specifically the microorganism codified as BP3-5. Likewise, the most representative of the group of samples, i.e. BP3-5, BP3-7, BP3-10, BP3-14, BP3-16, were selected for the enzyme activity test.

RESULTS AND DISCUSSION

Microbiological Study

Influence of the presence of the BP-3 and of compost in the count of the microbiota of soil

The percentage of log UFC/g of soil obtained in soils without compost treatment (Figure 2 (a)) was different from the surface samples (100%), with a decrease in microorganisms observed of 8.1% and of one 21.8% for samples collected at 30 and 60 cm, respectively.

In soils amended with compost and contaminated with BP-3 (figure 2 (b)), was decreasing, with a reduction of 0.5 log in the CFU count ground/g along the test period. If both figures are compared, a downward trend of the counting is observed as the soil depth increases; this behavior is repeated in all the tests performed over time (0, 15 and 30 days).

In the counts of both control parcels and BP-3 samples, no significant variations in surface counts (2 centimeters) were observed, indicating that population levels remained quantitatively constant throughout the study. Likewise, both counts at 30

and 60 centimeters of depth indicated that there were no significant variations in time in relation to the number of bacteria present in the control soil samples as in the treated ones, obtaining very similar values in the number of microorganisms in samples collected at 30 centimeters as well as those at 60 centimeters deep.

According to the results obtained, an increase in the absolute number of microorganisms in the presence of compost with respect to the unamended soil was observed, since the composted material not only provides chemical nutrients, but also increases the microbial wealth. Therefore, there is greater microbial development in the amended parcels.

This fact would be very useful in promoting an improvement in the characteristics of the compost, isolating the microorganisms of greater interest and supplementing the material with them.

Morphological characterization of the isolated microorganisms of the study plots

Of the 20 to 25 microorganisms isolated from the parcel treated with BP-3, 10 were selected and their morphological characterizations of the colonies and their morphological criteria were performed (Table 4). The microorganisms observed in the soils treated with BP-3 were isolated five Gram-positive bacilli, two Gram-negative bacilli and three Gram-positive cocci.

Table 4. Morphological characteristics of microorganisms isolated in soils treated with different compounds.

BP-3			
Identification	**Gram**	**Morphology**	**Grouping**
BP-3-1	+	Bacillus	Isolated
BP-3-5	+	Bacillus	In string
BP-3-7	+	Coco	In string
BP-3-10	+	Coco	In string
BP-3-11	+	Bacillus	Isolated
BP-3-14	+	Bacillus	In string
BP-3-16	+	Coco	In string
BP-3-18	-	Bacillus	Isolated
BP-3-20	-	Bacillus	Isolated
BP-3-21	+	Bacillus	Isolated

Biochemical characterization of the microorganisms selected

The use of API 50 CH kits for carbohydrate metabolism and API ZYM for enzymatic activity allowed the characterization of numerous microorganisms.

From the results obtained in the processes of oxidation and fermentation of carbohydrates, it was concluded that each microorganism metabolized them differently. This data, together with the results of enzymatic activity, which reports on the presence or absence of a certain activity of an enzyme, group of them or a certain metabolic pathway, allowed to discriminate the microorganisms studied and to select those that were different from the metabolic point of view.

Growth kinetics in the presence of BP-3 as a source of C/E. Degradation of the compounds

Figures 3 (a) and (b) show an example of growth kinetics of different microorganisms in the presence of the contaminant studied.

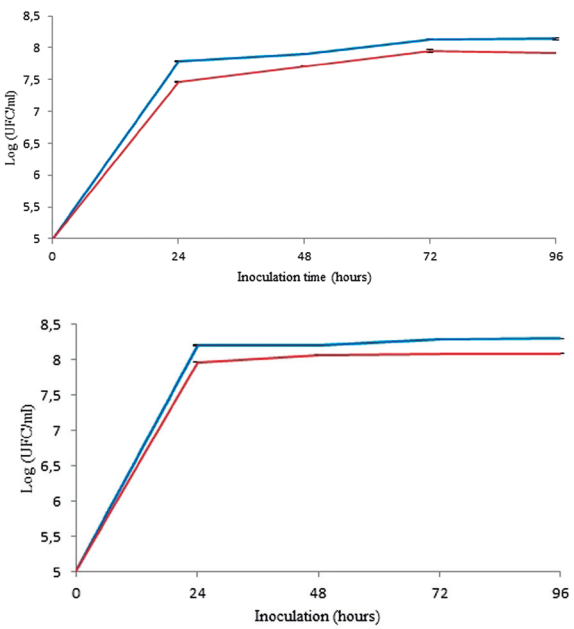

Figure 3. Kinetic growth examples of the microorganism encoded as BP-3-1 **(a)** and BP-3-5 **(b)** in a culture medium containing 5 ppm of BP-3. Blue line: growth of the microorganism in TSB 1/10 medium supplemented with BP-3. Red line: control, growth of the microorganism in TSB medium 1/10.

Table 5. Relationship between cultures of different microorganisms in the presence and absence of BP-3 at 48 h incubation. **A**: cultivation in the presence of compound. **B**: cultivation in the absence of compound. The results are expressed as log CFU mL $^{-1}$.

Microorganism	A - B (Log CFU mL $^{-1}$)	Microorganism	A - B (Log CFU mL $^{-1}$)
BP-3-1	0.19	BP-3-14	0.41
BP-3-5	0.14	BP-3-16	0.33
BP-3-7	**0.76**	BP-3-18	0.21
BP-3-10	0.26	BP-3-20	0.33
BP-3-11	0.31	BP-3-21	0.16

The results showed that the selected microorganisms were able to grow in the presence of the contaminant and that this growth was superior to that obtained in the base medium in the absence of the compound (control).

As a result of the growth kinetics of the selected microorganisms, the growth difference (log UFC mL^{-1}) between the cultures in the presence of BP-3 and the control cultures without compound after 48 h of test is shown in Table 5. The greatest difference in growth between both cultures was observed in the case of the test with the microorganism BP-3-7, this being 10.0%.

Degradation of BP-3 in the presence of different microorganisms

The amount of the degraded compound in the presence of the different microorganisms was similar in all tests performed. In Table 6 it is observed that after 96 h of culture, all the microorganisms tested degraded the compound in a concentration higher than 75%, with almost total degradation in the case of microorganism BP-3-1 (99.3%) and BP- 3-14 (99.9%).

The microorganisms that degraded the compound in a lower percentage were BP-3-7 (78.0%) and BP-3-11 (76.3%).

CONCLUSION

As a result of this research the following main conclusions have been obtained:

The counting of the cultivable heterotrophic microbiota of the soils treated with BP-3 and of the compost-amended soils and treated with this pollutant showed that, as the soil depth increases, there is a progressive decrease in the number of viable microorganisms. On the other hand, it was observed that the greatest microbial development occurred in the parcels amended with compost.

The study of the morphological and staining characteristics of the microorganisms isolated from the soils treated with benzophenone 3 showed a higher proportion of Gram (+) and Gram (-) bacilli.

Table 6. Final concentration of BP-3 (ppm) determined using LC-MS/MS. Initial concentration 5 ppm. **To:** final concentration (ppm) of BP-3 detected after 96 h of culture with different microorganisms isolated from MPB treated soils. **B:** Concentration of degraded BP-3 (ppm after the end of the test.

Microorganism	To (ppm)	B (ppm)
BP-3-1	0.03	4.05
BP-3-5	0.43	3.65
BP-3-7	0.89	3.18
BP-3-10	0.36	3.72
BP-3-11	0.96	3.12
BP-3-14	$1.0 . 10^{-3}$	4.08
BP-3-16	0.59	3.48
BP-3-18	0.53	3.55
BP-3-20	0.27	3.81
BP-3-21	0.68	3.39
CONTROL	**4.08**	

The isolated compound was degraded to a greater or lesser proportion by the selected microorganisms, with degradation rates above 70% in all cases.

The results obtained demonstrate the growth ability of the microorganisms selected in the presence of this compound as a source of C/E.

Therefore, these microorganisms could be used in bioremediation processes of environments contaminated with benzophenone-3.

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