

BIOSTIMULATION OF DOMESTIC WATER CONTAMINATED BY A MIXTURE OF HYDROCARBONS

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Juan Manuel Sánchez-Yañez

Environmental Microbiology Laboratory,
Institute of Chemical-Biological
Research). University City. Universidad
Michoacana de San Nicolás de Hidalgo,
Fco J. Mujica S/N Col Felicitas del Rio, ZP
34,000,Morelia, Michoacan, México
0000 0002 1086-7180

Gladys Juárez-Cisneros

CONACYT, El Colegio de Michoacan/
LADIPA, La Piedad Michoacan, México.
0000-0002-2918-0805

Juan Luis Ignacio de la Cruz

Environmental Microbiology Laboratory,
Institute of Chemical-Biological
Research). University City. Universidad
Michoacana de San Nicolás de Hidalgo,
Fco J. Mujica S/N Col Felicitas del Rio, ZP
34,000,Morelia, Michoacan, México
0000-0002-5638-3720

ABSTRACT: Water is a critical factor for life, currently the recovery of domestic water impacted by waste motor oil (WMO) for reuse involves biological methods such as biostimulation. The objective of this work was to analyze the biostimulation of domestic water impacted by 12,000 ppm

of WMO with two mineral solutions 1 and 2. For these 2 mineral solutions were used to eliminate the WMO mediated by the production of CO₂ and the disappearance of the WMO by gas chromatography. The results show that it was possible to remove the WMO from the water with one of the mineral solutions based on the concentration that facilitated the rapid oxidation of the WMO. It is concluded that the biostimulation of domestic water with mineral solutions is viable for the possible reuse of domestic water to avoid using clean water in industrial irrigation without environmental health risk.

KEYWORDS. water, bioremediation, hydrocarbon oxidation, intelligent use of water.

INTRODUCTION

Its estimated that 240 million liters of automotive lubricating oil are sold annually in Mexico (1-4). At the end of its useful life in automobiles, with the oil change, an undetermined amount is illegally thrown into the drain by individuals and mechanical workshops, with the consequent contamination of domestic water (5,6).

Used crankcase oil, or waste residual oil (WMO), is a brown to black liquid that is removed from automobile engines (1,2,7). WMO is made up of 80% aliphatic hydrocarbons, 20% simple and polycyclic aromatics (4,5,) and trace metals: aluminum, chromium, copper, iron, lead, manganese, nickel, silica and tin; from engine parts that wear out (1,3,4,8). According to the United States Environmental Protection Agency (7), one liter of WMO seeps into an aquifer can contaminate up to one million liters of water (8,9). An alternative to restore the environmental damage caused by the inadequate disposal of WMO, is bioremediation through biostimulation (10,11) with inorganic and organic compounds of N (nitrogen), P (phosphorus) and K (potassium) that are basic elements of the life, those that induce the native heterotrophic aerobic microorganisms, to the mineralization of hydrocarbons similar to those found in the WMO (9-10). The biostimulation of environments impacted by WMO, as occurs in the soil, is based on the use of various strategies that induce heterotrophic microbial activity, WMO oxidation (11-14). In contrast, there are few reports on the biostimulation of domestic water impacted by WMO, especially because to eliminate aliphatics, one of the most common constituents of this mixture (2-4), it is essential to enrich it with an adequate and balanced mineral solution at basis of N, P and K; along with a sufficient concentration of O₂ (oxygen) for its oxidation and conversion into H₂O and CO₂ products that are innocuous to the environment (15-17). In this regard, report that the biostimulation of an environment impacted by WMO it is essential to start with a detergent that emulsifies them, followed by enrichment with a mineral solution and O₂ that allows the rapid oxidation of the WMO. In general, the information on the dynamics of biostimulation in aquatic environments impacted by WMO is scarce (7, 18-20). Therefore, the objective of this work was to analyze the biostimulation of domestic water contaminated by 12,000 ppm of residual automotive oil with two mineral solutions.

MATERIALS AND METHODS

Domestic water contaminated with WMO was collected from a car wash and grease shop in Morelia, Mich., Mexico. For the biostimulation analysis of domestic water contaminated by WMO, 500 mL Bartha flasks were used with 100 mL of WMO diluted 1:100 (final concentration 12,000 ppm of WMO); those two mineral solutions were tested, mineral solution1 (g/L): K₂HPO₄ 5, KH₂PO₄ 4, MgSO₄ 3, NH₄NO₃ 10, CaCO₃ 1, KCl 2, ZnSO₄ 0.5, CuSO₄ 0.5, FeSO₄ 0.2; and mineral solution 2 (g/L): K₂HPO₄ 1.17, NaHPO₄ 4.30, MgSO₄ 0.22, NH₄NO₃ 8.1, CaCl 0.27, FeCl₃ 0.22, NH₄Cl 1.9, NaMoO₄; EDTA 8 and detergent tween 20 (0.5mM). As absolute control: WMO was used domestic water diluted non biostimulated by mineral solution; as relative control 1: WMO diluted in distilled water biostimulated with mineral solution; as relative control 2: WMO in domestic water biostimulated with mineral solution.

These flasks were shaken at 100 rpm; then were incubated at room temperature

for 18 days; as a control, a relative 1 was used: a Bartha flask with domestic water diluted polluted by WMO, non biostimulated with mineral solution; as relative 2: a Bartha flask with distilled water, biostimulated with mineral solution; as relative control 2 was sterile control: a Bartha flask with domestic water polluted by WMO biostimulated with mineral solution sterilized at 121°C / 15 min. The Bartha flasks were incubated under shaking at 100 rpm at room temperature for 18 days. To quantify the generation of CO₂ derived from the mineralization of the WMO, due to biostimulation by mineral solution, were taken 10 mL of 0.1 N KOH was added to one of the arms of the Bartha flask, every 24 h /18 day. The remaining alkali was determined by titration with 0.1 N HCl (2,4,7). All experimental data were analyzed by ANOVA and Tukey ($P \leq 0.05$), to establish the minimum significant difference (11).

RESULTS AND DISCUSSION

In domestic water, the composition of aliphatic of the WMO was analyzed before biostimulation, shown in the chromatogram of figure 1. The estimated concentration of WMO estimate of 12,000 ppm, a level higher than what environmental standards in Mexico and the world consider risky for environmental health (1, 2, 20, 21). What forces to look for effective methods of remediation that allow reuse (10-15) especially due to the problem of scarcity of drinking water for the population and needs of the countryside, the cities of the world (3,4-7).

In the figure 2, the biostimulation of domestic water impacted by WMO by the mineral solution 1 induced to native microbiota heterotrophic aerobic to start the mineralization of WMO at the sixth day achieving the maximum at fifteenth day to the highest oxidation of the WMO (17-19) due to enrichment by the optimal concentration of the inorganic compounds of N, P and K (12-14) which was detected by the production of CO₂ with a maximum of 5.52 mmol mL⁻¹. In evident contrast with the biostimulation with the mineral solution 2 when the concentration level of the inorganic compound of N, P, K and other were not enough to induce the activity of the mineralization native microbiota of domestic water to oxidate the WMO (17-19).

In contrast to domestic water non biostimulated with mineral solution there the minimum amount of CO₂ was detected with 0.224 mmol•mL⁻¹; these results were consistent with the mass-coupled gas chromatography analysis, showed in figure 4 that indicates with certainty that the nutritional factor was the main limitation of the biostimulation of domestic water impacted by WMO, since the native heterotrophic aerobic microbiota of the domestic water was unable to eliminate the WMO (11,12), due to the imbalance of essential minerals for the oxidation of the WMO (13-16) that explains why certain concentration of the inorganic compounds of N and phosphates of the mineral solution1 (11,15) are important since due to biostimulation the diverse heterotrophic microbial populations of the water was possible

to oxidate the main aliphatic of the WMO (2,3,20). While the microbiota did not respond positively to the components of the mineral solution 2, that did not provide the enough concentration of the essential nutrients required to oxidize these WMO aliphatics (12, 17) consequently the amount of CO₂ generated was similar to that detected in the mixed distilled water with WMO (C); where are not basic nutrients and neither detergent and enough number of microorganisms to mineralize of WMO. While when WMO was emulsified with tween 20 even the biostimulation by the mineral solution, the sterilization kills all microorganisms able to eliminate the WMO (D); in the other side even that WMO was emulsified with tween 20 in distilled water had two main problems not biostimulation by the mineral solution 1 and not enough diversity and number of microorganisms able to eliminate the WMO (E). Supported by a statistical difference observed among the results of CO₂ produced according each experiment done (16,18). These assays explain why domestic water polluted by WMO is a source of wide diversity of microorganisms that depends on the biostimulation with tween 20 that emulsified of WMO and then due the specific mineral solution in terms of concentration and availability of basic nutrients was necessary to induce the maximum of elimination of WMO for domestic water recovery polluted by this type of mixing hydrocarbons (12,14-16).

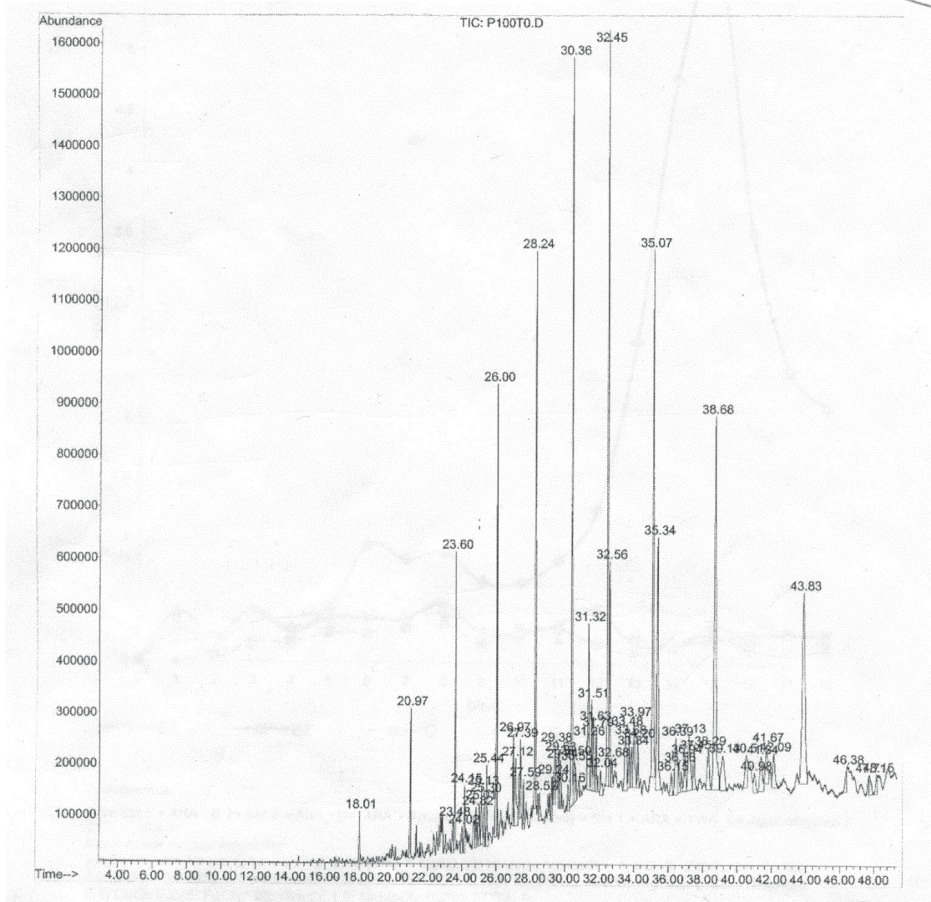


Figure 1. Chromatogram of the domestic water polluted by 12,000 ppm of waste residual at time zero of the biostimulation with mineral solutions.

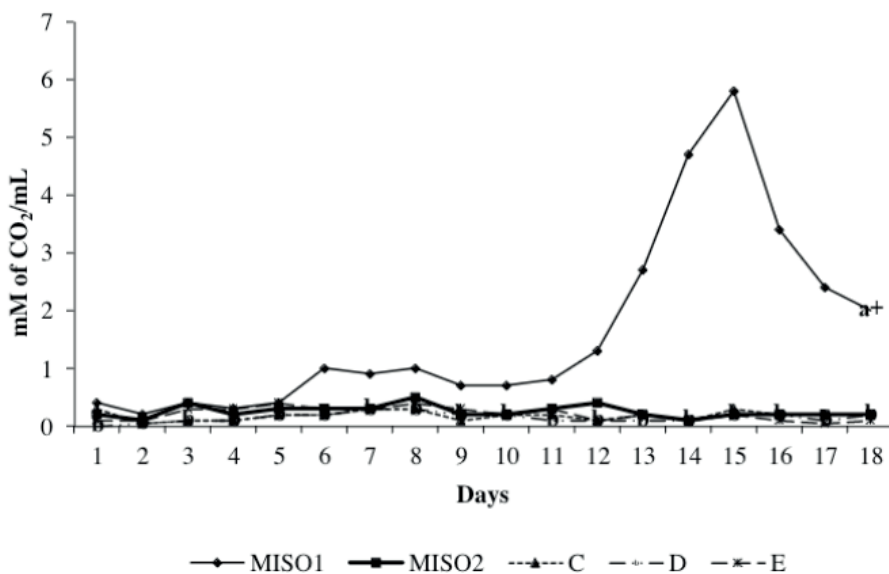


Figure 2. CO₂ production by the microbiota of domestic water polluted by 12,000 ppm of waste residual oil, biostimulated with two mineral solutions

Treatments:

B1=mineral solution 1 (**MISO1**) + WMO; **B2**= mineral solution 2 (**MISO2**) +WMO; **C**=WMO+ distilled water; **D (sterile)**=mineral solution 1+WMO+Tween 20; **E**= distilled water +

Tween 20 +WMO

WMO=waste motor oil

a+ and **b** had statistically different according to ANOVA, Tukey ($P \leq 0.05$).

MISO1= mineral (g/L): K₂HPO₄: 5; KH₂PO₄: 4; MgSO₄: 3; NH₄NO₃: 10; CaCO₃: 1; KCl: 2; ZnSO₄: 0.5; CuSO₄: 0.5; FeSO₄: 0.2;

MISO2= mineral solution (g/L): K₂HPO₄: 1.179; Na₂HPO₄: 4303; MgSO₄: 0.225; NH₄NO₃: 8.1; CaCl: 0.275; FeCl₃: 0.25; NH₄Cl: 1.9; NaMoO₄: 0.250; EDTA: 8, Tween 20 (0.5mM).

Figure 3 shows the analysis of the response of the microbiota in domestic water impacted by WMO biostimulated with mineral solutions 1 and 2 in the mineralization of the WMO, where a statistical difference was observed in the nutritional induction of the heterotrophic microbiota with the mineral solution1 (15,20,21), in the oxidation of the WMO, with a CO₂ value of 5.7 mM /mL, in contrast to the microbiota of domestic water impacted by the WMO without biostimulation by the concentration of the basic components of the mineral solution 2 that did not allow the WMO oxidation with numerical value (19,22,23), non-statistical difference compared to the CO₂ concentration by the microbiota of domestic water impacted by WMO, without biostimulation with the mineral solution used as negative control (23). The positive effect of biostimulation on the heterotrophic aerobic microbiota of WMO-impacted domestic water to oxidize it (11, 12) were consistent with mass-coupled gas chromatography analysis (22,23) that specifically demonstrated the aliphatic types that were removed (5). In comparison with the domestic water impacted by WMO non

biostimulated that supports the concentration of essential minerals to induce microbial consortia of domestic water for mineralization of the aliphatics of the WMO with mineral solution 1 (13,14,15).

Figure 4 shows the biostimulation of domestic water impacted by WMO with mineral solution 1 in the mineralization of the aliphatics of the WMO 18 days after the start of the assay here, a drastic decrease in the concentration (observe the axis and the relative scale of abundance) of the aliphatics and aromatics of the WMO was observed due to the mineralization capacity of the aerobic heterotrophic microbiota of domestic water induced by the sufficient concentration of N salts, P and K of mineral solution 1 (14,16,20) that allowed a reduction of the initial concentration of 12,000 ppm of WMO to a value lower than 1000 ppm, according to concentration of hydrocarbons remaining, that implies the possibility of reusing this water for irrigation of gardens and industrial processes without risk to the environment (13,18,19).

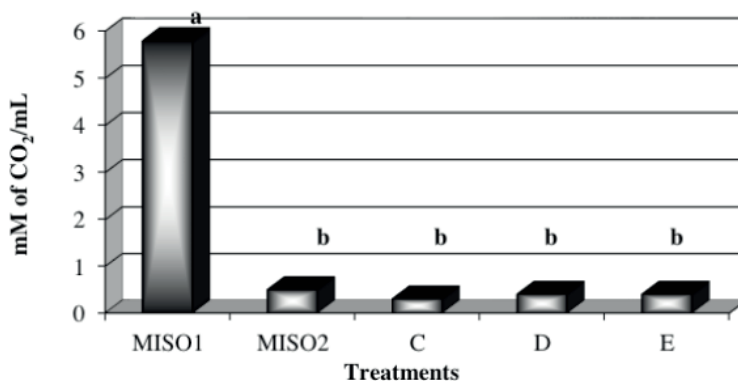


Figure 3. CO₂ production in domestic water impacted by 12,000 ppm of waste motor oil biostimulated with two mineral solutions

Treatments:

MISO1=mineral solution 1+ WMO + domestic water; **MISO2**= mineral solution 2+WMO + domestic water **a** and **b** had statistically different according to ANOVA, Tukey ($P \leq 0.05$)

C=WMO+ distilled water; **D** (sterile)=mineral solution 1+WMO+Tween 20; **E**= distilled water + Tween 20 +WMO

WMO=waste motor oil

MISO1= mineral (g/L): K_2HPO_4 : 5; KH_2PO_4 : 4; $MgSO_4$: 3; NH_4NO_3 : 10; $CaCO_3$: 1; KCl: 2; $ZnSO_4$: 0.5; $CuSO_4$: 0.5; $FeSO_4$: 0.2;

MISO2= mineral solution (g/L): K_2HPO_4 : 1.179; Na_2HPO_4 : 4303; $MgSO_4$: 0.225; NH_4NO_3 : 8.1; CaCl: 0.275; $FeCl_3$: 0.25; NH_4Cl : 1.9; $NaMoO_4$: 0.250; EDTA: 8, Tween 20 (0.5mM).

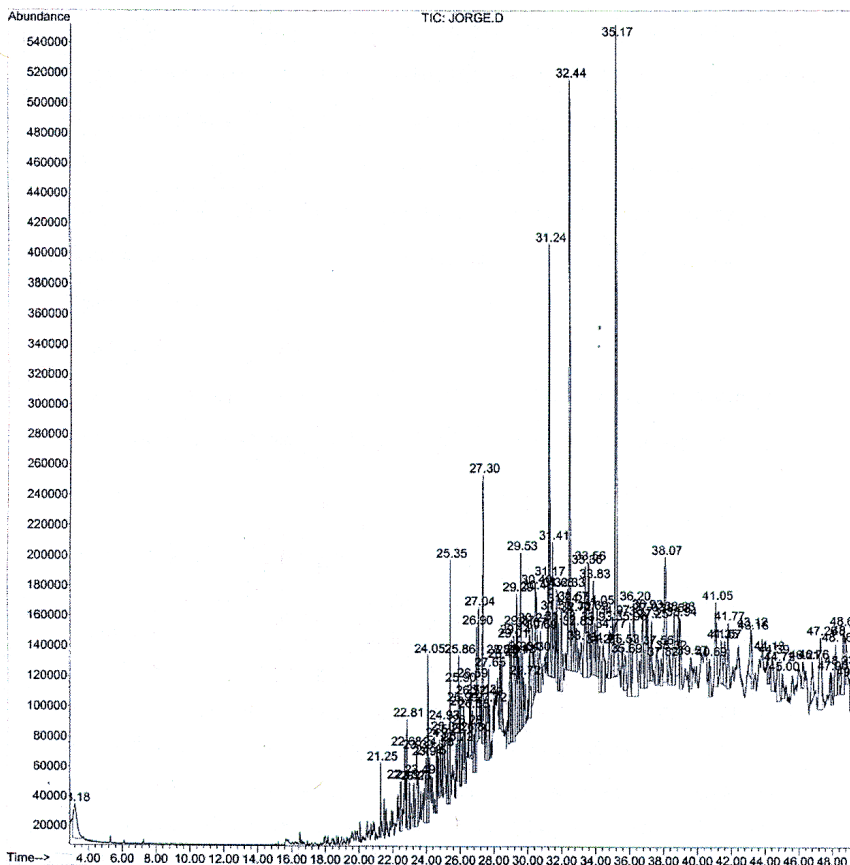


Figure 4. Chromatogram of the remaining hydrocarbons from biostimulation with two mineral solutions of domestic water impacted by 12,000 ppm of waste motor oil

CONCLUSION

The microbiota of the domestic water biostimulated with the mineral solution1, improve the aliphatic elimination of the WMO by an equilibrium concentration of the mineral salts related to the N, P, C of the domestic water polluted by WMO. That induced the aerobic heterotrophic microbiota to mineralize the aliphatics from the WMO. These results support that biostimulation with a mineral solution in nutritional balance is a viable strategy for the recovery of domestic water impacted by WMO, with a high percentage of oxidation of more than 66% of toxic hydrocarbons of this mix.

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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