

Research Article





Biorecovery of sewage polluted by waste motor oil

Abstract

An acute problem in México and everywhere is the reutilization of sewage polluted by hydrocarbon, such as waste motor oil (O), a toxic waste according to the General Law of Ecological Balance and Environmental Protection and NOM-001- SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003-ECOL-1997, indicate that the maximum permissible limits of 25 ppm of hydrocarbons in sewage 75 ppm of wastewater to systems of urban sewage and 15 ppm for treated wastewater for public reuse, respectively, which, when exceeding the total of these values, inhibiting the treatment of that domestic sewage. An alternative solution is biostimulation with detergent, minerals and O, (oxygen) that induce the aerobic heterotrophic microbial population in the sewage to eliminate WMO and reuse it. The objective of this work was the biostimulation of domestic sewage contaminated by AWO until it decreased to a value lower than the maximum of the NOM-001-SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003-ECOL-1997. For this, the sewage impacted by WMO was diluted and biostimulated with the detergent Tween 80, a mineral solution (MS) and H2O2 as a source of O2, using the response variables: i) CO2 production due to the mineralization of AWO in sewage, ii) determination of the decrease in the concentration of WMO in sewage by gas chromatography coupled to mass (GC-MS) and by Soxhlet, the experimental data was analyzed by ANOVA/Tukey HSD ($P \le 0.05$). The results indicate that the BIS of the water impacted by WMO with Tween 80, MS and H₂O₂, reduced the concentration to a value of 10 ppm, lower than that established by the NOM-001-SEMARNAT-1996, the NOM-002-ECOL -1997 and NOM-003-ECOL-1997, due to mineralization of the WMO and the evidence of its disappearance according to the CG-EM analysis. This demonstrated the biorecovery of water contaminated by WMO allow industrial and/or recreational reuse.

Keywords: water reautilization, hydrocarbons, bioremediation, public and enviornmental health

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Introduction

In Mexico, waste motor oil (WMO), a product of the petrochemical industry, is a complex mixture of linear, branched, and polycyclic aromatic aliphatic hydrocarbons, due to lubrication cycle of automobile engines that at the end of its useful life is not properly disposed of by mechanical workshops that change the oil,2 WMO throw in the drain and avoid reuse treatment,3 in partly because components of WMO are toxic to life.4 When WMO impacts surface water and groundwater the General Law of Ecological Balance and Environmental Protection (GLEBEP), which classifies it as a hazardous waste; underline in swage due to regulations: NOM-001-SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003-ECOL-1997. The spillage of WMO in the drainage system is real problem because it is not cleaning up by sewage treatment, causing other enviornmental due to the scarcity of water for reutilization when level of WMO is over 25 ppm for, 75 ppm for urban sewage system and 15 ppm for reutilization of this type of water in public service. An ecological way to eliminate WMO impacted sewage is the biostimulation with detergent that emulsifies hydrocarbons, followed by biostimulation with a mineral solution (MS), with minerals base in: N (nitrogen), P (phosphorus), etc, and also demands O₂ (oxygen) source to improve of WMO mineralization.⁵⁻⁷ However, cleaning up of sewage impacted by petroleum derivatives, it is not enough understood.8 Based on the above, the objective of this research was the biostimulation of sewage contaminated by WMO to eliminate at lower level accepted by NOM-001-SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003. -ECOL-1997.

Materials and methods

This research at was conducted in microcosmos of Bartha respirometer at the environmental microbiology laboratory, Instituto

de Investigaciones Químico-Biológicas at Universidad Michoacana de San Nicolás de Hidalgo, Morelia, Mich., México.

Biostimulation of sewage polluted by waste motor oil

In that sense 500 mL Bartha flasks were used (Figure 1), with 100 mL of swage polluted by WMO (from auto mechanic shop in Morelia, Mich, Mexico) diluted 1:100, equivalent to 10,000 ppm biostimulated with 0.01% of Tween 20, following by mineral solution (MS) with this chemical composition (g•L-1): K₂HPO₄, 5.0; KH₂PO₄, 4.0; MgSO₄, 3.0; NH₄NO₅, 10.0; CaCO₅, 1.0; KCl, 2.0; ZnSO₄, 0.5.0; CuSO₄; 0.5; FeSO₄, 0.2, and EDTA 8. The flasks were shaken at 100 rpm and incubated at 30 ± 2 °C/3 weeks, the experiment was carried out in 4 repetitions. As a relative control, we used: a flask with sewage sludge impacted by diluted WMO without biostimulation; as absolute control a flask with distilled water, biostimulated with Tween 20 and MS; as a sterile control, a flask with sterilized water (121°C/15 min), with Tween 20, impacted by WMO biostimulated with MS. As treatment 1, a flask with sewage impacted by WMO, biostimulated with Tween 20 and MS; as treatment 2 a flask with sewage impacted by WMO, biostimulated with Tween 20, MS and H₂O₂, as shown in Table 1. To demonstrate the mineralization of WMO in the sewage by biostimulation, in each of the arms 10 mL of 0.1 N KOH was added to the flask to capture the CO₂, every 24 h the 0.1 N KOH was taken from each flask, the CO2 production was quantified by titration using 0.1 N HCl.9

Quantification of WMO aliphatic hydrocarbons in biostimulated sewage

In sewage polluted by WMO biostimulated with Tween 20, SM and H₂O₂, it was measured by GC-MS analysis, and by the Soxhlet method. For this purpose, a Hewlett-Packard (Waldbroon, Germany)





6890 series gas chromatograph coupled to a 5792 A series mass spectrophotometer was used, with a 30 m long HP-5MS capillary column, with an internal diameter of 0.25 mm and a film thickness of 0.25 mm, the injection was split mode, the carrier gas was He (helium), with a flow rate of 37 cm/sec⁻¹, the oven temperature was 40°C/8 min, with an increase to 180°C/6°C min⁻¹ and the injector temperature was 250°C.¹⁰

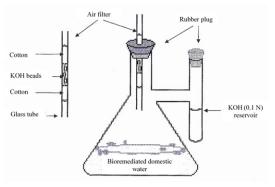


Figure I Bartha respirometer to measure biostimulation in sewage impacted by waste motor oil.

Table I Experimental design for the biostimulation of sewage polluted by waste motor oil with Tween 20, with a mineral solution and H_2O_2 under agitations

Variables	Treatments*				
	Relative control	Absolute control	Sterile control	T-I	T-2
WMO	+	-	-	+	+
Sterile WMO	-	-	+	-	-
Distilled water	-	+	-	-	-
Mineral solution*	-	+	+	+	+
Tween 20: 0.01%	+	+	+	+	+
Agitation: 100 rpm	+	+	+	+	+
H2O2: 25 ppm	-	+	-	-	+

^{*}Number of repetitions (n) =3; added (+); not added (-).

Statistical analysis

All results data, were subjected to ANOVA analysis of variance and Tukey comparison of means ($P \le 0.05$), by Statgraphics Centurion statistical program.¹¹

Results and discussion

Figure 2 shows the production of CO₂ during the biostimulation of sewage with Tween 20, MS and H₂O₂ in agitation, where the mineralization of WMO was detected, measured indirectly by amount of CO, releasing with a maximum 3.5 mmol/mL at 24 h. Some research done related with supports that, in the sewage, the limiting factor of WMO elimination depends enough basic minerals to supply for native microorganism in sewage. In balance, allows the diversity aerobic heterotrophic microorganism to mineralize WMO, in a relatively short time, due that aliphatic are the main hydrocarbons of WMO, according to Soxhlet analysis was 10 ppm, a value lower than the maximum accepted by NOM-001-SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003-ECOL-1997. In compared to the assay of sewage polluted by WMO without biostimulation with MS where the heterotrophic microorganisms native of the sewage were unable to eliminate the hydrocarbons of the WMO, due, lowest production of CO₂. While it is shown that WMO consumed because the capacity of aerobic heterotrophic microorganisms, since that the sterilization of sewage destroyed and thus suppressed the generation of CO₂, despite biostimulation with MS; this CO₂ values were statistically different, compared to biostimulating sewage with MS, supporting that the nutritional is the limiting factor of the bioremediation of the sewage polluted by WMO. In contrast to the trial where sewage impacted by WMO, biostimulated with Tween 20, without MS. It was clear that the detergent is necessary only for the emulsification of WMO, in that sense without biostimulation by MS there was no CO₂ production, the main evidence of WMO mineralization. A critical point of the research is to understand the dynamic of bioremediation of sewage polluted by WMO. Therefore, the data shown in this research is supporting why the bioremediation of swage, as an ecological strategy for its reuse. As is reported by Gopinath indicating that bioremediation of sewage impacted by WMO that got an elimination percentage of 92.5%.

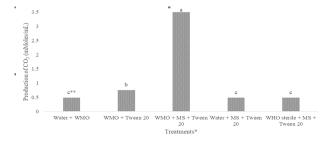


Figure 2 CO₂ production in sewage (water) contaminated by waste motor oil biostimulated with Tween 20, mineral solution, H₂O₂ at 100 rpm.

*n =4.WMO: sewage polluted by waste motor oil, diluted 1:100, biostimulated with MS: mineral solution, Tween 20 at 0.01% temperature: 30+-2° C. agitation: 100 rpm. **Distint letters indicate stadistically differences according to ANOVA/Tukey HSD ($P \le 0.05$).

In Figure 3, the biostimulation of sewage contaminated by WMO with Tween 20, MS plus intermittent of $\rm H_2O_2$ kept concentration of $\rm O_2$ available for a longer time and decreasing rapid volatilization, in consequence was induced a maximun mineralization of the WMO and simultaneously the production of $\rm CO_2$, up to a value of 5.88 mmoles/mL at 24 h; in comparison with the sewage polluted by WMO biostimulated with the MS; but without $\rm H_2O_2$, due the aerobic heterotrophic microorganisms of the WMO without sufficient $\rm O_2$ produced less $\rm CO_2$, with 3.5 mmoles/mL, supports the mineralization depends on the $\rm H_2O_2$ as a limiting factor for the effective elimination of the WMO.

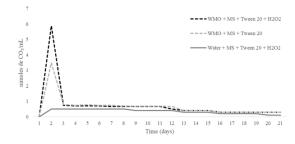


Figure 3 CO_2 production during the biostimulation of sewage (water) contaminated by waste motor oil with Tween 20, mineral solution and intermittent application of H_2O_2 while agitation at 100 rpm.

In Figure 4, it is shown that CO_2 production during the biostimulation of sewage contaminated by WMO with Tween 20 emulsifies aliphatic hydrocarbons of WMO while MS due its chemical composition with basic minerals of nitrogen, phosphates etc, induced that native microorganisms to oxide WMO under aerobic condition accelerated by the application of $\mathrm{H_2O}_2$ in consequences registered the

maximum generation of $\rm CO_2$ of 5.88 mmoles/mL, numerical value statistically different in comparison. with the sewage polluted by WMO biostimulated just by the Tween 20 and the MS generated 3.5 mmoles/mL of $\rm CO_2$ while sewage no polluted by WMO biostimulated with the Tween 20, plus the MS and the $\rm H_2O_2$ registered the lowest value with 0.5 mmoles/mL of $\rm CO_2$.

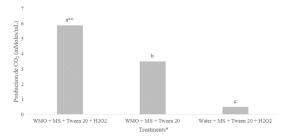


Figure 4 CO_2 production during biostimulation of sewage contaminated by waste motor oil or WMO with Tween 20, mineral solution and regular application of H_3O_3 .

*n =4.WMO: sewage contaminated by waste motor oil diluted 1:100 (10,000 ppm) with MS: mineral solution, temperature: $30+-2^{\circ}$ C. agitation: 100 rpm. **Distint letters indicate stadistically different according to ANOVA/Tukey HSD (P \leq 0.05).

Figure 5 shows the chromatographic profile of sewage impacted by WMO biostimulated with Tween 20, mineral solution, $\rm H_2O_2$ and agitation at 100 rpm, where it was detected that the main aliphatic hydrocarbons of WMO were chains between $\rm C_{11}$ and $\rm C_{20}$, was used as absolute control, in that case aliphatic hydrocarbons concentration of WMO was similar (data not shown).

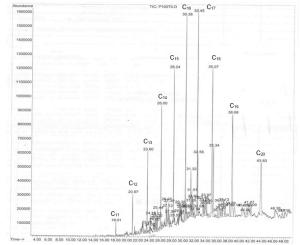


Figure 5 Chromatogram of sewage non-sterilized contaminated by waste motor oil before biostimulation with Tween 20, mineral solution and H₂O₃.

Figure 6 shows the biostimulation of sewage impacted by WMO with Tween 20, SM and $\rm H_2O_2$, with evident elimination of 100% of the aliphatic hydrocarbons of chains between 11, 12 and 20 carbons data different it has been reported in the literature that support that hydrocarbon of WMO with the least number of carbons are the first to oxide, due that short-chain alkanes mineralize faster than long-chain ones. The results shown that sewage impacted by WMO, face a wide diversity of aerobic heterotrophic microorganisms able to mineralize 96% of the aliphatic ones from WMO having of 13-19 carbons, based in the chromatographic analysis after biostimulation of sewage with Tween 20, MS, $\rm H_2O_2$, are necessary for the mineralization of the different aliphatic WMO, at the same time the Soxhlet analysis indicated that final concentration of the WMO it was close to 10

ppm, a lower value than the maximum permissible limits by NOM-001-SEMARNAT-1996, NOM-002-ECOL-1997 and NOM-003-ECOL-1997, ¹⁴ confirmed of waste recovery to be reused for garden and industrial irrigation. To improve sewage cleaning up form WMO bioaugmentation could be apply to accelerate WMO mineralization under environment condition close to what happens in the common sewage. ¹⁵

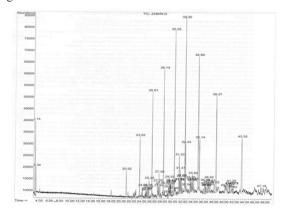


Figure 6 Chromatogram of sewage contaminated by waste motor oil after biostimulation with Tween 20, mineral solution and $\rm H_2O_2$ agitation at 100 rpm at 30°C.

Figure 7 shows the chromatogram of the biostimulation of sewage contaminated by WMO with Tween 20, MS and $\rm H_2O_2$, in agitation up to 100 rpm at 30°C on the mineralization of aliphatic with carbon chains between $\rm C_{11}$ - $\rm C_{20}$. This supporting that aerobic heterotrophic microbial native consortium have the ability in sewage to eliminate until 96% in the 21 days of the assay was evident the mineralization of 100% of the short-chain aliphatic hydrocarbons of WMO was detected mainly: undecane, dodecane, in opposite way to what was detected for the $\rm C_{13}$ - $\rm C_{19}$ WMO aliphatic in a sewage concentration of 4% at the end of the experiment supported by other reports related to bioremediation of environmental pollutes by hydrocarbons like WMO.^{4,6,16–18}

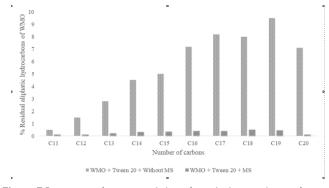


Figure 7 Percentage of remaining aliphatic from the biostimulation of sewage impacted by waste motor oil (WMO) with Tween 20, mineral solution, H_2O_2 .

Conclusion

The results of this research support biostimulation is strategy for the recovery of sewage impacted by WMO, biostimulation through actions to restore the physicochemical environment to induce microorganisms to oxidize WMO, eliminating its main aliphatic and aromatic hydrocarbons that allow industrial and/or recreational reuse.

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Conflicts of interest

The authors declare no conflict of interest.

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