MICROBIAL BIODEGRADATION OF POLYETHYLENE OF LOW DENSITY, UNDER CONTROLLED THERMAL CONDITIONS IN AIR LIFT BIO-REACTOR

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ABSTRACT

The present investigation seeks to identify new mechanisms that serve as tools for the mitigation of plastic contamination through the biodegradation of low density polyethylene using microorganisms of the species Pseudomona aeruginosa (bacteria) and Aspergillus brazilensis (fungus) under controlled thermal conditions in an airlift bioreactor. The methods used were 2 samples of low density polyethylene with concentrations of 50 mg/L and 2 samples of 100 mg/L deposited in an airlift bioreactor under controlled thermal conditions with a duration of 7 days. As a result it was obtained that the species Pseudomona aeruginosa (bacteria) reduced the low density polyethylene sample by 2% with a concentration of 49 mg/L at a temperature of 21.8ºC with a pH of 6.5 and dissolved oxygen (OD) of 6.8 mg/L, likewise the species Aspergillus brazilensis (fungus) reduced the low density polyethylene sample by 7% reaching a concentration of 93 mg/L at 22.1ºC of temperature, 7.14 of pH and 7.45 of dissolved oxygen (OD).

KEYWORDS

Air lift bioreactor, Biodegradation, Thermal conditions, Low density polyethylene (LDPE).

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1. INTRODUCTION

As we know, plastics are a big problem nowadays, since we live in a world where people do not have an adequate environmental conscience, unfortunately this generates a very negative impact to our planet. Plastics have a low economic value in the market and are the most used, therefore, the amount of plastic waste increases. In addition, he tells us that "In 2010, 8.07% of plastic waste was generated in our country and in 2011 9.85%, this indicates that every year there is an increase in plastic waste. Plastic bags (low-density polyethylene) take 150 years to degrade, which is why we have been looking for alternatives to reduce the life span of plastic, such as incineration, which causes health problems because it generates the famous greenhouse gases (GHGs), in addition to the release of dioxins and furans (POPs) that are highly carcinogenic according to the World Health Organization (WHO) (Barlow *et al.*, 2019).

Plastic waste is a worldwide problem since people use the bags only once, throw them away and buy others again without realizing it; this is how pollution begins, causing damage mainly to marine ecosystems, since fauna often confuses them with food. A study conducted for MINAM in June 2012 indicates that Peruvians generate approximately 23,260 tons of solid waste, which gives us a per capita production of 800 grams (Córdova *et al.*, 2020).

A study released in 2015 tells us that sea turtles have been ingesting 52% of garbage, with plastic waste such as bags being the most common (Inforegion, 2015).

Finally, according to the Peruvian newspaper El Comercio (2018), 79% of plastic waste globally is found in dumps or thrown on the roads; only 9% is recycled and 12% is incinerated.

2. MATERIALS AND METHODS

2.1. PLACE OF STUDY AND SAMPLING

In this stage the place to work was selected and was an informal dump located in the town center of Santa Clara in the district of Ate in Lima in April, which presented a large amount of plastic in an apparent state of degradation (Catto *et al.*, 2016).

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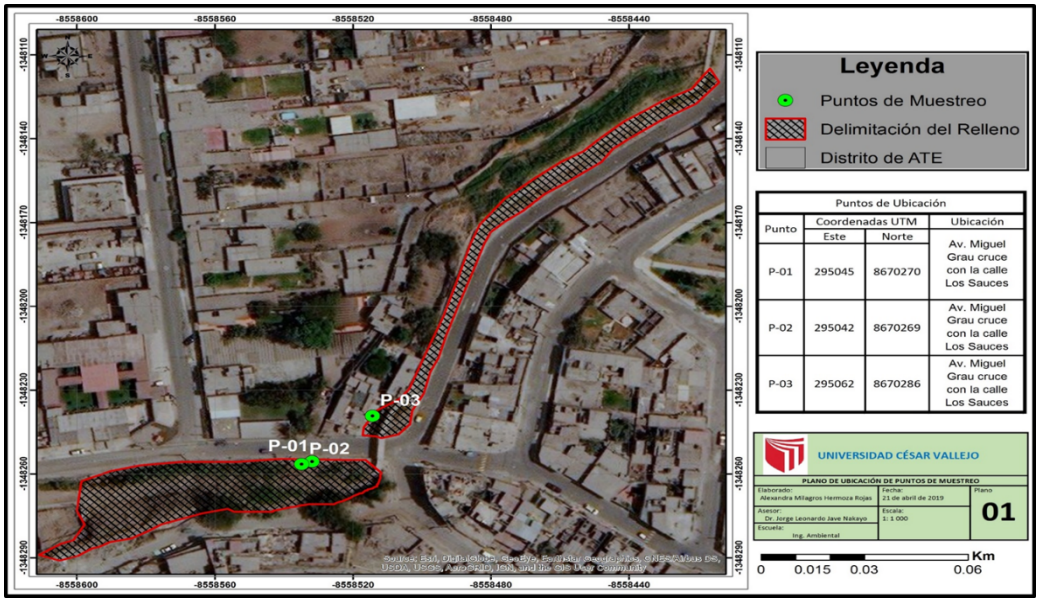

Figure 1. Map of sampling points

2.2. ELABORATION OF AIR LIFT BIOREACTORS

The bioreactors are made in the laboratory, they have a cylindrical glass culture chamber of 100 mm diameter by 200 mm height with a small pipe to take the samples, a deflector tube or central distribution of 80 by 28 mm, a venting filter attached to a regulating valve for air output, control valves for air input/output and air flow connectors attached to the tank pumps; which are the main source of oxygen for the bioreactor (Paço *et al.*, 2017). The culture chamber and deflector tube, the bioreactor accessories such as the top cover (airtight lid), air inlet/outlet control valves and air flow connectors are sterilized by immersion in 70% medicinal alcohol for 15 minutes and rinsed with purified water.

2.3. SOWING OF MICROBIAL CULTURES

The microorganisms chosen for the biodegradation process were a bacterium (speudomona aeruginosa) which was seeded on Trypticase Soy agar and incubated for 24 hours at a temperature of 30oc to 35oc and a fungus (Aspergillus brasiliensis) grown on Saburo Dextrose agar, incubated for 48 hours at a temperature of 20oc to 25oc (Giacomucci *et al.*, 2019).

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It should be considered that for the process to be more effective, the strains should be used within the first two hours after leaving the incubator.

2.4. BIOREACTOR OPERATION OF THE CULTURES

The microbial colonies kept inactive in refrigeration are reactivated in new plates with half Trypticase Soy and Saburo Dextrose forming in seed inocula for the bioreactors. The proportion is 15% of the total volume (1000ml). The operation starts with the ignition of the air pumps, the total opening of the vent valve for a period of at least 7 days (Soto, 2016).

2.5. CONCENTRATION OF LOW DENSITY POLYETHYLENE BAGS

The degradation of the low density polyethylene *(*LDPE) is determined with the decrease of concentrations placed in the bioreactor, for which we work with 50mg/L for the type A bags, 100 mg/L for the type B bags, as concentrations for the beginning of operation, these samples are measured again after three days. To find the final concentration, the whole culture medium is autoclaved for 15 minutes, then the low density polyethylene (LDPE) samples are extracted, rinsed with purified water and taken to the oven for 12 to 24 hours at 80°c. Finally, the weighing is done in the analytical balance.

Table 1. Table of concentrations of low density polyethylene (LDPE) type A.

Bioreactors	Concentrations of LDPE type A (mg/L)		
	DAY 0	DAY 3	DAY 7
pseudomonas 1	50	49.000623	49
aspergillus 2	50	45.000023	45

2.6. DETERMINATION OF TEMPERATURE, DISSOLVED OXYGEN AND PH

The determination of the internal temperature was obtained at the beginning, after three days and at the end after 7 days by means of a thermometer, for dissolved oxygen in the same time interval it will be determined by an oximeter in the unit's mg/L and finally for the determination of the pH in the same time interval it was obtained by means of a potentiometer (Janczak *et al.*, 2019).

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2.7. STATISTICAL ANALYSIS

The statistical test Anova was used to compare the low density polyethylene (LDPE) groups in a quantitative variable where $p < \alpha$, p-value $=0.04$ to $\alpha = 0.05$, T for the samples in the different groups, since the group of individuals sampled is less than 30.

2.8. RESULTS AND DISCUSSION

The results obtained in low density polyethylene (LDPE) type A using the concentration of 50 mg/L in relation to the determination of concentrations had a decrease of 1% for pseudomonas and 5% for aspergillus, taking into account the higher efficiency of the latter, according to the variation of the temperature of 1°c. influenced in the efficiency due to the fact that these temperatures are within the environmental thermal conditions. However, the concentrations of dissolved oxygen increased by 4% pseudomonas and 20% for aspergillus during the days of experimentation, as opposed to a reduction of pH by 17% for pseudomonas and 13% for aspergillus (Sadhukhan *et al.*, 2019).

Table 2. Table of concentrations of PEBD type B.

Biorreactors	Concentrations of PEBD type B (mg/L)		
	DAY 0	DAY 3	DAY 7
pseudomonas 1	100	98.001246	98
aspergillus 1	100	93.000064	93

The results obtained in low density polyethylene (LDPE) type B using the concentration of 100 mg/L in relation to the determination of concentrations had a decrease of 2% for pseudomonas and 7% for aspergillus, taking into account the higher efficiency of the latter, according to the variation of the temperature of 1°c. influenced in the efficiency due to the fact that these temperatures are within the environmental thermal conditions. However, the concentrations of dissolved oxygen increased by 8% pseudomonas and 4% for aspergillus during the days of experimentation, as opposed to a reduction of pH by 19% for pseudomonas and 11% for aspergillus.

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3. DISCUSSION

Microorganisms are capable of biodegrading low density polyethylene (LDPE) under certain controlled thermal conditions. On day zero, two samples of low density polyethylene (type A) with concentrations of 50 mg/L and two samples with 100mg/L of low density polyethylene with biodegradable additives (type B) were taken. On the last day of the process, sample 1 of type A had a final concentration of 49mg/L and sample 2 reached a concentration of 45mg/L. The first type B sample reached a concentration of 98mg/L and the second one a concentration of 93mg/L, which was the sample where the highest degradation was obtained with a temperature of 22.1°c.

For an efficient degradation by microorganisms, a favorable thermal condition is needed. Native bacteria present in worm, horse and chicken humus biodegraded polyethylene terephthalate and oxo-polyethylene efficiently at a temperature of 22°c in 35 days. For this research, an average temperature of 22.1°c was obtained on the last day of the process, this being the most efficient temperature for the biodegradation of low-density polyethylene with Aspergillus brazilensis (Perpetuo *et al.*, 2020).

The chemical conditions are important for microbial biodegradation, the pH of the samples was determined during the process. The most efficient pH for the sample with Pseudomona aeruginosa was 6.49 and for the sample with Aspergillus brazilensis was 7.14. This value was the most efficient for the whole process. For the optimum pH for the polyethylene sample with bacteria such as Pseudomonas sp was 5.5 and for the sample with fungi (unidentified yeast) was 7 (Alvarado *et al.*, 2020).

Finally, a percentage of 1% of degradation was obtained with Pseudomona aeruginosa for sample 1 of polyethylene type A and for the degradation with Aspergillus brazilensis for sample 2 of the same type of polyethylene a percentage of 5% was obtained. For sample 1 of polyethylene type B with Pseudomona aeruginosa a percentage of 2% was obtained and for sample 2 of the same type of polyethylene with Aspergillus brazilensis a result of 7% of degradation was obtained. For NOVOTNÝ, C (2015) in a period of six weeks and with a temperature of 28°c the samples of PETP/LA with Pseudomona aeruginosa had mass reductions of up to 5-10%.

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4. CONCLUSIONS

Microbial biodegradation of low-density polyethylene was evaluated in Santa Clara in an air lift type bioreactor and the results were 1% and 2% for low density polyethylene (LDPE) type A and B samples with Pseudomona aeruginosa bacteria under controlled thermal conditions.

For the low density polyethylene (LDPE) type A and B sample with Aspergillus brazilensis fungus are 5% and 7% respectively at controlled thermal conditions. The thermal condition favorable for microorganisms to degrade the low density polyethylene (LDPE) efficiently is 22.1°C since a degradation of 7% was obtained for the sample of low density polyethylene (LDPE) type B with Aspergillus brazilensis.

The chemical conditions necessary for the microorganisms to degrade the low density polyethylene (LDPE) efficiently were pH and dissolved oxygen (OD), being the most efficient pH value for biodegradation 7.14 in 7 days of duration of the process and the most efficient dissolved oxygen (OD) value is 7.45mg/L for the sample 2 type B with Aspergillus brazilensis.

REFERENCES

Alvarado, K., Esenarro, D., Rodriguez, C., & Vasquez, W. (2020). Lemna minor influence in the treatment of organic pollution of the industrial effluents. *3C Tecnología. Glosas de innovación aplicadas a la pyme, 9*(3), 77-97. https://www.3ciencias.com/wp-content/uploads/2020/09/art-4-3c-tecno-ed.-35-vol.-9-n.-3-1.pdf

Catto, A. L., Montagna, L. S., Almeida, S. H., Silveira, R. M. B., & Santana, R. M. C. (2016). Wood plastic composites weathering: Effects of compatibilization on biodegradation in soil and fungal decay. *International Biodeterioration & Biodegradation, 109,* 11-22. https://doi.org/10.1016/j.ibiod.2015.12.026

Córdova, A., Amaya, P., Esenarro, D., & Rodriguez, C. (2020). Vegetable Contamination by Heavy Metal Contained in Effluents from Wastewater Plant in the Totora Community, Ayacucho –Peru. *Journal of Green Engineering, 10*(7), 3484–3497. http://www.jgenng.com/volume10-issue7.php

https://doi.org/10.17993/3ctecno.2020.specialissue6.179-189

Giacomucci, L., Raddadi, N., Soccio, M., Lotti, N., & Fava, F. (2019). Polyvinyl chloride biodegradation by Pseudomonas citronellolis and Bacillus flexus. *New Biotechnology, 52*, 35-41. https://doi.org/10.1016/j.nbt.2019.04.005

Hung, C.-S., Barlow, D. E., Varaljay, V. A., Drake, C. A., Crouch, A. L., Russell, J. N., Nadeau, L. J., Crookes-Goodson, W. J., & Biffinger, J. C. (2019). The biodegradation of polyester and polyester polyurethane coatings using Papiliotrema laurentii. *International Biodeterioration & Biodegradation, 139*, 34-43. https://doi.org/10.1016/j.ibiod.2019.02.002

Inforegión. (2015, January 25). *El 46% de los residuos sólidos en playas del Perú son plásticos*. http://www.inforegion.pe/196519/el-46-de-los-residuos-solidos-en-playas-del-peru-son-plasticos/

Janczak, K., Hrynkiewicz, K., Znajewska, Z., & Dąbrowska, G. (2019). Use of rhizosphere microorganisms in the biodegradation of PLA and PET polymers in compost soil. *International Biodeterioration & Biodegradation, 130*, 65-75. https://doi.org/10.1016/j.ibiod.2018.03.017

Novotný, C., Erbanová, P., Sezimová, H., Malachová, K., Rybková, Z., Malinová, L., Prokopová, I., & Brožek, J. (2015). Biodegradation of aromatic-aliphatic copolyesters and polyesteramides by esterase activity-producing microorganisms. *International Biodeterioration & Biodegradation, 97, 25-30.* https://doi.org/10.1016/j.ibiod.2014.10.010

Paço, A., Duarte, K., da Costa, J. P., Santos, P. S. M., Pereira, R., Pereira, M. E., Freitas, A. C., Duarte, A. C., & Rocha-Santos, T. A. P. (2017). Biodegradation of polyethylene microplastics by the marine fungus Zalerion maritimum. *Science of The Total Environment, 586,* 10-15. https://doi.org/10.1016/j.scitotenv.2017.02.017

Perpetuo, E. A., da Silva, E. C. N., Karolski, B., & Oller, C. A. (2020). Biodegradation of diethyl-phthalate (DEP) by halotolerant bacteria isolated from an estuarine PERPETUO, environment. *Biodegradation*. https://doi.org/10.1007/s10532-020-09913-y

https://doi.org/10.17993/3ctecno.2020.specialissue6.179-189

Sadhukhan, J., Martinez-Hernandez, E., Amezcua-Allieri, M. A., Aburto, & Honorato, J. A. (2019). Economic and environmental impact evaluation of various biomass feedstock for bioethanol production and correlations to lignocellulosic composition. *Bioresource Technology Reports, 7,* 100230. https://doi.org/10.1016/j.biteb.2019.100230

Soto, M. (2016, June 3). *Contaminación por plástico es nuevo reto ambiental del país.* Diario La Nación. http://www.nacion.com/vivir/ambiente/Contaminacion-plastico-nuevo-ambiental-pais_0_1564643544.html

https://doi.org/10.17993/3ctecno.2020.specialissue6.179-189

https://doi.org/10.17993/3ctecno.2020.specialissue6.179-189