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Efficiency Of the Microalgal Consortium in the Depuration of Wastewater from Pig Farms

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ABSTRACT

This study evaluated in vitro the ability of microalgae consortia to remove a higher percentage of pollutants present in wastewater from pig farming. The wastewater samples were collected from a local pig farm. The biomasses of the microalgal consortia were bioaugmented in 1.5 L phycoreactor, in a nutrient medium, 2 % v/v culture medium and sterilized distilled water. Growth was measured using a Spectroquant pharo 300 spectrophotometer at a wavelength of 647 nm. The assays were set up in triplicate and the reduction efficiency of these microorganisms to remove the highest amount of contaminants present in the piggery wastewater was evaluated before and after the experiment. The results obtained show that the microalgae consortia showed higher efficiency in pollutant removal. At the end of the trial it was observed that those treatments with microalgae consortia had a higher remediation effect as they presented a higher percentage of removal of heavy metals cadmium, arsenic and lead of 100%, followed by a reduction in the amount of total and faucal coliforms, total solids and nitrates. Wastewater treatment with microalgae is a method that is being widely studied as this technology is environmentally friendly, does not require energy consumption and is low cost to maintain.

Key words. Microalgae, consortium, wastewater, removal.

1. INTRODUCTION

Intensive livestock practices generate a large amount of wastewater that pollutes the environment. Pig farming is growing, with annual production volumes expected to reach 21.7 million tons by 2030 (Statista, Pork consumption volume EU-28 2015-2030). According to Eurostat in the European Union, 1.7% of pig farms have more than 400 pigs and raise 48.6% of sows and 77.9% of other pigs. In Colombia, national pork production reached 468,880 tons by 2020, registering a positive growth of 5%, compared to the previous year's supply. Intensive farming and rearing produces large quantities of waste that cannot be disposed of according to traditional practices

As indicated by Boursier et al., (2005); Choi et al., (2005), wastewater from pig herds is rich in organic matter and nutrients and its spreading on land can lead to the leaching of these nutrients into water, the emission of greenhouse gases and odors, as well as the spread of pathogens and antibiotic residues which, in turn, can promote the development of antibiotic resistance genes. One of the most important waste products in the world in terms of production, geographical distribution and polluting potential is pig waste, aggravated in some places by the concentration of animals in industrial farms with large numbers of animals and the consequent accumulation of waste.

As stated by Malaviya et al., (2020); Sharma and Malaviya, (2021), to minimize these environmental risks, various physico-chemical (i.e. screening, flotation, neutralization, oxidation, flocculation, etc.) and biological methods (such as oxidation ponds, aeration ponds, lagoons, activated sludge, biological filters, constructed wetlands, rain gardens, phytoremediation, mycoremediation, phyco-remediation, phycoremediation) have been used.

As pointed out by (Singh et al., (2016), physical and chemical methods have drawbacks such as: cost, sludge formation, high energy consumption, skilled manpower, production of toxic end products and their disposal, etc. Microalgae-based phyco-remediation is currently important because it offers attractive advantages such as low energy requirements, higher cost-effectiveness, reduction of pollutants and pathogens, nutrient recycling, less sludge formation, reduction of greenhouse gases, recovery of nutrients in the form of valuable biomass (Mishra et al., 2019).

In the same vein, microalgae not only treat wastewater, but their harvested biomass serves as a substrate for the production of bioenergy or other bio-based products (Oyebamiji et al, 2019; Singh et al., 2021). Between 45-75% of total energy costs are spent in the aeration process of wastewater treatment plants, and also approximately 1 kg of CO2 is produced in the energy generation process (Udaiyappan et al., 2017). In contrast, microalgae do not require any energy input, produce 1 kWh of energy through methane production, use CO2 as a carbon source, and release oxygen in return, thereby reducing aeration costs (Fernandez et al., 2021). Moreover, the sludge formed during microalgae treatment, the microalgae biomass itself, can be processed into valuable products such as fertilizers, biodiesel and nutrients (Ghoneim et al., 2014).

One of the activities that affect the environment is agriculture and livestock farming, from which significant quantities of pollutants are released, mainly from the pig farming sector. For this reason, the objective was to evaluate the use of microalgal consortium as a tool to mitigate the environmental pollution generated by wastewater from pig farming and the applicability of this biotechnology in situ in pig farms in the department of Sucre.

2. MATERIALS AND METHODS

Wastewater from pig farms. The sample was obtained from pig farms located at 9°150'01.1‘’ N, 75°41'0.06‘’ W and was not treated in any way. One-litre amber bottles were used to collect the samples, which were refrigerated until they were transported to the laboratory for physical, chemical and microbiological analysis.

Photosynthetic microorganisms. The microalgal consortia were obtained from the germplasm bank collection of the company Agro biotech Solution S.A.S. The cells were maintained in Nutrifoliar culture medium (Colinagro 4.0), with 200 g/l total nitrogen, 100 g/l phosphorus and nutrients such as K, Mg, S, Cl, Fe, Cu, Zn, Mn and Mo (Vitola Romero et al., 2021); this culture medium was diluted at 2 % v/v in distilled water and sterilized in an autoclave.

Growth curves. Microalgal growth was carried out in 1 litre phyco-reactors with a working volume of 500 mL of 2% v/v culture medium and sterilized distilled water, concentration 1 x10^6 cells of initial inoculum. Microalgal biomass growth was measured every 24 hours for 21 days using a Spectroquant pharo 300 spectrophotometer at a wavelength of 647 nm and compared to the control microalgae assay with the culture medium.

Phyco-remediation test. In this experiment, piggery wastewater was used to evaluate the pollutant removal capacity of the wastewater. The tests were set up in triplicate, for which treatments were used. The tests were carried out in vitro in glass phyco-reactors. The treatments to be used: T0: BBM medium (Bold's Basal Medium) and pig wastewater; T1: pig wastewater plus 50 mL of the consortium identified in the water of the samples concentrated at 120 cells/mL, the tests will be carried out in triplicates.

Parameters to be evaluated. The parameters evaluated during the experiment were: temperature, salinity, dissolved oxygen, pH, turbidity, total solids, BOD, NO_3, phosphorus, faecal and total coliforms, COD and heavy metals (arsenic, cadmium and lead).

Statistical analysis: The results were expressed as the mean ± Standard Deviation, an analysis of variance was carried out using a completely randomized design, with a 2x3 factorial arrangement; previously determining the normality criterion by means of the Shapiro Wilk test (5%). Significant statistical differences were determined by Tukey's test ($p < 0.05$). All experiments were performed in triplicate and analyzed in the free version of InfoStat software.

3. RESULTS AND DISCUSSION

Figure 1 shows the growth of each strain of microalgae that make up the consortium used in the phycoremediation trial to remediate wastewater from pig farming. The microalgal consortia used in this study were Chlorella vulgaris, Scedesmus sp., Desmodesmus sp. and Crococcus sp. species (figure 1). In figure 1, it is observed that all the species used in the consortium showed growth up to 17 days and after this time they entered the stationary phase. It can also be seen that Chlorella vulgaris had the highest growth rate compared to the other microalgae species.

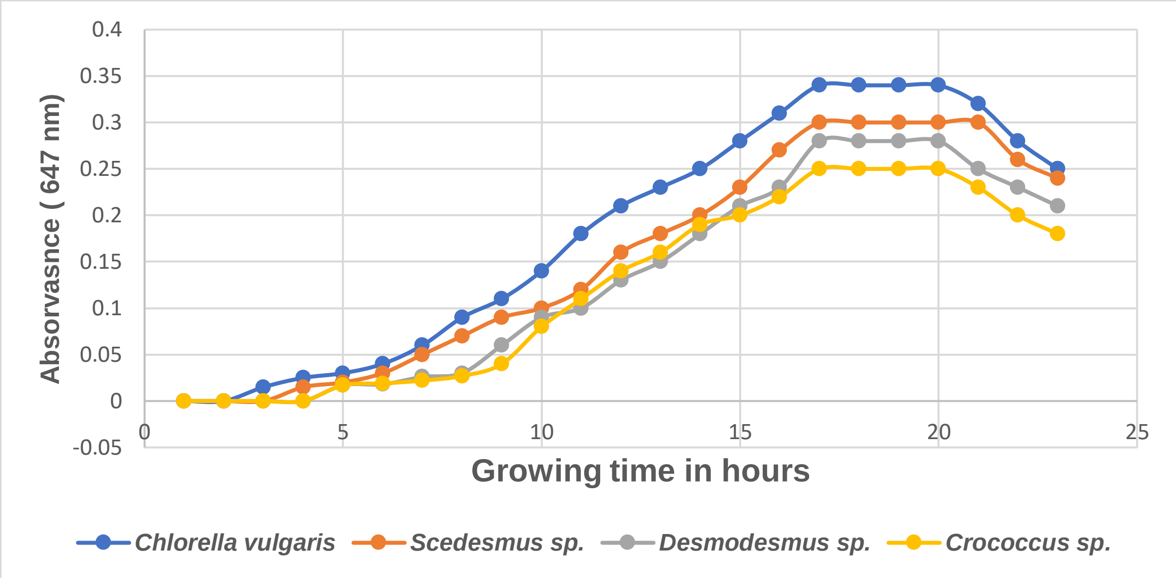

Figure 1. Growth curve of microalgae strains used in piggery wastewater remediation trials.

The efficiency of the removal percentage was given 15 days after the start of the experiment, showing the highest removal for the heavy metals cadmium, arsenic and lead of 100%, followed by a reduction in the amount of total coliforms (99.34), faecal coliforms (98.4), total solids (98.3) and nitrates (93.56%). The removal of total phosphorus corresponded to 90.19%, while for total phosphorus it was 80.48%. It is also observed that the biological oxygen demand (BOD5) was reduced by 78.15% and the biochemical oxygen demand (COD) by 75.77% with respect to the initial values found in the pig wastewater reported before the experiment (table 1). The same table shows that the pH went from acidic to alkaline, the dissolved oxygen increased and the temperature decreased to 37°C.

Wastewater from pig farms contains micro- and macronutrients, organic and inorganic carbon, heavy metals and harmful compounds (Daneshvar et al., 2018). Microalgae consume nutrients from piggery wastewater for growth and produce dissolved oxygen (DO) during photosynthesis. CO2 released during degradation is used by microalgae in autotrophic metabolism (Garcia et al., 2019). The treated wastewater can be released into watercourses and the harvested biomass can be used to recover more manageable and valuable products (Fernández et al., 2021).

Table 1. Pig wastewater characteristics and removal rates during treatment with microalgal consortium.

Indicators	**Initial values (Vi)**	**Final values microalgae consortium test (Vf)**	**% of phycoremotion ([Vi-Vf/Vi]* 100)** (Rajesh et al., 2021)
Temperature	42	37	Decrease
pH	4,3	8,1	Alcalinice
Dissolved Oxygen (In situ)	0	5,78	Increases
Turbidity	46	5	89,13
Total Solids	649	11	98,31
Salinity	0,927	0,1	89,21
BOD_5	55	12	78,18
COD	115,6	28	75,78
Nitrates	1,553	0,1	93,56
Orthophosphate	3,075	0,600	80,49
Total Phosphorus	2,04	0,2	90, 20
Faecal Coliforms	12400	200	98,39
Total Coliforms	23000	150	99,35
Arsenic	0,04	0	100

Cadmium	0,02	0	100
Lead	0,117	0	100

According to the studies carried out by Wang et al., (2015; Luo et al., (2016); Raeisossadati et al., (2019), they conclude that the use of microalgae to remove toxic compounds present in swine wastewater found that Chlorella sp., Desmodesmus sp., Scenedesmus sp. are the most used species for the treatment of swine wastewater, due to their high capacity of tolerance to extreme environmental stress and their potential for biomass and lipid accumulation.

The microalgae species Desmodesmus sp. CHX1 can effectively remove 87.3% TN, <95.1% $NH4+$ and 93.1% TP from swine effluent (Cheng et al., 2013). C. vulgaris JSC-6 can remove up to 60-76% COD and 40-91% $NH3$ with the production of 3.96 g/L biomass (Wang et al., 2015). Coelastrella sp. QY01 can remove 94% and 100% of TP and $NH3$, respectively (Luo et al., 2016). Luo et al. (2018) evaluated the performance of Desmodesmus sp. CHX1 and found 78.46% and 91.66% nitrogen and phosphorus reduction. Furthermore, when they experimented with optimal conditions including light intensity of 150 μmol/m2/s, 24 h photoperiod, cell inoculation concentration of 30 and 35 ◦C temperature, the efficiencies were 88.26% and 95.06%, for nitrogen and phosphorus, respectively. When two or more microalgae are grown together, they show even higher removal efficiencies. For example, Chlorella sp. and Scenedesmus sp. recorded a high removal rate of 1.97 gm2/d and 5.83 gm2/d for ammonia and COD, respectively (Raeisossadati et al., 2019), compared to 60.7% COD, 81.5% TN and 64.6% TP removal by Scenedesmus sp. Individually (Chen et al., 2018) Chlamydomonas sp. QWY37 is able to remove 81% COD, 96% TN and 100% TP (Cheng et al., 2020

4. CONCLUSION

The results of the swine farm wastewater remediation trial allow inferring that the effectiveness of organic load reduction in the wastewater was reached in the in vitro experiment 15 days after the beginning of the experiment, showing the highest elimination for heavy metals cadmium, arsenic and lead of 100%, followed by a reduction in the amount of total coliforms (99.34), fecal coliforms (98.4), total solids (98.3) and nitrates (93.56%). Total phosphorus elimination corresponded to 90.19%, while for total phosphorus it was 80.48%. It was also observed that the biological oxygen demand (BOD5) was reduced by 78.15% and the biochemical oxygen demand (COD) by 75.77% with respect to the initial values found in the swine wastewater reported. In the present trial it was demonstrated that microalgae have the capacity to eliminate a large part of the pollutants in the water from a pig farm, which makes them a very good technology for purifying this type of water. The biotechnological package composed of the microalgae consortium implemented in this trial is a good option for the remediation of wastewater from pig farming activities in the department of Sucre-Colombia.

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6. AUTHOR CONTRIBUTION. Alexander Perez Cordero: experiment execution, data analysis. Donicer Montes V and Yelitza Aguas M, conceptualization, writing - revision and editing. All authors have read and approved the manuscript.

7. CONFLICT OF INTEREST. All the authors of the manuscript declare that they have no conflictof interest

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