Entry to Stockholm Junior Water Prize 2021

“Biophyte, a life-giving weed”

Lisania Karina Monzón Fernández
Sofía Tress González

MEXICO
BIOPHYTE: A LIFE-GIVING WEED

SUMMARY

The San Pedro River, located in the border of Mexico and Guatemala, is of great cultural importance to the Mayan culture, connects a wide wetland network, and is a revenue source to local fishers. However, pollution and the growth of aquatic weeds, if uncontrolled, might harm fish and fisheries. Moreover, the rural border communities of “Cuatro Poblados” and “San Pedro”, Balancán, Tabasco, Mexico, are also confronted with a scarcity of commercial fertilizers, making it harder for their integration into the governmental program “Sembrando Vida” (Sowing Life). This research aims to determine the potential of the use of *Eichhornia Crassipes*, *Pistia Stratiotes*, and *Salvinia Molesta* in the production of a biofertilizer with the ability to promote germination and growth of seedlings, maintain soil fertility and increase crop productivity. To corroborate this, the vegetable biomass of aquatic weeds was submitted to a process of aerobic composting under controlled conditions (by monitoring pH, temperature, and humidity) for 77 days. Afterward, a physicochemical characterization (soil texture, organic matter, K, Ca, Mg, Na, Fe, Zn, Mn, Cu, B, S, N-NO3) and an evaluation of toxic elements (As, Cd, Pb) in the substratum was carried out. A test of efficiency was also completed (germination and growth trials) with seeds of *Capsicum Chinense* and lentil (*Lens Esculenta*) in the presence of clay soils. We confirmed that Biofita is a biofertilizer that, aside from possessing suitable levels of fertility and null toxicity, can act as a germination and growth promoter, assuring its continuous use by farmers as an alternative substratum.
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ACKNOWLEDGEMENTS

We are grateful for the support provided by USAER N° 48 and USAER N° 152 of the municipality of Balancán, Tabasco. We also appreciate the time and unconditional support of our advisor, Alan Cupil Diaz, a teacher at our institution, “General Luis Felipe Domínguez” State High School (EMSAD No. 4), Tabasco, who has guided us and acted as our advisor during these past 2 years.
INTRODUCTION

In 2019 the Mexican Government launched the program “Sembrando Vida” (Sowing Life) that would contribute to the social wellbeing of the rural communities from 19 states of the Mexico, namely Campeche, Chiapas, Chihuahua, Colima, Durango, Guerrero, Hidalgo, Michoacan, Morelos, Oaxaca, Puebla, Quintana Roo, San Luis Potosi, Sinaloa, Tabasco, Tamaulipas, Tlaxcala, Veracruz, and Yucatan. These states are known for having high poverty rates and social lag, and for concentrating most of the forest resources. Therefore the establishment of productive systems in these areas would not only recover the country’s forest cover, but also improve the economic income and food self-sufficiency of the community producers (Bienestar, 2019).

The objective is to be met through the plantation of native tree and fruit species; produced in forest nurseries owned by the National Defense Secretary (SEDENA) or by users (communal or commercial nurseries), which would contribute to the fulfillment of the objectives and benefits of the program (Job, 2019). Nonetheless, in some regions, the lack of commercial fertilizers puts the success of the program at risk. Such is the case of the rural communities of Cuatro Poblados and San Pedro, located in the border zone between Mexico and Guatemala, in the municipality of Balancan, Tabasco. Here the producers face the scarcity of commercial fertilizers, due to the long distance of the communities to the main distribution centers as well as their cost. In addition, the dominance of unproductive soils (low fertility) translates into higher demand for fertilization.

On the other hand, these communities are close to the San Pedro River, a tributary flow of Usumacinta River. The San Pedro River is of great cultural importance to the Mayan culture, connects a wide wetland network, and is a revenue source to local fishers. However, the growth of aquatic weeds, if uncontrolled, might harm fish and fisheries.

The present document proposes the preparation of a non-conventional biofertilizer made from plant biomass: aquatic lily (Eichhornia crassipes), water lettuce (P. stratiotes), and mouse-tail (Salvinia molesta). The biomass is to be obtained from the anaerobic composting of said aquatic weeds, based on ancient knowledge of the Aztec civilization, who employed "swamp peat", a kind of unconsolidated organic sediment as natural fertilizers in their traditional chinampas. Finally, the use of these weeds as raw material.
offers not only an economic alternative for the production of biofertilizers to be used in the program “Sowing Life”, but also a control strategy to this aquatic vegetation whose presence in the San Pedro River (on the Usumacinta River basin) represents a risk for the quality of water and the availability of the country’s water resource.

Image 1. “Sowing Life” program

HYPOTHESIS
“The species *Pistia stratiotes*, *Eichhornia crassipes* and *Salvinia molesta* after being subjected to an aerobic composting process meet the established quality requirements to be used as raw material in the elaboration of biofertilizers for the program Sowing Life”.

MATERIALS AND METHODS

STUDY AREA
The Campamento San Pedro Lagoon is a body of water located on the banks of the San Pedro River in the community of San Pedro, Balancan, Tabasco, which also belongs to the Usumacinta River basin, the most important freshwater reserve in the country. See Image 2.

WEED COLLECTION
For the elaboration of the biofertilizer, 30 kg of the following plant material were collected from the Campamento San Pedro Lagoon: *Pistia stratiotes*, *Eichhornia crassipes* and *Salvinia molesta*. The materials were finely chopped to obtain small particles (> 2.0 cm), that were finally scattered and dried for three days (Ramos, 2014). See Image 3.
PLANT MATERIAL COMPOSTING

A modified version of the methodology described by Restrepo (2007) was followed to produce the biofertilizer. The method consisted of the aerobic composting of the plant biomass of *Pistia stratiotes*, *Eichhornia crassipes* and *Salvinia molesta* for 77 days. During this time, temperature, pH, and humidity were carefully monitored. The temperature and the pH were verified with a soil pH-meter, while the humidity was measured according to the Ferruzi (1986) method, which consists of hand-compressing a
fistful of the material and proving that being completely wet, it doesn’t release water (humidity 70-80%). In addition, to optimize the fermentation process, the team decided to add molasses and bread yeast, which were diluted and applied when moistening the mound.

DETERMINATION OF THE FERTILITY AND PRESENCE OF HEAVY METALS OF THE BIOFERTILIZER

Once the biofertilizer production process was finished, two compound samples of the substrate were taken. Samples were tested for fertility (OM, P Bray, K, Ca, Mg, Na, Fe, Zn, Mn, Cu, B, S, and N-NO₃) and heavy metals (As, Cd, and Ps). The analyses were carried out by personnel from FERTILAB Laboratories in the city of Celaya, Guanajuato, Mexico. Once the results of the physicochemical analysis were obtained, they were compared to the Mexican Official Standard NOM-021-SEMARNAT-2000 which establishes the specifications of fertility, salinity, and soil classification. Also, the results from the As, Cd, and Pb analysis were interpreted according to the EPA 503 and EPA 2006 methods. These activities allowed us to evaluate the fertility and safety of the aquatic weeds as raw materials for the elaboration of the biofertilizer.

DETERMINATION OF THE EFFICIENCY OF THE BIOFERTILIZER

The efficiency of the biofertilizer was estimated from a germination test and a vegetable growth test. For the first test, the germination of habanero pepper seeds (*Capsicum chinense*) was carried out under a completely randomized experimental design in 3 treatments (in triplicate) (Table 1). For the growth test, lentil seeds (*Lens esculenta*) that had been previously disinfected with 1% (v/v) NaClO commercial sodium hypochlorite (Resendiz, 2011) solution were used for posterior pre-germination. Then, the germinated seeds (radicle of 5 mm) were planted in the treatments shown in Table 2. These treatments were replicated by triplicate under a completely random treatment (Lara et al., 2011) during a period of growth of 20 days.
Table 1. Habanero pepper germination test treatment

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Constitution</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>Clay</td>
</tr>
<tr>
<td>T2</td>
<td>Biofita</td>
</tr>
<tr>
<td>T3</td>
<td>50% Biofita + 50% Clay</td>
</tr>
</tbody>
</table>

Table 2. Plant growth test treatment

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Constitution</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>Clay</td>
</tr>
<tr>
<td>T2</td>
<td>Dark soil</td>
</tr>
<tr>
<td>T3</td>
<td>25% Biophyte + 25 Clay</td>
</tr>
<tr>
<td>T4</td>
<td>50% Biophyte + 50 Clay</td>
</tr>
<tr>
<td>T5</td>
<td>75% Biophyte + 25% Clay</td>
</tr>
<tr>
<td>T6</td>
<td>100% Biophyte</td>
</tr>
</tbody>
</table>

STATISTIC ANALYSIS

The obtained results of the germination and growth tests were analyzed with the statistical program JMP. A variance analysis (ANOVA) and the Post-Hoc Tukey HSD test (p=0.05) were carried out to identify the significant differences in each of the treatments performed in triplicate. Charts were elaborated using Microsoft Excel.

RESULTS

FERTILITY. The results of the physicochemical characterization, based on the Mexican Official Standard NOM-021-SEMARNAT-2000, show that the substrate obtained from the process of composting has adequate levels of fertility for agricultural purposes, as required by the “Sowing Life” program. (See Image 3).
Table 3. Determination of fertility based on NOM-021-SEMARNAT-2000

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Obtained Value</th>
<th>Class obtained according to NOM-021-SEMARNAT-2000</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>8</td>
<td>Moderately alkaline</td>
</tr>
<tr>
<td>MO</td>
<td>5.17%</td>
<td>High</td>
</tr>
<tr>
<td>P-Bray</td>
<td>121 ppm</td>
<td>High</td>
</tr>
<tr>
<td>Ca</td>
<td>4584 ppm</td>
<td>High</td>
</tr>
<tr>
<td>Mg</td>
<td>328 ppm</td>
<td>Medium</td>
</tr>
<tr>
<td>K</td>
<td>456 ppm</td>
<td>High</td>
</tr>
<tr>
<td>Fe</td>
<td>36.4 ppm</td>
<td>Adequate</td>
</tr>
<tr>
<td>Cu</td>
<td>1.81 ppm</td>
<td>Adequate</td>
</tr>
<tr>
<td>Zn</td>
<td>7.32 ppm</td>
<td>Adequate</td>
</tr>
<tr>
<td>Mn</td>
<td>8.27 ppm</td>
<td>Adequate</td>
</tr>
<tr>
<td>N-NO3</td>
<td>59.6</td>
<td>High</td>
</tr>
<tr>
<td>CIC</td>
<td>27.1 meq/100g</td>
<td>High</td>
</tr>
</tbody>
</table>

**HEAVY METALS.** According to methods EPA 503, EPA 2006 and the Mexican Official Standard NOM-021-SEMARNAT-2000, concentrations of As, Cd, and Pb as reported from the chemical analysis of the biofertilizer were found to be within the maximum permissible levels of the reference norms (Table 4).

Table 4. Maximum permissible limits of heavy metals

<table>
<thead>
<tr>
<th>Heavy Metals</th>
<th>Substrate mg kg -1</th>
<th>EPA 503 Norm</th>
<th>EPA 2006</th>
<th>NOM-021-SEMARNAT-2000</th>
</tr>
</thead>
<tbody>
<tr>
<td>As</td>
<td>2.12</td>
<td>54</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Cd</td>
<td>0.01</td>
<td>18</td>
<td>400</td>
<td>3-5</td>
</tr>
<tr>
<td>Pb</td>
<td>NA</td>
<td>300</td>
<td>300</td>
<td>10-300</td>
</tr>
</tbody>
</table>

**GERMINATION.** The results obtained in this test allow us to observe that an adequate 50% substrate mixture in poorly productive soils (clay-like) can lead to a higher percentage of seed germination efficiency, in addition, its direct use acts like a peat that holds in moisture and is rich in organic matter.
GROWTH TEST. The statistical analysis for elongation of stem and elongation of radicle showed significant differences (p ≤ 0.05). It was noted that substrate acts as a growth promoter, as the substrate treatments at 75% and 50% favored aerial and root elongation of the plants (Image 4).

CONCLUSIONS
The biofertilizer has adequate levels of fertility and heavy metals (Cd, Pb and As) that ensure its quality and safe use as a fertilizer in the "Sembrando Vida" program. In addition, the substrate acts as a promoter of the germination and growth of vegetable seedlings in clay soils, which optimizes their use within community nurseries and promotes the survival of plant species during the transplant stage in the field (plots).
Finally, the collection and use of aquatic weeds as raw material in the production of the biophyte fertilizer serves as a control strategy to this aquatic vegetation whose presence in the San Pedro River pose a risk for the quality of water and its biodiversity.

**BIBLIOGRAPHICAL REFERENCES**

- Norma Oficial Mexicana NOM 021-SEMARNAT-2000. [http://cort.as/-PvGU](http://cort.as/-PvGU)
CITED LITERATURE


ADDITIONAL PICTURES