Chemistry Research Journal, **2024, 9(6):6-14**
Available online www.chemrj.org

Research Article

ISSN: 2455-8990
CODEN(USA): CRJHA5

Valorization of *Moringa Oleifera* Leaf Powder and its Activated Carbon for the Detoxification of Fish Ponds: Case of the Pond of the Research Station on the Diversification of Fish Farming of the Wetlands Research Laboratory (Benin)

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Abstract
Water quality degradation in fish ponds, often due to the accumulation of organic matter and algal blooms, is a major threat to fish health and aquaculture production. Traditional detoxification methods, often chemical, have drawbacks in terms of environmental impact and residues in aquaculture products. Our study focused on a more ecological and sustainable approach: the use of Moringa oleifera powder, rich in antioxidants, vitamins and minerals, Moringa oleifera being recognized for its purifying and regenerating properties.
The main aim of this research is to evaluate the effectiveness of Moringa powder and its activated carbon in detoxifying fish ponds, using a cytotoxicity test based on inhibiting root growth in cultured onions.
The cytotoxicity test carried out on the untreated fish pond samples showed that the Effective Concentration at which chemical effects are observed in 50% of the individuals tested (EC50) is 12%. This result shows that the fish pond studied is chemically polluted. These fish pond samples were depolluted with crude bioadsorbent and activated carbon, synthesized from Moringa leaf powder. Our research revealed that fish water samples treated with crude bioadsorbent had an Effective Concentration (EC50) of 24%, while those treated with activated carbon based on Moringa leaf powder had an EC50 of 32%. These results reveal the effectiveness of bioadsorbents prepared with Moringa leaf powder and activated carbon on fish farm water samples from the Research Laboratory's fish farm diversification research station on the Wetlands of the UAC. They represent a promising alternative to traditional methods of fish pond detoxification.

Keywords: Valorization, moringa, detoxification, fish farming

1. Introduction

Demography is undoubtedly one of the major issues of our century. While one billion human beings are already suffering from malnutrition, the world's population continues to grow, reaching around 8.6 billion by 2030 and 9.8 billion by 2050 [1]. At the same time, fish populations are declining due to overexploitation and water pollution, as well as growing public demand for healthy, tasty and affordable food, which has encouraged fish farming under controlled conditions. Despite the considerable contribution of fish farming as an important source of fish supplies for human consumption, numerous environmental constraints limit the expansion of this business sector.

To survive, man has learned to master water, but his human activities have rendered it unfit for use and polluted. Indeed, water is a fragile element, easily denatured by pollution. Polluted, it becomes a threat to life and an obstacle to the health and progress of populations. For this reason, water treatment is necessary to guarantee water quality and avoid the degradation of ecological balances [2].

Fish production in fish farming systems can accumulate pollutants in the water, including particulate organic matter (uneaten food, faeces, microorganisms, bacteria, algae) and dissolved organic matter. These suspended particles irritate the gills and external tissue of fish; degrade in the water, contributing to toxic nitrogen waste; and can harbor pathogenic bacteria and toxic algae [3].

To limit the disastrous effects of these pollutants on the environment, the use of methods to maintain the quality of fish breeding water while protecting the aquatic environment and water resources is therefore necessary. Thus, multiple water treatment methods, particularly physicochemical, are implemented, in particular the technique of adsorption using porous materials that is easily applicable in the field and more profitable in terms of performance / cost [4], the Growing demand for adsorbent materials for water treatment and environmental protection processes is leading to additional research into their manufacture from non-conventional materials, particularly plants. The valorization of these plants makes it possible on the one hand to eliminate them and on the other hand to optimize the yield and production costs of farms. They are widely available, inexpensive and inherently a potential source of environmentally friendly renewable materials [5]

Among these plants, Moringa oleifera is used for its various medicinal and nutritional properties. It is a tropical tree native to Asia, and is now widespread on the African continent [6]. The many valuable properties of this plant make it a highly interesting subject for study. In addition, the seeds and leaves can be used for wastewater clarification and purification [7]. Several studies have been carried out on fish ponds and the use of Moringa in the wastewater treatment process. As none of these studies have established a link between the detoxifying power of Moringa leaf powder and the detoxification of fish ponds, we decided to investigate this topic.

The overall aim of this study is to evaluate the potential of Moringa oleifera leaf powder as a natural detoxifying agent, and that of its activated carbon, to improve the water quality of fish ponds, while offering a more ecological and economical alternative to conventional methods.

Specifically, we will study the adsorbent properties of Moringa oleifera leaf powder and its activated carbon, treat ponds with bioabsorbent and activated carbon made from Moringa oleifera leaf powder, and assess the effectiveness of activated carbon and crude bioabsorbent from Moringa oleifera leaf powder in depolluting the treated pond.

The aim of our research is to detoxify the treated pond with bioadsorbents synthesized from Moringa oleifera leaf powder and activated carbon. The use of these natural depollutants to detoxify fish ponds will boost the productivity of fishery resources by using more ecological means to preserve our environment.

2. Materials and methods

Preparation of Moringa powder

• Washing

Once removed from the branches, the leaflets are washed once with distilled water to remove dust. They are then washed in a 1% saline solution for about 15 minutes to eliminate germs. Finally they are rinsed a second time with distilled water.

• Draining and drying

The leaflets are then drained before being dried. The method used for this study is solar drying; the leaflets were spread on a container for four hours per day and for four days.

• Grinding and sieving

The dry leaves are then crushed using a mortar and pestle and then sifted to obtain a fine Moringa leaf powder.

Preparation of activated carbon from Moringa leaf powder

A sample of Moringa leaf powder was transformed into activated carbon according to the experimental protocol optimized in several research works on activated carbons [8]. The chemical activation of biomass by H_3PO_4 is carried out in two stages, impregnation followed by pyrolysis:

- A mass of Moringa leaf powder was impregnated with 40% phosphoric acid (H3PO4) solution with a mass ratio of 1:2;
- The mixture obtained is placed in an oven (Memmert model) for 8 hours at 100°C;
- The product obtained was placed in porcelain crucibles then placed in an oven (Nabertherm model) whose oven temperature evolves increasingly with heating rates of 10 °C/min, then an isotherm level of 60 min at 450°C;
- In order to eliminate possible carbonization residues, the charcoal obtained was washed with a 0.1 N hydrochloric acid solution [9].
- The hydrochloric acid was removed by washing with distilled water several times until a constant pH was obtained;
- After rinsing, the washed carbon was dried in an oven at 100°C for a period of 8 hours;
- The activated carbon obtained is stored in airtight bottles for subsequent studies.

Characterization of adsorbents

Determination of mass yield of activated carbon

Yield is an important quantitative characteristic for activated carbons. It reflects the mass loss of biomass during its pyrolysis [5].

It is an important measure of the feasibility of producing activated carbon and an indicator of the performance of a preparation method, especially on an industrial scale [10].

Humidity rate

The humidity level provides information on the dry matter content (actual mass of material) placed in contact with the effluent [11].

The method used is desiccation by evaporation or gravimetric method. The sample is steamed at 100°C until a constant mass is obtained. The moisture content is obtained by differential weighing before and after drying [12].

Sampling

The fish pond samples were taken at the fish farming diversification research station of the Wetlands Research Laboratory. Using a bucket, a strainer, and two 5L cans.

The physicochemical parameters of the samples taken are: Temperature: 29.75°C; pH: 5.2; Conductivity: 3.84 µs/cm; Total Dissolved Solids (TDS): 10.21 mg/l.

The cans were hermetically closed and then transported to the Laboratory of Inorganic Environmental Chemistry (LaCIE) for analyses.

Cytotoxicity test

The onions used for the toxicity test of the fish pond samples were purchased in Abomey-Calavi at the Kpoôta market. The first dry layers of the onions were removed as well as all the roots. The weight of the onions varies between 40g and 80g and their diameters vary between 4.8cm and 6.7cm.

3. Results

Mass yield

The mass yield of activated carbon prepared for this study is 33.53%. This result is comparable to the results of previous work carried out by Master students of ADRAR University on water treatment using the pods of the Moringa oleifera plant who found a yield of 34.46% for the preparation of activated carbon from Moringa oleifera powder [13].

Humidity rate

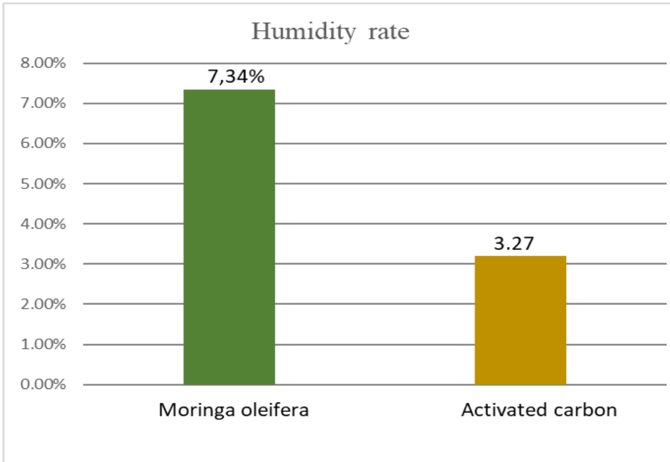

Figure 1: Humidity rate of the two adsorbents

The humidity level values obtained are within the standards and are less than 10% according to [14]. The humidity rate of Moringa powder is higher than that of activated carbon.

Parameters based on root length of onions grown in fish pond water samples

An inhibition of onion root growth was observed in samples from the fish pond analyzed with the cytotoxicity test carried out on onion roots. We noticed that the roots of the onions cultivated did not grow well like those of the 0% concentration onions. We can therefore conclude that the fish pond studied is polluted.

The bioadsorbent (BA) and activated carbon (AC) obtained from Moringa leaf powder made it possible to depollute water samples taken from the fish pond. A comparative study of the toxicity of water samples treated with the crude bioadsorbent and its activated carbon was carried out. The results obtained showed the effectiveness of the bioadsorbents used to depollute fish water samples.

The average root lengths of the onions grown in the fish pond samples are shown in (Figure 2).

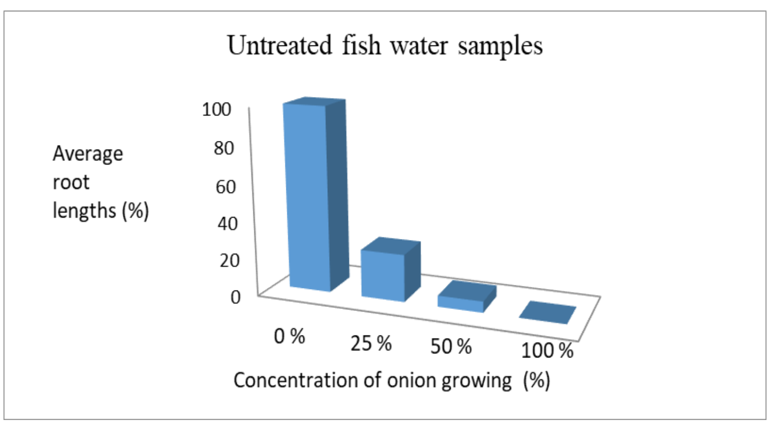

Figure 2: Average root lengths of onions grown in samples from the fish pond.

Parameters based on the length of the roots of onions grown in fish water samples decontaminated by the crude bioadsorbent (BA) based on Moringa oleifera powder

Samples treated with bioadsorbent (BA)

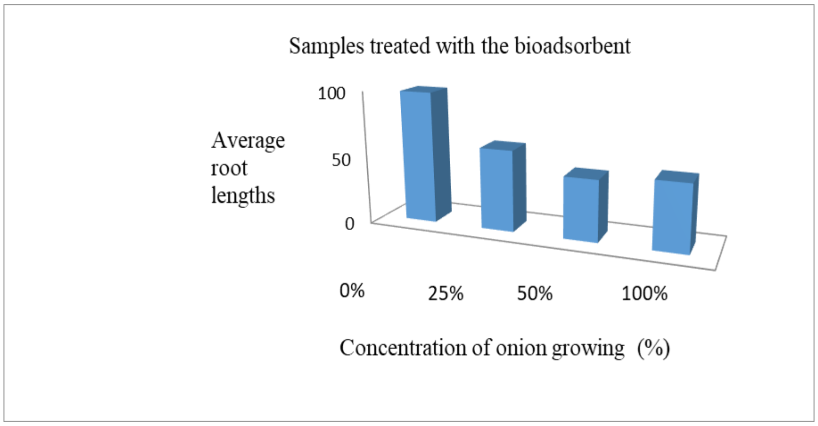

Figure 3: Average root lengths of onions grown in fish water samples treated with bioadsorbent (BA)

The average root lengths obtained in (Figure 3) are respectively equal to 60.82% and 45.83% for the 25% and 50% concentrations. For a concentration of 100%, it increases to 50%. We note that the roots of onions grown in water samples treated with bioadsorbent (BA) grew more than onions grown in untreated fish water samples. We can therefore conclude that the bioadsorbent used made it possible to detoxify the fish water samples.

Fish water samples treated with Activated Carbon (AC) from Moringa oleifera leaf powder

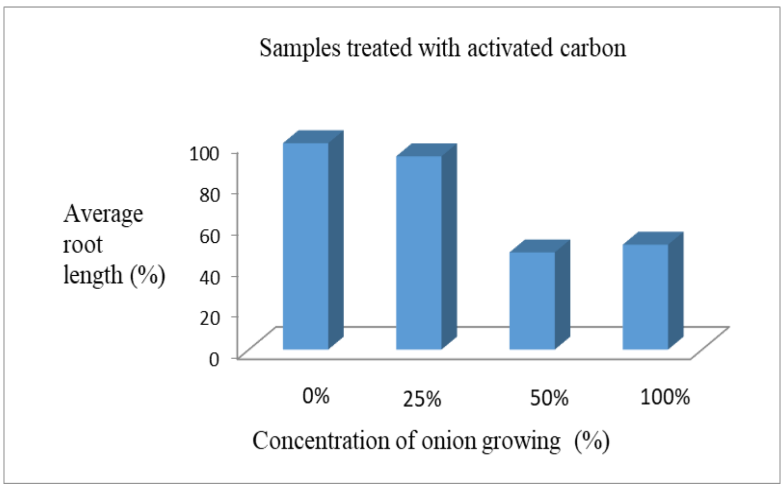

Figure 4: Average root lengths of onions grown in fish water samples treated with activated carbon from Moringa oleifera powder

The cytotoxicity results (Figure 4) show that the average root lengths for the 25%, 50%, and 100% concentrations are respectively 93.65%; 47.08% and 50.83%. These results indicate a decrease in root length when the concentration varies from 25% to 50% followed by root growth when the concentration of culture media increases from 50% to 100%. But we note that the averages of the roots obtained are all higher than those obtained after using the crude bioadsorbent (BA) as a detoxifier. It is therefore clear that activated carbon was more effective in detoxifying fish water samples than the bioadsorbent (BA).

Table 1: Average root lengths of onions grown in different concentrations

	Average onion root lengths (%)		
Concentrations of treated effluent samples	**Untreated fish water samples**	**Fish water samples treated by BA**	**Fish water samples treated by AC**
0 %	100	98,77	100
25 %	25,55	60,83	93,65
50 %	6	45,83	47,08
100 %	0	50	50,83

For untreated fish water samples, root length averages decrease to 0%, when concentrations vary from 25% to 100% (Table 1). It is deduced that the untreated fish pond water samples prevent the roots of the onions from growing, which shows that the fish pond water is toxic.

Parameters based on onion root growth inhibitions

We call Effective Concentration (EC50), the one for which chemical effects are observed for 50% of the individuals tested. Inhibition of onion root growth prevents onion roots from growing and is indicative of parameters based on effective concentration.

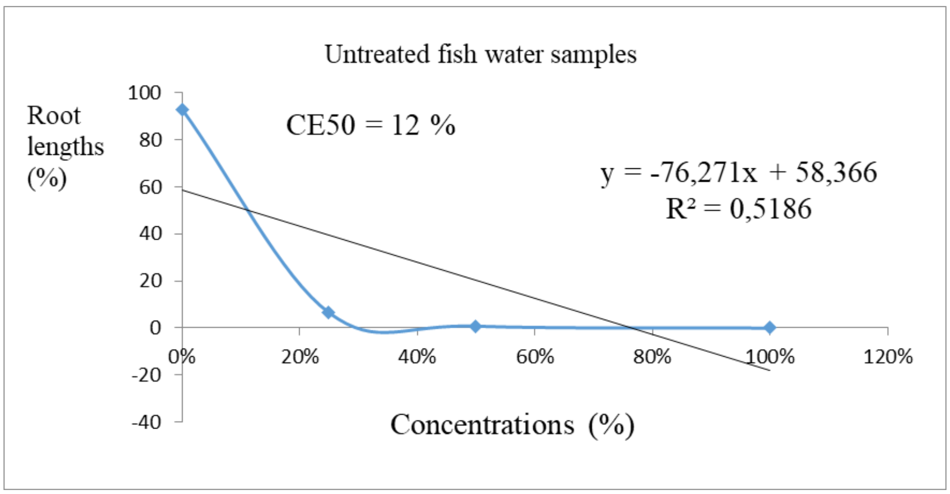

Figure 5: EC50 of untreated fish water samples

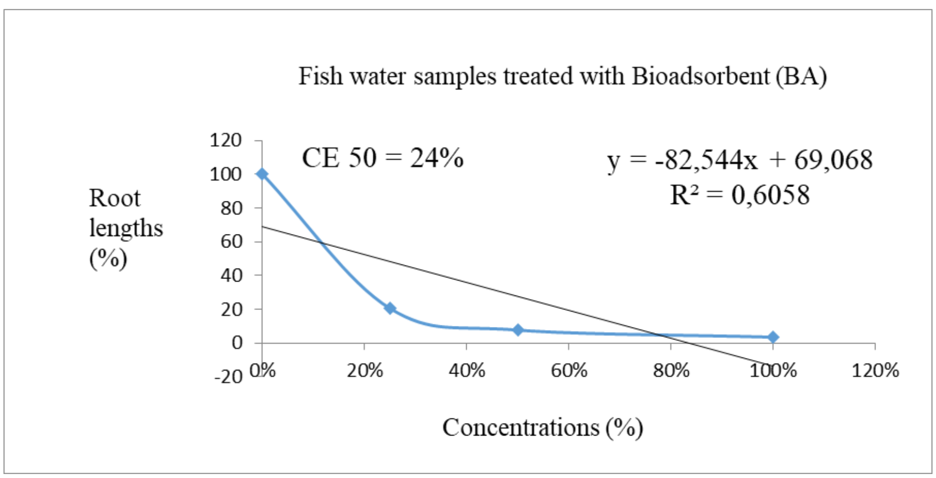

Figure 6: EC50 of fish water samples treated with the bioadsorbent (BA)

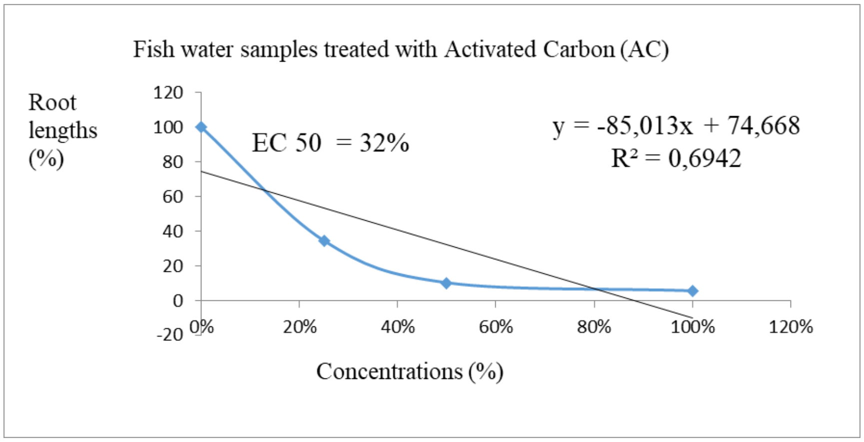

Figure 7: EC50 of fish water samples treated with Activated Carbon (AC)

The toxicity of the fish water samples studied was evaluated by the EC50 Effective Concentrations obtained. The effective EC50 concentration of untreated water samples is 12% (figure 5). Two depollutants based on Moringa oleifera leaf powder and its activated carbon were used for the detoxification of fish water samples. The effective EC50 concentration obtained when the fish water samples were treated with the bioadsorbent is equal to 24% (Figure 6). The EC50 obtained after the treatment of the samples with Activated Carbon obtained from Moringa oleifera powder is 32% (Figure 7). All EC50 values obtained are shown in table (2).

Table 2: EC50 of fish water samples

	Untreated fish water samples	Fish water samples treated with raw BA	Fish water samples treated by AC
EC50	12 %	24 %	32 %

4. Discussion

The humidity rate represents an important indicator of product quality [15]. The higher the humidity content, the more carbon is likely to stimulate fungi; so the moisture content affects the storage and shelf life of activated carbon. Too high a humidity level can encourage the development of mold and bacteria, thus degrading the properties of Moringa oleifera powder and its activated carbon. Microorganisms can also multiply in the pores blocking them and thus reducing the carbon adsorption capacity. The studies that have been carried out have shown that lower moisture content increases the adsorption rate of contaminants [16].

Yield is also a very important parameter in the characterization of bioadsorbents and their activated carbons. That obtained for the activated carbon synthesized from Moringa oleifera leaf powder is very close to those obtained by other researchers who have carried out work on Moringa oleifera powder.

The cytotoxicity test carried out is based on the number and length growth of roots of onions grown in the different concentrations of fish pond samples. The greater the number and length of the roots, the less polluted the growing medium.

The concentrations carried out for this study are: 0%; 25%; 50% and 100%. They are obtained by diluting the samples taken with distilled water [17]. Each concentration is repeated four times; we therefore have sixteen (16) culture media for each analysis.

The raw adsorbent synthesized from Moringa oleifera powder and its activated carbon with low humidity levels are ideal for the storage of pollutants and therefore have high contaminant adsorption rates.

The histogram of the average root length of the onions grown in the samples from the fish pond is shown in (figure 2). The smallest root is 0.1 cm long and the largest is 2.7 cm. For onions grown in concentrations 0% and 25%, the average root lengths are 100% and 25.55%, respectively. In culture media concentrated at 50% and 100%, the average root lengths are 6% and 0%, respectively. When the concentration increases in the culture medium, we see that the average length of the roots decreases and that no root has grown 100%. We can therefore deduce that the roots were prevented from growing by the waters of the fish pond.

When the concentration varies from 0% to 100%, we see that the average lengths of the onion roots obtained after treatment with Activated Carbon are greater than those of the samples treated with the bioadsorbent (BA) then those of the untreated samples. These results show that Activated Carbon (AC) makes it possible to decontaminate the fish pond analyzed.

The highest effective concentration obtained during this study is equal to 32% (Table 2). It is deduced that Activated Carbon (AC) prepared from Moringa oleifera powder detoxifies fish water samples more effectively than the bioadsorbent (BA). The EC50 of BA and AC depollutants have values higher than that of untreated fish water samples which is equal to 12%. We can therefore conclude that the two depollutants based on Moringa oleifera powder used for this study allowed detoxification of the waters of the pond studied.

5. Conclusion

A comparative study on the detoxifying power of the crude bioadsorbent and its activated carbon obtained from Moringa oleifera powder on fish pond samples taken in the research station on the diversification of fish farming of the Wetlands Research Laboratory was made.

Assessment of the toxicity of these fish water samples using the onion root growth inhibition based cytotoxicity test reveals that the tested samples prevented onion roots from growing. This inhibition was observed completely in the 100% concentration, that is to say that the roots of the onions practically did not grow at the level of the pond samples tested. We can therefore conclude that the fish pond studied is chemically polluted.

These water samples from the pond studied were then depolluted firstly with Moringa oleifera BA powder and secondly with its Activated Carbon (AC). The different EC50 values obtained after 72 hours of exposure of onion roots in the different concentrations of the pond samples showed that root growth varies depending on the concentrations used. These values of effective concentrations made it possible to demonstrate that the EC50 of BA and AC depollutants are respectively equal to 24% and 32%. We note that these values are higher than that obtained with untreated fish water samples which is equal to 12%.

We can therefore conclude that the two depollutants based on Moringa oleifera powder used for this study allowed detoxification of the waters of the pond studied, and that the Activated Carbon (AC) prepared from Moringa oleifera powder detoxified the samples more effectively of fish water than the bioadsorbent (BA).

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