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**Phytochemistry and Potential bio-sorbent of
Euphorbia prostrata extract to remove Cu, Pb, Cd
from Ouidah coastal lagoon waters in Benin**

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Abstract

The use of plant-based biosorbents in water treatment is one of the scientific challenges. This present work contributes to the valorization of *Euphorbia prostrata* as a biosorbent of certain metals and its application for the treatment of water in the coastal lagoon of Ouidah. The content of Cu^{2+} , Pb^{2+} , Cd^{2+} and Zn^{2+} of water taken from five (05) sites around the coastal lagoon was measured by the atomic absorption spectrophotometer method. The phytochemistry of the aqueous and ethanolic extract (70°) of *Euphorbia prostrata* indicates the presence of flavonoids, leucoanthocyanins, coumarins, reducing compounds and that they are rich in alkaloids and tannins, both catechic and gallic. From the ETM analysis, it appears that the lagoon water contains high levels of heavy metals of Cu^{2+} (8.16 – 10.32 mg/L), Pb^{2+} (5.02 – 14.59 mg/L) and Cd^{2+} (6.52 – 8.71 mg/L) from one site to another. The removal of metals from the samples by the different extracts revealed that they have an absorbent action where the ethanolic extract (70°) of *Euphorbia prostrata* expressed an efficiency of: Cu (96.07%), Pb (97.01%) and Cd (59.86%). It makes this plant a good bio-sorbent candidate for heavy metals in water treatment.

Keywords: Tannins, alkaloids, biosorbent, heavy metals, *Euphorbia prostrata*, Ouidah lagoon

Introduction

In recent years, the use of biocoagulants, biosorbents that are biodegradable and have negligible side effects on human health has been a quest in many water treatment studies.

Literature reports that some plants such as *Moringa oleifera*, a plant used for the prevention and treatment of more than 300 diseases in Indian Ayurvedic medicine (Ghazali and Abdulkarim, 2015; Dzuvo et al., 2021) has been shown to be interesting in water treatment. The seeds of this plant have shown effectiveness, on the one hand, on water turbidity compared to polyaluminium chloride (Yarahmadi et al., 2009) and on the other hand, on raw water bacteria without chlorination compared to aluminum sulfate (Mohamed et al., 2019).

In the many studies of biosorbent plants in water treatment, the literature does not mention *Euphorbia prostrata*.

Euphorbia prostrate Commonly called red spurge, is a plant that grows in gardens, disturbed areas, cultivated land and roadsides, especially on sandy soils (Mosango, 2008). It has a wide range of interesting biological activity such as antibacterial, anti-inflammatory (Single et al., 1989; Syed et al., 2013; Sultana et al., 2016,

Voukeng et al. 2017) and antioxidants (Sundara Prabha and Beschi Antony Rayan, 2018).

The present work reports the biosorbent potential of extracts from the leaves of *Euphorbia prostrata* acclimatized in Benin in the removal of heavy metals and its application in the treatment of water from the coastal lagoon of Ouidah in Benin.

Materials and Methods

Plant material and Preparation of the Plant Extracts

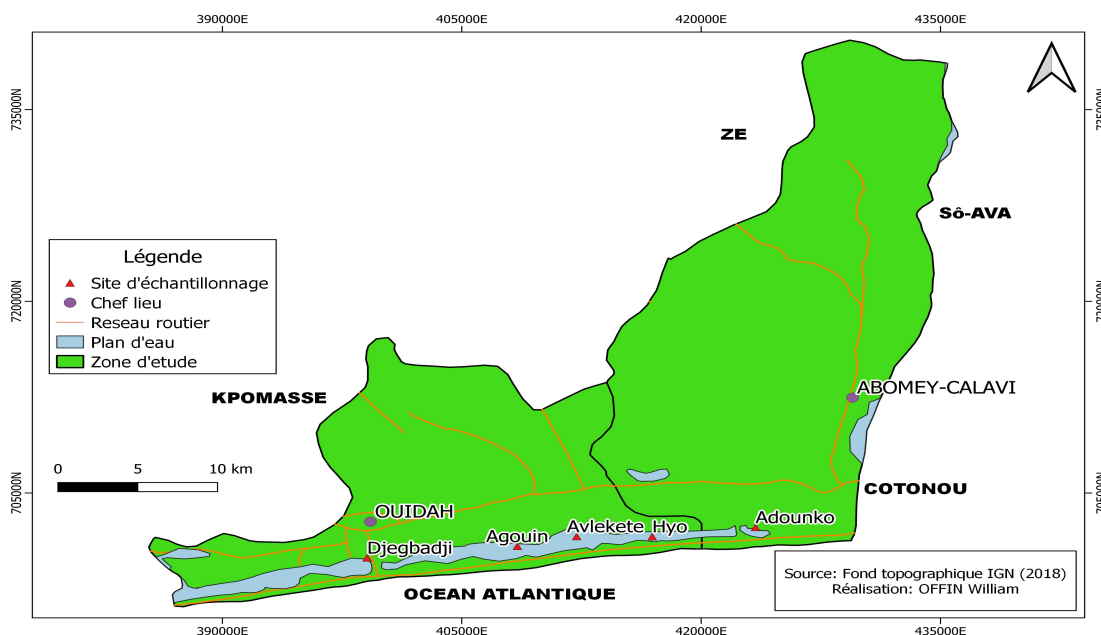
Euphorbia prostrate plant comes in two ecotypes: red and green. The red form was collected in Glo Gjigbé in the commune of Abomey-Calavi (Benin). The leaves, washed and dried at a temperature of 18°C in the laboratory for a few days, are placed in an oven at 60°C for one hour before being ground.

The aqueous extract was obtained by decoction. 20 g of the powder was put in 400 ml of distilled water and boiled for 30 minutes. As for the hydroethanolic extract, it was obtained by maceration, 10 g of the powder was put in 150 ml of the organic solvent and then stirred for 24 hours. After filtration, all the extracts were condensed using a rotary evaporator (IKA HB 10 basic) and dried in an oven at 60 ° C. The extracts obtained were kept cold at -4 ° C before use.

Description of study area

The Ouidah coastal lagoon is upstream of the entire coastal lagoon that is included in the western complex of wetlands of southern Benin (Ramsar site no. 1017). It is located between the parallels $6^{\circ}27'01''$ and $6^{\circ}19'40''$ and between the meridians $2^{\circ}20'48''$ and $2^{\circ}04'57''$ and extends over an area of 4000 hectares. It takes its source from the Grand-Popo lagoon and the Ahô channel, then ends its course at the level of the village Togbin (Viaho et al. 2020).

Five (05) sampling sites in the municipalities of Abomey-calavi and Ouidah were chosen for general representativeness and coverage of all agricultural activities in the lagoon. These sites S1, S2, S3, S4, S5 are located and delimit respectively the villages Togbin ($6^{\circ}27'$ North - $2^{\circ}04'$ East); Hyo Houta ($6^{\circ}20'$ North - $2^{\circ}14'$ East); Agouin ($6^{\circ}20'$ North - $2^{\circ}10'$ East); Avlékété ($6^{\circ}21'$ North - $2^{\circ}12'$ East); Djègbadji ($6^{\circ}19'$ North - $2^{\circ}04'$ East) (Figure 1).



Phytochemical screening

The presence of compounds belonging to different classes of secondary metabolites in different extracts was determined according to the methods described by Houghton & Raman (1998) and by Shaikh & Patil (2020), reviewed and supplemented.

Copper absorption experiments

The copper test experiment was carried out to identify the extract having a high copper absorption capacity; and its use in the treatment of water samples from Ouidah coastal lagoon. The Bicinchoninate method described by Nakano & Yakugaku (1962) and adapted to laboratory conditions was used.

A solution of copper sulfate at 1.84 mg/L was prepared and divided into 15 ml in different beakers. In each beaker, only 0.2% or 0.03 g of a single given extract was added, then labeled. The beaker without the addition of extract constitutes a control. The solutions of copper sulfate with the extract were stirred for 2 hours, decanted and filtered into tubes. In the 10 ml tubes of each of the labeled solutions and the control solution, a sachet of the HACH brand Kit (Permachem Reagents. Cat. 2105869 pk/100) was added, respectively. The readings indicating the amount of copper Co in the control solution and the amount of copper C present in each tube were made using the DR/890 colorimeter, after 2 minutes of the addition.

The percentage of Cu absorption was calculated from the following formula: Co: amount of copper in the control solution; C: amount of uncomplexed copper present in the extract tube.

$$R(\%) = 100 \left(\frac{Co - Cf}{Co} \right)$$

Processing and analysis of water samples

Water samples from the different sites were collected using 1.5 L plastic cans. The rinsed cans were immersed in water 20 cm from the surface and filled to the brim to avoid the entry of air

bulbs. The extract absorption experiment was carried out at laboratory temperature according to the methods described by Assaad(2006) and Siew-Teng et al., (2012), reviewed and adapted under laboratory conditions. Control tubes (tubes without addition of the extract) from each site were made beforehand. In 100 ml of the water samples from each site distributed in tubes, 0.5 mg of the extract was added. These tubes were stirred for 3 hours, then filtered on filter paper after 30 minutes of decantation. Copper, zinc, lead and cadmium were analyzed by a flame atomic absorption spectrophotometer (AAS).

Results and Discussion

Table 1: Phytochemical composition of *Euphorbia prostata* extracts

Phytochemical composition	Eaq	EHE
Alkaloids	+	+
catechismTannins	+	+
gallic tannins	+	+
Flavonoids	+	+
Anthocyanins	-	-
Anthraquinones	+	+
Leuco-anthocyanin	+	+
Coumarins	+	+
Saponins	+	+
Terpenoids	+	+
Reducing compounds	+	+

(+): Present; (-) : Absent ; Eaq: Aqueous extract; EHE: Hydroethanolic extract (70°)

Table 2: Copper test by the bicinchoninate method

Extracts	Remaining copper content: Cf (mg/l)	Yield (%)
Eaq	1.60	13.04
EHE	0.07	96.19
Witness (Co,mg/l)	1.84	

Eaq: Aqueous extract; EHE: Hydroethanolic extract (70°)

Table 3: Content heavy metals in samples before and after treatment

	Cu (mg/l)			Zn (mg/l)		Pb (mg/l)			Cd (mg/l)		
	T0	TEP	P %	T0	TEP	T0	TE	P %	T0	TE	P %
Togbin (Site 1)	10.32	0.703	93.18	00	00	5.27	0.598	93.51	00	00	
HyoHouta (Site 2)	8.16	0.366	95.51	00	00	0.69	0.312	97.01	6.52	2.617	59.86
Avlekete (site 3)	10.02	0.449	95.51	00	00	5.02	0.382	92.39	7.73	3.218	58.36
Agouin (site 4)	13.05	0.512	96.07	00	00	14.59	0.435	54.78	8.71	3,673	57.83
Djègbadji (site 5)	9.96	0.606	93.91	00	00	7.96	0.516	88.65	5.29	4.357	17.63

P%: absorption percentage, $P (\%) = 100 \left(\frac{T_0 - T_{EP}}{T_0} \right)$

The phytochemical composition of the extracts recorded in Table 1 revealed that in all the extracts, the presence of alkaloids, flavonoids, leucoanthocyanins, coumarins, reducing compounds and tannins both catechic and gallic. This composition is similar to that found by Kengni et al (2013) in the aqueous extract, by Kamganget al (2007) in the hydroethanolic extract (80°) and by Farooq et al (2016) for the methanolic extract.

Furthermore, Sharma et al (2012) and Voukeng et al (2017), in their phytochemical investigation work of *Euphorbia Prostrata* have reported an absence of alkaloids in the aqueous, ethanolic and methanolic extracts, respectively.

Our phytochemical study showed that the aqueous and hydroethanolic extracts of *Euphorbia Prostrata* acclimatized in Benin are rich in alkaloids. It could be explained by biological and environmental factors influencing genetic variability and the production of secondary metabolites of a given species (Gouvea et al., 2012), thus resulting in plants with different chemical compositions and therefore sometimes different ones depending on the collection region (Cunha, 2010).

Analysis of water samples from the various sites revealed the presence of certain heavy metals including Cu, Pb and Cd and an absence of Zn (Table 3). The treatment of water from the Ouidah lagoon with an extract of *Euphorbia Prostrata* acclimatized to Benin revealed an interesting absorbent potential in ETM (Table 3).

Indeed, the hydro-ethanolic extracts of *Euphorbia Prostrata* showed an absorbing action on Cu, Pb and Cd (Table 3) with an absorption of Cu (96.07%), Pb (97.01%) and Cd (59.86%) for some samples. The absence of trace of zinc after treatment of the samples indicates that the extracts are not rich in zinc.

The high potency of the hydro-ethanolic extract of *Euphorbia prostrata* confirmed the results of the copper analysis test by the bicinchoninate method where this extract showed a high rate of decrease in copper from 1.84 to 0.07 mg/l or 96.19% compared to the aqueous extract (table 2).

This sorbent potential of the different extracts could be explained by the presence of tannins contained in the extracts. Indeed, according to the literature, tannin-rich plants have been widely accepted and applied as biocoagulant, biosorbent

in wastewater treatment with potential pollutant removal power including turbidity, dyes, suspended solids, chemical oxygen demand, algae and heavy metals (Kavitha et al., 2020; Leiviskä and Santos 2023).

Edogbanya et al (2016; 2017), in his studies on bioremediation *Adansonia digitata* L. oil cake residues in the removal of Pb and Cd from water, showed interesting efficiency up to lead (99.99%) and cadmium (74.27%). In addition, Subramaniam et al (2011) showed that *Moringa* seeds eliminate the heavy metals Cu, Pb, Cd, Cr and Zn with interesting elimination percentages: Cu (90%), Pb (80%), Cd (60%), Zn (50%) and Cr (50%).

Our present study showed that *Euphorbia prostrata* has an interesting biosorbent potential for the elimination of heavy metals Cu, Pb, Cd and would have a better efficiency than moringa and *Adansonia digitata* seeds.

Conclusion

This study allows, on the one hand, to contribute to the valorization of the natural resources of our country and on the other hand, the possibility of introducing new biosorbent in the water treatment process. The hydroethanolic extract of *Euphorbia prostrata*, a medicinal plant has been shown to have interesting absorbent potential in bioremediation of Trans metallic elements (TME) such as Pb, Cd, Cu. The results obtained can serve as a preliminary test for other extracts of this plant and also other plants in their biosorbent capacity. This performance can be improved by the extraction and purification of tannins; this can be studied. The excerpts of *Euphorbia prostrata* and other plants will therefore have a strong possibility of being an alternative to coagulants and chemical flocculants.

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Conflict of Interest Statement

The authors declared no conflict of interest.

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