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Evaluation of sodium chloride (NaCl) effects on water hyacinth *Eichhornia crassipes* development : Preliminary results

Evaluation des effets du chlorure de sodium (NaCl) sur le développement de la jacinthe d'eau *Eichhornia crassipes*: Résultats préliminaires

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NADEGE CHIMENE GUEZO[1,2], EMILE DIDIER FIOGBE[1], MAHUNAN TOBIAS CÉSAIRE AZON[1], PAUL ESSETCHI KOUAMELAN[2] & ALLASSANE OUATTARA[3]
(1) Research laboratory on Wetlands (URZH)
University of Abomey-Calavi
Benin 01 B.P. 526 Cotonou
Email : edfiogbe@ yahoo.fr** Or **azonmahunan@gmail.com
(2) Ecotoxicology and hydrobiology laboratory
University Félix Houphouët - Boigny
Ivory Coast - 22BP : 582 Abidjan
***Email :** kessetch2012@gmail.com.*
(3) Laboratory of environment and Aquatic Biology
University Nangui Abrogoua
Ivory Coast - 02 BP 821 Abidjan
Email : allassane_ouattara@yahoo.fr

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Abstract Ability of water hyacinth to live in different basins (lakes and lagoons) tropical regions makes this species an adventitious by excellence. Its high multiplication capacity poses enormous problems to the fisheries sector due to the dense carpet that it forms on the surface of the water thus preventing navigation, the penetration of light, the dissolution of oxygen and causing death of some aquatic organisms. This plant strictly subject to freshwater is considered as aquatic species among the most harmful in the world because of its excellent reproductive power. Salinity generally limited its development in coastal and estuarine areas. The present study examines the development of water hyacinth subjected to a salinity gradient.

Experiments were conducted in buckets of 15 L plastic at the rate of ten (10) treatments including one (01) control, repeated four times.The salinity gradient consisted of ten levels : 0 g/L, 5 g/L, 9 g/L, 13 g/L, 15 g/L, 18 g/L, 21 g/L, 24 g/L, 27 g/L and 30 g/L.

The results of these studies showed, firstly, that for the direct supply of salt in the Middle, water hyacinth can't survive at a salinity of the environment greater than or equal to 9 g/L. On the other hand, the effective salinity for transfoliaire spraying of Eichhornia crassipes is 24 g/l.

MOTS CLES
Lutte, chlorure de sodium, *Eichhornia crassipes*

Résumé L'habileté de la jacinthe d'eau *Eichhornia crassipes* à vivre dans différents bassins (lacs et lagunes) des régions tropicales fait de cette espèce un adventice par excellence. Sa forte capacité de multiplication pose d'énormes problèmes au secteur halieutique du fait du tapis dense qu'elle forme à la surface de l'eau empêchant ainsi la navigation, la pénétration de la lumière, la dissolution

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de l'oxygène et provoquant la mort de certains organismes aquatiques. Cette plante strictement inféodées aux eaux douces est considérée comme l'espèce aquatique parmi les plus nuisibles au monde de par son excellent pouvoir de reproduction. La salinité limite généralement le développement de l'espèce dans les zones côtières et estuariennes. La présente étude examine le développement de la jacinthe d'eau soumise à un gradient de salinité.

Les expériences ont été réalisées dans des seaux en plastique de 15 L à raison de dix (10) traitements dont un (01) témoin, répétés quatre fois. Chaque seau contenait de l'eau douce fertilisée à base du fumier de porc et trois spécimens de jacinthe d'eau. La première étape a consisté à l'adduction directe de sel dans le milieu dans le but de déterminer la dose de sel pouvant accélérer la sédimentation de la jacinthe d'eau et la deuxième au traitement par pulvérisation transfoliaire de solutions salées. Le gradient de salinité était constitué de dix niveaux : 0 g/L, 5 g/L, 9 g/L, 13 g/L, 15 g/L, 18 g/L, 21 g/L, 24 g/L, 27 g/L et 30 g/L.

Les résultats de ces études ont montré d'une part que pour l'adduction directe de sel dans le milieu, la jacinthe d'eau ne peut survivre à une salinité du milieu supérieure ou égale à 9 g/L. D'autre part, la salinité efficace pour la pulvérisation transfoliaire de la jacinthe d'eau *Eichhornia crassipes* est de 24 g/L.

1. Introduction

Water Hyacinth is a floating aquatic plant that can live and reproduce on the surface of freshwater. Its size varies from a few centimeters to a meter in height depending on the environment [1. Its proliferation rate, under certain circumstances, is extremely rapid and can spread to large water surface. [2] reported that hyacinth originated in Amazon basin and has been introduced in many parts of the world as an ornamental plant in ponds because of its beauty. It has proliferated on every continent outside Europe. The plant is an aquatic perennial herb that belongs Pontederiaceae family [3]. Reproduction is usually asexual and takes place at a rapid pace in conditions preferential. Over the past ten decades, rapid spread of water hyacinth in many parts of Africa has become very alarming. According to [4], ten plants in eight months can produce 655.330 individuals, underscoring invasive potential of the plant. Similarly, [5] received 30 clones from two (02) adulte plants in 23 days.

Environment in general and aquatic biodiversity balance in particular are threatened by the spread of this plant. Environment and aquatic biodiversity balance are threatened by the spread of this plant. Indeed, infestation of water causes not only significant or total reduction of autochthonous aquatic plants; but it causes also the installation of a dense herbaceous carpet that interferes with rice crops, irrigation systems, hydroelectric production and obstructs bridges, canals. It also causes inaccessibility of humans, livestock and wild animals to water for their consumption. It promotes implentation of suitable habitats of water borne diseases vectors (malaria, bilharzias, cholera and onchocerciasis), blocking light penetration and navigation [6]. Several methods of fighting against this plant have been adopted, but none of them has produced a satisfactory result. Of course it's physical struggles (manual and mechanical) consisting in manual removal of plants as well as using heavy equipment [7]; chemical fights based on using of herbicides; biological struggles based on using of natural enemies (insects, aquatic herbivores) of the plant in order to create a permanent pressure on it [8].

This study evaluates the effect of sodium chloride (NaCl) known as first-order enemy of water hyacinth. It consisted of monitoring the development of water hyacinth subjected to various salinities and testing salinity levels above tolerance limit, by spraying.

The success of this method can serve as an alternative to a significant reduction of hyacinth plants in infested water.

2. Material and Methods

2.1. Equipment

Wetland Research Laboratory (LRZH) of Abomey-Calavi University (Benin) served as a framework for the realization of this study. The experimental device consists of plastic buckets 30 cm in diameter and 15 liters of water as a useful volume. Hyacinth plants collected in situ were used as biological material, dry pig manure was used to

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fertilize culture medium, and fine grain cooking salt (NaCl) was chemical material used.

2.2. Methodology

Buckets received 10L of freshwater fertilized pork manure. The concentration of the different media in nitrogen and phosphorus after fertilization was respectively 5.5 and 1.06 mg/l [9]. Three days after fertilization, news plants were then rinsed before being introduced into the buckets, in order to avoid proliferation of possible small water lenses. The plants were selected according to their number's leaves. Thus, the plants-girls had between 02 and 03 leaves; young plants between 05 and 06 leaves and adult plants b07 and 09 leaves. They had been in buckets of fertilized water for a week.

2.2.1. Treatment of plants by salt adduction

Saline solutions were prepared from the contents of each bucket following a concentration gradient. For this purpose, we have ten (10) levels of salinities namely: 0 g/L; 5 g/L; 9 g/L; 13 g/L; 15 g/L and 18 g/L; 21 g/L; 24 g/L; 27 g/L and 30 g/L. Each level of salinity corresponded to a treatment. For saline solutions preparation, 2L of water were taken from each bucket and were used to dissolve the amount of salt required for each treatment. After salt total dissolution, the salt solution obtained was poured back into the bucket. The contents were homogenized and water levels were also readjusted on a daily to compensate evapotranspiration losses. Plants were treated for a week. The buckets were installed at natural photoperiod and in open air. A "Copper pegler" brand sprayer having a maximum pressure of three (03) bar with a maximum flow of 1.7L was used to spray hyacinth plants.

2.2.2. Plants treatment by spraying

Another sampling was done following same methodology but now we have seven (07) treatments repeated twice. Saline solutions were prepared in plastic bottles of one liter (1 L) capacity following a gradient of seven levels namely 13 g/L, 15 g/L, 18 g/L, 21 g/L, 24 g/L, 27 g/L and 30 g/L. Each saline solution corresponded to a treatment. These solutions were applied to water hyacinth plants by spraying. The spraying was done mainly on plant's leaves and this only once a day for two (02) weeks. The daily amount of saline spray was 250 mL per observation. The moment chosen for this exercise is period of strong sunshine of the day one (01) o'clock pm. Assessment of sodium chloride effect on water hyacinth was done by measuring growth parameters (leave's number, their length and width) and by taking of physic-chemical parameters of water. Water physic-chemical parameters were taken at regular intervals of four days while plant growth parameters have been taken once a week.

2.2.3. Data analyzes

The remaining number of leaves at each observation was obtained from the following formula : $N_r = N_i - N_f$

Where Nr is the remaining number of leaves ; Nf the final number of leaves (after the experiments) and Ni the initial number of leaves (that is to say before the experiments).

The same technique was also used for the length and width of the leaves. STATISTICA version 6 were used to make data analyzes.

Variance analyse (ANOVA) was performed to assess any significant differences between the averages. The application of FISHER's 'Less Significant Differences' statistical test (FISHER LSD) enabled us to make comparisons between treatments.

3. Results

3.1. Plants treatment by salt adduction

One day after adding salt to the medium, *Eichhornia crassipes* leaves began to soften and fold from T2 treatment to T9 treatment. Furthermore, four (04 days after salt adduction, all the plants were decimated from T3 to T9 treatment. For plants submited to T2 treatment, it is on the 7th day that the plants mortality were observed. We also noted that at the level of each treatment, the mortality started with the daughter-plant, then the young plant and finally adult plants. However, no mortality was recorded at the T0 and T1 treatments; we noted that some leaves were destroyed at T1 treatment but plants were living normally. Variance Analysis (ANOVA) revealed significant differences between the means of different treatments ($p<0.05$).

3.2. Plants treatment by spraying

After two weeks of spraying, all daughter plants were decimated from T1 to T7 treatment. As for young plants, mortalities were recorded from T3 to T7 treatment. Adult plants were decimated from T5 treatment to T7 treatment. No persistent leaves were observed at the level of the different treatments after spraying (Pictures A,B,C,D,E,F,G,H).

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Picture: A

Picture: B

Picture: C

Picture: D

Picture A shows plants condition before spraying; those B, C and D show their state respectively at T5 24 (g / L), T6 27 (g/L) and T7 (g/L) treatments after two weeks of spraying.

Picture: E

Picture: F

Picture: G

Picture: H

Source: Personal work

Pictures E, F, G and H show the state of the plants respectively at T1 (13g / L), T2 (15g / L) T3 (18g / L) and (T4 21g / L) treatments after two weeks of spray.

Table 1 shows average value of leaves number recorded at the level of each treatment before and after spraying.

Table 1: Average value of leaves number recorded at the level of each treatment.

	Adult plants		Young plants		Daughter plants	
	Ni_{-1}	**Nf_{-1}**	**Ni_{-2}**	**Nf_{-2}**	**Ni_{-3}**	**Nf_{-3}**
T1 (13 g/L)	16± 04	6±01	08±03	3±01	4±01	-
T2 (15 g/L)	14± 02	03±01	12±03	01	05±02	-
T3 (18 g/L)	17±04	02	15±03	0	06±01	-
T4 (21 g/L)	18±03	01	14±02	-	07±01	-
T5 (24 g/L)	18±01	-	12±04	-	06±01	-
T6 (27 g/L)	12±03	-	12±04	-	06±02	-
T7 (30 g/L)	18±04	-	12±03	-	07±01	-

Source: Personal work

Ni : Initial leaves number (before spraying) ; Nf : Final leaves number (after spraying)

Statistical analyzes revealed significant differences between the means ($p < 0.05$).

Comparisons between different treatments using the FISHER LSD test are summarized in table below.

Table 2: Comparisons between different treatments using the FISHER LSD test Probabilities less than 0.05 show significant differences (number of leaves sprayed)

	13 g/L (T_1)	15 g/L (T_2)	18 g/L (T_3)	21 g/L (T_4)	24 g/L (T_5)	27 g/L (T_6)	30 g/L (T_7)
13 g/L (T_1)		0,058	0,088	0,011	0,003	0,017	0,002
15 g/L (T_2)	0,058		0,786	0,296	0,088	0,425	0,058
18 g/L (T_3)	0,088	0,786		0,201	0,058	0,296	0,038
21 g/L (T_4)	0,011	0,296	0,201		0,425	0,786	0,296
24 g/L (T_5)	0,003	0,088	0,058	0,425		0,296	0,786
27 g/L (T_6)	0,017	0,425	0,296	0,786	0,296		0,201
30 g/L (T_7)	0,002	0,058	0,038	0,296	0,786	0,201	

Source: Personal work

Overall, no significant difference was found between T1, T2 and T3 treatments (p>0.05). But significant differences exist between the T4; T5; T6 and T7 treatments (p <0.05). Measurement of physicochemical parameters of each treatment before and after spraying is presented in table (3).

Table 3: Average value of physico-chemical parameters at each treatment before and after spraying

	Before spraying				**After spraying**			
	Salinity (‰)	**Temperature**	**Conductivity (µs/cm)**	**Ph**	**Salinity (‰)**	**Temperature (°C)**	**Conductivity (µs/cm)**	**pH**
T1	**0,37±0,16**	38,44± 0,33	690,15±17,6	4,04± 0,17	**1,48± 0,56**	37,79±0,14	2813,5±76,54	4,61±0,32
T2	**0,40± 0,23**	38,47±0,16	742,35±31,45	4,02±0,43	**1,56±0,32**	38,50±0,56	2950±32,67	4,25± 0,31
T3	**0,40± 0,62**	37,86±0,74	741,16±21,65	4,07± 0,23	**1,51±0,12**	38,52±0,62	2835±67,21	4,14± 0,34
T4	**0,33±0,54**	37,39± 0,15	616,5±12,78	4,03±0,54	**1,55±0,34**	38,73±0,23	2965±39	4,22±0,16
T5	**0,38± 0,17**	37,9± 0,76	709±23,5	4,03± 0,43	**1,60±0,12**	38,89±0,31	3060±47,54	4,14±0,21
T6	**0,39±0,21**	37,65± 0,62	730,55±10,76	4,03± 0,23	**1,79±0,23**	38,96±0,53	3485±56,32	4,34± 0,23
T7	**0,37±0,13**	39,19±0,43	688,3± 34,34	4,06± 0,54	**1,95±0,67**	39,16±0,19	3780±63,2	4,45±0,12

Source: Personal work

As water hyacinth is a plant subject to freshwater, we have considered particularly water salinity, especially after spraying, because significant increase of this paramater could impact the environment. According to this table, we noted after spraying a slight modification of the salinity resulting in 1,11; 1,15; 1,11; 1,21; 1,21; 1,39; 1,58 respectively for T1, T2, T3, T4, T5, T6 and T7 treatments.

The plants stayed in the buckets for seven days before spraying. Indeed, the station on which the tests were conducted is fed by a newly designed borehole whose water initially had a pH less than 5.

If despite these values water hyacinth plants were able to ensure their vital functions it confirms literature data that E. crassipes can tolerate large changes in water pH between 4 and 10 [10].

4. Discussion

4.1. Treatment of plants by salt adduction

At T0 and T1 treatments, no plant mortality was observed. These results are consistent with those obtained by [11] according to which water hyacinth is only able to survive under freshwater and oligosaline conditions (0 and 5 g / L).

If, despite the necrosis of certain leaves at the beginning, the plant has been able to survive and grow normally at 5 g/L, there is an adaptation of water hyacinth to this salinity dose. Salinity tolerance limits of water hyacinth vary from 0 to 5 g/L. Water hyacinth mortality was

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observed from T2 to T9 treatment. These results confirm the work of [11] who observed necrosis and mortality of hyacinth in mesosaline and polysaline conditions (5 and 18g/L and more). Similar research conducted in France revealed that hyacinth plants collected in southern France and cultivated at 9.2g/L and 13.7g/L salinity have all resulted in necrosis; from 9g/L, water hyacinth completely disappears from the medium [12]. At the level of T3, T4, T5, T6, T7, T8 and T9 treatments, it is at the fourth day that the mortality was observed. We can then conclude that at salinity levels between 13 g / L and 30 g / L, the time required for Eichhornia crassipes mortality is four (04) days. It is also the ideal time for salt to cross until causing necrosis of the leaves. In fact, during this period, symptoms such as leaf folding, softening of the stems and yellowing of the plant appear and announce necrosis. Indeed, salt disrupts the physiology of the plant. Necrosis and drying of the leaves prevent photosynthesis and respiration of plant, hence the mortality. Mortality began with daughter-plant, then young plant and finally adult plant. We can then remember that the sensitivity of water hyacinth to salinity is decreasing according to ontogeny; daughter plants are more sensitive to salinity than younger plants, which in turn are more sensitive than adult plant.

4.2. Treatment of plants by spraying

After two weeks of spraying all plants were decimated from T5 to T7 treatment. After spraying, there is a rapid drying of saline solution on the leaves through the sun. This drying of saline solution allows recrystallization of the salt which directly attacks the leaves. The latter harboring the respiratory and photosynthetic organs of the plant, their attack by salt disrupts physiology of the plant. It is at this moment that we observe symptoms as folding and yellowing which announce necrosis of the leaves and finally mortality of the plant. These results are consistent with the work of [13] who treated water hyacinth plants using copper sulphate and chelate, which are not selective herbicides that allowed after spraying, complete destruction of water hyacinth.

The slight changes in water salinity (Table 1) observed in these treatments show that small change is obtained at T5(24 g/L) treatment with an increase of 1.21 g/L compared to T6 (27g /L) and T7 (30g/ L) treatments which are respectively 1.39 g/L and 1.58 g/L. These results show that T5 treatment influences the environment less than T6 and T7 treatments.

Treatments T1 (13 g/L), T2 (15g/L), T3 (18g/L) and T4 still show persistent leaves after two weeks of spraying. These salinity rates don't allow effective control of water hyacinth because evergreen leaves can still allow the plant to come back to life. Thus, we can deduce that the effective salinity dose for the Eichhornia crassipes is obtained at T5 (24 g/L) treatment.

5. Conclusion

Water hyacinth is a problematic invasive plant worldwide. Its propagation in fresh water is very fast and forms a dense carpet that causes enormous socio-economic, environmental and even sanitary damage.

The results of our studies confirm that salt is a natural enemy for water hyacinth Eichhornia crassipes. Treatment of water hyacinth by saline is an effective control method that can be used to significantly reduce hyacinth carpets in water surface. As a result of our work, minimum salinity dose that can be used to remove water hyacinth plants from spray is 24 g/L.

As water yacinth is an floating plant the realization of this technique in natural environment can't give the certainty to spray the same plant over a period of two weeks. Thus, experiments resumption based on the reduction of the number of treatment days and the variability of spraying frequency would be an adequate technique to combat proliferation of this plant. Moreover, after spraying of the NaCl solution, a study based on physiological aspect of the plant opposite to NaCl solution appears to be essential; which can bring out the mechanisms by which NaCl could destroy the plant.

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