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Influence of Mercuric Chloride on Survival and Suitability for *In Vitro* Regeneration of Three Sweet Potato Landraces (*Ipomoea batatas* L.) Produced in Benin

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Abstract

Ipomoea batatas L. is tuberous root plant of great nutritional and economic importance in Benin. *In vitro* culture of this plant allows in a short period time, to produce a planting material through the establishment of efficient disinfection and micropropagation protocol. This work aims to study the survival and *in vitro* regeneration of three sweet potato landraces ("Dokoun", "Vobodouaho" and "Djlodou"). Four doses of mercuric chloride (C1: 0.05%, C2: 0.1%, C3: 0.5%, C4: 1%) were tested for disinfection and two Murashige and Skoog (MS) media, M0 (MS without growth regulator) and M1 (MS + 1mg/l of BAP + 0.01 mg/l of NAA) was used to evaluate the regeneration of the plantlets. The results showed that 0.5% mercuric chloride had the best survival rate (83.33% for "Djlodou", 61.08% for "Vobodouaho" and 52.75% for "Dokoun"). Of the two media used, M0 has more adapted to landraces "Vobodouaho" and "Dokoun" while M1 contributed to a better development of the landraces "Djlodou".

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Introduction

Sweet potato (*Ipomoea batatas* L.), family of Convolvulaceae, is the seventh largest crop in the world and the fifth crop in developing countries after rice, wheat, maize and cassava with an annual production of 104 MT (FAO, 2013). The tubers and leaves were the most important parts in are human and animal food and in industry (Doliński and Olek, 2013). Color flesh sweet potato varieties contain large amounts of β-carotene, vitamin A precursor (Adelia, 2007). Sweet potato has an interesting agronomic capacities and a wide climatic and edaphic adaptation for several varieties (Lebot et al., 2009). This constitutes major asset to deal with the challenge of food security in a global climate change context (Glato et al., 2014). In sub-Saharan Africa particulary in Benin, sweet potato is the third tuber crop after yam and cassava. It is an important source of income for the producers and is used during the lean season (Paraïzo et al., 2013). It is existing in some

landraces rich in vitamin A and used for the manufacture of infant flours capable of combating malnutrition in children (Sanoussi et al., 2013 and 2016). Alongside these advantages of sweet potatoes, it should be noted that its culture in African countries of which Benin is confronted with several abiotic and biotic constraints. Among other abiotic constraints noted in Benin, the insufficiency of planting material performant varieties, the decline of productivity, the irregularity of rains, the pests and diseases. Indeed, the work carried out by Zinsou et al., 2010 showed the existence of several fungal and viral diseases. The most known are Alternaria caused by *Alternaria bataticola* and moderate marbling which is a viral disease called *Sweet Potato Mild Mottle Virus* (SPMMV). Concerning the seed system, it is purely traditional and remains informal, unstructured with seeds of lower phytosanitary qualities than in other African countries such as Burkina Faso, Rwanda and Uganda where there existing the specialized structures in the production and distribution of improved and healthy sweet potato seeds (Gibson et al., 2009). The using the of previous season cuttings, traditional way of production or acquisition of planting material, favors transmission of the virus to the next generation and cause an annual yield losses varying between 50% and 98% (Ngailo et al., 2013). This sometimes results an abandonment followed by the disappearance of certain sweet potatoes landraces. Faced with this finding, *in vitro* culture techniques (cell culture or plant tissue) are an important way to produce healthy planting material in quantity and free to viruses, bacteria, and nematodes (Lassois et al., 2009), but also to preserve *ex situ* the local genetic diversity. In Benin, *in vitro* culture techniques are already successfully used for sanitation, micropropagation and *ex situ* conservation of certain root and tuber crops including yams (Ahanhanzo et al., 2010; Agbidinoukoun et al., 2013) and cassava (Cacaï et al., 2013). However, since the work of Houndonougbo (1989) on the influence of different concentrations of indole-3-acetic acid, naphthalene acetic acid, 2,4-dichlorophenoxyacetic acid and kinetin on callogenesis and *in vitro* organogenesis of inter-node fragments of two sweet potatoes landraces, no work has been reported on the *in vitro* culture of sweet potato in Benin as part of the improvement of the seed system. For the establishment of *in vitro* culture, bacteria and fungi are a cause of contamination of explants. The success of *in vitro* culture therefore requires adequate disinfection of the explant using chemicals such as sodium hypochlorite, mercury lauryl and mercuric chloride. The use of this last proved more effective for the disinfection of the woody species like teak, herbaceous plants such as yams (Ahanhanzo et al., 2008 and 2010) and sweet potatoes (Hammond et al., 2014). Thus, the development of an effective disinfection protocol will, in a long term, reduce the cost of producing plantlets for better accessibility to producers.

The present work is part of the optimization of productivity by the micropropagation of three sweet potato landraces. The present study has been aimed to determine (1) the optimal dose of mercuric chloride for better survival of the explants, (2) to analyze the *in vitro* regeneration capacity of three sweet potatoes landraces on two different media.

Materials and methods

Plant material and production of mother plants

The plant material for this study constituted three sweet potato landraces (Djlodou, Vobodouaho and Dokoun) collected from the Department of Oueme during September 2015. "Dokoun" with white skin and yellow flesh coming from the Municipality of Dangbo; "Vobodouaho" with white skin and white flesh coming from the Municipality of Adjohoun and "Djlodou" with red skin and white flesh coming from the Municipality of Bonou. These sweet potato landraces were selected on the basis of the availability of their cuttings as seed and their tubers which are the most produced. Cuttings of the three sweet potato landraces were grown in polyethylene pots filled with sterilized soil. The pots were maintained under greenhouse of Genetic and Biotechnology Department of Central Laboratory of Plant Biotechnology and Breeding Plant (LCBVAP) of the Faculty of Sciences and Techniques (FAST) of the University of Abomey-Calavi (UAC). Young shoots were then obtained as mother plants after two months of culture.

Culture media

For the study of the influence of mercuric chloride on the survival of plantlets of selected landraces of sweet potato, the culture medium used was Murashige and Skoog (MS) (1962) without growth regulator. The study relative to the aptitude for *in vitro* regeneration, two culture media were used. The MS without a growth regulator (Glato et al., 2014) serving as a control and the MS supplemented with Benzylaminopurine (1mg/l) and α-Naphthalene Acetic Acid (0.01 mg/1) (Sivparsad and

Gubba, 2012). The pH of the media was adjusted to 5.7 ± 0.1. The media supplemented with 30 g/l of sucrose and 8 g/l of agar were distributed in tubes and then sterilized for 15 minutes in autoclave at 121°C and transferred to the culture chamber.

Disinfection of explants

For *in vitro* initiation, the disinfection protocol applied was that used by Ahanhanzo et al. (2008) on the yam with some modifications. The explants taken from the mother plants of the three varieties after eight weeks in the greenhouse were fragments of uninodal stems. The fragments were cleared of their leaves and rinsed with tap water. They were then immersed in 70% alcohol for 1 minute, then dipped into solutions of mercuric chloride containing two drops of tween 80 at different concentrations (C1 : 0.05% ; C2 : 0.1% ; C3 : 0, 5% ; C4 : 1%). The immersion time used was 10 minutes. The explants were rinsed three times with sterilized distilled water for five minutes per rinsing.

Culture and *in vitro* culture conditions

The disinfected explants were rid of their extremities necrosed by a scalpel. Each explant of 3 cm to 4 cm was placed in a test (25 mm × 150 mm) tube containing the medium. Each tube was sealed with sterile cotton and sealed with parafilm. The seeded media were placed in the culture chamber at 27±1°C. The chamber was subjected to a photoperiod of 12 hrs per day under a light intensity of 5000 lux provided by Philips TLD18W and Sibalec lamps. The relative humidity was maintained at 80%.

Evaluation parameters

For the study of the influence of mercuric chloride on the survival of plantlets, daily observations on the explants initiated were made for 30 days. Infection, necrosis and survival rates were estimated. These parameters were reported daily after seeding to evaluate the effectiveness of the disinfection protocol. For the study of the capacity in the *in vitro* regeneration, after inoculation, observations were made every two days over 30 days. The formation of the roots, the number of nodes and leaves formed were evaluated.

Statistical analysis

The experimental device used in establishing the *in vitro* culture was a completely random block. In each treatment, twelve (12) explants were seeded with three replicates. To evaluate the influence of four doses of mercuric chloride on survival explant and the regenerative capacity of the three sweet potato landraces, an analysis of variance (ANOVA) was realized. The RYAN-JOINER and LEVENE tests were previously applied to verify the normality and the equality of the variances. The Student-Newmann-Keuls test at the 5% threshold was also used to compare the averages. Since the "necrosis" variable did not satisfy the requirements of a parametric test, the Kruskal-Wallis test was used. Statistical analyzes of the results were carried out using GenStat software (V9.2.0.153).

Results

Influence of mercuric chloride on the survival of plantlets

The use of different concentrations of mercuric chloride on the survival of plantlets made it possible to observe some explants infected or necrosed while others proved to be healthy and survived. The analysis of variance for infections (Table 1) shows that the difference is highly significant (p<0.001) between the varieties on the one hand and the different treatments on the other hand. The interaction between "Varieties and Treatments" is significant (p<0.05).

Table 1. Results of the analysis of variances of infections.

Source of variation	Degree of freedom	Sum of squares	Average squares	Variance	Probability
Varieties	2	43.1667	21.5833	22.20	<.001
Treatments	3	542.8889	180.9630	186.13	<.001
Varieties × Treatments	6	24.6111	4.1019	4.22	0.005
Error	24	23.3333	0.9722		
Total	35	634.0000			

Fig. 1 presents the percentages of infections of three landraces as a function of four doses of mercuric chloride. The dose of 0.05% mercuric chloride allowed the highest infection rates to be recorded, while the 1% yielded the lowest infection rates for all varieties. Furthermore, the dose of 0.05% of mercuric chloride involved more infection for the variety "Vobodouaho" (94.41%) compared to the variety "Dokoun" (80.58%) and the variety "Djlodou" (69.41%). Similarly, the 1% mercuric chloride dose resulted in no infection with the varieties "Djlodou" and "Dokoun" compared to the variety "Vobodouaho" (5.58%).

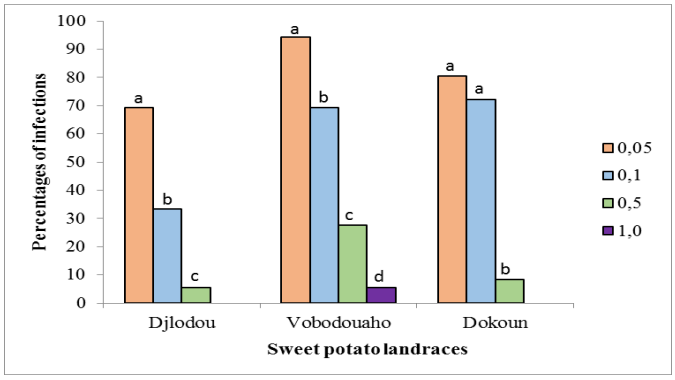


Fig. 1: Comparison of the percentage of infections of three sweet potato landraces as a function of four doses of mercuric chloride. Histograms with the same letter are not significantly different according to the Student, Newman and Keuls averaging test at the 5% threshold.

After disinfection of explants, cases of necrosis were observed. Statistical analysis shows that there is no significant difference between local varieties (p=0.766). However, the difference is highly significant (p<0.001) between doses of mercuric chloride.

Fig. 2 presents the percentages of necroses of three sweet potato landraces as a function of four doses of mercuric chloride. The dose of 1% mercuric chloride produced the most necrosis compared to the dose of 0.05% which resulted in less or no necrosis for three landraces. Moreover, the highest rate of necrosis was recorded for the variety "Dokoun" with 1% mercuric chloride whereas 0.05% mercuric chloride did not cause necrosis in the varieties "Djlodou" and "Dokoun".

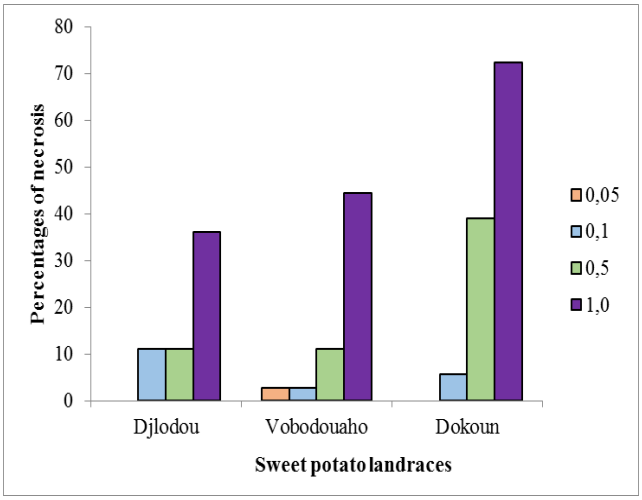


Fig. 2: Comparison of the percentage of necrosis of three sweet potatoes landraces as a function of four doses of mercuric chloride.

For the evaluation of survival compared with different doses of mercuric chloride, explants neither infected nor necrosed were considered. Analysis of variance (Table 2) showed a significant difference (p<0.001) between the varieties and between the different treatments. Similarly, the difference is significant (p<0.05) for the interaction between the two factors "varieties × treatments".

Table 2. Results of the analysis of variances of survivals.

Source of variation	Degree of freedom	Sum of squares	Average squares	Variance	Probability
Varieties	2	68.3889	34.1944	39.71	<0.001
Treatments	3	158.3333	52.7778	61.29	<0.001
Varieties × Treatments	6	20.5000	3.4167	3.97	0.007
Error	24	20.6667	0.8611		
Total	35	267.8889			

Fig. 3 shows the percentages of survival of three landraces as a function of four doses of mercuric chloride. The concentration of 0.05% mercuric chloride gave the lowest percentages of survival regardless of the variety. However, the best survival percentages were obtained with 0.5% mercuric chloride. Besides, the variety "Djlodou" obtained the highest percentage of survival compared to the other varieties for 0.5% mercuric chloride.

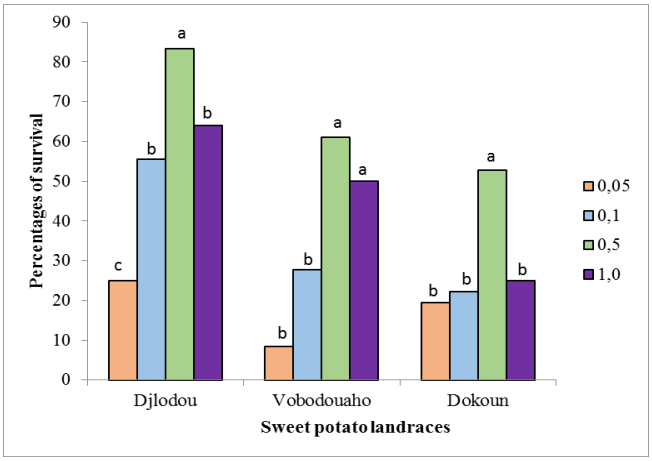


Fig. 3: Comparison of the percentage of three sweet potatoes landraces as a function of four doses of mercuric chloride. Histograms with the same letter are not significantly different according to the Student, Newman and Keuls averaging test at the 5% threshold.

In vitro regeneration capacity of explants

The growth of sweet potato plantlets was evaluated on a control medium M0 (medium MS without growth regulator) and a medium M1 (medium MS + 1mg/l BAP + 0.01 mg/1 NAA). The formation of roots, node and leaves are the parameters evaluated. Table 6 makes a synthesis of the different averages as a function to the parameters considered and the varieties. Fig.4 shows plantlets of the three sweet potato landraces grown in Benin.

Roots

The analysis of variance (Table 3) of "Varieties" and "Medium" factors gives a significant difference in the number of roots formed. The interaction between these two factors is highly significant (p<0.001). The control medium M0 yielded better results for the varieties "Dokoun" and "Vobodouaho" with respectively the averages of 1.47 and 2.09 of roots formed against respectively 0.69 and 1.01 for the medium M1. At the level of the variety "Djlodou", it was noted that the medium M1 which gave the highest average of roots formed (1.88) compared to the control medium M0 (1.45). The Student, Newman and Keuls averaging test at the 5% threshold shows a difference between the M0 and M1 media for the varieties "Dokoun" and "Vobodouaho" while no differences were observed between these two media for the variety "Djlodou" (Table 6).

Nodes

The analysis of variance (Table 4) of the factors "Variety" and "Medium" gives a highly significant difference (p<0.001) for the number of nodes formed. The interaction between these two factors is also highly significant (p<0.001). The varieties "Djlodou" and "Vobodouaho" had the highest average of node with the medium M1 (9.63 and 3.77 respectively) and the control medium M0 (respectively 3.38 and 4.14). The variety "Dokoun" obtained the lowest average of nodes with the media M0 and M1 (respectively 2.83 and 3.20). It has been observed that the production of nodes for the variety "Djlodou" has almost multiplied by three (03) for the medium M1 compared to the medium M0 (Table 6). The test of classification of Student, Newman and Keuls at the 5% threshold shows that there is no difference between the M0 and M1 media for the varieties "Dokoun" and "Vobodouaho". A difference was observed between these two media for the variety "Djlodou".

Table 3. Results of root analysis of variance.

Source of variation	Degree of freedom	Sum of squares	Average squares	Variance	Probability
Varieties	2	1.1527	0.57635	9.8484	0.003
Medium	1	1.02245	1.02245	17.4711	0.001
Varieties × Treatments	2	1.91123	0.955617	16.3291	<0.001
Error	12	0.702267	0.0585222		
Total	17	4.78865			

Table 4. Results of nodes analysis of variance.

Source of variation	Degree of freedom	Sum of squares	Average squares	Variance	Probability
Varieties	2	39.1834	19.5917	122.815	<0.001
Medium	1	19.5938	19.5938	122.828	<0.001
Varieties × Treatments	2	39.5247	19.7623	123.885	<0.001
Error	12	1.91427	0.159522		
Total	17	100.216			

Table 5. Results of leaves analysis of variances.

Source of variation	Degree of freedom	Sum of squares	Average squares	Variance	Probability
Varieties	2	31.1234	15.5617	115.405	<0.001
Treatments	1	11.9235	11.9235	88.4239	<0.001
Varieties × Treatments	2	28.741	14.3705	106.571	<0.001
Error	12	1.61813	0.134844		
Total	17	73.406			

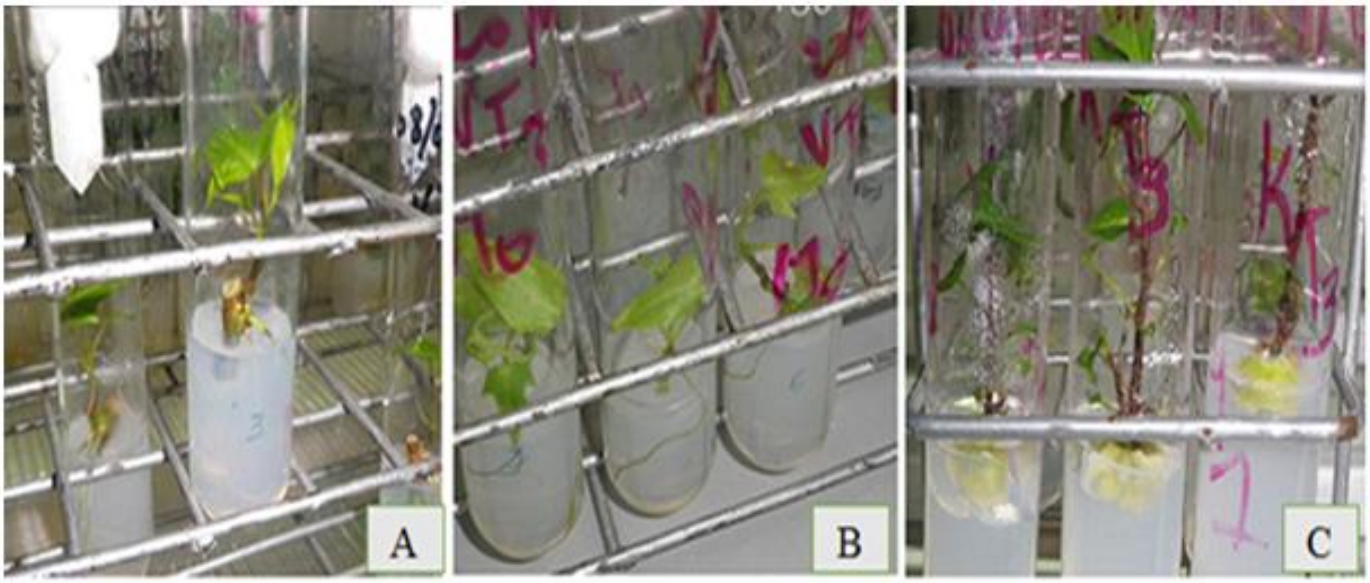
Leaves

Analysis of variance (Table 5) of "Variety" and "Medium" factors gives a highly significant difference (p<0.001) for the number of leaves formed. The interaction between these two factors is also highly significant (p<0.001). The variety "Djlodou" yielded an average of leaves production (6.75) four (4) times higher on the medium M1 compared to the averages (1.60) of the MS medium without regulator (M0). At the level of the variety "Dokoun", it was the medium M0 that gave the best average of leaves (1.4) compared to the average (0.72) of the medium MS with regulators (M1). As for the variety "Vobodouaho", it has given averages of 2.11 and 1.7 for the media M1 and M0 respectively. The Student, Newman and Keuls average ranking test at the 5% threshold shows that there is no difference between the M0 and M1 media for the variety "Vobodouaho". A difference was observed between these two media for the varieties "Djlodou" and "Dokoun" (Table 6).

Table 6. Effect of M0 and M1 media on the formation of Roots, Nodes and Leaves after 30 days of culture.

Parameters and Media		Average of roots		Average of nodes		Average of leaves	
		M0	M1	M0	M1	M0	M1
Varieties	Djlodou	1.45 ± 0.04a	1.88 ± 0.11a	3.38 ± 0.21a	9.64 ± 0.40b	1.60 ± 0.14a	6.75 ± 0.23b
	Vobodouaho	2.09 ± 0.19a	1.02 ± 0.14b	4.14 ± 0.20a	3.77 ± 0.15a	1.70 ± 0.24a	2.11 ± 0.07a
	Dokoun	1.48 ± 0.19a	0.69 ± 0.07b	2.83 ± 0.09a	3.20 ± 0.18a	1.40 ± 0.33a	0.73 ± 0.13b

The averages in the boxes were compared by the Student, Newman and Keuls averaging test at the 5% threshold. M0 = MS without regulators; M1= MS + 1 mg.l^{-1} BAP + 0.01 mg.l^{-1} NAA.



A- Plantlets of local variety "Djlodou" on the medium M1

B- Plantlets of local variety "Vobodouaho" on the medium M0

C- Plantlets of local variety "Dokoun" on the medium M0

Fig. 4: Plantlets of the three sweet potato landraces grown in Benin.

Discussion

The application of different doses of mercuric chloride to disinfect the explants of three sweet potatoes landraces cultivated in Benin allowed obtaining different results. Indeed, the results of Figs. 2 and 3 have shown that infection and necrosis are the main problems whose control is indispensable for establishment of to *in vitro* culture as demonstrated by Ahanhanzo et al.(2008) on yam and Hammond et al. (2014) on sweet potato. These observations revealed that the explants were infected with fungi and bacteria. Omamor et al. (2007) and Hammond et al. (2014) have shown that the contamination by fungi and bacteria can compromise *in vitro* culture. The results showed that, more the concentration of mercuric chloride (0.05%, 0.1%, 0.5% and 1%), more the decrease of average infection and more the increase of average necrosis in all the three varieties. One per cent (1%) concentration was effective for explant disinfection for 10 min immersion, however, it induced a high rate of necrosis. The work of Hammond et al. (2014) showed that the concentrations of 0.4% and 0.6% mercuric chloride for the same duration were not effective to control the infections. The concentration of 1% $HgCl_2$ was considered effective for the disinfection of sweet potatoes explants according to the same author. This result could be explained by the conservation conditions of mother plants before sampling the explant and the genotypes of the plants. Indeed, the statistical analysis showed a significant difference in the interaction of treatment-genotype. This shows that the effect of the treatment and that of the genotype can not be dissociated. The expression of the treatment effect depends on the genotype and vice versa. The percentage of survival is the most important parameter to be considered into account identify the most suitable mercuric chloride concentration for good disinfection (Ahanhanzo et al., 2008).

Uninfected and non-necrotic explants are those which developed and whose percentages were considered for the survival. The average of survival is proportional to the concentration of mercuric chloride. The best average of survival is obtained with 0.5% of $HgCl_2$ for the three sweet potatoes landraces (83.33% for "Djlodou", 61.08% for "Vobodouaho" and 52.75% for "Dokoun"). This difference of survival average between the varieties may be genotypic origin. Hammond et al. (2014) and Doukouré (2000) also used 1% mercuric chloride to have high survival rates in several sweet potato and yam genotypes respectively. It is important to note, however, that most authors prefer to use sodium hypochlorite which is less toxic to the explants (Sivparsad and Gubba, 2012 on sweet potato). However, these authors have acknowledged that with sodium hypochlorite, internal infections of the plant material can not be eliminated by superficial disinfection. This is because the degree of disinfecting power of sodium hypochlorite depends on the quantity of free chlorine and pH.

The introduction in *in vitro* culture of the sweet potato landraces allowed to compare the control medium M0 (medium MS without growth regulator) with the medium M1 (medium MS + 1mg/l of BAP + 0.01 mg/l of NAA). Thus, for root formation, the results showed that the control medium M0 is better than the M1 medium for the three varieties. In the obtaining of nodes, the medium M1 is more adapted to "Djlodou" while the Medium M0 is more suitable for "Vobodouaho" and "Dokoun". For the leaves, "Vobodouaho" and "Dokoun" gave a good result with the M0 where as for "Djlodou" the medium M1 is more efficient than the medium M0. These results show that for the same parameter, the hormonal balance allowing the best result varies from one cultivar to another. This could be explained by the presence of endogenous auxins which interferes specifically with these varieties. Similar kind of results have also been observed by Sivparsad and Gubba (2012) who showed that cytokinins and auxins interact to produce different morphological responses depending on the endogenous concentration of hormones and the ratio of the concentration of exogenous hormones (cytokinin and auxin). The results obtained for the *in vitro* regeneration of the three sweet potatoes landraces show that the response to the medium depends on the variety and that for each variety a specific treatment must be done. This is in agreement with the work of Triqui (2009) and Sivparsad and Gubba (2012) where they noted regeneration dependence on genotypes.

Conclusion

The results obtained in the present study helped to identify the mercuric chloride with a concentration of 0.5% as the optimal concentration which allows efficient disinfection and the survival of three sweet potato landraces cultivated in Benin. Furthermore, for the ability to regenerate *in vitro*, the response to the culture medium has varied according to the three varieties. The MS medium without growth regulator is more suitable for "Vobodouaho" for all the parameters considered. The opposite effect was observed for the local variety "Djlodou".

Conflict of interest statement

Authors declare that they have no conflict of interest.

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References

Adelia, C.B., 2007. Sweet potato a review of its past, present and future role in human nutrition. J. Agric. Food Chem. 50(6), 56-60.

Agbidinoukoun, A., Ahanhanzo, C., Adoukonou-sagbadja, H., Adjassa, M., Agassounon Djikpo-Tchibozo, M., Agbangla, C., 2013. Impact of osmotic dehydration on the encapsulated apices survival of two yams (*Dioscorea* spp.) genotypes from Benin. J. Appl. Biosci. 65, 4999-5007.

Ahanhanzo, C., Agbangla, C., Dangou, J., Toukourou, F., Montcho, D., 2008. Influence du chlorure mercurique et de la cytokinine sur la survie et la morphogenèse *in vitro* d'explants de différents génotypes d'ignames (*Dioscorea* spp.). Ann. Sci. Agron. 11(1), 33-47.

Ahanhanzo, C., Gandonou, C. B., Agbidinoukoun, A., Dansi, A., Agbangla, C., 2010. Effect of two cytokinines in combination with acetic acid Q-naphthalene on yams (*Dioscorea* spp.) genotypes' response to *in vitro* morphogenesis. Afr. J. Biotechnol. 9(51), 8837-8843.

Cacaï, G., Ahanhanzo, C., Adjanohoun, A., Houédjissin, S., Azokpota, P., Agbangla, C., 2013. Hormonal influence on the *in vitro* bud burst of some cassava varieties and accessions from Benin. Afr. J. Biotechnol. 12(13), 1475-1481.

Doliński, R., Olek, A., 2013. Micropropagation of sweet potato [*Ipomoea batatas* (L.) Lam.] from node explants. Acta Sci. Pol. Hortorum Cultus. 12(4), 117-127.

Doukouré, S., 2000. Amélioration de la production de l'igname par bouturage *in vitro*, chez les cultivars Florido et Brazo fuerte de *D. alata* L. Thèse de Doctorat Ingénieur, Université de Cocody, Côte d'Ivoire. 123p.

FAO, 2013. FAO Statistical Databases. Food and agriculture organization of the United Nations. http://faostat.fao.org

Gibson, R.W., Mwanga, R.O., Namanda, S., Jeremiah, S.C., Barker, I., 2009. Review of sweet potato seed systems in East and Southern Africa. International Potato Center (CIP), Lima, Peru. Integrated Crop Management Working Paper. 58p. http://cipotato.org/wp-content/uploads/2014/08/004730.pdf

Glato, K., Aïdam, A., Odah, K., Tozo, K., Attoh-Mensah, M., Etse, K., 2014. Régénération *in vitro* par organogenèse directe de pousses à partir de boutures de trois cultivars de patate douce (*Ipomoea batatas*) originaire du Togo. Eur. Scient. J. 10(27), 276-291.

Hammond, R., Buah, J.N., Asare, P.A., Acheampong, S., 2014. Optimizing sterilization condition for the initiation of sweet potato (*lpomoea batatas*) culture *in vitro*. Asian J. Biotechnol. 6(2), 25-37.

Houndonougbo, B., 1989. Influence de différentes concentrations d'acide indole-3-acétique, d'acide naphtalène-acétique et de kinétine sur la callogenèse et l'organogenèse *in vitro* de fragments d'entre-nœuds de deux variétés de patate douce (*Ipomoea batatas* L.). Agronomie. 9, 653-660.

Lassois, L., Busogoro, J. P., Jijakli, H., 2009. La banane: de son origine à sa commercialisation. Biotechnol. Agron. Soc. Environ. 13(4), 575-586.

Lebot, V., Champagne, A., Malapa, R., Shiley, D., 2009. NIR determination of major constituents in tropical root and tuber crop flours. J. Agric. Food Chem. 57, 10539-10547.

Murashige, T., Skoog, F., 1962. A revised medium for rapid growth and bio assays with tissue culture. Physiol. Plantarum. 15, 473-497.

Ngailo, S., Shimelis, H., Sibiya, J., Mtunda, K., 2013. Sweet potato breeding for resistance to sweet potato virus disease and improved yield : progress and challenges. Afr. J. Agric. Res. 8(25), 3202-3215.

Omamor, I.B., Asemota, A.O., Eke, C.R., Eziashi, E.I., 2007. Fungal contaminants of oil palm tissue culture in Nigeria Institute for Oil Palm Research (NIFOR). Afr. J. Agric. Res. 2, 534-537.

Paraïzo, A., Tokoudagba, S.F., Sanni, A., Olodo, P.G., Yegbemey, R.N., Gutmetzoe, M., 2013. Sweet potato (*Ipomoea batatas* L.) production determinants in North-East Benin: The commune of Gogounou as a case in point. Int. J. Sci. Adv.

Technol. 3(1), 14-20.

Sanoussi, A. F., Dansi, A., Ahissou, H., Adebowale, A., Sanni, L.O., Orobiyi, A., Dansi, M., Azokpota, P., Sanni, A., 2016. Possibilities of sweet potato [*Ipomoea batatas* (L.)] value chain upgrading as revealed by physico-chemical composition of ten elites landraces of Bénin. Afr. J. Biotechnol. 15(13), 481-489.

Sanoussi, A., Dansi, A., Bokossa-yaou, I., Dansi, M., Egounlety, M., Sanni, L.O., Sanni A., 2013. Formulation and biochemical characterization of sweet potato (*Ipomoea batatas*) based infant flours fortified with soybean and sorghum flours. Int. J. Curr. Microbiol. Appl. Sci. 2(7), 22-34.

Sivparsad, B. J., Gubba, A., 2012. Development of an efficient plant regeneration protocol for sweet potato (*Ipomoea batatas* L.) cv. Blesbok. Afr. J. Biotechnol. 11(84), 14982-14987.

Triqui, Z. A., 2009. Contribution à l'amélioration de la patate douce [*Ipomoea batatas* (Lam.)] par application des biotechnologies : Embryogénèse somatique et transformation génétique. Thèse de Doctorat d'Etat Université Mohammed V-Agdal, Faculté des Sciences, Rabat. 143p.

Zinsou, V., Paraïso, A., Thomas-Odjo, A., Ahohuendo, B., 2010. Identification des principaux agents pathogènes de la patate douce (*Ipomoea batatas* Lam.) au Nord du Bénin. Ann. Sci. Agron. 14(2), 241-255.

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