

Adaptation of plants derived from cultivated yam of *Dioscorea cayenensis*-*D. rotundata* complex species seeds germination in two agroecological zones of Benin

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Abstract

Vegetative propagation of plants promotes the accumulation of viruses in plant material; this causes the loss of vigor and consequently the drop of vegetable yield. Keep up productivity level of vegetatively propagated plants; it is therefore important to regenerate genetic material by sexual reproduction to improve the biodiversity. The main objective of this study is to improve yam seeds by sexual way, specifically to assess the response of seedlings from yam seeds in two agroecological areas, area IV (Djougou), in Sudanese climate and area V (Bantè), in transition climate. Seedling were transplanted in this agroecological areas using a completely Randomized design with three replications. Data was analyzed using one way ANOVA at 5% level of significance and a mean comparison test. Seedling survival rate is significantly different, 55.41% in zone IV and 23.60% in zone V, but the rate of tuberization of the two areas is similar, 55.63%; of zone IV and 57.62%; of zone V. The average of the tubers length and their weight are respectively 13.21 cm and 24.87 ± 19.22 g in zone IV and 8.95 cm and 10.30 ± 9.91 g in zone V. This results show that microtubers produced from the seeds show a high degree of heterogeneity, and, varieties creation and improvement is possible from the seeds. This technique would promote the selection of varieties adapted to current climatic conditions.

Keywords: Yam seed; Germination; Agroecological zone; Microtuber; Selection

1. Introduction

Yam's place in the tropical zone is justified by the climatic and edaphic conditions. Climatic variation in general and especially drought have negative impacts on several components, which condition success of agricultural production in general, and mainly of yam [1, 2, 3, 4], occasion loss of yam's diversity and yields drop. Moreover, degradation of soil fertility which occasion around 3% of drop of yield in the world [5], thus affecting food self-sufficiency [6, 7]

However, the quality of the seed favors the adaptation of plants to these new conditions. For food security and sovereignty, it is therefore urgent to compensate diversity loss and yield decrease-installing varieties adapted to different climatic and soil conditions.

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Preserve food security and sovereignty, need to have good quality of seed, which depend plants adaptation of environment variation. Sexual way can carry out these kinds of seed. Yams of the *Dioscorea cayenensis-D. rotundata* species complex produce fruit and seeds, able to sprout after sowing [8, 9, 10, 11, 12, 13]. In North of Benin [14], in North of Côte d'Ivoire [15] and in center of Benin [16], this authors shown that in each yam's field around 4.800 to 27.000 seeds is yield in average each year.

Farmer integrated tuber of seedling from seed germination in nature in their cultivated gene pool unconsciously [17, 18, 19]. Moreover, studies of germination of yam seeds from farmer's field have shown germination rates ranging from 30 to 80% [8, 11, 20, 21]. Improve genetic diversity of the yam by sexual reproduction require to observe the behavior of the seedlings resulting of yam's seeds germination. This study aims to assess the response of seedlings produced from yam seeds in two agroecological zones of Benin.

2. Material and methodology

2.1. Material

Vegetal material is seedlings resulting from the germination of seeds collected from ten (10) cultivated yam, in the fields of farmers in three (03) Municipalities of Benin (Table 1).

Table 1 Characteristics and origins of the female parents of the seeds collected.

Name	Cycles	Villages	Municipalities
Dodo A	Late	Agoua	Bantè
Katala	Intermediate	Agoua	Bantè
Mafobo	precocious	Agoua	Bantè
Ewourou	Intermediate	Agoua	Bantè
Agatou	Intermediate	Agoua	Bantè
Gnidou	Intermediate	Adjiro	Bassila
Dodo I	Late	Idaho	Dassa
Kpakala	Late	Idaho	Dassa
Adigbili	Precocious	Idaho	Dassa
Moroko	precocious	Soclogbo	Dassa

2.2. Methodology

2.2.1. Study environment

The seeds germination was implement on the experimental site of the Department of Genetics and Biotechnologies of the University of Abomey-Calavi, located in the agro-ecological zone VI (Nitisols land area), at 15 km in north of Cotonou (6°30' N and 2°24' E). Its climate is of the subequatorial type with a bimodal rainfall regime. The mean annual precipitation is 1,168.3mm and the mean annual temperature is around 27.75 °C, with an amplitude of 4 °C. Temperature extremes are recorded in March for the high and in August for the weak [22].

The seedlings from yam's seeds were transplanted into two (2) agro-ecological zones; agro-ecological zones IV and V described by the National Action Program for Adaptation to Climate Change of Benin [23] as zones offering propice climatic conditions of yams growing. The first site is located in Kpayerou in the Municipality of Djougou (agro-ecological zone IV, West-Atacora zone), with a Sudanese-type climate and the second in Agoua in the Municipality of Bantè (agro-ecological zone V, cotton zone of Center-Benin) with Sudano-Guinean climate (Figure 1, Figure 2).

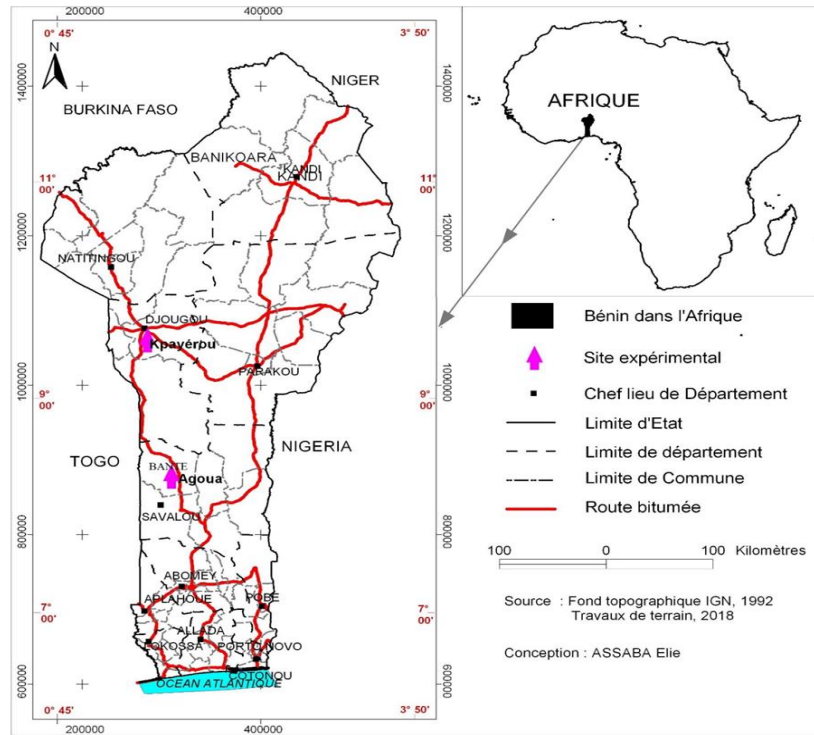


Figure 1 Map of republic of Benin in West Africa

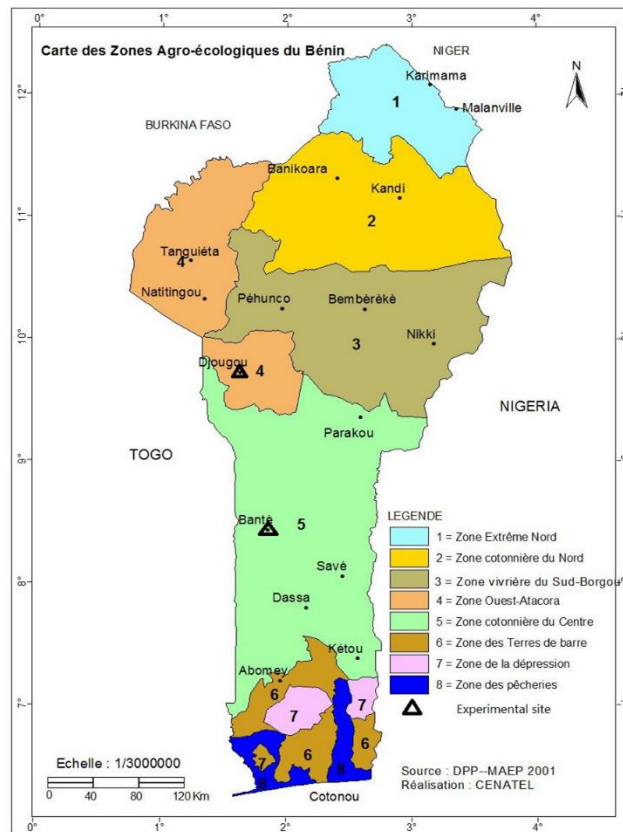


Figure 2 Map of Benin showing the two experimental sites in two different agroecological zones

2.3. Sampling of producers

The farmer's selection criteria are based on their knowledge and expertise of yam cultivation, being voluntary and available to follow the seedlings in the field and access to the field.

2.4. Seeds collection and transplantation

The fruits collected in the farmers' fields in December 2015 were sundried for two weeks. After dry, the seeds were extracted and stored at room temperature. In February 2016, the seeds extracted from dry fruits were sown in tank of 50 cm long, 25 cm wide and 15 cm deep, filled with mould. In each tank, 30 seeds are sown, tanks are been order by a completely randomized design with three replications per cultivar [24], germinations are observed after two (02) weeks and this up to eight (08) weeks (Figure 3a).

Seedling are been transferred to tanks on pot (Figure 3b), and three weeks after, 582 seedlings were throwed to two producer's fields, respectively 265 seedlings in agro-ecological zone IV and 317 seedlings in agro-ecological zone V. In the two agro-ecological zones, the seedlings were transplanted in a completely randomized design with three replications per cultivar.

Each block is made up of 10 rows of mounds, and each row is made up of ten mounds (agro-ecological zone IV) or a ten-hole ridge (agro-ecological zone V) according to the cultivation techniques adapted to each agro-ecological zone and the know-how of concerned Farmers. The spacing between the rows is 80 cm and between the mounds or pockets are 60 cm apart. In each mound or pocket, a single seedling is transplanted there (Figure 2c). Cultivation in the different agro-ecological zones was carried out in July 2016.



Figures 3a to 3d Different stages of yam development: (a) Yam seed, (b) yam seedlings at germination stage, (c) yam seedlings transplanted in pots, (d) yam seedling of two (02) months age after transplantation

2.5. Data collection

On each experimental site, two (02) months after transplantation, the number of survival seedlings was noted per cultivar (hybrid descendant), and the number of seedlings which have tuberized is determined.

At the end of the vegetative cycle, the tubers were harvested and ten morphological parameters were observed. These are three (03) quantitative parameters (weight, length and circumference) and seven (07) qualitative parameters (shape of the tuber, color of the skin of the tuber, appearance of the tuber, presence or not of rootlets on the tubers, location of the rootlets on the tubers, presence or not of stripes and presence or not of ramified on the tubers).

2.6. Data analysis

The survival or recovery rate which is the ratio of the number of seedling which having complete their vegetative cycle by the total seedling transplanted was calculated, and the rate of tuberization which is the ratio between the number of seedlings having given a tuber and the total number of seedlings having completed their vegetative cycle. The variation in the rate of recovery and tuberization were subjected to an analysis of variance (ANOVA) according to the Generalized Linear Model (GLM) in fishtail. The quantitative parameters that follow normality were subjected to an ANOVA and the Kruskal Wallis test for those, which do not flow normality. To perform the Pearson correlation test, the shape of the

tuber, the color of its skin, the presence or absence of rootlets and stripes on the tuber were been coded according to the yam descriptor standard [25].

3. Results

3.1. Seedling recovery rate

In agro-ecological zone IV, survival rate is between 42.55% and 77.5% respectively in the seedling having as female parents the cultivars Dodo I and Gnidou, with an average of 57.72%. The best survival rates were observed in the descendants of female cultivars Gnidou, Katala, Mafobo and Agatou with respective rates of 77.5%, 74.07%, 63.63% and 62.5% (Figure 3). A highly significant difference was observed between survival rate of the different accessions at the 5% threshold with a probability ($P = 0.036$) in this agro-ecological zone.

Of the 250 seedlings transplanted in agro-ecological zone V, the recovery rate varies from 12.5% to 40.74% respectively in seedling having as female parents the cultivars Moroko and Dodo A. The descendants of female cultivars Gnidou, and Kpakala yielded 34.61% and 31.03% respectively (Figure 4). There is no significant difference ($P = 0.079$) between the recovery rates of the different hybrids in this agroecological zone.

The variation in the recovery rate in the two agro-ecological zones is very significantly difference at the 5% level, with an associated probability of <0.0001 .

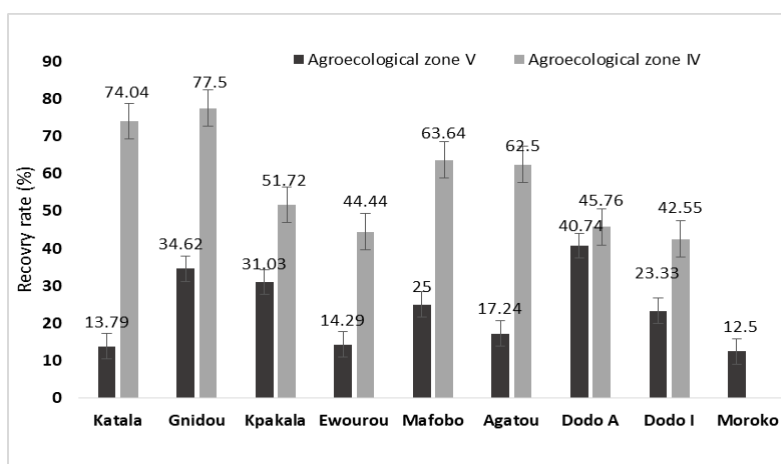


Figure 4 Seedling recovery rate (%) in agroecological zones IV and V

3.2. Tuberization rate

Seedlings tuberization rate in the agro-ecological zone IV fluctuate 0% obtained in in the offspring of female parents Moroko and Mafobo to 85% obtained in the cultivar Dodo I with an average rate of 55.41%. The best performing hybrids in this area are those having as female parents the cultivars Dodo A with 59.26%, Katala with 60%, Ewourou with 75%, and Dodo I with 85% (Figure 5).

The tuberization rates in the agro-ecological zone V varied from 25% to 80% respectively observed in hybrids from cultivars Katala and Agatou with an average rate of 57.62%. The best performing hybrids in this area are the descendants of cultivars Gnidou with 55.56%, Dodo I with 57.14%, Dodo A with 72%, Kpakala with 77.78% and Agatou with 80% (Figure 5).

The average rate of tuberization was 57.62% in agro-ecological zone V and 55.41% in agro-ecological zone IV. No significant difference ($P = 0.46$) is observed between the average rates of tuberization in the two agro-ecological zones.

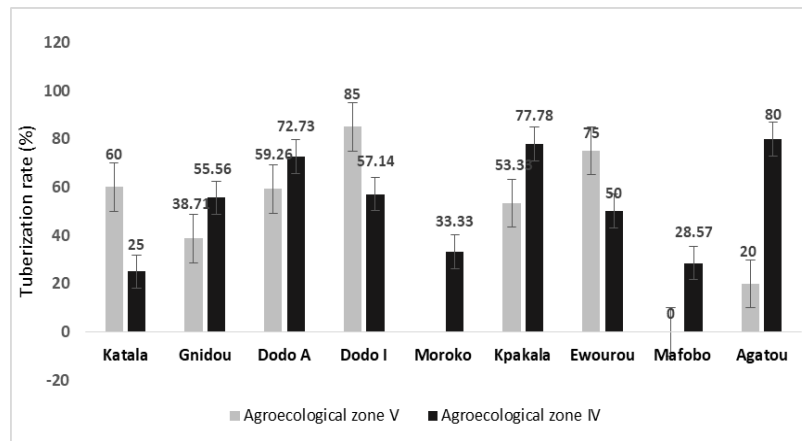


Figure 5 Tuberization rates (%) of accessions in the two agro-ecological zones V and VI

The rate of tuberization of hybrids from the cultivar Katala in agro-ecological zone V (25%) was lower than that of agro-ecological zone IV which was 60%. The same observation was made for the genotype produced by the cultivar Agatou which had a tuberization rate of 20% in agro-ecological zone IV, against 80% in agro-ecological zone V.

3.3. Agromorphological characteristics of the first generation tubers

In the agro-ecological zone IV, the microtubers length varies between 2.4 and 30 cm respectively in the seedlings from the cultivars Katala and Kpakala, with an average length of 13.01 ± 5.84 cm (Figure 6). The probability associated to the means of this variable ($P = 0.13$) not reveal any significant difference between accessions of this area.

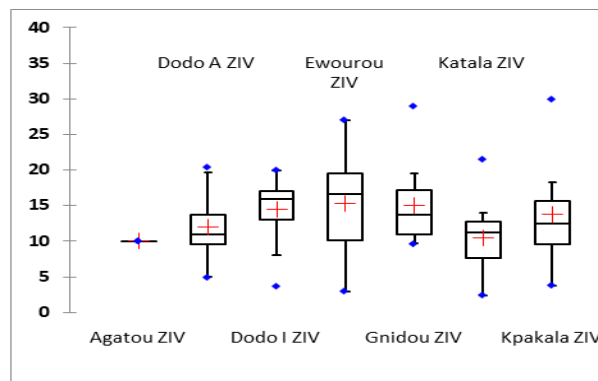


Figure 6 Length (cm) of the first generation tubers of accessions in the agroecological zone IV

The length of the first generation tubers of the agro-ecological zone V is between 1.5 and 32 cm observed respectively in the descendants of the cultivars Moroko and Mafobo, with an average of 8.95 ± 7.4 cm (Figure 7). The difference of the microtubers length is not statistically significant ($P = 0.44$) at the 5% level.

In agro-ecological zone IV, the circumference of the microtubers varied from 2.5 to 10 cm respectively in the genotypes carried by the cultivars DodoA and Katala, with an average circumference of 5.93 ± 1.38 cm (Figure 8). The probability linked to this mean ($P = 0.71$) shows a similarity between the circumference of the first generation tubers in zone IV.

The first generation tubers circumference in the agro-ecological zone V is between 1.2 cm and 6.8 cm observed respectively in the descendants of cultivars Kpakala and Agatou, with an average of 4.34 ± 0.88 cm. The very low value of the standard deviation reflects a small variation between the circumference of the first generation tubers, and this is confirmed by the probability $P = 0.47$ associated with this mean (Figure 9).

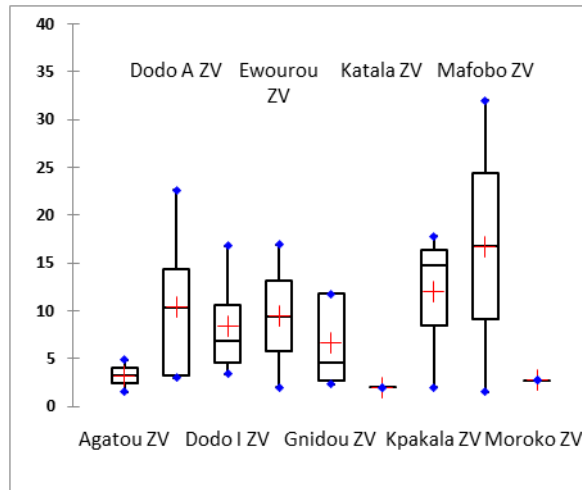


Figure 7 Length (cm) of the first generation tubers of accessions in the agroecological zone V

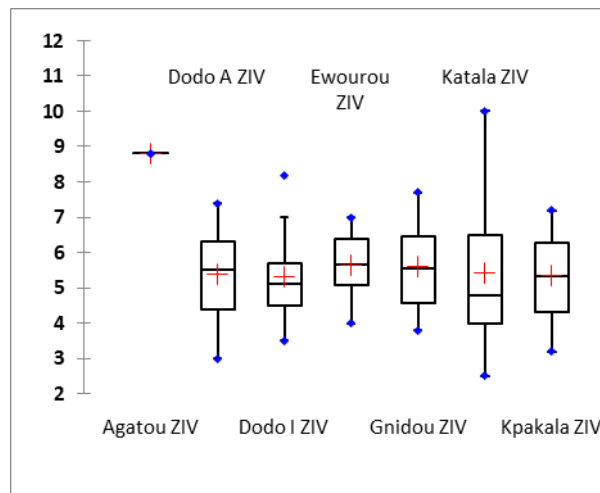


Figure 8 Circumference (cm) of the tubers of accessions in the agroecological zone IV

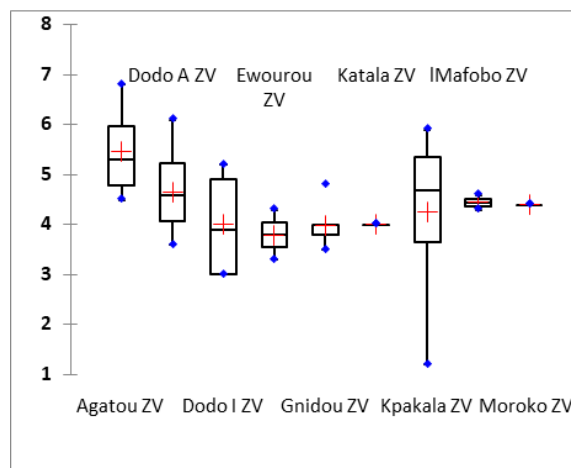


Figure 9 Circumference (cm) of the tubers of accessions in the agroecological zone V

The weight of the first generation tubers is between 3 g and 113 g, respectively in the genotypes from cultivars Dodo I and Gnidou, with an average of 24.87 ± 19.22 g in agro-ecological zone IV (Figure 10). The value of the standard deviation (19.22) shows variability of the different hybrid's microtubers weight, but not statistically difference ($P = 0.219$).

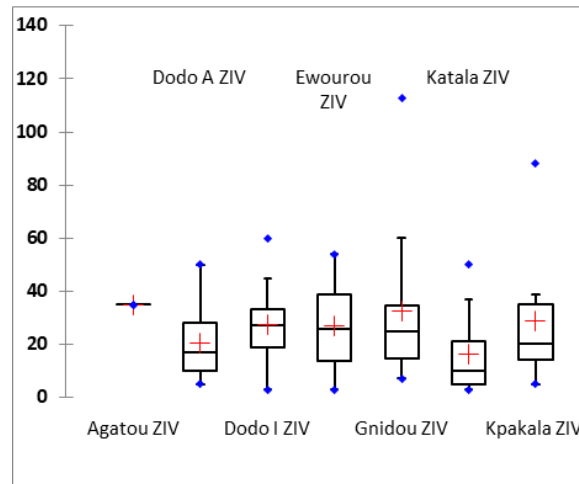


Figure 10 Weight (g) of the first generation tubers of accessions in agro-ecological zone IV

In agro-ecological zone V, the weight of the microtubers obtained is between 2 and 37 g, observed in the hybrids carried by cultivars Gnidou, Ewourou, Katala and Kpakala and Mafobo with an average of 10.30 ± 9.91 g (Figure 11). The probability associated with this comparison of mean ($P = 0.439$) does not reveal a significant difference in microtubers weight between cultivars.

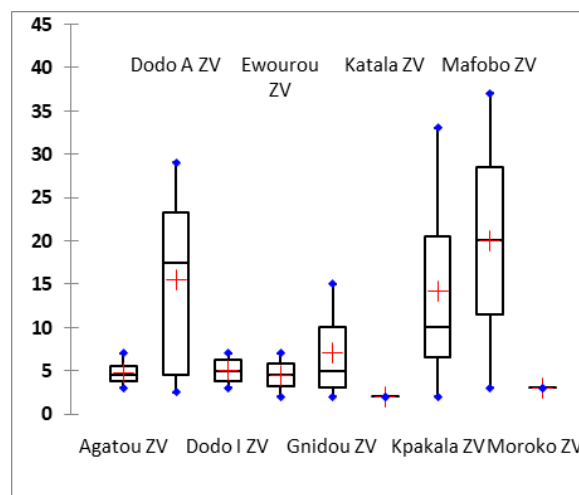


Figure 11 Weights (g) of the first generation tubers of accessions in agro-ecological zone V

The means comparison test revealed a very highly significant difference between the length of the tubers obtained of the two agro-ecological zones with $P = 0.0007$ (Figure 12).

The probability associated of means comparison test ($P = 0.0003$), revealed a very highly significant difference between the circumference of the first generation tubers of the two agro-ecological zones (Figure 12).

The probability associated with the comparison of the means ($P < 0.0001$), revealed a very highly significant difference between the mean weight of the microtubers obtained in the two agro-ecological zones (Figure 12).

Table 2 Relative importance (%) of the first generation tubers of accessions for morphological descriptors.

Agroeco-logical zones	Name	Shape of tuber		Color of tuber skin		Tuber appearance		Absence of rootlets on tubers	Location of the rootlets on tubers			Stripe			Digitated			
		Oval	Cylin-dric	Purple clair	Purple	smooth skin	Rough		Top of tuber	Base of the tuber	Whole tuber	Absent	Top	Whole tuber	Absent	Middle	Base	
IV	Dodo I	0	100	45,45	54,54	13,63	86,37	77,27	9,09	0	13,63	0	4,54	95,45	100	0	0	
	Dodo A	14,28	85,92	35,91	64,29	64,29	35,91	85,92	7,14	0	7,14	85,92	14,28	0	100	0	0	
	Ewourou	20	80	20	80	60	40	60	40	0	0	100	0	0	100	0	0	
	Katala	28,57	71,43	42,86	57,14	64,29	35,91	78,57	0	0	27,27	0	0	100	100	0	0	
	Kpakala	30	70	10	90	70	30	80	20	0	0	0	30	70	90	0	10	
	Gnidou	0	100	81,80	18,20	100	0	54,55	0	0	18,17	27,28	100	0	0	100	0	0
	Agatou	100	0	0	100	0	100	100	100	0	0	0	100	0	0	100	0	0
Great mean		27,55	72,48	33,72	66,31	53,17	46,88	76,62	10,89	2,60	10,76	55,13	6,97	37,92	98,57	0,00	1,43	
V	Kpakala	14,29	85,71	0	100	57,15	42,85	0	42,85	0	57,15	57,15	0	42,85	100	0	0	
	Gnidou	60	40	60	40	40	60	0	20	0	80	100	0	0	100	0	0	
	Dodo A	44,44	55,55	0	100	66,67	33,33	11,11	33,33	0	55,56	0	11,11	88,89	100	0	0	
	Mafabo	50	50	100	0	0	100	0	50	0	50	50	50	0	100	0	0	
	Dodo I	60	40	20	80	20	80	0	40	0	60	60	0	40	100	0	0	
	Ewourou	50	50	100	0	100	0	0	50	50	0	0	50	50	100	0	0	
	Agatou	100	0	75	25	25	75	0	50	0	50	100	0	0	100	0	0	
Great mean		54,10	45,89	50,71	49,29	44,12	55,88	1,59	40,88	7,14	50,39	52,45	15,87	31,68	100,00	0,00	0,00	

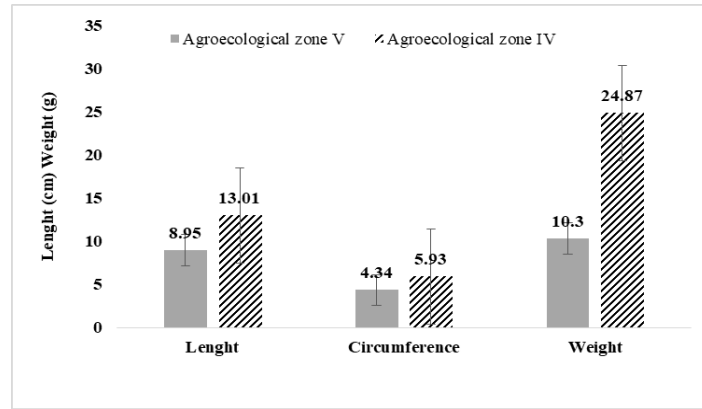


Figure 12 Means of length (cm), circumference (cm) and weight (g) of first generation tubers by agroecological zone

In agroecological zone IV, 72% of the first generation tubers were cylindrical, 66.31% had purple skin color and 53.17% had smooth skin. The rootlessness was found on 76.62% of these tubers which are all (100%) scratch-free, with 1.43% branching observed in the first generation tubers of Kpakala seedlings (Table 2).

In agroecological zone V, the first generation tubers were oval (54.10%), light purple (50.71%) and rough (55.88%) in color. The 98.41% of these tubers had roots, either over their entire surface or at the top; stripes were observed on 52.45% of the tubers and all of them (100%) unbranched (Table 2).

Pearson's correlation shows that the presence or absence of roots is positively correlated with agroecological zones ($r = 0.96$). In the two agro-ecological zones, morphological heterogeneity of the first generation tubers was observed within each cultivar seedling and between all cultivars of seedlings (Figures 13a, 13b, 14a, 14b, 15a, 15b, 16, 17, 18 and 19).



Figure 13a First generation tubers obtained from seedlings of the accession Dodo in agroecological zone IV

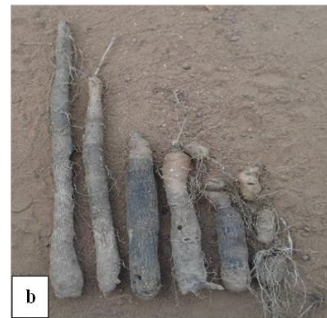


Figure 13b First generation tubers obtained from seedlings of the accession Dodo in agroecological zone V



Figure 14a First generation tubers obtained from seedlings of the accession Gnidou in agroecological zone IV



Figure 14b First generation tubers obtained from seedlings of the accession Gnidou in agroecological zone V



Figure 15a First generation tubers obtained from seedlings of the accession Katala in agroecological zone IV



Figure 15b First generation tubers obtained from seedlings of the accession Katala in agroecological zone V



Figure 16 First generation tubers obtained from seedlings of the accession Kpakala in agroecological zone IV



Figure 17 First generation tubers obtained from seedlings of the accession Agatou in agroecological zone IV



Figure 18 First generation tubers obtained from seedlings of the accession Ewourou in agroecological zone V



Figure 19 First generation tubers obtained from seedlings of the accession Mafobo in agroecological zone V

3.4. Correlation between agro-morphological parameters

The weight of the tubers is positively correlated with their circumference ($r = 0.58$) and their length ($r = 0.80$). Tuber shape is negatively correlated with length ($r = -0.59$) and weight ($r = -0.38$). The presence of stripes on a tuber is correlated by its color ($r = 0.40$). The more strongly colored was the tuber; more the stripe appeared on the colored part (Table 3).

Table 3 Correlation between the agro-morphological parameters of tubers

Variables	Lenght (cm)	Circumference(cm)	Weight (g)	Shape	Tuber skin color	Rootlet	Stripe
Lenght (cm)	1						
Circumference (cm)	0,26	1					
Weigh (g)	0,80	0,58	1				
Shap	-0,59	0,04	-0,38	1			
Tuber skin color	0,10	0,20	0,15	-0,01	1		
Rootlets	0,19	-0,09	0,13	-0,19	-0,11	1	
Stripes	0,17	0,12	0,13	-0,12	0,40	0,03	1

4. Discussion

Two (02) months after transplantation, the seedling recovery rate is 55.41% in agro-ecological zone IV and 23.6% in agroecological zone V. The recovery rates of *Dioscorea cayenensis-Dioscorea rotundata* species complex obtained in the same study area is 55.41% [21]. These recovery rates of *Dioscorea cayenensis-Dioscorea rotundata* species complex seedlings are lower than the response obtained in *Dioscorea alata* seedlings in Vanouatou, which is 100% after transplantation [26]. This difference could be explained by the vigor of the seedlings before transplanting. In the present study, *Dioscorea cayenensis-Dioscorea rotundata* species complex seedlings were transplanted at a development stage of 12 weeks after sowing while those of *Dioscorea alata* were transplanted at 24 weeks of development. The high recovery rate (56%) obtained in the agroecological zone IV result of the environment and climatic conditions of this zone, which are similar with these of the seed-sowing zone. The recovery rate of seedling is acceptable (more than 50%) when there is not significant difference between climatic and environment conditions of seed sowing zone and seedling transplanted zone. Humidity of the transplanted site is very important for the development of seedlings, because the transplants are very vulnerable of water deficit the first weeks after their transplantation [27] ; transplants are very vulnerable to water deficit in the first weeks after their transplantation [Zoundjihékpou, 2020, pers. comm].

The tuberization rate is similar in the two studies area, 57.63% in agroecological zone IV and 55.64% in agroecological zone V. Nevertheless, between seedlings performance, difference is very significant, with respectively 24.9 ± 19.2 g for agroecological zone IV and 10.30 ± 9.91 g for agroecological zone V.

Seedlings produce from cultivated yam's seeds have received allelic combinations, which allow them to adapt in the both agro-systems, like cultivated yam's [19, 28] or wild yams, which disperse by sexual way in their natural ecosystems, are adapted in this hostile environment [29, 30].

In the agroecological zone V, 43.65% of the first generation tubers are branched and various shape are been observed in the both agroecological zones. These various shapes of the first generation tubers had been mentioned by more authors in the pass [11, 20, 21, 26]. The ramifications or digitations and roots observed on the first generation tubers are wild characters which will disappear over time, as during domestication of wilds ancestors [31] to have better agronomic and morphological performance. Concerning rootlet, 88, 89% of Dodo A tuber of agroecological zone V have rootlets, but in the agroecological zone IV 85,92% of tubers of this accession are without rootlets. This result show that this character expression is under environment and climatic conditions.

These wild characters are the expression of some genes, which were repressed in the cultivated germplasm by domestication process. These types of natural hybridizations between cultivated and wild clones have been observed in the other species such as cassava germplasm [32]. These types of hybrids between cultivated and wild yams found in forests and savannas are considered wild, for that, some authors have argued that not all wild yams are wild [33]. The stability of the pollen grains size between *Dioscorea cayenensis-D. rotundata* complex species [34], could promote gene flow between cultivated and wild yam.

Sexual reproduction served to develop new varieties of cassava (*Manihot esculenta*) [17] and in yams (*Dioscorea alata*) in the West Indies [26, 35]. It offers possibilities for obtaining new high-yielding varieties adapted to current climatic conditions, as new varieties adapted to different agroecological zones and tolerating peasant cropping constraints [33]. Without that, the agro-biodiversity food, humanity food reservoir [36] which contributes of rural population's income and socio life [37] is threatened.

5. Conclusion

This study show that the yam's seeds produced by sexual reproduction can be used of yams production. The seeds offer a great varietal genetic diversity of yams. The strong heterogeneity of tubers obtained by this method shows that seeds can be used to create varietal diversity. It's a strength way to improve cultivated yam's germplasm, and supply the genetic erosion caused by unsuitable climatic conditions and the impoverishment of the soil. Seedlings can outlive in various climatic conditions. However, it is preferable to sow the seeds directly in the transplantation areas to avoid the influence of climatic variations between the germination site and the seedling-transplanted site.

Compliance with ethical standards

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Disclosure of conflict of interest

All authors declare no conflict of interest.

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