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RESEARCH ARTICLE

IN-SITU MORPHOLOGICAL CHARACTERIZATION OF CASSAVA LANDRACES (*MANIHOT ESCULENTA* CRANTZ) FROM CÔTE D'IVOIRE

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Abstract

Cassava provides food security for millions of people worldwide. In Côte d'Ivoire, it is the second most important food crop. The objective of this study is to evaluate the structure and genetic variability of certain qualitative traits in targeted cassava landraces in order to identify the most widespread cassava landraces. Thus, prospecting missions were carried out in different regions of Côte d'Ivoire to characterize cassava landraces, from which 180 cassava landrace accessions were characterized using 14 morphological markers. Multiple Correspondence Analysis (MCA) showed that among the 14 markers, 10 were relevant for describing the total variability within accessions. In addition, the Hierarchical Ascending Classification (HAC) grouped the Ivorian landraces into five homogeneous groups and also revealed that this morphological diversity is not structured according to the landraces' geographical origin. The study highlighted 131 distinct cultivars among the 180 accessions characterized. Moreover, of the 131 distinct cultivars, four were the most widespread. These are the local cultivars: *Yacé*, *Six mois*, *Blébou* and *Trogla*. Thus, the study has definitively shown that breeders have a wide choice of parents for developing improved cassava cultivars adapted from the populations found in Côte d'Ivoire.

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Introduction:-

Cassava (*Manihot esculenta* Crantz), a Euphorbiaceae native to the Amazon, is the only domesticated species of the genus *Manihot* comprising 98 species (Ugent *et al.*, 1986; Olsen & Schaal, 2001). Introduced to Africa by the Portuguese in the 16th century (Allem, 1994), it is cultivated today in tropical and subtropical countries (Nassar & Ortiz, 2007; Ortiz *et al.*, 2016).

Cassava provides food security for millions of people worldwide. It is grown primarily for its starch-rich tuberous roots and is the world's second largest source of starch after corn (Norton, 2014). It has the ability to grow on poor soils and in areas with unpredictable rainfall (El-Sharkawy, 1993; De Tafur *et al.*, 1997). In addition, it is adaptable

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to several agrosystems (Doubi *et al.*, 2016). Thus, cassava is an important plant for food security in the context of climate change and arable land reduction.

Several studies on cassava genetic diversity have been conducted to guide varietal selection. These studies have been conducted both morphologically (N'Zué *et al.*, 2014; Lebot *et al.*, 2015) and molecularly (Kawuki *et al.*, 2013; Peña-Venegas *et al.*, 2014).

Cassava exhibits high morphological variability that reveals intraspecific diversity as perceived by farmers. Furthermore, according to Elias *et al.* (2001), leaf, stem and tuberous root color remains virtually unaffected by the environment thus allowing for distinction between cultivars. Indeed, farmers distinguish between cultivars based primarily on qualitative traits such as organ color (Lebot *et al.*, 2015). Also, organ color can be used as a marker for genetic characterization in different ecological zones.

In Côte d'Ivoire, this crop represents the second most important food crop after yam (FAO, 2018). However, studies of the *in situ* genetic diversity held by producers have yet been to be conducted. Yet, study cultivar morphological diversity is essential for developing new varieties adapted to different agro-ecological zones for subsequent adoption by producers. The objective of this work is to evaluate the morphological diversity of cassava on the basis of qualitative characteristics. Specifically, the aims were to (i) analyze morphological variability, (ii) establish the genetic structure of cassava accessions originating from Côte d'Ivoire and (iii) identify the most widespread cultivars in order to include them in cassava genetic improvement programs.

Material And Methods:-

Study area

Côte d'Ivoire has a topography that has few contrasting features overall. However, three main sub-topographies can be distinguished. These are the plains that extend to the south of the country, the plateaus that extend the plains to the north and the mountainous massifs located in the west of the country (Avit, 1999).

As for the vegetation of Côte d'Ivoire, it is divided into two large parts: the Guinean domain and the Sudanese domain. The Sudanese domain, located in the northern part of the country, is characterized by savannah. This area is under the influence of the Sudanese climate, which corresponds to a tropical climate with an average annual rainfall of between 1200 and 1600 mm. The Guinean domain, characterized by the forest zone, is located in the southern half of the country and is under the influence of three climates. These are the sub-equatorial climate, the humid tropical climate and the mountain climate. The sub-equatorial climate or 'Attéen climate', includes the coastline and a large part of the forest area with an annual rainfall of between 1200 and 2200 mm. The humid equatorial climate or 'Baouléen' climate with 1000 to 1900 mm of rainfall per year extends from the east (*Bouna* and *Bondoukou* regions) to the forest and mountainous regions of the west, passing through the center of Côte d'Ivoire (*Bouaké* region). The mountainous climate is located in the mountainous massif of western Côte d'Ivoire in the regions of *Man*, *Toulepleu* and *Danané*, with an average annual rainfall of between 1600 and 2000 mm (Goula *et al.*, 2007).

Plant Material

The plant material consists of 180 cassava cultivars from 13 regions of Côte d'Ivoire. Figure 1 shows the different localities surveyed.

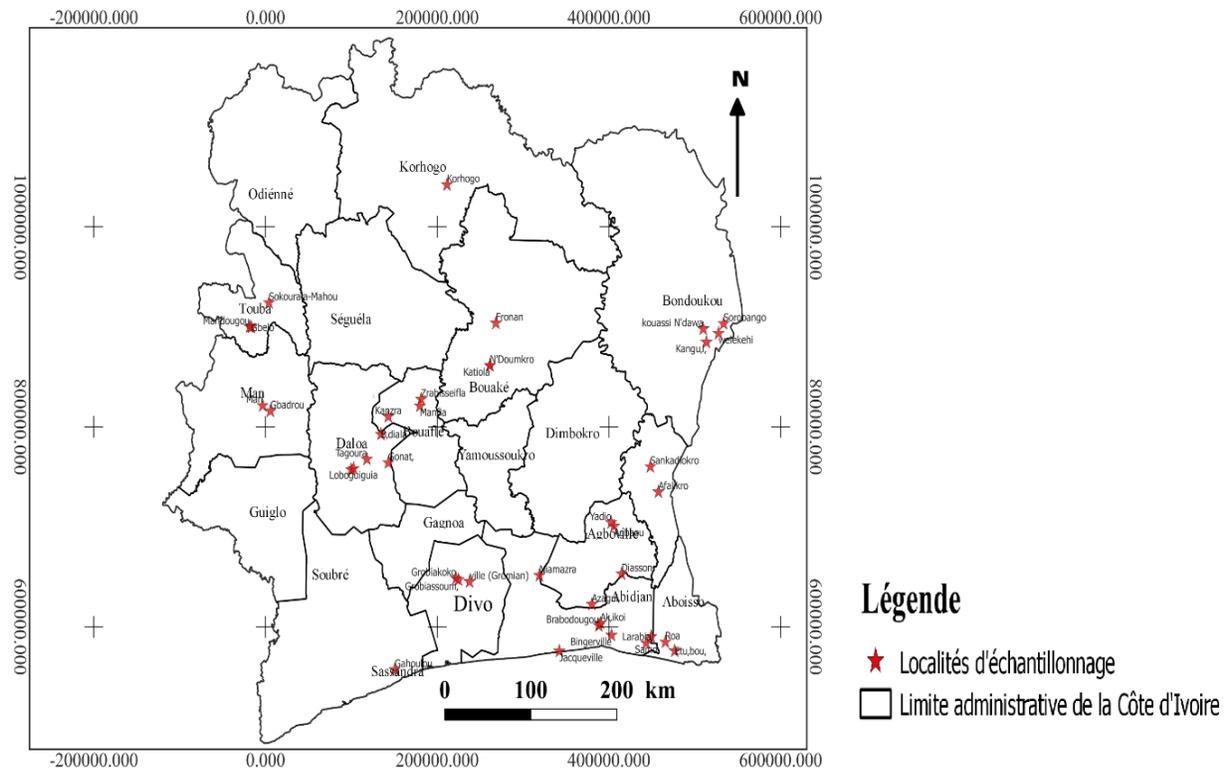


Figure 1: Area of characterization of different cassava accessions in Côte d'Ivoire.

Methods:-

Morphological characterization

Morphological characterization consisted of *in situ* diversity analysis of cassava cultivars at least 12 months old based on 14 cassava descriptors (N'Zué *et al.*, 2014). On average three fields at least 10 km apart were assessed per locality. During the missions, interviews were conducted with farmers. Information collected included cropping pattern, vernacular name and household use of cassava cultivars. A total of 297 farmers were interviewed.

Frequency of qualitative traits

The variability of the 14 qualitative traits studied was assessed by determining the frequency of occurrence (P_i) of phenotypes of each trait. These relative frequencies were used to determine the mode of each trait.

Statistical analysis of the data

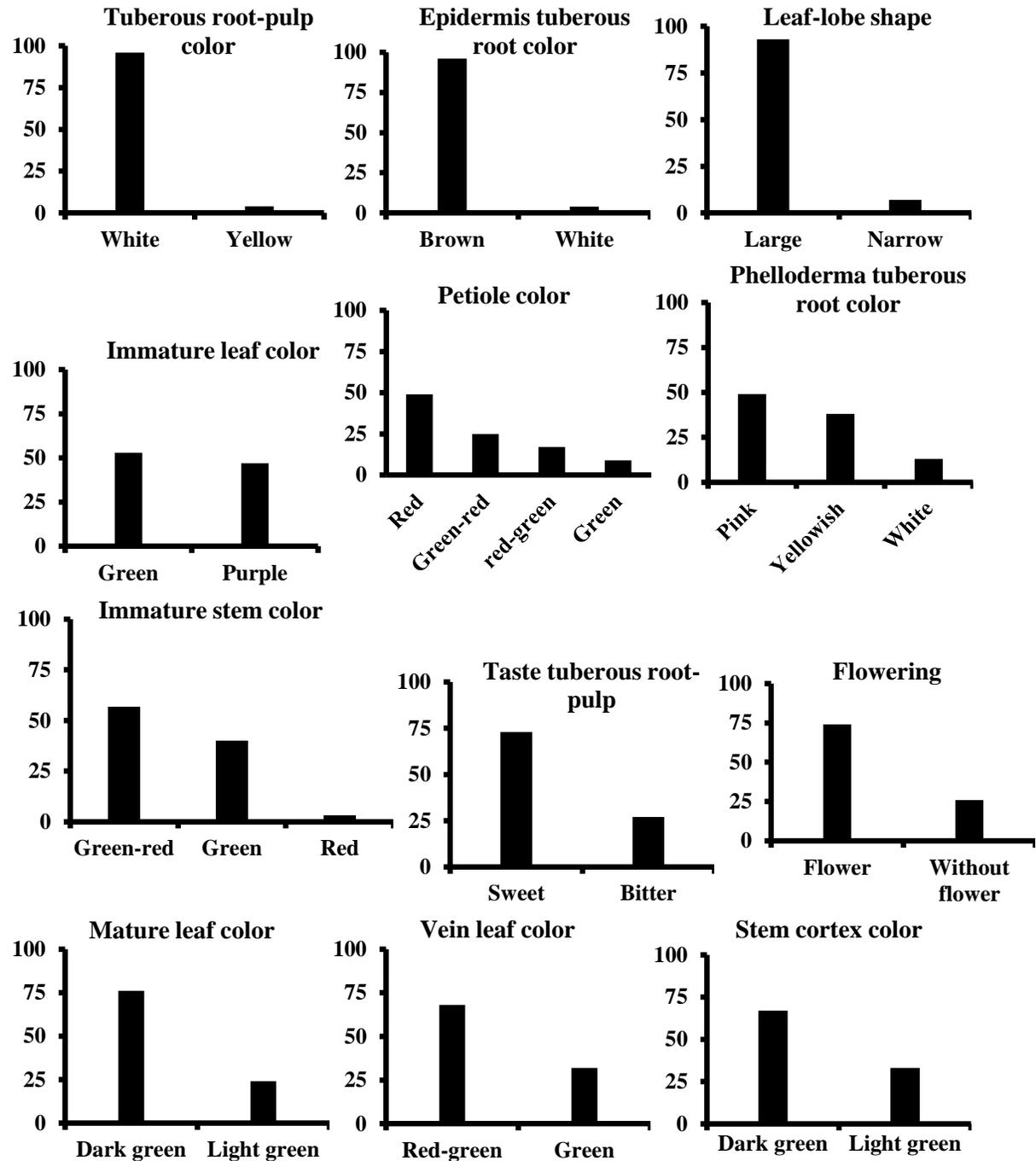
A Multiple Correspondence Analysis (MCA) was used to reveal the most discriminating traits and the relationships between traits in a two-dimensional space. Hierarchical Ascending Classification (HAC) was performed using Ward's method to highlight homogeneous groups and duplicates. Descriptive statistics, MCA and HAC were performed using STATISTICA software version 7.1 (StatSoft Inc., France). Histograms were generated with Excel 2010 software.

Results:-

Descriptive analysis of local cultivars

Analysis of phenotypic frequencies of traits revealed significant variability within each trait (Figure 2). However, low variability was noted in the characters tuberous root-pulp color, tuberous-root epidermis color and leaf-lobe shape. Indeed, more than 90% of the accessions evaluated showed white root pulp, brown-skinned root and broad leaves. Accessions with yellow pulp, white-skinned tubers, and thin-lobed leaves represented less than 10% of all accessions. The number of cultivars with green immature leaves was approximately equal to those with purple immature leaves. In addition, nearly half of the cultivars had red petiole leaves, pink phelloderma tuberous-root, and bicolored (green-red) immature stems. Regarding the taste of tuberous root-pulp, 73% were sweet versus 27% bitter.

In addition, nearly ¾ of the local cultivars flowered and showed leaves with dark green coloration. Furthermore, in 66% of the local cultivars, the leaf veins were bi-colored (red-green) and the stem cortex was dark green in color.



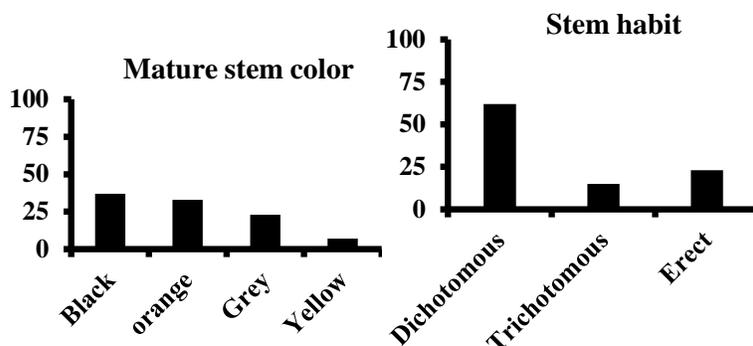


Figure 2:- Phenotypic frequencies of morphological descriptors of 180 cassava accessions in Côte d'Ivoire.

Relevance of morphological descriptors

The MCA highlighted the traits that best explained variability among cassava cultivars. The first five factorial axes accounted for 52.73% of the variation (Table 1). Also, based on the contributions of each trait to the formation of the first five factorial axes, 10 out of 14 traits best explained morphological variation (Table 2). The first five factorial axes are defined as follows:

Axis 1 was defined by the characters petiole coloration, young stem and the character tuberous root taste. It explained 18.19% of the total variation. The characters petiole color and tuberous root taste were negatively correlated with this axis. On the other hand, the character color of the young stem was positively correlated to it (Figure 3). Thus, axis 1 pitted cultivars with red leaf petiole and bitter taste of tuberous-root against those with green immature stem.

Axis 2 explained 10.35% of the total variability (Table 1). It is characterized by leaf-lobe shape, mature stem coloration, immature stem coloration, phelloderma tuberous-root coloration and stem cortex coloration (Table 2). This axis pitted cultivars with reddish immature stem, blackish mature stem, and narrow leaves against cultivars with white phelloderma tuberous-root and light green stem cortex (Figure 3).

Axis 3 explained 9.54% of the total variability (Table 1). The traits petiole color, epidermis tuberous-root color, phelloderma tuberous root color, and stem color contributed to this component (Table 2). Axis 3 pitted cultivars with green-dominant bicolored petioles against cultivars with tuberous roots with white epidermis and phelloderma, red-dominant bicolored leaf petiole, and gray-colored mature stems (Figure 4).

Axis 4 accounts for 8.07% of total cultivar variability (Table 1). The characters that contributed to its formation are: the color of the mature leaf, petiole, apical stem and phelloderma of tuberous roots (Table 2). This axis contrasts cultivars with a light-green leaf and cultivars with a bicolored (red-green) leaf petiole with red dominance, a red apical stem and a white phelloderma (Figure 5).

Axis 5 is characterized by leaf petiole color, mature stem color, stem habit (Table 2). On this axis, cultivars with a dichotomous habit are opposed to those with green leaf petiole, yellowish mature stem and a trichotomous habit (Figure 6). Axis 5 explains 6.58% of the total variability of the cultivars (Table 1).

Table 1:- Eigenvalues of the factorial axes and their percentages of inertia

Factorial axes	Axe 1	Axe 2	Axe 3	Axe 4	Axe 5
Eigenvalue	0,273	0,155	0,143	0,121	0,099
Percentage of inertia	18,187	10,347	9,541	8,074	6,581
Cumulative percentage	18,187	28,534	38,076	46,150	52,730

Table 2:- Contribution of the traits to the formation of the first six factorial axes

Axes	Qualitative traits	Contribution of traits (%)
Axe 1	Leaf petiole color	14,51
	Immature stem color	14,27
	Taste tuberous root-pulp	13,96

Axe 2	Immature stem color	10,34
	Phelloderma tuberous root color	14,14
	Mature stem color	28,9
	Leaf-lobe shape	11,26
	Stem cortex color	11,14
Axe 3	Leaf petiole color	18,76
	Epidermis tuberous-root color	22,11
	Phelloderma tuberous-root color	9,96
	Mature stem color	16,93
Axe 4	Leaf petiole color	14,61
	Mature leaf color	23,47
	Immature stem color	11,29
	Phelloderma tuberous-root color	9,17
Axe 5	Leaf petiole color	25,05
	Stem color	15,17
	Stem habit	32,35

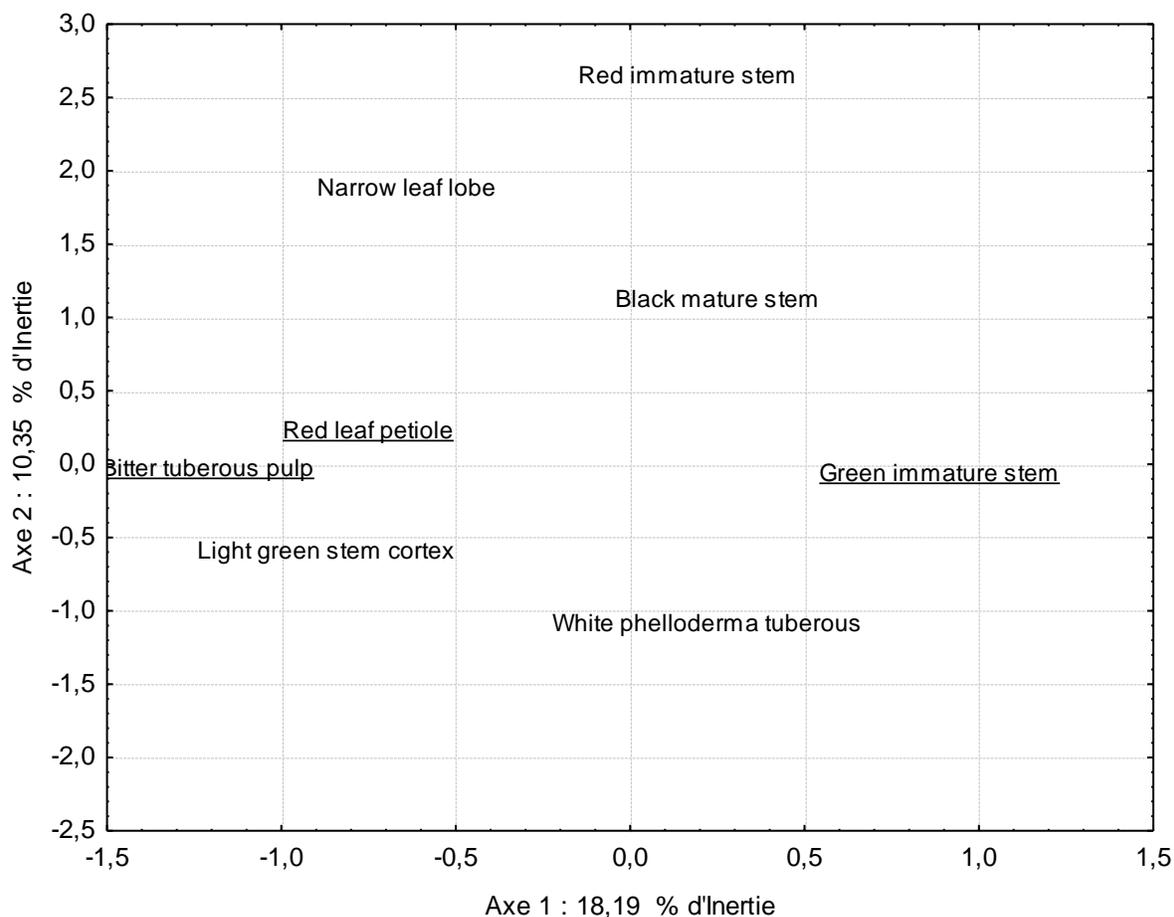


Figure 3:- Projection in the factorial plane 1-2 of the MCA of the qualitative traits analyzed in 180 cassava cultivars in Côte d'Ivoire. Underlined and not underlined are the modalities that contributed to the formation of axis 1 and axis 2 respectively.

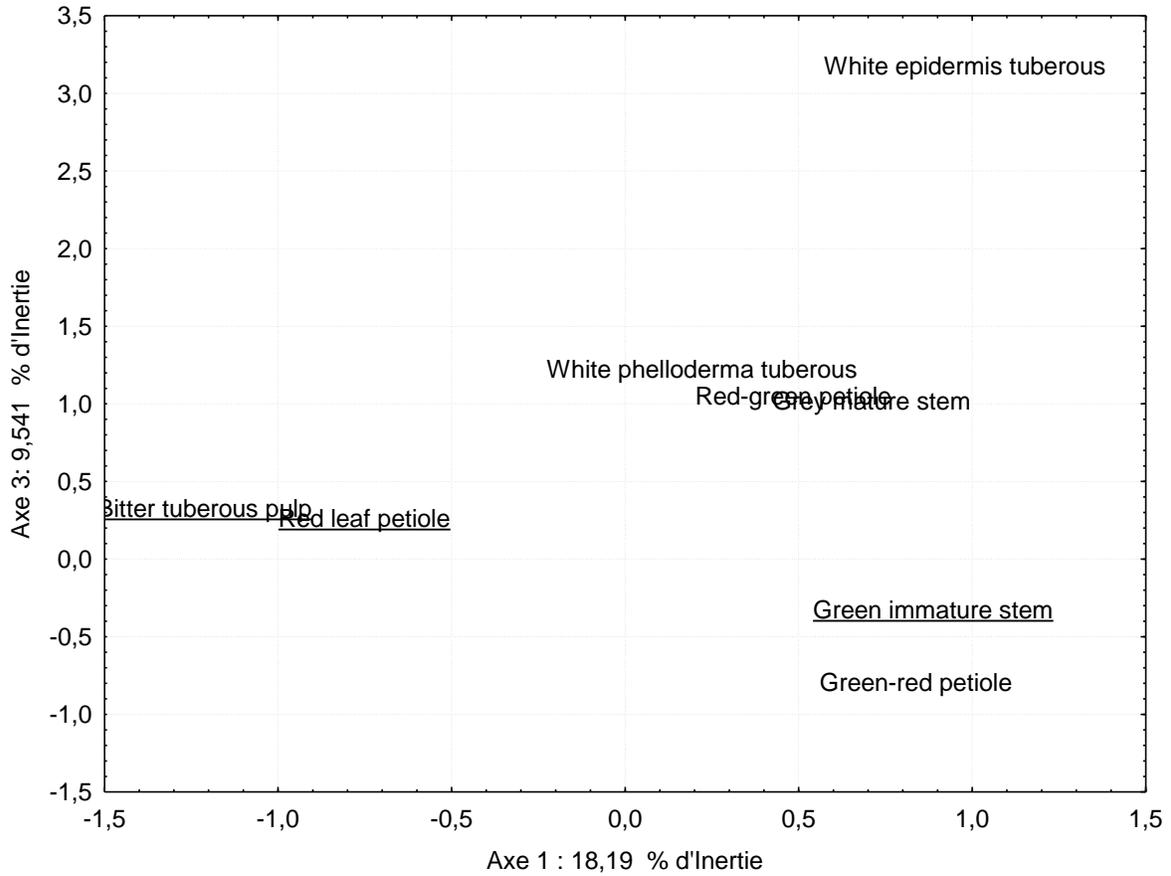


Figure 4:- Projection in the factorial plane 1-3 of the MCA of the qualitative traits analyzed in 180 cassava cultivars in Côte d'Ivoire. Underlined and not underlined are the modalities that contributed to the formation of axis 1 and axis 3 respectively.

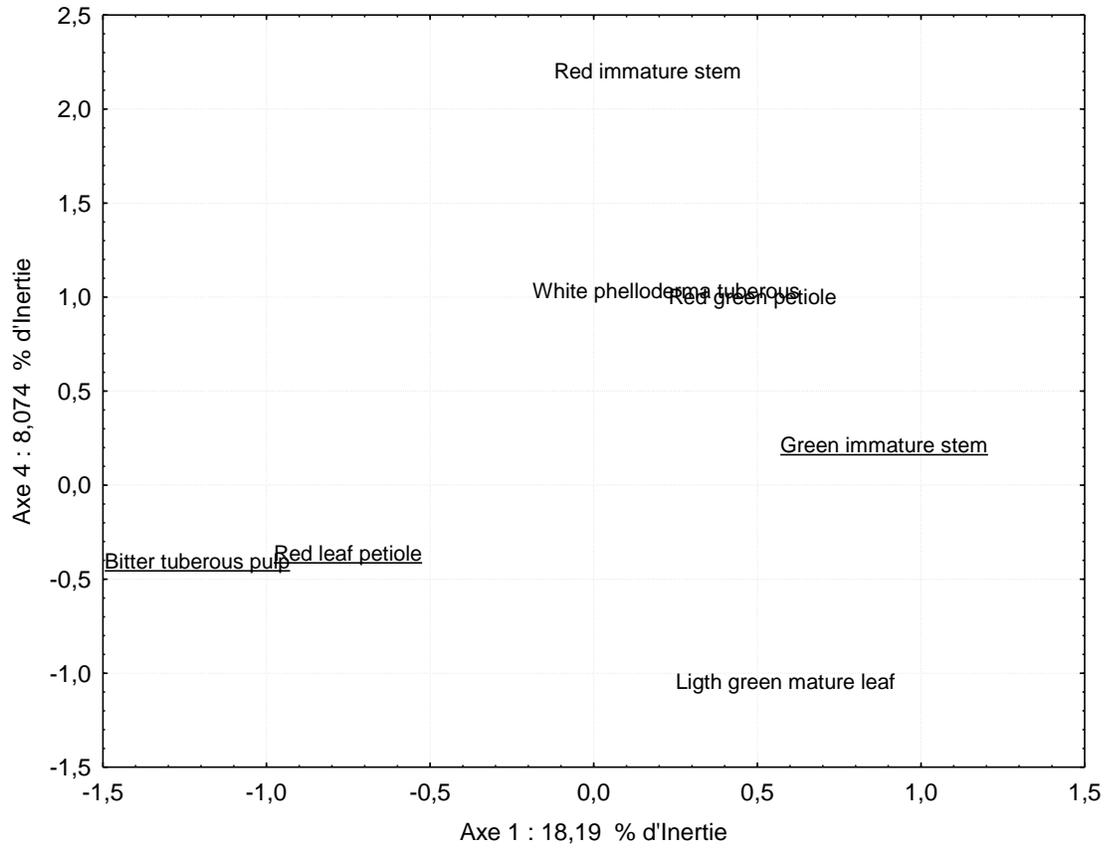


Figure 5:- Projection in the factorial plane 1-4 of the MCA of the qualitative traits analyzed in 180 cassava cultivars in Côte d'Ivoire. Underlined and not underlined are the modalities that contributed to the formation of axis 1 and axis 4 respectively.

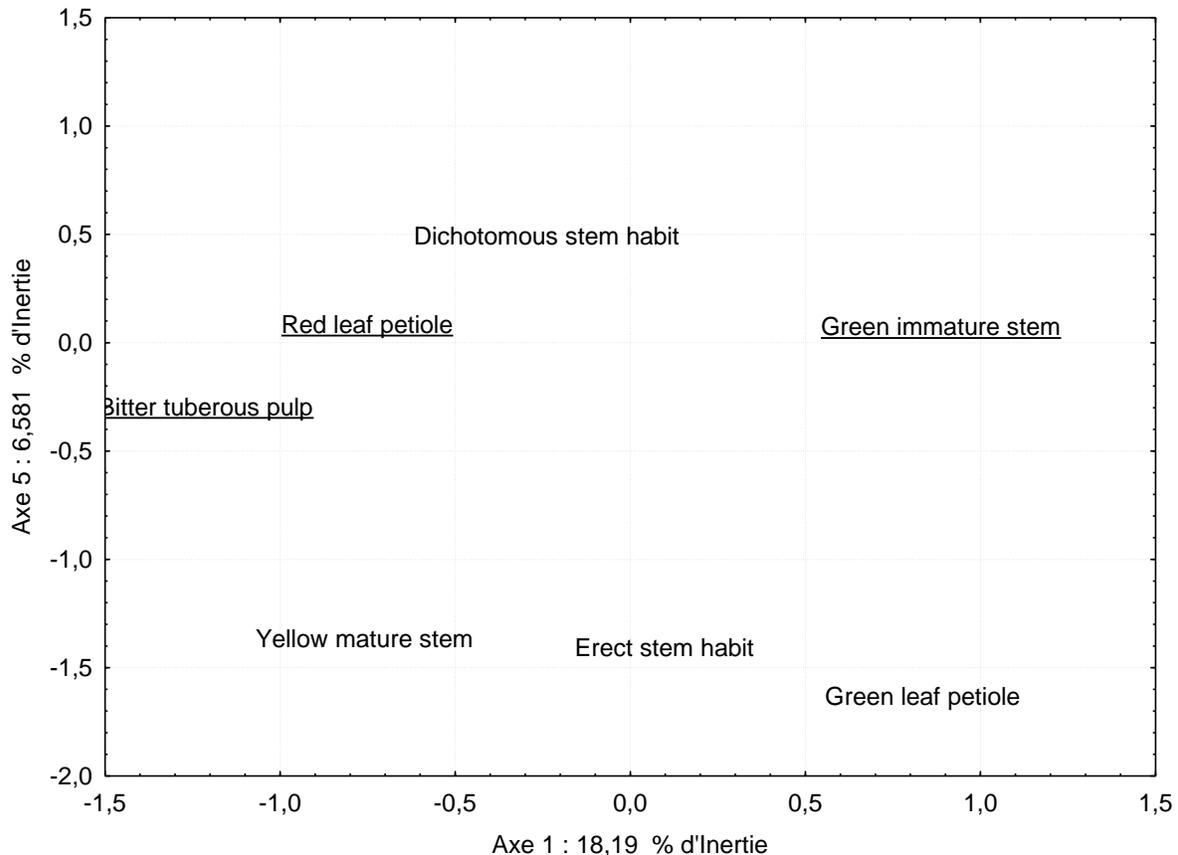


Figure 6:- Projection in the factorial plane 1-5 of the MCA of the qualitative traits analyzed in 180 cassava cultivars in Côte d'Ivoire. Underlined and not underlined are the modalities that contributed to the formation of axis 1 and axis 5 respectively.

Structuring the diversity of local cultivars

Hierarchical Ascending Classification (HAC) based on the qualitative traits examined in the 180 cassava cultivars was performed. A split at similarity level 24 of the dendrogram resulting from the HAC defined five homogeneous groups (Figure 8). The different groups 1, 2, 3, 4, and 5 contain 70, 22, 35, 19, and 34 cultivars respectively. The HAC revealed a structuring of cultivars independently of these cultivars' geographical origin.

Group 1 contained the largest number of cultivars. It represented 38.89% of the cultivars and was composed mainly by sweet cultivars. The cultivars of this group presented young green stems with leaves with wide lobes and the mature leaves of dark green color. In addition, the cortex of the stems was dark green in color. The tuberous roots possessed a brown epidermis and white pulp.

Group 2 contained 12.22% of the cultivars. The cultivars of this group were characterized by soft tuberous roots with a brown epidermis and with leaves possessing bicolored petioles, with wide lobes and a dark-green color. The stem cortexes were light-green.

Group 3 contained 19.44% of cultivars, characterized by soft tuberous roots with a brown epidermis, pink phelloderma and white pulp. Leaves showed broad lobes with red petioles. The stem cortexes were dark-green.

Group 4 with 10.56% of the cultivars contained the bitter cultivars with tuberous roots possessing a brown epidermis, white pulp, young bicolored stems, and leaves with bicolored veins and red petioles. Cultivars in this group also had tuberous roots with a yellowish phelloderma and stems with a dark-green cortex.

Group 5 consisted of 18.89% of the cultivars and grouped bitter and light-green stem cortex cultivars. The cultivars in this group were characterized by leaves with red petioles, dark green blades, broad lobes and bicolored veins.

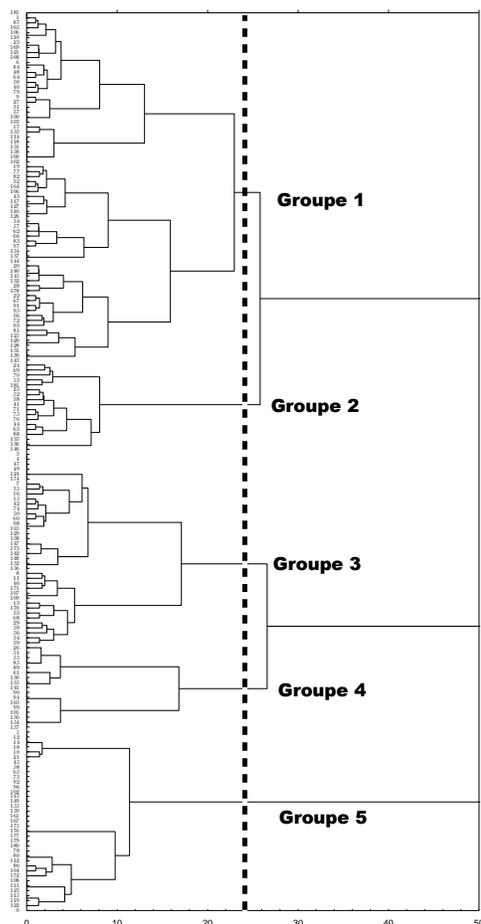


Figure 8:- Hierarchical ascending classification (HAC) using Ward's method based on Euclidean distances. Group 1 (70 local sweet cultivars, dark-green cortex), group 2 (22 local sweet cultivars, light-green cortex), group 3 (35 local sweet cultivars, red petiole), group 4 (19 local bitter cultivars, dark-green cortex), group 5 (34 local bitter cultivars, light green-cortex).

Identification of duplicates within cassava accessions

The traits studied allowed for the distinction between local cassava cultivars. The Euclidean distance matrix was used to identify duplicates within each cultivar group. Indeed, accessions with different names and a zero Euclidean distance were considered as identical cultivars. Thus, the cultivars *Blannin*, *Déhikachia*, *Six Mois*, *Egypt* and *Sanka* are duplicates. In addition, the cultivars *Blêbou*, *Israël* and *Agbabalama* on the one hand, and the cultivars *Totocla*, *Clapkan*, *Trogl*, *Gnananmininan* and *Kalowôrô* on the other, would be duplicates (Table 3). In the end, of the 180 cassava cultivars, 131 distinct cultivars and duplicates accounted for 27.22%. In addition, cultivars recorded under the same name of *Yacé* in different areas were effectively identical for all traits. In contrast, *Bonoua* and *Anader* designated different cultivars because they exhibited different characters. In analyzing the distribution of cultivars, *Yacé* was the most common, followed by *Trogl*, *Six Mois* and *Blêbou* (Table 3).

Table 3:- Vernacular names and location of duplicates in the cassava cultivar list.

Groups	Numbers	Vernacular names of duplicates	Assessment areas
Group 1	70	<i>Six Mois</i> , <i>Egypte</i> , <i>Blannin</i> , <i>Déhikachia</i> , <i>Sanka</i>	South, Southeast, Central-West, West, East
Group 4	19	<i>Blêbou</i> , <i>Israël</i> , <i>Agbabalama</i>	Central-West, Southeast, Northeast
		<i>Trogl</i> , <i>Totocla</i> , <i>Clapkan</i> ,	West Central, Central, West, Northwest,

		<i>Gnananmininan, Kalowôrô</i>	Northeast
Group 5	34	<i>Yacé</i>	South, Southeast, Southwest, Central, West, West, Northeast

Discussion:-

Morphological characterization of plant material is the preliminary phase of any plant breeding program. In this study, *in situ* morphological variability assessment missions were carried out in Côte d'Ivoire, using qualitative markers to identify the most-cultivated cassava cultivars. Indeed, according to Elias *et al.* (2001), qualitative traits such as organ color, leaves, stem and roots in cassava are highly heritable and are good markers that help discriminate between cultivars.

This study indicated significant variability in quality traits among the different local cassava cultivars encountered. However, the analysis revealed that the majority of cultivars exhibited tuberous roots with a brown epidermis and broad-lobed leaves. These results are similar to those of Lebot *et al.* (2015) in Vanuatu and Nadjiam *et al.* (2016) in Chad. Furthermore, 74% of local cassava cultivars flowered, compared to 22% in Chad (Nadjiam *et al.*, 2016) in a collection of 59 accessions. This difference in flowering rates could be explained by the low frequency of upright local cultivars, 23% versus 85% in the work of Nadjiam *et al.* (2016). Indeed, according to Medard (1973), cassava cultivars that do not branch do not flower either. Furthermore, 4% of local cassava cultivars had yellow pulp. N'Zué *et al.* (2009) also reported low proportions of yellow-pulped cassava cultivars in Côte d'Ivoire. Low proportions of yellow-pulped cassava have been reported in Chad (Nadjiam *et al.*, 2016), Sierra Leone (Karim *et al.*, 2020), Vanuatu (Lebot *et al.*, 2015) and Brazil (Mezette *et al.*, 2013), among others. For Emperaire *et al.* (1998), cassava with yellow pulp is rarer in Africa than in the Amazon. This yellow coloration of the pulp is however, an indicator of high carotenoid and amino acid content (Nassar *et al.*, 2007). In addition, the yellow-orange pulp cultivar reported by Ferreira *et al.* (2008) and Nassar *et al.* (2007) in Brazil was not collected in the present study. On the other hand, nearly half of the cassava cultivars showed tubers with pink phelloderma and leaves with red petioles. In contrast, in Chad and Vanuatu cassava cultivars largely had tuberous roots with cream-colored phelloderma (Nadjiam *et al.*, 2016; Lebot *et al.*, 2015).

Based on the 14 qualitative traits, the 180 local cassava cultivars were grouped into five homogeneous groups. In similar studies, Karim *et al.* (2020) in Sierra Leone, grouped 102 accessions into five groups while N'Zué *et al.* (2014) in Côte d'Ivoire identified for 159 accessions three groups. This difference in homogeneous groups could be explained by the level of truncation, which was 28 in our study versus 102 in that of N'Zué *et al.* (2014). Furthermore, in this study, the structure of morphological diversity was not superimposed on geographical origins. In other words, cultivars are found in different areas. In addition, the relatively high percentage of duplicates (27.22%) could be explained by the extent of exchange of cuttings between populations in different regions of Côte d'Ivoire. This large flow of cuttings exchange between populations has been noted in Vanuatu (Lebot *et al.*, 2015) and French Guiana (Elias *et al.*, 2000). On the other hand, nomenclature could also explain this level of duplication. Indeed, in peasant nomenclature, several names can be assigned to the same cultivar depending on the region and ethnicity (Elias *et al.*, 2001). Thus, the cultivar named *Totocla* by the *Baoulé* in the center is called *Clapkan* by the *Wobé* in the West, *Kalowôrô* by the *Koulango* in the Northeast, *Gnananmininan* by the *Malinké* in the Northwest, and *Troglâ* by the *Gouro* in the Centre-West. The cultivar *Six mois* is so named in the South, but *Egypt* in Adiaké, *Blannin* in Gohitafla, *Déhikachia* in Fakobly and *Sanka* in Abengourou. Also, the cultivar called *Blêbou* in the Center-West is named *Israel* in the South-East and *Agbabalama* in the North-East. On the other hand, the cultivar *Yacé* was given the same name in different regions. This study showed that the most widespread cultivars are *Yacé*, *Troglâ*, *Six mois* and *Blêbou*. However, the *Troglâ* cultivar, although widespread, is not widely cultivated because of its very bitter taste, probably due to a very high level of cyanide in the pulp. Indeed, this study shows that people in Côte d'Ivoire prefer sweet cassava cultivars.

Conclusion:-

This study focused on the morphological diversity of local cassava cultivars in Côte d'Ivoire. This description highlighted the morphological diversity that exists within the cassava cultivars held by farmers. This diversity is best explained by 10 of the 14 characters used. The local cassava cultivars are subdivided into five different morphological groups. Four local cassava cultivars were identified as the most widespread: *Yacé*, *Six mois*, *Blêbou* and *Troglâ*. In addition, the study showed that diversity is not geographically structured. There would therefore be a

significant flow of genes favored by farmers, which would prove their role in preserving and maintaining genetic diversity *in situ*. The sustainable management of the resources of this crop depends on the farmers.

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